Draft Assessment Report

Evaluation of Active Substances

Plant Protection Products

Prepared according to **Regulation (EC) 1107/2009**
as it applies in Great Britain

Bixlozone (F9600)

Volume 3 – B.8 (AS)

Environmental Fate & Behaviour

Great Britain

July 2022

Version History

When	What
July 2022	Initial DAR
September 2022	Updated post July 2022 Expert Committee on Pesticides (ECP) meeting Independent Scientific Advice (ISA)

Table of contents

CA.B.8. ENVIRONMENTAL FATE AND BEHAVIOUR

The fate and behaviour of the active substance bixlozone (development code: F9600) is presented in this report. In soil, there is one major aerobic metabolite, 2,4-dichlorobenzoic acid (2,4-DBA) observed in field dissipation studies, and an additional major anaerobic metabolite, bixlozone-3-OH-propanamide (3-OH). These two metabolites are also major metabolites in water/sediment. There are also two further major water/sediment metabolites, dimethyl malonamide and 4-carboxy-bixlozone; the metabolic pathway is summarised in Figure CA.B.**8-1**. Laboratory route of degradation studies were undertaken with both bixlozone rings radiolabelled (phenyl and carbonyl positions).

Some of the key bixlozone physical and chemical properties are summarised in Table CA.B.**8-1** (see Vol. 3 CA, B1,2-4 for further information). The representative formulation used in the Vol. 3 CP is bixlozone 4-SC. Soil dissipation studies were conducted using this formulation, as well as another formulation, F9600-21 CS. The representative uses of bixlozone are (to be applied by broadcast sprayer): maize (spring application), winter oilseed rape (autumn application) and winter cereals (autumn application).

Figure CA.B.8-1: Bixlozone metabolic pathways in soil and water

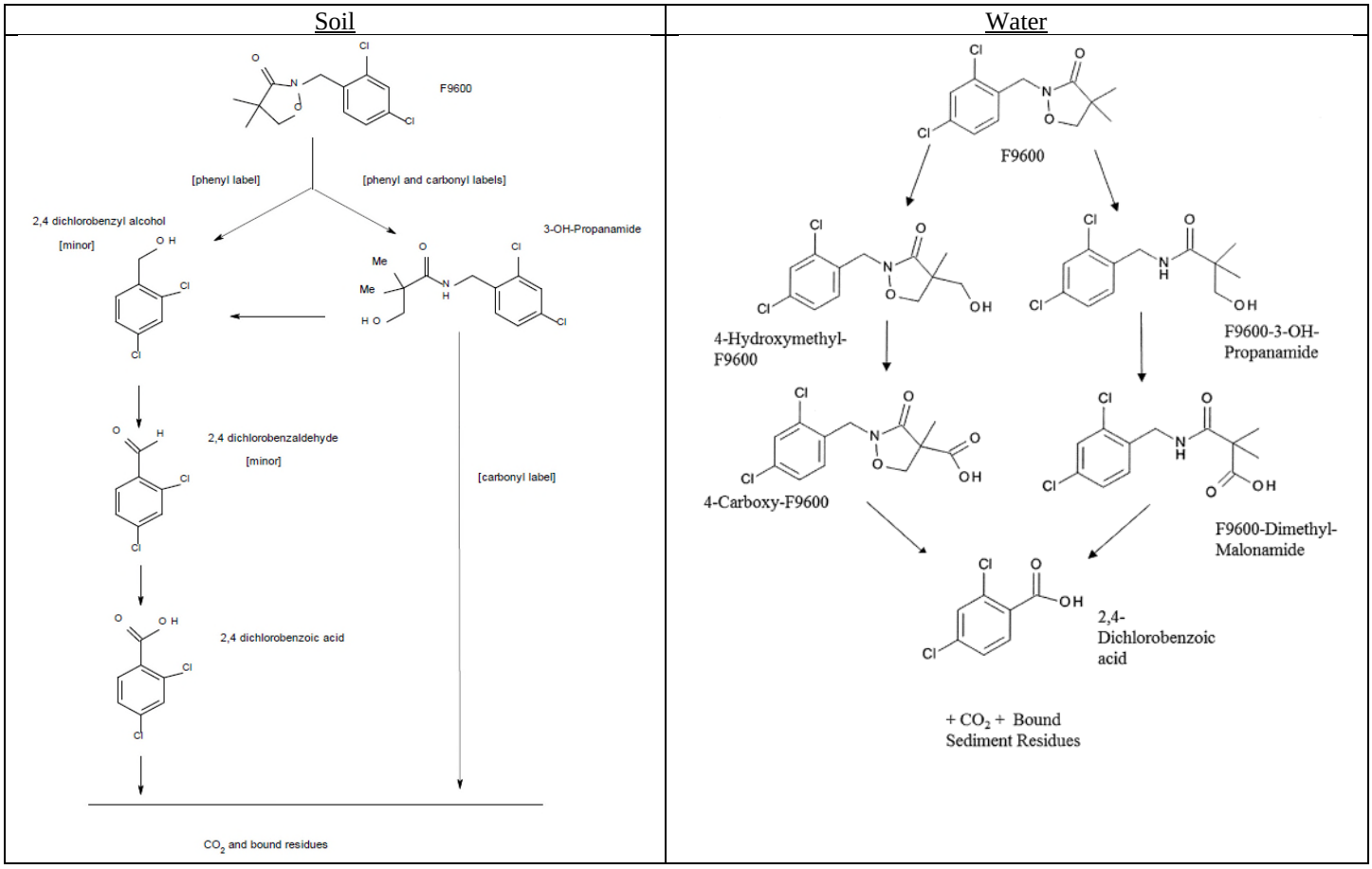


Table CA.B.8-1: Summary of key bixlozone physical and chemical properties

Molar mass (g/mol)	274.14	Vapour pressure (Pa)	20 °C	1.1×10^{-3}
Solubility in water, pH 7, 20 °C (mg/L)	40		25 °C	2.3×10^{-3}
Henry's Law constant, 20 °C (Pa m^3 mol)	7.2×10^{-3}	Log Pow, pH 7, 20 °C		3.3
Dissociation constant	Bixlozone does not contain any groups that are ionisable within an environmentally relevant pH range			

CA.B.8.1. FATE AND BEHAVIOUR IN SOIL

CA.B.8.1.1. Laboratory route and rate of degradation in soil

CA.B.8.1.1.1. *Aerobic route of degradation*

CA.B.8.1.1.1.1. Aerobic route of degradation of the active substance

Report:	KCA 7.1.1.1 Simmonds, R., (2015a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]-F9600: Route and Rate of Aerobic Degradation in Seven Soils at 20°C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/001; FMC Tracking no. 2013EFT-ISX1021
Guidelines:	OECD Guideline 307 (April 2002); US EPA OPPTS Guideline 835.4100 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

CA comments	The CA notes for one soil (CA-SL) pesticides had been used 3 and 4 years prior to sampling. However, as none of the pesticides used were structurally analogous to bixlozone, the CA does not consider this to have significantly impacted on the outcomes of the study. **This study is relied upon.**

INTRODUCTION

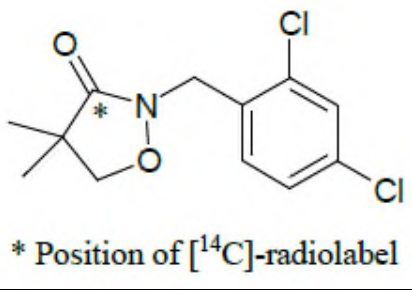
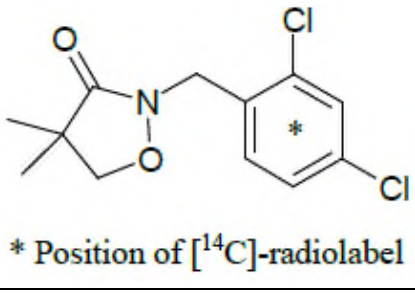
This study was conducted to investigate the route and rate of degradation of bixlozone (active substance development code: F9600) in four European and three US soils over 120 days. The study was conducted in accordance with OECD 307 guidelines.

MATERIALS

Test substances

The chemical properties of the test substances are summarised in Table CA.B.**8.1.1.1.1-1**.

Table CA.B.8.1.1.1.1-1: Summary of study chemical properties

Substance	Bixlozone	
Chemical name (IUPAC)	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-1,2-oxazolidin-3-one	
Formula	$C_{12}H_{13}Cl_2NO_2$	
Weight	274.14 g/mol	
CAS No	81777-95-9	
Test substance	[carbonyl-^{14}C]-bixlozone	[phenyl-U-^{14}C]-bixlozone
Structure	 * Position of [^{14}C]-radiolabel	 * Position of [^{14}C]-radiolabel

Specific activity	7.56 MBq/mg	7.56 MBq/mg
Radiochemical purity	98.45% (from HPLC)	98.57% (from HPLC)

Soil

Fresh topsoil was collected from four European and three US sites. These soils all broadly fall into the 'representative soil' defined in the OECD guideline (pH 5.5 – 8.0, organic carbon content of 0.5 – 2.5 %). Lufa 2.2 has a slightly lower pH than 5.5 (5.4) and CA-SL has an organic carbon slightly lower than 0.5 % (0.3 %), however, these minor deviations were considered acceptable by the CA. All soils were sieved to 2 mm and stored at 4°C in the dark for a maximum of 2 months prior to the initiation of the experiment. A summary of the physical and chemical properties of the soils is provided in Table CA.B.**8.1.1.1.1-2**. Microbial biomass was also measured at the initial and final time point of the study using the chloroform fumigation method, accepted as an alternative to the Substrate Induced Respiration (SIR) method in OECD 307 and confirmed that soils remained microbially viable throughout the study.

In line with the OECD 307 guidelines, no pesticides were used in at least the last 4 years for the Iowa, LAD-SCL-PF and European soils. For soil CA-SL, no pesticides had been used in the last 2 years, however, pesticides had been used in the years prior to that; details of the pesticides used are summarised in Table CA.B.**8.1.1.1.1-3**Table CA.B.. However, the CA does not consider the active substances to be analogues to the active substance and, therefore, do not contravene the OECD 307 guidelines.

Table CA.B.8.1.1.1.1-2: Soil Physiochemical Properties

Soil Characterisation	**Lufa 6S**	**Lufa 5M**	**Lufa 2.2**	**Refesol 02-A**	**CA-SL**	**Iowa**	**LAD-SCL-PF**
Sampling location	Siebel-dingen Germany	Mechter-sheim Germany	Hanhofen Germany	Schmal-lenberg Germany	Hughson USA	Jackson USA	Fremont USA
Particle size distribution							
Sand (%)	29	56	84	22	77	15	27
Silt (%)	26	27	9	61	18	62	26
Clay (%)	45	17	7	17	5	23	47
Textural classification (USDA)	Clay	Sandy loam	Loamy sand	Silt loam	Loamy sand	Silt loam	Clay
pH (0.01M $CaCl_2$)	6.9	7.2	5.4	6.1	6.9	6.8	8.0
pH (water)	7.1	7.5	5.7	6.3	7.4	7.2	8.1
% Organic matter	3.6	2.2	2.6	2.1	0.6	3.6	1.8
% Organic carbon†	2.1	1.3	1.5	1.2	0.3	2.1	1.0
CEC (meq/100g)	21.0	9.7	7.3	11.2	5.5	13.6	31.1
Bulk density, disturbed (g/cm^3)	1.20	1.13	1.23	1.10	1.33	1.02	1.08
% Moisture at pF2.0	31.0	20.8	11.3	37.0	13.4	42.3	40.9
% Moisture at pF2.5	26.1	13.1	8.4	18.3	7.2	30.0	29.7
% Moisture maintained in experiment	26.5	18.0	11.5	27.8	11.5	31.8	29.7
Soil biomass (µg C/g soil)							
Initial	344.8	213.9	326.8	156.5	56.4	365.9	349.3
Final	293.5	204.3	323.1	113.7	57.1	252.3	289.0

† % Organic carbon = % Organic matter / 1.724

Table CA.B.8.1.1.1.1-3: Pesticide application history for soil CA-SL

Date of application	Product	Active ingredient	CAS number

Date of application	Product	Active ingredient	CAS number
2013	none	-	-
2012	Ridomil Gold 1.5 pt/A	Mancozeb Metalaxyl-M	8018-01-7 70630-17-0
2011	Prowl 3.3 EC 2pt/A	Pendimethalin	40487-42-1
2010	Ridomil Gold EC 1pt/A	Mefenoxam	70630-17-0
2010	Trifluralin 4 EC 1.25 pt/A	Trifluralin	1582-09-8

STUDY DESIGN

Experimental conditions

Collected soil samples were stored for a maximum of 2 months prior to use. Soil samples (100 g oven-dry weight equivalent) of each of the seven soils were weighed into individual incubation vessels and were adjusted to soil moisture contents between pF2.0 and pF2.5, if necessary. The soils were allowed to acclimatise under study conditions prior to the addition of test substance.

[phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone were prepared by evaporating an aliquot of the supplied test item to dryness under a stream of nitrogen and re-dissolving in 20 mL acetonitrile. The final concentration of the [phenyl-U-^{14}C]-bixlozone solution was 1.01 mg/mL and the [carbonyl-^{14}C]-bixlozone was 1.07 mg/mL, determined by Liquid Scintillation Counting (LSC).

The two radiolabelled forms of the test substance, [phenyl-U-^{14}C]- and [carbonyl-^{14}C]-bixlozone were applied at a nominal rate of 1.06 mg/kg soil (equivalent to 400 g a.s./ha) to duplicate vessels for each of the six time points for each soil. Treatment solutions were added dropwise to the soil surface ensuring an even distribution. For each soil and radiolabel there were eight surplus treated vessels. The total treated vessels per radiolabel per soil totalled twenty. For each soil, additional untreated vessels were prepared for microbial biomass determination at the beginning and end of the study. All vessels were maintained in the dark at 20 ± 2°C for 120 days, with aerobic conditions maintained by the constant passage of moist air through the sample flasks and out through the trap solutions. Volatile organic compounds and CO_2 were trapped in ethylene glycol and 2M KOH traps.

Sampling

Duplicates of each soil were removed for analysis immediately after test substance application. Duplicate incubation vessels and their associated traps were removed for analysis at intervals of 7, 14, 30, 75/76, and 120 days after treatment (DAT).

The initial extraction of all soil samples was conducted on the day that they were collected. Samples and extracts were stored refrigerated (*ca* 5°C) between extraction steps. Extracts generated during the study were generally profiled chromatographically within 10 days of generation. Soil residues were stored refrigerated (*ca* 5°C), while extracts were stored frozen (*ca* -20°C). Initial HPLC profiles of the extracts were obtained with 10 days of the sample generation and therefore the applicant did not consider that stability on storage was a significant factor in the study. Consequently no storage stability study was conducted.

For samples taken at 0 days after treatment (DAT), soil samples were extracted four times, except the LAD-SCL-PF soil which was extracted three times, at ambient temperature with acetonitrile/water (80:20 v/v). LAD-SCL-PF soil was additionally extracted with acetonitrile/water/formic acid (50:50:1 v/v/v) followed by one final soxhlet extraction at elevated temperature with acetonitrile/water (80:20 v/v). All soil samples from day 7 onwards were extracted four times at ambient temperature – one extraction with acetonitrile, two successive extractions with acetonitrile/water (80:20 v/v), and one extraction with acetonitrile/water/formic acid (50:50:1 v/v/v). All soils were then subjected to one final soxhlet extraction (6 hours) at elevated temperature with acetonitrile/water (80:20 v/v); the extracts from each extraction were pooled together for analysis.

Methods of analysis

The radioactivity in the combined ambient extracts and soxhlet extracts from each soil sample were determined directly by liquid scintillation counting (LSC). Extracts were further characterised and quantified by gradient elution reverse phase high performance liquid chromatography (HPLC) with UV detector (LOQ = 0.06% of applied radioactivity), using a Zorbax RX-C18 column, 0.01 % acetic acid:water (v:v) and 0.01 % acetic acid:acetonitrile (v:v) solvents.

Identification was performed by LC-MS using a Kromasil C18 column, and a gradient elution of 100:0.01 % water:acetic acid (v:v) and 100:0.01 % acetonitrile:acetic acid solvents and a micromass Quattro-LC mass spectrometer monitoring an ion transition of 274-276 m/z. Substances analysed from the samples were compared against reference standards.

Samples which had >10% of applied radioactivity associated with the soil non-extractable residues were subjected to two further solvent extraction steps repeated three times each using a medium (tetrahydrofuran) and low polarity solvent (hexane).

After extraction, non-extractable soil residues were determined in air-dried soil by combustion and direct analysis via LSC (LOQ < 0.001% of applied radioactivity). Fractionation of non-extractable residues was performed on selected 75/76 DAT samples. The CA notes that for some soils and radiolabels, there were more non-extracted residues in the 120 DAT samples, however, use of the 75/76 DAT time points is acceptable for giving a representative analysis. 0.5M NaOH was added to samples and shaken for 24 hours at room temperature, after which extracts were centrifuged at 2000 rpm for 10 minutes. An aliquot of the aqueous extract and washings of the soil with 0.5 M NaOH and distilled water was taken for analysis via LSC. The remaining aqueous extract was then acidified with HCl and centrifuged at room temperature at 2000 rpm for 10 minutes. An aliquot of the supernatant was taken for analysis via LSC, and the precipitate was redissolved in 0.5M NaOH and analysed by LSC.

The radioactivity present in the volatile traps was quantified via LSC. Radioactivity in the 2M KOH trap solutions was confirmed as $^{14}CO_2$ by barium chloride precipitation.

Microbial biomass was measured using a chloroform fumigation method. Four of the seven initial samples and all seven final samples were subject to delays prior to analysis due to additional time needed for drying and sieving samples. These delays were 2 days for the four initial samples and 4-5 weeks for the final samples, however, no significant decline in the biomass was observed between initial and final samples. Therefore, these delays are not expected to have affected the outcome of the test.

RESULTS AND DISCUSSION

The total mass balance, distribution of radioactive residues, and the characterisation of the extractable residues are presented in Table CA.B.**8.1.1.1.1-4** to Table CA.B.**8.1.1.1.1-10** for the seven soils treated with [phenyl-U-^{14}C]-labelled bixlozone and in Table CA.B.**8.1.1.1.1-11** to Table CA.B.**8.1.1.1.1-17** for the seven soils treated with [carbonyl-^{14}C]-labelled bixlozone.

Table CA.B.8.1.1.1.1-4: Percent recovery of applied radioactivity in Lufa 6S soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
1	0	96.59	95.73	<LOQ	<LOQ	<LOQ	0.86	1.90	NA	98.49
2	0	96.61	96.61	<LOQ	<LOQ	<LOQ	0.00	2.08	NA	98.69
Mean		**96.60**	**96.17**	**<LOQ**	**<LOQ**	**<LOQ**	**0.43**	**1.99**	**NA**	**98.59**
4	7	96.05	94.88	0.96	<LOQ	0.12	0.08	0.90	0.54	97.50
6	7	95.33	94.11	1.07	<LOQ	0.15	0.00	1.32	0.07	96.72
Mean		**95.69**	**94.49**	**1.02**	**<LOQ**	**0.14**	**0.04**	**1.11**	**0.31**	**97.11**
10	14	91.40	87.87	2.42	0.85	0.26	0.00	2.73	1.97	96.09
11	14	92.74	88.19	2.53	1.11	0.26	0.66	2.34	1.82	96.91
Mean		**92.07**	**88.03**	**2.47**	**0.98**	**0.26**	**0.33**	**2.53**	**1.90**	**96.50**
8	30	85.59	80.00	3.43	1.78	0.39	0.00	5.63	4.18	95.41
9	30	83.99	81.75	1.74	<LOQ	0.25	0.24	5.86	5.25	95.09
Mean		**84.79**	**80.88**	**2.59**	**0.89**	**0.32**	**0.12**	**5.74**	**4.72**	**95.25**
13	75	67.51	65.10	0.45	1.03	0.50	0.44	9.90	17.46	94.87
14	75	67.60	64.87	0.51	1.02	0.64	0.57	9.90	14.81	92.32
Mean		**67.56**	**64.98**	**0.48**	**1.02**	**0.57**	**0.51**	**9.90**	**16.14**	**93.60**
15	120	54.23	50.37	0.15	1.73	0.59	1.38	13.11	22.79	90.12
17	120	57.33	52.95	0.54	1.99	0.48	1.36	12.93	20.87	91.14
Mean		**55.78**	**51.66**	**0.35**	**1.86**	**0.54**	**1.37**	**13.02**	**21.83**	**90.63**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1] Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2] 2,4-dichlorobenzoic acid

[3] 2,4-dichlorobenzyl alcohol

[4] 2,4-dichlorobenzaldehyde

[5] Non-extractable radioactivity from soil

[6] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-5: Percent recovery of applied radioactivity in Lufa 5M soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
41	0	97.90	97.90	<LOQ	<LOQ	<LOQ	<LOQ	0.25	NA	98.15
42	0	97.62	97.62	<LOQ	<LOQ	<LOQ	<LOQ	0.25	NA	97.87
Mean		**97.76**	**97.76**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.25**	**NA**	**98.01**
44	7	95.20	94.15	1.06	<LOQ	<LOQ	<LOQ	1.63	0.59	97.42
45	7	95.47	94.78	0.69	<LOQ	<LOQ	<LOQ	2.02	0.55	98.04
Mean		**95.33**	**94.46**	**0.87**	**<LOQ**	**<LOQ**	**<LOQ**	**1.53**	**0.57**	**97.73**
46	14	92.98	88.85	3.26	0.87	<LOQ	<LOQ	3.05	1.71	97.74
47	14	92.35	88.37	3.10	0.64	0.25	<LOQ	2.92	1.57	96.85
Mean		**92.67**	**88.61**	**3.18**	**0.75**	**0.12**	**<LOQ**	**2.99**	**1.64**	**97.30**
49	30	85.67	79.15	4.81	1.70	<LOQ	<LOQ	5.56	4.60	95.82
51	30	84.49	78.00	4.91	1.58	<LOQ	<LOQ	5.68	4.96	95.13
Mean		**85.08**	**78.58**	**4.86**	**1.64**	**<LOQ**	**<LOQ**	**5.62**	**4.78**	**95.48**
53	75	71.39	65.47	2.34	1.45	1.00	1.14	11.24	11.55	94.18
54	75	72.14	64.77	4.42	1.70	0.54	0.69	9.60	10.12	91.86
Mean		**71.76**	**65.12**	**3.38**	**1.57**	**0.77**	**0.92**	**10.42**	**10.83**	**93.02**
48	120	46.90	42.12	0.00	2.22	0.63	1.93	14.46	29.95	91.31
59	120	49.56	45.29	0.00	1.75	0.41	2.11	15.28	27.02	91.85
Mean		**48.23**	**43.71**	**0.00**	**1.99**	**0.52**	**2.02**	**14.87**	**28.48**	**91.58**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2]2,4-dichlorobenzoic acid

[3]2,4-dichlorobenzyl alcohol

[4]2,4-dichlorobenzaldehyde

[5] Non-extractable radioactivity from soil

[6] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-6: Percent recovery of applied radioactivity in Lufa 2.2 soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
81	0	98.19	98.19	<LOQ	<LOQ	<LOQ	<LOQ	0.33	NA	98.52
82	0	98.35	98.35	<LOQ	<LOQ	<LOQ	<LOQ	0.32	NA	98.67
Mean		**98.27**	**98.27**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.32**	**NA**	**98.59**
83	7	95.96	95.96	<LOQ	<LOQ	<LOQ	<LOQ	1.19	1.29	98.44
84	7	96.42	96.42	<LOQ	<LOQ	<LOQ	<LOQ	0.99	1.13	98.54
Mean		**96.19**	**96.19**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**1.09**	**1.21**	**98.49**
85	14	91.89	90.84	<LOQ	1.04	<LOQ	<LOQ	2.05	3.03	96.97
86	14	94.21	93.68	<LOQ	0.53	<LOQ	<LOQ	1.64	1.41	97.25
Mean		**93.05**	**92.26**	**<LOQ**	**0.79**	**<LOQ**	**<LOQ**	**1.85**	**2.22**	**97.11**
87	30	89.57	84.19	<LOQ	3.43	0.19	1.76	2.49	4.06	96.12
89	30	89.67	87.10	<LOQ	2.25	0.31	<LOQ	2.64	2.23	94.54
Mean		**89.62**	**85.65**	**<LOQ**	**2.84**	**0.25**	**0.88**	**2.56**	**3.14**	**95.33**
90	75	84.52	81.2	<LOQ	2.48	0.47	0.38	3.63	6.72	94.87
91	75	84.47	81.83	<LOQ	2.00	0.29	0.35	3.30	7.48	95.25
Mean		**84.49**	**81.51**	**<LOQ**	**2.24**	**0.38**	**0.36**	**3.47**	**7.10**	**95.06**
93	120	79.01	74.25	<LOQ	2.41	0.33	2.02	3.26	10.79	93.06
99	120	81.02	76.50	<LOQ	2.77	0.33	1.41	4.29	10.02	95.32
Mean		**80.01**	**75.38**	**<LOQ**	**2.59**	**0.33**	**1.72**	**3.78**	**10.40**	**94.19**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2]2,4-dichlorobenzoic acid

[3]2,4-dichlorobenzyl alcohol

[4]2,4-dichlorobenzaldehyde

[5] Non-extractable radioactivity from soil

[6] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-7: Percent recovery of applied radioactivity in RefeSol 02-A soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
121	0	97.43	97.43	<LOQ	<LOQ	<LOQ	<LOQ	0.35	NA	97.78
122	0	97.77	97.77	<LOQ	<LOQ	<LOQ	<LOQ	0.38	NA	98.15
Mean		**97.60**	**97.60**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.37**	**NA**	**97.97**
123	7	93.65	92.08	1.57	<LOQ	<LOQ	<LOQ	2.28	1.34	97.27
124	7	94.62	92.43	2.18	<LOQ	<LOQ	<LOQ	2.28	1.04	97.93
Mean		**94.13**	**92.26**	**1.88**	**<LOQ**	**<LOQ**	**<LOQ**	**2.28**	**1.19**	**97.60**
125	14	88.69	87.98	-	0.71	<LOQ	<LOQ	4.67	2.99	96.35
126	14	89.14	86.55	0.91	1.69	<LOQ	<LOQ	3.89	2.23	95.27
Mean		**88.92**	**87.27**	**0.45**	**1.20**	**<LOQ**	**<LOQ**	**4.28**	**2.61**	**95.81**
128	30	83.60	77.05	<LOQ	2.73	0.56	3.27	5.73	2.74	92.06
132	30	84.38	80.77	0.18	2.54	<LOQ	0.88	5.69	5.42	95.49
Mean		**83.99**	**78.91**	**0.09**	**2.63**	**0.28**	**2.08**	**5.71**	**4.08**	**93.78**
134	75	73.24	69.75	<LOQ	2.42	0.90	0.17	7.66	12.32	93.22
135	75	75.07	72.60	<LOQ	2.15	0.32	<LOQ	7.35	10.01	92.43
Mean		**74.16**	**71.18**	**<LOQ**	**2.29**	**0.61**	**0.08**	**7.50**	**11.16**	**92.82**
130	120	72.62	69.56	<LOQ	2.51	0.41	0.13	7.92	12.96	93.50
133	120	69.21	65.54	<LOQ	2.22	0.90	0.54	8.62	14.92	92.74
Mean		**70.92**	**67.55**	**<LOQ**	**2.37**	**0.66**	**0.34**	**8.27**	**13.94**	**93.12**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2]2,4-dichlorobenzoic acid

[3]2,4-dichlorobenzyl alcohol

[4]2,4-dichlorobenzaldehyde

[5] Non-extractable radioactivity from soil

[6] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-8: Percent recovery of applied radioactivity in CA-SL soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
161	0	98.07	97.40	<LOQ	<LOQ	<LOQ	0.68	0.11	NA	98.18
162	0	97.67	97.67	<LOQ	<LOQ	<LOQ	<LOQ	0.09	NA	97.76
Mean		**97.87**	**97.53**	**<LOQ**	**<LOQ**	**<LOQ**	**0.34**	**0.10**	**NA**	**97.97**
163	7	97.24	95.06	2.18	<LOQ	<LOQ	<LOQ	0.71	0.39	98.34
164	7	96.90	95.01	1.89	<LOQ	<LOQ	<LOQ	0.83	0.31	98.04
Mean		**97.07**	**95.04**	**2.03**	**<LOQ**	**<LOQ**	**<LOQ**	**0.77**	**0.35**	**98.19**
165	14	95.10	91.83	3.27	<LOQ	<LOQ	<LOQ	1.82	1.15	98.07
166	14	93.42	87.13	6.29	<LOQ	<LOQ	<LOQ	1.92	1.26	96.61
Mean		**94.26**	**89.48**	**4.78**	**<LOQ**	**<LOQ**	**<LOQ**	**1.87**	**1.21**	**97.34**
167	30	89.15	84.37	4.64	0.08	<LOQ	0.06	4.01	3.21	96.37
168	30	90.48	85.86	4.62	<LOQ	<LOQ	<LOQ	3.71	3.17	97.37
Mean		**89.81**	**85.12**	**4.63**	**0.04**	**<LOQ**	**0.03**	**3.86**	**3.19**	**96.87**
169	75	77.06	70.91	4.92	1.08	<LOQ	0.15	6.91	9.48	93.46
170	75	72.31	67.94	3.06	1.21	<LOQ	0.10	7.86	11.18	91.35
Mean		**74.69**	**69.43**	**3.99**	**1.15**	**<LOQ**	**0.12**	**7.38**	**10.33**	**92.40**
174	120	60.35	58.62	<LOQ	1.11	<LOQ	0.62	17.40	22.38	100.14
176	120	56.37	54.74	<LOQ	0.73	0.34	0.55	17.10	23.62	97.08
Mean		**58.36**	**56.68**	**<LOQ**	**0.92**	**0.17**	**0.58**	**17.25**	**23.00**	**98.61**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]2,4-dichlorobenzoic acid
[3]2,4-dichlorobenzyl alcohol
[4]2,4-dichlorobenzaldehyde
[5] Non-extractable radioactivity from soil
[6]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-9: Percent recovery of applied radioactivity in Iowa soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns**	**NER[5]**	**CO_2[6]**	**Mass Balance**
201	0	96.21	96.21	<LOQ	<LOQ	<LOQ	<LOQ	1.89	NA	98.09
202	0	96.05	96.05	<LOQ	<LOQ	<LOQ	<LOQ	1.68	NA	97.73
Mean		**96.13**	**96.13**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**1.78**	**NA**	**97.91**
203	7	93.22	91.06	2.00	<LOQ	0.15	<LOQ	2.96	1.02	97.20
204	7	93.48	92.03	1.45	<LOQ	<LOQ	<LOQ	2.85	2.22	98.55
Mean		**93.35**	**91.55**	**1.73**	**<LOQ**	**0.08**	**<LOQ**	**2.90**	**1.62**	**97.87**
206	14	87.04	81.46	2.33	1.85	0.44	0.95	5.07	4.96	97.07
208	14	86.65	80.14	3.74	2.76	<LOQ	<LOQ	5.46	5.10	97.21
Mean		**86.84**	**80.80**	**3.04**	**2.31**	**0.22**	**0.47**	**5.27**	**5.03**	**97.14**
211	30	75.68	73.02	0.64	1.74	<LOQ	0.27	7.87	9.28	92.82
212	30	73.31	69.61	1.68	1.71	0.30	<LOQ	9.09	11.73	94.13
Mean		**74.49**	**71.32**	**1.16**	**1.73**	**0.15**	**0.13**	**8.48**	**10.5**	**93.48**
213	75	46.37	42.98	<LOQ	0.90	0.53	1.97	14.30	36.39	97.06
214	75	44.25	41.58	0.27	0.76	0.33	1.31	15.27	38.70	98.22
Mean		**45.31**	**42.28**	**0.13**	**0.83**	**0.43**	**1.64**	**14.78**	**37.55**	**97.64**
215	120	26.43	24.23	<LOQ	<LOQ	0.31	1.89	18.13	47.17	91.72
214	120	27.79	24.86	<LOQ	0.51	0.57	1.85	18.22	47.65	93.66
Mean		**27.11**	**24.54**	**<LOQ**	**0.26**	**0.44**	**1.87**	**18.18**	**47.41**	**92.69**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2]2,4-dichlorobenzoic acid

[3]2,4-dichlorobenzyl alcohol

[4]2,4-dichlorobenzaldehyde

[5] Non-extractable radioactivity from soil

[6] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-10: Percent recovery of applied radioactivity in LAD-SCL-PF soil following application of [phenyl-U-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity								
		Total extracted[1]	**Bixlozone**	**RRT: 0.45[2]**	**RRT: 0.81[3]**	**RRT: 0.95[4]**	**Total unknowns[5]**	**NER[6]**	**CO_2[7]**	**Mass Balance**
241	0	97.56	97.56	<LOQ	<LOQ	<LOQ	<LOQ	0.01	NA	97.57
242	0	97.46	97.46	<LOQ	<LOQ	<LOQ	<LOQ	0.02	NA	97.48
Mean		**97.51**	**97.51**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.02**	**NA**	**97.52**
243	7	98.77	98.77	<LOQ	<LOQ	<LOQ	<LOQ	0.57	0.14	99.48
245	7	97.67	97.55	<LOQ	<LOQ	0.12	<LOQ	0.31	0.14	98.11
Mean		**98.22**	**98.16**	**<LOQ**	**<LOQ**	**0.06**	**<LOQ**	**0.44**	**0.14**	**98.80**
246	14	97.21	93.61	3.61	<LOQ	<LOQ	<LOQ	1.09	0.26	98.57
247	14	97.09	95.98	1.12	<LOQ	<LOQ	<LOQ	1.31	0.23	98.64
Mean		**97.15**	**94.79**	**2.36**	**<LOQ**	**<LOQ**	**<LOQ**	**1.20**	**0.25**	**98.60**
248	30	95.50	83.83	2.35	<LOQ	0.32	9.01	1.11	1.00	97.62
249	30	94.30	84.66	2.15	<LOQ	0.18	7.29	1.26	0.39	95.95
Mean		**94.90**	**84.25**	**2.25**	**<LOQ**	**0.25**	**8.15**	**1.19**	**0.70**	**96.78**
251	75	85.06	75.49	2.51	<LOQ	0.17	6.90	4.41	4.20	93.68
253	75	84.11	75.53	2.47	<LOQ	0.14	5.97	4.72	3.25	92.08
Mean		**84.59**	**75.51**	**2.49**	**<LOQ**	**0.15**	**6.44**	**4.56**	**3.72**	**92.88**
256	120	71.41	59.62	1.91	<LOQ	0.09	9.78	7.75	11.49	90.65
259	120	70.50	60.59	0.68	<LOQ	0.25	8.97	6.56	13.47	90.52
Mean		**70.95**	**60.11**	**1.29**	**<LOQ**	**0.17**	**9.38**	**7.15**	**12.48**	**90.59**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 0 onwards) analysis combined
[2]2,4-dichlorobenzoic acid
[3]2,4-dichlorobenzyl alcohol
[4]2,4-dichlorobenzaldehyde
[5]No individual >3.6% of applied radioactivity at any one time point
[6]Non-extractable radioactivity from soil
[7] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-11: Percent recovery of applied radioactivity in Lufa 6S soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
21	0	97.62	97.62	<LOQ	<LOQ	1.09	NA	98.71
22	0	96.60	96.60	<LOQ	<LOQ	0.69	NA	97.29
Mean		**97.11**	**97.11**	**<LOQ**	**<LOQ**	**0.89**	**NA**	**98.00**
23	7	93.19	92.81	0.16	0.21	1.71	1.90	96.81
24	7	93.34	93.04	0.15	0.15	1.34	1.39	96.07
Mean		**93.27**	**92.93**	**0.16**	**0.18**	**1.53**	**1.65**	**96.44**
25	14	88.11	86.29	0.99	0.82	2.65	3.10	93.86
27	14	89.36	88.09	0.73	0.54	2.24	4.04	95.65
Mean		**88.74**	**87.19**	**0.86**	**0.68**	**2.45**	**3.57**	**94.76**
28	30	79.86	78.06	1.44	0.36	3.46	13.53	96.85
29	30	81.93	81.31	<LOQ	0.62	3.53	9.19	94.65
Mean		**80.89**	**79.68**	**0.72**	**0.49**	**3.50**	**11.36**	**95.75**
30	76	65.52	63.15	1.11	1.27	7.16	21.08	93.76
31	76	67.05	64.13	1.55	1.36	6.76	21.08	94.89
Mean		**66.28**	**63.64**	**1.33**	**1.32**	**6.96**	**21.08**	**94.33**
32	120	54.44	52.53	0.80	1.10	9.09	27.73	91.25
33	120	51.47	49.68	0.69	1.10	10.18	30.11	91.76
Mean		**52.95**	**51.10**	**0.75**	**1.10**	**9.63**	**28.92**	**91.51**

NA = not analysed, LOQ = limit of quantification (0.06% AR)

[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined

[2] Non-extractable radioactivity from soil

[3] Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-12: Percent recovery of applied radioactivity in Lufa 5M soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns[2]**	**NER[3]**	**CO_2[4]**	**Mass Balance**
61	0	100.16	100.16	<LOQ	<LOQ	0.31	NA	100.47
62	0	98.26	98.26	<LOQ	<LOQ	0.29	NA	98.55
Mean		**99.21**	**99.21**	**<LOQ**	**<LOQ**	**0.30**	**NA**	**99.51**
63	7	93.01	92.84	<LOQ	0.17	1.94	2.48	97.42
64	7	92.90	92.90	<LOQ	<LOQ	2.2	2.01	97.10
Mean		**92.95**	**92.87**	**<LOQ**	**0.08**	**2.07**	**2.25**	**97.26**
67	14	87.76	86.89	0.87	<LOQ	3.48	4.59	95.84
68	14	89.28	88.39	0.89	<LOQ	3.01	3.31	95.60
Mean		**88.52**	**87.64**	**0.88**	**<LOQ**	**3.25**	**3.95**	**95.72**
69	30	81.83	80.13	1.26	0.44	4.86	9.85	96.54
70	30	83.14	81.33	1.31	0.50	4.25	7.24	94.63
Mean		**82.49**	**80.73**	**1.29**	**0.47**	**4.56**	**8.54**	**95.59**
73	76	63.73	60.53	1.69	1.51	8.16	21.33	93.21
74	76	65.05	60.89	2.63	1.53	7.98	20.48	93.51
Mean		**64.39**	**60.71**	**2.16**	**1.52**	**8.05**	**20.90**	**93.36**
77	120	53.91	50.45	1.28	2.17	9.62	28.37	91.89
78	120	50.85	48.66	0.72	1.47	10.77	29.10	90.72
Mean		**52.38**	**49.55**	**1.00**	**1.82**	**10.19**	**28.74**	**91.31**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]No individual >1.8% of applied radioactivity at any one time point
[3]Non-extractable radioactivity from soil
[4]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-13: Percent recovery of applied radioactivity in Lufa 2.2 soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
101	0	98.47	98.47	<LOQ	<LOQ	0.36	NA	98.82
102	0	97.84	97.84	<LOQ	<LOQ	0.39	NA	98.23
Mean		**98.15**	**98.15**	**<LOQ**	**<LOQ**	**0.37**	**NA**	**98.53**
103	7	95.90	95.90	<LOQ	<LOQ	1.10	1.30	98.30
104	7	95.75	95.58	0.10	0.06	1.03	0.26	97.04
Mean		**95.82**	**95.74**	**0.05**	**0.03**	**1.06**	**0.78**	**97.67**
105	14	94.26	94.26	<LOQ	<LOQ	1.41	2.52	98.19
106	14	93.65	92.83	0.36	0.47	1.64	2.36	97.64
Mean		**93.95**	**93.55**	**0.18**	**0.23**	**1.52**	**2.44**	**97.92**
107	30	91.88	90.61	1.17	0.11	1.89	2.41	96.18
108	30	90.58	89.69	0.64	0.25	1.94	4.75	97.28
Mean		**91.23**	**90.15**	**0.90**	**0.18**	**1.92**	**3.58**	**96.73**
109	76	85.23	83.21	1.80	0.22	2.81	9.03	97.07
110	76	83.84	80.33	2.69	0.82	2.87	9.60	96.31
Mean		**84.53**	**81.77**	**2.25**	**0.52**	**2.84**	**9.32**	**96.69**
118	120	79.89	75.83	2.37	1.68	3.38	11.62	94.89
119	120	79.86	75.82	2.39	1.65	3.22	11.66	94.74
Mean		**79.87**	**75.83**	**2.38**	**1.67**	**3.30**	**11.64**	**94.81**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]Non-extractable radioactivity from soil
[3]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-14: Percent recovery of applied radioactivity in RefeSol 02-A soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
141	0	97.37	97.37	<LOQ	<LOQ	0.34	NA	97.71
142	0	98.77	98.77	<LOQ	<LOQ	0.40	NA	99.17
Mean		**98.07**	**98.07**	**<LOQ**	**<LOQ**	**0.37**	**NA**	**98.44**
143	7	91.99	91.03	0.70	0.26	2.91	0.03	94.93
145	7	91.60	90.79	0.82	<LOQ	3.17	2.74	97.51
Mean		**91.80**	**90.91**	**0.76**	**0.13**	**3.04**	**1.38**	**96.22**
146	14	88.40	87.69	0.71	<LOQ	3.87	4.09	96.36
148	14	86.82	85.88	0.54	0.40	5.46	5.14	97.42
Mean		**87.61**	**86.78**	**0.63**	**0.20**	**4.67**	**4.61**	**96.89**
147	30	84.33	82.48	1.33	0.53	4.45	9.81	98.59
149	30	84.26	82.26	1.48	0.52	3.98	8.51	96.75
Mean		**84.30**	**82.37**	**1.40**	**0.52**	**4.22**	**9.16**	**97.67**
151	76	74.49	71.25	2.92	0.32	7.27	14.97	96.73
152	76	76.15	73.34	1.95	0.86	6.17	14.45	96.77
Mean		**75.32**	**72.29**	**2.43**	**0.59**	**6.72**	**14.71**	**96.75**
157	120	70.44	66.72	2.72	1.00	6.60	17.40	94.44
158	120	70.29	65.92	2.66	1.72	7.24	17.07	94.61
Mean		**70.37**	**66.32**	**2.69**	**1.36**	**6.92**	**17.23**	**94.52**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]Non-extractable radioactivity from soil
[3]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-15: Percent recovery of applied radioactivity in CA-SL soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
181	0	98.38	98.38	<LOQ	<LOQ	0.12	NA	98.50
182	0	98.72	98.72	<LOQ	<LOQ	0.13	NA	98.85
Mean		**98.55**	**98.55**	**<LOQ**	**<LOQ**	**0.13**	**NA**	**98.68**
183	7	94.59	94.47	<LOQ	0.12	1.26	2.06	97.91
186	7	95.31	95.15	<LOQ	0.16	1.01	1.28	97.60
Mean		**94.95**	**94.81**	**<LOQ**	**0.14**	**1.13**	**1.67**	**97.75**
184	14	93.95	93.95	<LOQ	<LOQ	1.46	2.18	97.59
188	14	91.45	91.45	<LOQ	<LOQ	2.09	3.68	97.22
Mean		**92.70**	**92.70**	**<LOQ**	**<LOQ**	**1.77**	**2.93**	**97.40**
187	30	93.47	91.68	1.69	0.11	1.20	1.98	96.66
189	30	86.57	86.57	<LOQ	<LOQ	1.29	6.61	94.47
Mean		**90.02**	**89.13**	**0.84**	**0.05**	**1.25**	**4.30**	**95.56**
194	76	71.31	70.23	<LOQ	1.07	5.15	18.14	94.61
196	76	72.01	70.71	<LOQ	1.30	5.19	17.80	95.01
Mean		**71.66**	**70.47**	**<LOQ**	**1.19**	**5.17**	**17.97**	**94.81**
198	120	56.56	54.82	1.10	0.63	5.90	29.21	91.66
199	120	57.76	56.32	1.04	0.40	6.96	27.8	92.52
Mean		**57.16**	**55.57**	**1.07**	**0.52**	**6.43**	**28.50**	**92.09**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]Non-extractable radioactivity from soil
[3]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-16: Percent recovery of applied radioactivity in Iowa soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
221	0	97.47	97.47	<LOQ	<LOQ	0.68	NA	98.15
222	0	95.89	95.89	<LOQ	<LOQ	2.09	NA	97.98
Mean		**96.68**	**96.68**	**<LOQ**	**<LOQ**	**1.38**	**NA**	**98.07**
223	7	89.51	88.93	0.36	0.21	3.68	2.67	95.91
224	7	89.37	88.04	1.08	0.25	3.58	1.51	94.47
Mean		**89.44**	**88.49**	**0.72**	**0.23**	**3.63**	**2.09**	**95.17**
225	14	85.29	83.50	1.31	0.49	3.94	8.07	97.30
226	14	84.78	82.66	0.80	1.34	4.18	6.43	95.39
Mean		**85.04**	**83.08**	**1.06**	**0.92**	**4.06**	**7.25**	**96.34**
227	30	71.77	69.50	1.81	0.46	5.08	16.66	93.51
229	30	70.74	68.01	1.35	1.39	6.35	22.76	99.86
Mean		**71.25**	**68.75**	**1.58**	**0.93**	**5.72**	**19.71**	**96.68**
230	76	48.14	45.72	0.84	1.57	9.92	34.67	92.73
231	76	46.32	43.99	1.08	1.25	10.82	39.60	96.74
Mean		**47.23**	**44.86**	**0.96**	**1.41**	**10.37**	**37.14**	**94.74**
236	120	27.39	25.87	<LOQ	1.52	11.47	53.93	92.79
240	120	28.72	27.30	<LOQ	1.42	11.80	54.79	95.32
Mean		**28.06**	**26.59**	**<LOQ**	**1.47**	**11.64**	**54.36**	**94.05**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 7 onwards) analysis combined
[2]Non-extractable radioactivity from soil
[3]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

Table CA.B.8.1.1.1.1-17: Percent recovery of applied radioactivity in LAD-SCL-PF soil following application of [carbonyl-^{14}C]-bixlozone and their metabolites under aerobic conditions

Unit	Time (day)	% Applied radioactivity						
		Total extracted[1]	**Bixlozone**	**Unknown RRT: 0.81**	**Other unknowns**	**NER[2]**	**CO_2[3]**	**Mass Balance**
261	0	93.66	93.66	<LOQ	<LOQ	3.70	NA	97.36
262	0	99.02	99.02	<LOQ	<LOQ	4.05	NA	103.07
Mean		**96.34**	**96.34**	**<LOQ**	**<LOQ**	**3.88**	**NA**	**100.22**
263	7	96.83	96.83	<LOQ	<LOQ	0.76	1.11	98.69
264	7	97.48	97.36	<LOQ	0.12	0.79	1.12	99.39
Mean		**97.15**	**97.09**	**<LOQ**	**0.06**	**0.77**	**1.11**	**99.04**
266	14	95.55	95.44	<LOQ	0.11	0.56	1.80	97.91
267	14	95.25	95.25	<LOQ	<LOQ	0.82	1.40	97.48
Mean		**95.40**	**95.35**	**<LOQ**	**0.05**	**0.69**	**1.60**	**97.69**
269	30	90.45	90.45	<LOQ	<LOQ	1.13	3.25	94.83
271	30	90.83	90.20	<LOQ	0.64	1.09	3.72	95.64
Mean		**90.64**	**90.32**	**<LOQ**	**0.32**	**1.11**	**3.49**	**95.24**
265	76	74.51	73.41	<LOQ	1.11	5.86	13.96	94.34
272	76	73.85	72.68	0.30	0.87	6.54	11.34	91.74
Mean		**74.18**	**73.04**	**0.15**	**0.99**	**6.2**	**12.65**	**93.04**
273	120	61.89	61.02	0.18	0.69	6.38	24.76	93.03
275	120	64.57	63.79	<LOQ	0.78	6.69	19.41	90.67
Mean		**63.23**	**62.40**	**0.09**	**0.74**	**6.53**	**22.08**	**91.85**

NA = not analysed, LOQ = limit of quantification (0.06% AR)
[1]Ambient extracts 1-4 and soxhlet extract (from day 0 onwards) analysis combined
[2]Non-extractable radioactivity from soil
[3]Radioactivity in KOH traps confirmed to be $^{14}CO_2$

The material balance was acceptable in all soils at all time points with individual values ranging from 90.1–99.5% and 90.7–103.1% of applied radioactivity for the phenyl and carbonyl label treated soils, respectively. Between 3.8–18.2% and 3.3–11.6% of applied radioactivity remained non-extracted at 120 DAT for the phenyl and carbonyl labelled soils, respectively.

After 120 days of aerobic incubation, between 10.4 and 47.4% of carbon dioxide was evolved from the phenyl labelled soils and between 11.6 and 54.4% in the carbonyl labelled soils. ≤ 0.4% AR in any individual sample was observed in the ethylene glycol traps.

No metabolites were observed > 5% of applied radioactivity in any soil for either label, with three metabolites identified from the degradates of the [phenyl-U-^{14}C]-bixlozone, and no identified metabolites for [carbonyl-^{14}C]-bixlozone. It is noted that the applicant has separated data for unknown metabolite RRT 0.81 when reporting the results for the carbonyl-14C label, presumably because this metabolite represented the most significant proportion of all minor unknowns with this label. In the soils treated with phenyl labelled bixlozone, 2,4-dichlorobenzoic acid reached mean maximum levels ranging from 1.9 to 4.9% of applied radioactivity but was declining toward the end of the study. 2,4-dichlorobenzyl alcohol and 2,4-dichlorobenzaldehyde reached mean maxima of 2.8% and 0.8% of applied radioactivity, respectively. One unknown metabolite (RRT 0.81) was observed in all soils treated with carbonyl labelled bixlozone reaching a mean maximum value of 2.7% applied radioactivity. All other unknown metabolites accounted for <3.6% and <1.8% of applied radioactivity at any one time point for the phenyl and carbonyl label treated soils, respectively.

The organic matter fractionation results characterising unextractable residues are given in Table CA.B.**8.1.1.1-1** below.

Table CA.B.8.1.1.1-1: Characterisation of non-extractable residues by organic matter fractionation

Soil	Label	As % applied radioactivity			
		Fulvic Acid	Humic Acid	Humin	Total
Lufa 6S	[Phenyl-^{14}C]-bixlozone	2.28	1.38	6.24	9.90
	[Carbonyl-^{14}C]-bixlozone	2.69	1.86	2.61	7.16
Lufa 5M	[Phenyl-^{14}C]-bixlozone	2.41	1.43	7.41	11.24
	[Carbonyl-^{14}C]-bixlozone	3.16	1.97	3.03	8.16
Lufa 2.2	[Phenyl-^{14}C]-bixlozone	0.87	0.57	2.19	3.63
	[Carbonyl-^{14}C]-bixlozone	1.19	0.93	0.75	2.87
RefeSol 02-A	[Phenyl-^{14}C]-bixlozone	2.09	1.34	4.23	7.66
	[Carbonyl-^{14}C]-bixlozone	3.21	2.20	1.87	7.27
CA-SL	[Phenyl-^{14}C]-bixlozone	1.88	1.19	4.78	7.86
	[Carbonyl-^{14}C]-bixlozone	2.24	1.54	1.42	5.19
Iowa	[Phenyl-^{14}C]-bixlozone	2.37	2.26	10.64	15.27
	[Carbonyl-^{14}C]-bixlozone	4.34	3.01	3.46	10.82
LAD-SCL-PF	[Phenyl-^{14}C]-bixlozone	2.41	1.48	0.83	4.72
	[Carbonyl-^{14}C]-bixlozone	2.27	1.42	2.85	6.54

The soil microbial biomass analysis gave between 365.9 – 56.4 µg C/g soil for samples taken at the initial time point, and between 323.1 – 57.1 µg C/g soil for samples taken at the final time point. There was no significant decline of biomass over the study duration (and so levels maintained >1% organic carbon as per the guidelines), though the CA-SL soil had lower biomass than the other soils throughout the study.

CONCLUSION

In four European and three US soils, [phenyl-U-^{14}C]- and [carbonyl-^{14}C]-labelled bixlozone degraded to CO_2 (maximum 54.4% of applied radioactivity) and non-extractable residues. No metabolites were observed > 5% of applied radioactivity. 2,4-dichlorobenzoic acid and 2,4-dichlorobenzyl alcohol reached a mean maximum of 4.9% and 2.8% of applied radioactivity, respectively. 2,4-dichlorobenzaldehyde did not exceed 1% of applied radioactivity in any soil at any timepoint. All unknown metabolites individually accounted for less than 3.6% of applied radioactivity. Overall the CA concluded that none of the minor metabolites breached the triggers for further consideration in this study.

CA.B.8.1.1.1.2. Aerobic route of 3-OH-propanamide degradation

Report:	KCA 7.1.2.1.2-01 Göcer, M., (2016a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600-3-OH-Propanamide Aerobic Degradation in Three Soils at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01058, FMC Tracking no. 2016EFT-ISX2465
Guidelines:	OECD Guideline 307 (April 2002); OPPTS Guideline 835.4200 (October 2008) SANCO/3029/99 rev.4
GLP:	Yes (laboratory certified by German National Authority)

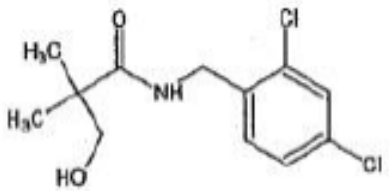
CA comments	This study has been submitted due to 3-OH being detected at levels >10% AR in the laboratory anaerobic degradation study (CA.B.8.1.1.2.1). No significant deviations from the guidelines occurred; further methods of analysis validation are presented in Vol 3CA, section B5. **This study is relied upon.**

INTRODUCTION

This study was conducted to investigate the aerobic soil degradation of bixlozone-3-OH-propanamide (hereby referred to as 3-OH) in three European soils over 48 days. The study was conducted in accordance with OECD 307 guidelines.

MATERIALS

Test substance Bixlozone-3-OH-propanamide



Lot/Batch no. ARD48P2
Purity 98.5% (w/w)
CAS No Not available

Soil

Topsoil was freshly collected from three sites in Germany and France where there had been no pesticide use in the last 5 years. These soils all broadly fall into the 'representative soil' defined in the OECD guideline (pH 5.5 – 8.0, organic carbon content of 0.5 – 2.5 %) . All soils were sieved to 2 mm and stored at *ca* 4°C in the dark for up to 2 months prior to the initiation of the experiment. A summary of the physical and chemical properties of the soils is provided in Microbial biomass was also measured at the initial and final time point of the study, using the substrate induced respiration (SIR) method, as recommended in the OECD guideline.

Table CA.B.8.1.1.1.2-1: Soil physiochemical properties

Soil Characterisation	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
Sampling location	Dudenhofen, Germany	Leimersheim, Germany	Herault, France
Particle size distribution			
Sand (%)	86.0	34.5	13.7

Soil Characterisation	LUFA 2.1	LUFA 2.4	St. Bauzille 12-060
Silt (%)	10.5	40.6	46.0
Clay (%)	3.5	24.9	40.4
Textural classification (USDA)	Loamy sand	Loam	Silty clay
pH (0.01M $CaCl_2$)	4.84	7.41	7.53
% Organic matter	1.17	3.25	3.61
% Organic carbon†	0.68	1.89	2.1
CEC (meq/100g)	4.3	32.0	19.0
Bulk density, disturbed (g/cm^3)	1.447	1.220	1.21
% Maximum water holding capacity (MWHC)	31.65	49.16	46.33
Moisture maintained in experiment	50±5% MWHC	50±5% MWHC	50±5% MWHC
Soil biomass ‡ (mg C/100 g soil)			
Initial	122.5	243.7	373.4
Final	137.8	201.2	334.4

† % Organic carbon = % Organic matter / 1.724

‡ For samples treated with application solution

METHOD

Experimental conditions

Soil samples (100 g oven-dry weight equivalent) of each of the three soils were weighed into individual incubation vessels and were adjusted to soil moisture contents of 50 ± 5% of the maximum water holding capacity as reported in Table CA.B.**8.1.1.1.2-1Error! Reference source not found.**. Stock solution of 3-OH was prepared by dissolving 10.8 mg of test item in 20 mL acetonitrile. Application solution was prepared by diluting 7.565 mL stock solution with acetonitrile/water (1/1, v/v) to a final volume of 100 mL. This application solution has a concentration of 40.25 mg/L.

28 flasks were treated with 3-OH, of which 8 were initially analysed. Application controls consisted of spiking an aliquot of 400 µL application solution into vessels and dilution to volume with acetonitrile/water (80/20, v/v). Concurrent recoveries with 440 µL (110 %) and 20 µL (LOQ) of the application solution were performed at time zero, 12, 24 and 48 hours after treatment to demonstrate extraction efficiency. Please refer to section Vol 3CA, B5 for method validation. 2 additional vessels were treated and analysed subsequently in order to capture residue data for 12 hours after treatment, and a further 4 were treated and analysed for 2 and 4 hours after application time points.

42 vessels contained untreated soil, of which 12 were analysed with one blank control and two concurrent recovery samples per original time point. A further 6 untreated vessels were analysed as controls for the additional 2,4 and 12 hours time points. 10 treated and 10 untreated vessels were used for the determination of soil microbial biomass. Samples were pre-incubated for at least 3 days at 20 ± 2 °C in the dark.

The application rate of the non-labelled test substance was 0.161 µg/g soil (dry weight). The incubation vessels were closed by a polyurethane plug and were maintained in the dark at 20 ± 0.7 °C under aerobic conditions for 48 hours. If necessary, moisture content of the individual soils was readjusted to these specific contents by addition of deionised water during the incubation. The CA notes that there were no trapping systems attached to vessels to collect volatile organic compounds which is recommended in the OECD guideline.

The applicant has used a closed biometer flask system in this study, with all soils being shown as similarly biologically active at both the first and last time point. The CA would ideally have liked to

see volatile trapping systems included in this set up. This study was conducted to find the rate of degradation of 3-OH, as the route of degradation study in section CA.B.8.1.1.1.1 shows 3-OH as a terminal metabolite in the applicant's proposed degradation scheme. Therefore the CA considers that these deviations do not affect the outcome of the study.

Sampling

Duplicate units of each soil were removed for analysis immediately after test substance application. Duplicate incubation units were removed for analysis at intervals of 2, 4, 6, 12, 24, and 48 hours after treatment.

Description of analytical procedures

Soil samples were immediately extracted and stored at <18°C for up to 1 day. Soil samples were extracted once with acetonitrile/water (80:20 v/v, *ca* 200 mL) at ambient temperature, agitated for 30 min and centrifuged at 2230 rpm for 4 min. This was repeated twice with acetonitrile/water (80:20 v/v, *ca* 100 mL). One final extraction was then performed under microwave conditions at elevated temperature (55°C) with acetonitrile/water (80:20 v/v, *ca* 100 mL) for 15 min. This was followed by agitation for 30 min after which all extracts were combined and centrifuged for 5 min.

An aliquot (200 µL) was transferred into a glass vial and diluted with acetonitrile/water (1/1; v/v) to a final volume of 1200 µL. 3-OH was analysed by reverse phase high performance liquid chromatography coupled with mass spectrometry (LC-MS/MS) using a Phenomenex Luna 5µ C_{18} 100A column and a gradient elution using water containing 5mM ammonium acetate and methanol containing 5mM ammonium acetate as solvents. The ion transition monitored by the mass spectrometer was 276.0 – 159.0 m/z, with a conformation run monitored at 276.0 – 89.1 m/z. The limit of quantification (LOQ) was 0.0081 mg/kg, equivalent to 5.0% of the applied test substance. Calibration was performed with internal reference standards. Please see section Vol 3CA, B5 for method validation.

RESULTS

Similar levels of microbial biomass were measured between initial and final time points in all the three soils.

3-OH degraded over the course of the study, with percentage recovery of 3-OH in the three soils treated with the non-labelled test substance is presented in Table CA.B.**8.1.1.1.2-2**

Table CA.B.8.1.1.1.2-2: Percent recovery in the three European soils following application of non-labelled 3-OH under aerobic conditions

	% Applied test substance		
Time (hours)	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
0	102.6	103.9	99.8
0	103.7	100.7	98.4
Mean	103.2	102.3	99.1
2	91.7	91.9	87.0
2	93.9	89.8	87.7
Mean	92.8	90.9	87.4
4	70.2	79.3	77.1
4	82.2	77.4	79.1
Mean	76.2	78.4	78.1
6	72.3	71.2	73.2
6	71.2	78.6	79.3
Mean	71.8	74.9	76.3
12	48.4	36.8	38.9
12	47.5	35.8	40.2
Mean	48.0	36.3	39.6
24	22.4	16.1	16.3
24	37.1	17.6	15.8
Mean	29.8	16.9	16.1
48	4.4*	3.6*	4.7*
48	7.1	8.1	4.9*
Mean	5.8	5.9	4.8

* <LOQ. The limit of quantification is 0.0081 mg/kg, equivalent to 5.0% of the applied test substance

Recoveries for samples on day 0 (0 hours) were between 98.4 % and 103.9 % demonstrating acceptable recovery for non-radiolabelled samples according to the OECD 307 guideline. Residues of 3-OH in blank samples were less than 30 % of the assigned LOQ of the test item.

Concurrent recoveries fortified at 110 % and 5 % (LOQ) samples were between 90.6 – 109.5 % for Lufa 2.1 (n=10, RSD 4.5 %), 92.2 – 107.2 % for Lufa 2.4 (n=10, RSD 3.2%) and 89.5 – 107.6 % for St Bauzille 12-060 (n=10, RSD 5.2 %).

KINETIC ASSESSMENT

The kinetic assessment of the results is presented in section CA.B.8.1.1.4.2.

CONCLUSIONS

The aerobic degradation of non-labelled 3-OH was investigated in three European soils incubated at 20 ºC and 50% MWHC. The study was performed over 48 days, with final mean quantities detected between 4.8% and 5.9% of the initial dose. The kinetic assessment is presented in section CA.B.8.1.1.4.2.

CA.B.8.1.1.1.3. Aerobic route of 2,4-dichlorobenzoic acid degradation

Report:	KCA 7.1.2.1.2-02 Göcer, M., (2016b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	2,4-Dichlorobenzoic Acid Aerobic Degradation in Three Soils at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01059, FMC Tracking no. 2016EFT-ISX2468
Guidelines:	OECD Guideline 307 (April 2002); OPPTS Guideline 835.4200 (October 2008) SANCO/3029/99 rev.4
GLP:	Yes (laboratory certified by German National Authority)

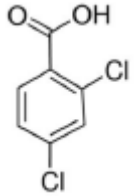
CA comments	This study has been submitted due to 2,4-DBA being detected at levels >10% AR in the soil dissipation studies (CA.B.8.1.2.1) and >5% and increasing at study end in the laboratory anaerobic degradation study (CA.B.8.1.1.2.1). No significant deviations from the guidelines occurred; further methods of analysis validation are presented in Vol 3CA, section B5.. **This study is relied upon.**

INTRODUCTION

This study was conducted to investigate the aerobic soil degradation of bixlozone-2,4-Dichlorobenzoic acid (hereby referred to as 2,4-DBA) in three European soils over 60 days. The study was conducted in accordance with OECD 307 guidelines.

MATERIALS

Test substance 2,4-Dichlorobenzoic acid



Lot/Batch no. S34634V
Purity 99.9% (w/w)
CAS No 50-84-0

Soil

Topsoil was freshly collected from sites in Germany and France where there had been no pesticide use in the last 5 years. These soils all broadly fall into the 'representative soil' defined in the OECD guideline (pH 5.5 – 8.0, organic carbon content of 0.5 – 2.5%). All soils were sieved to 2 mm and stored at *ca* 4°C in the dark for up to 2 months prior to the initiation of the experiment. A summary of the physical and chemical properties of the soils is provided in Table CA.B.**8.1.1.1.3-1**. Microbial biomass was also measured at the initial and final time point of the study, using the substrate induced respiration (SIR) method, as recommended in the OECD guideline.

Table CA.B.8.1.1.1.3-1: Soil physiochemical properties

Soil Characterisation	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
Sampling location	Dudenhofen, Germany	Leimersheim, Germany	Herault, France
Particle size distribution			
Sand (%)	86.0	34.5	13.7
Silt (%)	10.5	40.6	46.0
Clay (%)	3.5	24.9	40.4
Textural classification (USDA)	Loamy sand	Loam	Silty clay
pH (0.01M $CaCl_2$)	4.84	7.41	7.53
% Organic matter	1.17	3.25	3.61
% Organic carbon†	0.68	1.89	2.1
CEC (meq/100g)	4.3	32.0	19.0
Bulk density, disturbed (g/cm^3)	1.447	1.220	1.21
% Maximum water holding capacity (MWHC)	31.65	49.16	46.33
Moisture maintained in experiment	50±5% MWHC	50±5% MWHC	50±5% MWHC
Soil biomass ‡ (mg C/100 g soil)			
Initial	90.6	269.0	364.9
After 18 days	107	288	305.9
After 32 days	97.7	228.1	219.7
Final (after 60 days)	109.1	Not determined*	224.2

† % Organic carbon = % Organic matter / 1.724; ‡ For samples treated with application solution
*soil only sampled to 30d

These soils are also used in study CA.B.8.1.1.1.2 for investigating the metabolite bixlozone-3-OH-Propanamide.

METHOD

Experimental conditions

Soil samples (100 g oven-dry weight equivalent) of each of the three soils were weighed into individual incubation vessels and were adjusted to soil moisture contents of 50 ± 5% of the maximum water holding capacity as reported in **Error! Reference source not found.**. Samples were pre-incubated for at least 3 days at 20 ± 2 °C in the dark. Stock solution was prepared by dissolving 20.0 mg of 2,4-DBA in 20 mL acetonitrile. Application solution was prepared by diluting 1.401 mL of stock solution with acetonitrile/water (1/1, v/v) to a final volume of 100 mL.

24 vessels for each soil were treated with 2,4-DBA. For Lufa 2.4 16 of these were analysed, whereas for Lufa 2.1 and St. Bauzille 12-060 18 vessels were analysed. 42 vessels for each time point and each soil were not treated with 2,4-DBA and used as controls and concurrent recoveries. 24 of these were analysed for Lufa 2.4 and 27 were analysed for Lufa 2.1 and St. Bauzille 12-060. The remaining treated and untreated vessels for each soil were kept as reserves. An additional 10 vessels per soil treated with 2,4-DBA and 10 vessels treated with the same amount of solvent were used for the determination of microbial biomass at the start, during and end of the study. application controls consisted of spiking an aliquot of 400 µL application solution into vessels and dilution to volume with acetonitrile/water (80/20, v/v). Concurrent recoveries with 440 µL (110%) and 20 µL (LOQ) of the application solution were performed at each time point to demonstrate extraction efficiency.

The application rate of the non-labelled test substance 2,4-DBA was 0.056 µg/g dry weight. The incubation vessels were closed by a polyurethane plug and were maintained in the dark at 20 ±2 °C under aerobic conditions for 30 days (LUFA 2.4) or 60 days (LUFA 2.1, St. Bauzille 12-060). If necessary, moisture content of the individual soils was readjusted to these specific contents by addition of deionised water during the incubation. All the three soils remained microbially active throughout the incubation.

The applicant has used a closed biometer flask system in this study, with all soils being shown as similarly biologically active at both the first and last time point. The CA would ideally have liked to see volatile trapping systems included in this set up; although it is noted only CO_2 was detected in the parent study volatile traps (section CA.B.8.1.1.1.1, where <5% 2,4-DBA was detected in soil), indicating negligible volatilisation occurred. This study was conducted to find the rate of degradation of 2,4-DBA, as the route of degradation study in section CA.B.8.1.1.1.1 shows 2,4-DBA as a terminal metabolite in the applicant's proposed degradation scheme. Therefore the CA considers that these deviations do not affect the outcome of the study.

Sampling

Duplicate samples were analysed immediately after treatment 0.25, 1, 2, 4, 7, 15, 30 and 60 DAT. The Lufa 2.4 soil was not sampled at 60 days after treatment.

Description of analytical procedures

The initial extraction of all soil samples was immediately conducted. The extracts were stored in a freezer (< -18°C) and were analysed a maximum of five days later. The soil samples were extracted once with acetonitrile/water (80:20 v/v, *ca* 200 mL) at ambient temperature, agitated for 30 min and then centrifuged at 2230 rpm for 4 min. This was repeated twice with acetonitrile/water (80:20 v/v, *ca* 100 mL). A final extraction was performed under microwave conditions at elevated temperature (55°C) for 15 min with acetonitrile/water (80:20 v/v, *ca* 100 mL), followed by agitation for 30 min at ambient temperature. All extracts were combined, an aliquot centrifuged for 5 mins, and an aliquot of the supernatant (250 µL) was transferred into a glass vial and was diluted with water to a final volume of 1000 µL. 2,4-Dichlorobenzoic acid was analysed by reverse phase high performance liquid chromatography coupled with mass spectrometry (LC-MS/MS) using a Phenomenex Luna 5µ C_{18} 100A column and a gradient elution using water containing 5mM ammonium acetate and methanol containing 5mM ammonium acetate as solvents. The ion transition monitored by the mass spectrometer was 276.0 – 159.0 m/z, with a conformation run monitored at 276.0 – 89.1 m/z. The limit of detection (LOD) for this system was 0.00056 mg/kg and limit of quantification (LOQ) was 0.0028 mg/kg, equivalent to 1.0 and 5.0% of the applied test substance, respectively. Calibration was performed with internal reference standards. Please see Section CA 4.1.2 for the method validation.

RESULTS

The soils showed no significant decline in microbial activity over the study duration, though it started to decrease after 30-60 days in St Bauzille soil. Lufa 2.1, the soil with the lowest organic carbon, had lower levels of microbial biomass than the other two soils at study start. Based on the soil biomass data presented in Table CA.B.**8.1.1.1.3-1**, the CA considers that all soils were microbially viable for the duration of the study.

2,4-DBA degraded over the course of the study, with percentage recovery of 2,4-dichlorobenzoic acid in the three soils treated with the non-labelled test substance is presented in Table CA.B.**8.1.1.1.3-2**.

Table CA.B.8.1.1.1.3-2: Percent recovery in the three European soils following application of non-labelled 2,4-dichlorobenzoic acid under aerobic conditions

	% Applied test substance		
Time (days)	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
0	95.4	95.2	92.9
0	95.7	94.5	96.0
Mean	**95.6**	**94.9**	**94.5**
0.25	97.3	99.8	100.1
0.25	96.3	101.2	100.6
Mean	**96.8**	**100.5**	**100.4**
1	89.8	95.6	92.9
1	92.6	96.2	94.4
Mean	**91.2**	**95.9**	**93.7**
2	81.9	69.1	74.8
2	78.2	68.7	73.0
Mean	**80.1**	**68.9**	**73.9**
4	58.1	39.3	49.4
4	57.1	46.6	61.1
Mean	**57.6**	**43.0**	**55.3**
7	47.4	20.6	46.4
7	47.7	27.5	54.8
Mean	**47.6**	**24.1**	**50.6**
15	38.7	11.0	29.5
15	45.3	2.5*	34.4
Mean	**42.0**	**6.8**	**32.0**
30	5.9	< LOD **	2.6 *
30	11.2	< LOD**	14.5
Mean	**8.6**	**< LOD****	**8.6**
60	4.8 *	n.d.	5.5
60	5.2	n.d.	5.4
Mean	**5.0**	**n.d.**	**5.5**

n.d. = not determined
* <LOQ. The limit of quantification is 0.0028 mg/kg, equivalent to 5.0% of the applied test substance
** < LOD. The limit of detection is 0.00056 mg/kg, , equivalent to 1.0% of the applied test substance

Recoveries for samples on day 0 were between 92.9 and 96.0% which is between the acceptable levels of 70 – 110 % for a non-radiolabelled test substance cited in the OECD guideline.

The concurrent recoveries of fortified samples were between 88.7 and 92.8% for Lufa 2.1 (n=10, RSD 1.4 %), 90.1 – 107.2 % for Lufa 2.4 (n=10, RSD 4.8 %) and 89.7 – 97.9% (n=10, RSD 3.1%). Residues of 2,4-DBA in blank samples were less than 30 % of the LOQ of the test item.

KINETIC ASSESSMENT

The kinetic assessment of the results is presented in section CA.B.8.1.1.4.3.

CONCLUSIONS

The aerobic degradation of non-labelled 2,4-DBA was investigated in three European soils incubated at 20 ºC and 50% MWHC. The study was performed over 60 days, with final mean quantities detected between 1% and 5.5% of the initial dose. The kinetic assessment is presented in section CA.B.8.1.1.4.3.

CA.B.8.1.1.2. *Anaerobic route of degradation*

CA.B.8.1.1.2.1. Bixlozone route of anaerobic degradation

Report:	KCA 7.1.1.2 Simmonds, R., (2015b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Route and Rate of Anaerobic Degradation in Four Soils at 20°C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/002, FMC Tracking no. 2013EFT-ISX1022
Guidelines:	OECD Guideline 307 (April 2002); US EPA OPPTS Guideline 835.4200 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

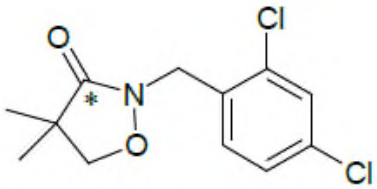
INTRODUCTION

This study was conducted to investigate the anaerobic degradation (over 150 days) of radio-labelled bixlozone in two European and two US soils. The study was conducted in accordance with OECD 307 guidelines.

MATERIALS

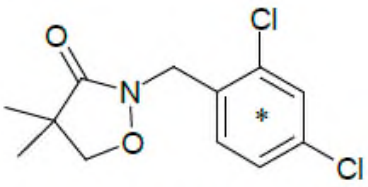
Test substances

Test substance [carbonyl-^{14}C]-bixlozone



* Position of [^{14}C]-radiolabel

[phenyl-U-^{14}C]-bixlozone



* Position of [^{14}C]-radiolabel

Purity [phenyl-U-^{14}C]-bixlozone: >98% (from HPLC)
[carbonyl-^{14}C]-bixlozone: >98% (from HPLC)

CAS No. 81777-95-9

Soil

Topsoil was freshly collected from two European sites and from two US sites. There had been no pesticide use in the last four years for the European soils and in the last two years for the US soil CA-SL (not recorded for soil Iowa). The selected soils fall broadly into the representative soil definition in the OECD guideline, (pH of 5.5 – 8.0). It is also noted that the pH levels of the soils all fall relatively closely between 6.8 and 7.3. A wider range of pH was tested in the aerobic degradation study, with no evidence of pH dependent degradation seen (see section CA.B.8.1.4); therefore, the CA accepts the pH of the soils used. Each soil was sieved to 2 mm and stored at 4 ± 2°C in the dark for up to 4 months prior to the initiation of the experiment. A summary of the physical and chemical properties of the soils is provided in Table CA.B.**8.1.1.2.1-1**. Microbial biomass was also measured at the initial and final time point of the study, using the chloroform fumigation method, as for the aerobic soil degradation study. The CA corresponded with the applicant to confirm pesticide use history in all soils. The CA can confirm that each soil had not been treated with pesticide for at least the previous 4 years, except the

soil CA-SL. The pesticide use history for CA-SL has been included in Table CA.B.**8.1.1.2.1-2** below. The CA can also confirm that the active substances from the history are not considered to be analogues to the active substance bixlozone and therefore do not contravene the OECD 307 guidelines.

Table CA.B.8.1.1.2.1-1: Soil physiochemical properties

Soil characterisation	**Lufa 6S**	**Lufa 5M**	**CA-SL**	**Iowa**
Sampling location	Siebeldingen, Germany	Mechtersheim, Germany	Hughson (CA), USA	Jackson (Iowa), USA
Sampling date	07/03/14	04/06/14	17/03/14	22/03/14
Particle size distribution				
Sand (%)	45	17	5	23
Silt (%)	26	27	18	62
Clay (%)	29	56	77	15
Textural classification (USDA)	Clay	Sandy loam	Loamy sand	Silt loam
pH (0.01 M $CaCl_2$)	6.9	7.3	6.9	6.8
% Organic matter	3.6	1.94	0.6	3.6
% Organic carbon†	2.1	1.1	0.3	2.1
CEC (meq/100g)	21.0	9.0	5.5	13.6
Bulk density (g/cm^3)	1.20	1.18	1.33	1.02
% MWHC	26.5	17.5	12.2	31.0
% moisture at pF2.0	31.0	19.7	13.4	42.3
Microbial biomass (µg C/g soil)				
Initial	334.4	158.5	70.6	319.2
Final	53.3	41.8	45.8	183.6

† % Organic carbon = % Organic matter / 1.724

Table CA.B.8.1.1.2.1-2: Pesticide application history for soil CA-SL

Date of application	Product	Active igredient	CAS number
2013	none	-	-
2012	Ridomil Gold 1.5 pt/A	Mancozeb Metalaxyl-M	8018-01-7 70630-17-0
2011	Prowl 3.3 EC 2pt/A	Pendimethalin	40487-42-1
2010	Ridomil Gold EC 1pt/A	Mefenoxam	70630-17-0
2010	Trifluralin 4 EC 1.25 pt/A	Trifluralin	1582-09-8

STUDY DESIGN

Experimental conditions

Soil samples (100 g oven-dry weight equivalent) of each of the four soils (Lufa 6S, Lufa 5M, CA-SL, and Iowa) were weighed into individual incubation vessels and were adjusted to soil moisture contents between pF2.0 and pF2.5, if necessary. Samples were allowed to acclimatise under study conditions prior to the addition of test substance.

[phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone were prepared by evaporating an aliquot of the supplied test item to dryness under a stream of nitrogen and re-dissolving in 10 mL acetonitrile. The final concentration of the [phenyl-U-^{14}C]-bixlozone was 1.32 mg/mL and the [carbonyl-^{14}C]-bixlozone was 1.38 mg/mL, determined by Liquid Scintillation Counting (LSC).

The two radiolabelled forms of the test substance, [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone, were applied to separate vessels at a nominal rate of 1.07 µg a.s./kg soil (dry weight), (equivalent to 400 g a.s./ha) for each of the eight time points measured per soil. Vessels were maintained in the dark at 19.8 ± 1°C under aerobic conditions for 30 days which was established by a flow of moist air. Nitrogen purged de-ionised water was then added to the samples to an approximate depth of 2 cm above the soil surface to establish anaerobic conditions which were maintained for *ca* 120 days by a flow of nitrogen through the flasks. Volatile organic compounds and CO_2 were trapped in ethylene glycol and potassium hydroxide trapping systems.

The redox potential of the soil and water was monitored in order to determine that anaerobic conditions had been established. pH and oxygen levels of the water were also monitored.

Sampling

Duplicates of each soil and radiolabel were removed for analysis immediately after test substance application. Duplicate incubation vessels and their associated traps were removed for analysis at intervals of 14 and 30 days during the aerobic phase, and at 3, 14, 45, 90 and 120/122 days after waterlogging.

Description of analytical procedures

For anaerobic samples, water was decanted from the flasks. For both anaerobic and aerobic samples, soil samples were extracted once with acetonitrile (*ca* 125 mL), twice with acetonitrile/water (80:20 v/v) (*ca* 125 mL), and once with acetonitrile/water/formic acid (50:50:1 v/v/v), followed by a soxhlet extraction for 6 hours with *ca* 300 mL acetonitrile/water (80:20 v/v). Samples and extracts were stored refrigerated (*ca* 5°C) and analysed within 7 day of sample generation. To determine unextractable radioactivity, samples were combusted and the products absorbed in Carbosorb E scintillation cocktail for quantification by LSC.

The radioactivity in the water layer and soil extracts was determined by LSC. Components present in the combined water (where appropriate) and soil extracts were characterised and quantified by gradient elution reverse phase HPLC with UV detector (LOQ = 0.4% AR), using a Zorbax Rx-C18 column, water:0.01 % acetic acid and acetonitrile:0.01% acetic acid solvents. The HPLC was calibrated against a reference standard for bixlozone and its metabolites, and selected extracts were analysed by LC-MS using a Thermo Q-exactive Orbitrap mass spectrometer monitoring an ion transition of 50 – 600 m/z to provide confirmation of structural identity of metabolites.

Unextracted soil residues were determined in air-dried soil by combustion and LSC. Radioactivity present in the traps was determined directly by LSC and confirmed as CO_2 by barium chloride precipitation.

Microbial biomass was measured using a chloroform fumigation method. Microbial biomass declined over the study duration, but this is expected for anaerobic conditions. As with the aerobic study, the CA-SL soil showed the least microbial biomass both before and after the study. Samples were subject to a 3 week delay prior to analysis due to practicalities. Whilst not ideal, the data gathered from this analysis is sufficient to show that the soils used for the study were microbially active to begin with., and microbial biomass was shown to still be present at levels >1% within the soil at study end in line with the OECD 307 guidelines. Therefore, this is accepted by the CA.

Results and discussion

The total mass balance, distribution of radioactive residues, and the characterisation of the extractable residues are presented in Table CA.B.**8.1.1.2.1-3** to Table CA.B.**8.1.1.2.1-6** for samples treated with [phenyl-U-^{14}C]-bixlozone and in Table CA.B.**8.1.1.2.1-7** to Table CA.B.**8.1.1.2.1-10** for samples treated with [carbonyl-^{14}C]-bixlozone.

Table CA.B.8.1.1.2.1-3: Percent recovery of applied radioactivity in Lufa 6S soil following application of [phenyl-U-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity									
Unit	**Time (day)**	**Total extracted***	**Bixlozone**	**RRT 0.45†**	**RRT 0.73†**	**RRT 0.81†**	**RRT 0.95†**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase											
1	0	98.37	97.43	<LOQ	<LOQ	<LOQ	<LOQ	0.94	0.11	NA	98.48
2	0	98.11	97.46	<LOQ	<LOQ	<LOQ	<LOQ	0.66	0.10	NA	98.21
Mean		**98.24**	**97.44**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.80**	**0.10**	**NA**	**98.34**
4	14	86.73	83.93	2.13	<LOQ	0.67	<LOQ	<LOQ	2.54	1.63	90.91
5	14	91.24	88.11	2.20	<LOQ	0.92	<LOQ	<LOQ	2.90	1.72	95.85
Mean		**88.99**	**86.02**	**2.17**	**<LOQ**	**0.80**	**<LOQ**	**<LOQ**	**2.72**	**1.67**	**93.38**
3	30	86.39	83.71	1.59	<LOQ	<LOQ	0.28	1.10	4.65	3.85	94.90
7	30	84.55	80.64	2.02	<LOQ	<LOQ	0.28	1.90	4.93	4.89	94.37
Mean		**85.47**	**82.17**	**1.80**	**<LOQ**	**<LOQ**	**0.28**	**1.50**	**4.79**	**4.37**	**94.63**
Anaerobic phase											
6	3	85.32	81.49	1.25	<LOQ	1.69	0.33	0.89	5.35	3.05	93.72
9	3	84.87	81.79	1.85	<LOQ	1.11	0.11	0.11	4.08	5.38	94.33
Mean		**85.09**	**81.64**	**1.55**	**<LOQ**	**1.40**	**0.22**	**0.50**	**4.71**	**4.22**	**94.02**
12	14	83.93	81.37	1.58	<LOQ	0.98	<LOQ	<LOQ	5.30	5.62	94.85
14	14	82.61	78.75	1.98	<LOQ	1.88	<LOQ	<LOQ	5.66	6.54	94.82
Mean		**83.27**	**80.06**	**1.78**	**<LOQ**	**1.43**	**<LOQ**	**<LOQ**	**5.48**	**6.08**	**94.83**
15	45	79.78	75.08	1.75	<LOQ	1.49	0.75	1.47	4.87	5.39	90.04
16	45	82.46	78.77	1.78	<LOQ	0.87	1.04	1.04	4.67	5.65	92.78
Mean		**81.12**	**76.92**	**1.77**	**<LOQ**	**1.18**	**0.89**	**1.25**	**4.77**	**5.52**	**91.41**
17	90	79.19	69.65	3.70	2.08	1.42	0.71	2.35	5.32	6.88	91.40
19	90	78.77	70.30	2.75	2.51	1.59	0.40	1.63	8.84	7.50	95.11
Mean		**78.98**	**69.98**	**3.22**	**2.29**	**1.50**	**0.56**	**1.99**	**7.08**	**7.19**	**93.26**
11	120	76.82	50.54	6.31	12.78	1.64	1.50	5.55	9.34	4.61	90.77
22	120	74.11	47.35	5.29	16.74	1.73	1.62	2.99	9.15	7.70	90.96
Mean		**75.46**	**48.95**	**5.80**	**14.76**	**1.69**	**1.56**	**4.27**	**9.24**	**6.15**	**90.86**

NA = not analysed, LOQ = limit of quantification (0.4% AR)

***** Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined

† Relative retention time RRT 0.45=2,4-dichlorobenzoic acid; RRT 0.73= bixlozone-3-OH Propanamide; RRT 0.81=2,4-dichlorobenzyl alcohol; RRT 0.95=2,4-dichlorobenzaldehyde

‡ No individual >1.3% AR

¶ Unextracted radioactivity from soil

§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-4: Percent recovery of applied radioactivity in Lufa 5M soil following application of [phenyl-U-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity								
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**RRT 0.45†**	**RRT 0.81†**	**RRT 0.95†**	**Unknowns ‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase										
45	0	99.35	98.45	<LOQ	<LOQ	<LOQ	0.90	0.06	NA	100.07
46	0	99.23	98.20	<LOQ	<LOQ	<LOQ	1.03	0.06	NA	100.07
Mean		**99.29**	**98.32**	**<LOQ**	**<LOQ**	**<LOQ**	**0.97**	**0.06**	**NA**	**100.07**
47	14	92.89	88.98	3.04	0.70	0.11	0.07	2.41	0.55	95.85
48	14	93.66	89.87	2.22	0.92	<LOQ	0.65	2.31	1.17	97.14
Mean		**93.27**	**89.42**	**2.63**	**0.81**	**0.05**	**0.36**	**2.36**	**0.86**	**96.50**
49	30	86.61	78.63	4.15	1.09	<LOQ	2.73	5.37	2.46	94.43
51	30	84.85	80.30	3.40	1.04	<LOQ	0.10	5.66	4.69	95.20
Mean		**85.73**	**79.46**	**3.77**	**1.07**	**<LOQ**	**1.42**	**5.51**	**3.57**	**94.82**
Anaerobic phase										
54	3	86.71	81.88	3.66	1.18	<LOQ	<LOQ	5.37	3.48	95.57
55	3	87.87	82.36	3.55	1.33	0.17	0.47	4.13	3.18	95.18
Mean		**87.29**	**82.12**	**3.60**	**1.25**	**0.09**	**0.24**	**4.75**	**3.33**	**95.37**
56	14	84.77	78.89	4.03	1.11	0.74	<LOQ	5.14	4.08	93.99
57	14	84.20	78.63	4.30	1.27	<LOQ	<LOQ	4.88	3.87	92.95
Mean		**84.48**	**78.76**	**4.16**	**1.19**	**0.37**	**<LOQ**	**5.01**	**3.98**	**93.47**
50	45	85.32	79.15	4.83	1.19	0.15	<LOQ	5.03	2.62	92.97
52	45	81.36	73.61	4.61	1.14	1.37	0.63	5.95	5.29	92.60
Mean		**83.34**	**76.38**	**4.72**	**1.16**	**0.76**	**0.32**	**5.49**	**3.95**	**92.79**
53	90	80.97	71.49	1.24	0.70	2.34	5.20	5.49	5.00	91.46
58	90	81.42	70.42	3.75	1.33	2.35	3.59	6.55	4.84	92.81
Mean		**81.19**	**70.95**	**2.49**	**1.01**	**2.34**	**4.40**	**6.02**	**4.92**	**92.13**
61	120	75.80	71.21	<LOQ	1.52	2.40	0.65	6.90	10.86	93.56
64	120	82.91	67.23	3.57	<LOQ	2.19	9.92	6.61	4.75	94.27
Mean		**79.35**	**69.22**	**1.78**	**0.76**	**2.30**	**5.29**	**6.76**	**7.80**	**93.92**

NA = not analysed, LOQ = limit of quantification (0.4% AR)

* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined (time zero soxhlet extracts omitted, <1% AR extracted)

† Relative retention time RRT 0.45=2,4-dichlorobenzoic acid; RRT 0.81=2,4-dichlorobenzyl alcohol; RRT 0.95=2,4-dichlorobenzaldehyde

‡ No individual >2.9% AR

¶ Unextracted radioactivity from soil

§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-5: Percent recovery of applied radioactivity in CA-SL soil following application of [phenyl-U-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity								
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**RRT 0.45†**	**RRT 0.81†**	**RRT 0.95†**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase										
89	0	100.08	100.08	<LOQ	<LOQ	<LOQ	<LOQ	0.02	NA	100.22
90	0	101.81	101.81	<LOQ	<LOQ	<LOQ	<LOQ	0.03	NA	101.99
Mean		**100.95**	**100.95**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.03**	**NA**	**101.1**
91	14	95.18	93.05	1.45	<LOQ	<LOQ	0.68	0.91	0.35	96.44
92	14	95.13	92.94	2.02	<LOQ	0.08	0.09	0.83	0.33	96.29
Mean		**95.16**	**92.99**	**1.73**	**<LOQ**	**0.04**	**0.39**	**0.87**	**0.34**	**96.37**
94	30	92.59	88.73	2.98	<LOQ	0.19	0.70	2.17	0.76	95.52
95	30	90.67	89.23	1.26	<LOQ	0.18	<LOQ	2.58	0.42	93.66
Mean		**91.63**	**88.98**	**2.12**	**<LOQ**	**0.18**	**0.35**	**2.37**	**0.59**	**94.59**
Anaerobic phase										
93	3	95.24	92.04	2.31	0.89	<LOQ	<LOQ	1.59	0.35	97.18
97	3	90.93	89.90	1.03	<LOQ	<LOQ	<LOQ	3.64	0.55	95.12
Mean		**93.09**	**90.97**	**1.67**	**0.44**	**<LOQ**	**<LOQ**	**2.61**	**0.45**	**96.15**
98	14	88.72	85.04	3.60	<LOQ	<LOQ	0.09	3.59	0.26	92.57
99	14	88.62	83.75	3.70	0.71	<LOQ	0.46	2.29	0.89	91.81
Mean		**88.67**	**84.40**	**3.65**	**0.35**	**<LOQ**	**0.27**	**2.94**	**0.58**	**92.19**
96	45	87.93	83.93	2.83	0.66	<LOQ	0.50	2.06	0.50	90.48
101	45	87.33	84.55	2.25	0.49	0.03	<LOQ	2.45	0.47	90.25
Mean		**87.63**	**84.24**	**2.54**	**0.58**	**0.02**	**0.25**	**2.25**	**0.49**	**90.37**
102	90	89.37	83.78	3.71	0.51	<LOQ	1.36	5.65	0.62	95.64
106	90	88.49	83.71	4.06	<LOQ	0.49	0.22	3.08	0.68	92.25
Mean		**88.93**	**83.75**	**3.89**	**0.26**	**0.25**	**0.79**	**4.36**	**0.65**	**93.95**
105	120	90.06	84.11	5.91	<LOQ	<LOQ	0.05	1.87	1.19	93.12
109	120	86.62	81.02	5.53	<LOQ	<LOQ	0.07	7.30	0.59	94.51
Mean		**88.34**	**82.57**	**5.72**	**<LOQ**	**<LOQ**	**0.06**	**4.58**	**0.89**	**93.82**

NA = not analysed, LOQ = limit of quantification (0.4% AR)

* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined (time zero soxhlet extracts omitted, <1% AR extracted)

† Relative retention time RRT 0.45=2,4-dichlorobenzoic acid; RRT 0.81=2,4-dichlorobenzyl alcohol; RRT 0.95=2,4-dichlorobenzaldehyde

‡ No individual >0.5% AR

¶ Unextracted radioactivity from soil

§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-6: Percent recovery of applied radioactivity in Iowa soil following application of [phenyl-U-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity								
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**RRT 0.45†**	**RRT 0.81†**	**RRT 0.95†**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase										
133	0	100.05	100.05	<LOQ	<LOQ	<LOQ	<LOQ	0.13	NA	100.19
134	0	99.87	99.87	<LOQ	<LOQ	<LOQ	<LOQ	0.11	NA	99.97
Mean		**99.96**	**99.96**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.12**	**NA**	**100.08**
135	14	88.76	84.33	2.49	1.43	<LOQ	0.51	3.38	3.28	95.43
137	14	87.52	84.52	1.12	1.88	<LOQ	<LOQ	3.42	0.01	90.95
Mean		**88.14**	**84.43**	**1.81**	**1.66**	**<LOQ**	**0.26**	**3.40**	**1.65**	**93.19**
138	30	78.33	76.16	0.26	1.37	0.29	0.25	5.15	9.33	92.80
139	30	89.90	84.66	2.52	2.21	0.40	0.11	5.56	9.82	105.29
Mean		**84.12**	**80.41**	**1.39**	**1.79**	**0.34**	**0.18**	**5.36**	**9.58**	**99.05**
Anaerobic phase										
177	3	81.80	77.05	1.94	1.87	0.59	0.37	6.73	7.86	96.38
178	3	82.35	78.01	1.59	1.87	<LOQ	0.87	6.60	5.78	94.74
Mean		**82.07**	**77.53**	**1.77**	**1.87**	**0.29**	**0.62**	**6.66**	**6.82**	**95.56**
141	14	77.94	74.61	1.65	1.68	<LOQ	<LOQ	6.19	9.16	93.30
147	14	80.94	77.22	1.62	1.96	<LOQ	0.15	6.37	4.77	92.08
Mean		**79.44**	**75.91**	**1.63**	**1.82**	**<LOQ**	**0.07**	**6.28**	**6.97**	**92.69**
144	45	76.38	72.48	1.13	1.44	1.07	0.26	6.06	8.91	91.35
145	45	75.33	72.63	0.69	1.80	0.20	<LOQ	6.26	8.15	91.68
Mean		**75.86**	**72.55**	**0.91**	**1.62**	**0.63**	**0.13**	**6.16**	**8.53**	**91.52**
153	90	75.88	64.64	2.74	2.12	0.70	5.70	7.22	8.17	91.27
154	90	75.38	69.71	2.32	2.03	0.97	0.34	6.60	8.98	90.96
Mean		**75.63**	**67.17**	**2.53**	**2.07**	**0.83**	**3.02**	**6.91**	**8.57**	**91.11**
148	120	74.46	64.45	4.58	2.16	0.44	2.84	7.46	10.93	92.85
149	120	73.93	64.78	3.74	1.61	0.18	3.62	7.65	10.81	92.38
Mean		**74.20**	**64.61**	**4.16**	**1.89**	**0.31**	**3.23**	**7.55**	**10.87**	**92.62**

NA = not analysed, LOQ = limit of quantification (0.4% AR)

* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined

† Relative retention time RRT 0.45=2,4-dichlorobenzoic acid; RRT 0.81=2,4-dichlorobenzyl alcohol; RRT 0.95=2,4-dichlorobenzaldehyde

‡ No individual >2.2% AR

¶ Unextracted radioactivity from soil

§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-7: Percent recovery of applied radioactivity in Lufa 6S soil following application of [carbonyl-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity						
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**RRT 0.73†**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase								
23	0	97.64	96.82	<LOQ	0.82	0.01	NA	97.65
24	0	98.35	98.28	<LOQ	0.07	0.08	NA	98.43
Mean		**97.99**	**97.55**	**<LOQ**	**0.45**	**0.05**	**NA**	**98.04**
25	14	88.90	87.79	<LOQ	1.11	3.05	4.43	96.38
26	14	89.95	88.09	<LOQ	1.86	2.69	5.04	97.67
Mean		**89.43**	**87.94**	**<LOQ**	**1.48**	**2.87**	**4.73**	**97.03**
27	30	82.18	80.60	<LOQ	1.58	4.20	9.67	96.05
28	30	82.85	82.05	<LOQ	0.80	3.91	8.28	95.04
Mean		**82.52**	**81.32**	**<LOQ**	**1.19**	**4.05**	**8.98**	**95.55**
Anaerobic phase								
29	3	83.85	81.76	<LOQ	2.09	3.79	7.88	95.52
30	3	84.25	83.18	<LOQ	1.06	3.77	8.11	96.12
Mean		**84.05**	**82.47**	**<LOQ**	**1.57**	**3.78**	**7.99**	**95.82**
31	14	81.49	80.47	<LOQ	1.02	3.78	8.88	94.15
32	14	82.16	81.20	<LOQ	0.96	3.68	8.64	94.48
Mean		**81.83**	**80.83**	**<LOQ**	**0.99**	**3.73**	**8.76**	**94.32**
33	45	80.67	77.77	<LOQ	2.90	3.53	6.42	90.62
34	45	78.95	77.19	<LOQ	1.76	4.46	7.23	90.64
Mean		**79.81**	**77.48**	**<LOQ**	**2.33**	**4.00**	**6.83**	**90.63**
35	90	76.08	68.19	3.29	4.59	5.42	10.65	92.16
37	90	73.89	65.17	3.59	5.13	7.40	10.97	92.25
Mean		**74.98**	**66.68**	**3.44**	**4.86**	**6.41**	**10.81**	**92.20**
42	122	70.37	46.73	12.90	10.73	7.42	15.93	93.72
43	122	73.42	50.25	14.28	8.90	8.30	14.76	96.48
Mean		**71.90**	**48.49**	**13.59**	**9.82**	**7.86**	**15.34**	**95.10**

NA = not analysed, LOQ = limit of quantification (0.4% AR)
*Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined
† Relative retention time RRT 0.73= bixlozone-3-OH Propanamide
‡ No individual >3.6% AR
¶ Unextracted radioactivity from soil
§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-8: Percent recovery of applied radioactivity in Lufa 5M soil following application of [carbonyl-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity					
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase							
67	0	97.13	96.11	1.02	0.00	NA	97.49
68	0	96.62	96.08	0.54	0.02	NA	97.35
Mean		**96.88**	**96.10**	**0.78**	**0.01**	**NA**	**97.42**
69	14	91.01	90.97	0.04	2.32	3.99	97.32
70	14	90.14	89.52	0.62	2.41	3.83	96.38
Mean		**90.58**	**90.25**	**0.33**	**2.37**	**3.91**	**96.85**
71	30	82.92	81.51	1.40	4.37	8.47	95.75
72	30	83.42	81.83	1.60	4.07	4.84	92.33
Mean		**83.17**	**81.67**	**1.50**	**4.22**	**6.65**	**94.04**
Anaerobic phase							
73	3	82.81	81.29	1.52	3.73	6.93	93.47
75	3	83.92	82.52	1.40	4.02	7.70	95.64
Mean		**83.36**	**81.90**	**1.46**	**3.88**	**7.31**	**94.55**
81	14	79.72	79.72	0.00	4.15	7.76	91.64
82	14	79.96	78.39	1.57	4.23	6.54	90.73
Mean		**79.84**	**79.06**	**0.79**	**4.19**	**7.15**	**91.18**
79	45	75.95	73.38	2.56	3.80	10.59	90.34
83	45	79.84	77.10	2.74	4.29	8.41	92.54
Mean		**77.90**	**75.24**	**2.65**	**4.05**	**9.50**	**91.44**
84	90	73.22	68.43	4.79	5.93	12.78	91.93
85	90	73.78	68.53	5.26	6.02	12.67	92.48
Mean		**73.50**	**68.48**	**5.02**	**5.98**	**12.72**	**92.20**
87	122	76.43	72.73	3.70	5.99	12.17	94.58
88	122	76.81	72.56	4.24	6.33	12.84	95.98
Mean		**76.62**	**72.65**	**3.97**	**6.16**	**12.50**	**95.28**

NA = not analysed
* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined (T0 soxhlet extracts omitted, <1% AR extracted)
‡ No individual >2.5% AR
¶ Unextracted radioactivity from soil
§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-9: Percent recovery of applied radioactivity in CA-SL soil following application of [carbonyl-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity					
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase							
111	0	97.36	96.30	1.06	0.00	NA	97.46
112	0	97.17	96.41	0.76	0.01	NA	97.29
Mean		**97.27**	**96.36**	**0.91**	**0.00**	**NA**	**97.38**
113	14	94.22	94.13	0.10	1.24	0.86	96.32
114	14	94.75	94.68	0.07	1.02	0.55	96.32
Mean		**94.49**	**94.40**	**0.08**	**1.13**	**0.70**	**96.32**
116	30	88.40	87.76	0.64	2.69	6.48	97.57
117	30	87.93	87.80	0.14	1.85	4.48	94.27
Mean		**88.17**	**87.78**	**0.39**	**2.27**	**5.48**	**95.92**
Anaerobic phase							
125	3	91.44	91.44	<LOQ	1.59	3.69	96.72
126	3	93.63	93.55	0.08	1.12	2.53	97.29
Mean		**92.53**	**92.49**	**0.04**	**1.36**	**3.11**	**97.00**
118	14	85.56	85.56	<LOQ	1.94	3.29	90.79
119	14	87.00	86.04	0.96	1.82	4.87	93.69
Mean		**86.28**	**85.80**	**0.48**	**1.88**	**4.08**	**92.24**
121	45	84.30	83.22	1.09	1.75	4.94	91.00
122	45	84.63	83.27	1.36	2.28	6.31	93.22
Mean		**84.46**	**83.24**	**1.22**	**2.02**	**5.63**	**92.11**
120	90	83.54	80.83	2.71	1.53	7.19	92.25
123	90	82.74	81.50	1.24	1.55	8.69	92.98
Mean		**83.14**	**81.17**	**1.97**	**1.54**	**7.94**	**92.62**
124	122	79.42	78.85	0.57	2.20	9.72	91.34
130	122	82.46	81.13	1.33	3.47	9.95	95.87
Mean		**80.94**	**79.99**	**0.95**	**2.83**	**9.83**	**93.60**

NA = not analysed, LOQ = limit of quantification (0.4% AR)
* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined (T0 soxhlet extracts omitted, <1% AR extracted)
‡ No individual >1.0% AR
¶ Unextracted radioactivity from soil
§ All volatile radioactivity found in KOH traps

Table CA.B.8.1.1.2.1-10: Percent recovery of applied radioactivity in Iowa soil following application of [carbonyl-^{14}C]-bixlozone and its metabolites under aerobic and anaerobic conditions

		% Applied radioactivity					
Unit	**Time (day)**	**Total Extracted ***	**Bixlozone**	**Unknowns‡**	**UER¶**	**CO_2§**	**Mass Balance**
Aerobic phase							
155	0	97.52	97.52	<LOQ	0.12	NA	97.64
156	0	98.02	98.02	<LOQ	0.05	NA	98.07
Mean		**97.77**	**97.77**	**<LOQ**	**0.08**	**NA**	**97.85**
157	14	78.32	76.90	1.42	3.43	8.59	90.34
158	14	78.11	76.71	1.41	3.67	9.58	91.36
Mean		**78.21**	**76.8**	**1.31**	**3.55**	**9.08**	**90.85**
159	30	77.27	73.78	3.49	5.34	13.90	96.51
160	30	78.79	76.66	2.13	5.28	5.97	90.04
Mean		**78.03**	**75.22**	**2.81**	**5.31**	**9.93**	**93.28**
Anaerobic phase							
175	3	75.32	72.76	2.56	5.73	10.17	91.22
176	3	80.49	78.22	2.27	4.91	11.06	96.46
Mean		**77.91**	**75.49**	**2.42**	**5.32**	**10.61**	**93.84**
165	14	75.41	73.14	2.27	5.09	11.90	92.40
166	14	75.26	72.88	2.38	5.82	13.95	95.03
Mean		**75.34**	**73.01**	**2.33**	**5.45**	**12.92**	**93.71**
167	45	72.91	70.14	2.77	6.26	10.90	90.07
168	45	74.37	72.35	2.01	6.33	11.94	92.64
Mean		**73.64**	**71.25**	**2.39**	**6.30**	**11.42**	**91.35**
169	90	73.21	71.66	1.55	6.26	17.46	96.92
170	90	73.87	70.92	2.94	6.29	17.64	97.80
Mean		**73.54**	**71.29**	**2.24**	**6.27**	**17.55**	**97.36**
171	122	69.52	63.10	6.42	6.66	19.80	95.97
174	122	69.39	67.30	2.10	6.48	21.26	97.13
Mean		**69.46**	**65.20**	**4.26**	**6.57**	**20.53**	**96.55**

NA = not analysed, LOQ = limit of quantification (0.4% AR)
* Associated water, Extracts 1-4 and Soxhlet Extract 5 analysis combined
‡ No individual >2.2% AR
¶ Unextracted radioactivity from soil
§ All volatile radioactivity found in KOH traps

Anaerobic conditions were maintained throughout the anaerobic portion of the study, with redox measurements falling from +422 mV in water and +425 mV in soil (after 3-4 days under anaerobic conditions) to -100 mV in the water and -149 mV in the soil. The first negative redox potentials were measured on 61 days after anaerobic conditions were initiated; however, the CA notes anaerobic conditions could have been achieved earlier than this sampling occasion. Oxygen readings in the water remained <1.0 % over this period. There was an initial increase in the pH measurements of the water from 7.61 to 8.96 between 3 and 14 days anaerobic conditions, after which it remained between 8.3 and 9.03 for the remainder of the study.

The recoveries of applied radioactivity were acceptable and ranged from 90.34 to 98.43% and from 90.04 to 105.29% for the samples treated with [carbonyl-^{14}C]-bixlozone and [phenyl-U-^{14}C]-bixlozone, respectively.

The radioactivity in the potassium hydroxide trap solutions was confirmed as $^{14}CO_2$ by barium chloride precipitation. After 30 days of aerobic incubation, between 0.6-9.6% AR carbon dioxide was evolved from the phenyl label treated soils and between 5.5-9.9% AR in the carbonyl label treated soils. Carbon dioxide ranged from 0.8% to 10.9% AR in the phenyl label treated soils and from 9.8% to 20.5% AR in the carbonyl label treated soils at the end of the anaerobic phase of the study (day 120/122). Only very minor amounts of radioactivity (≤ 0.02% AR in any individual sample) were observed in the ethylene glycol traps.

No metabolites were observed > 5% AR during the aerobic phase in any soil for either label. For the anaerobic phase of the study, bixlozone-3-hydroxy-propanamide was the only metabolite in both labels which exceeded 10% AR, reaching a maximum mean level of 14.76 % AR in the Lufa 6S soil at the end of the study with the phenyl label. The metabolite 2,4-dichlorobenzoic acid was observed at concentrations >5% AR in the phenyl labelled samples only, achieving a maximum level of 5.8% AR in Lufa 6S soil and still increasing at study end. The metabolites 2,4-dichlorobenzaldehyde and 2,4-dichlorobenzyl alcohol were both only detected in the phenyl label at a maximum concentration of 2.4 % and 2.16 % AR, respectively. Two metabolite-dosed anaerobic studies of bixlozone-3-hydroxy-propanamide and 2,4-dichlorobenzoic acid are presented in sections CA.B.8.1.1.2.2 and CA.B.8.1.1.2.3 respectively.

It is noted that unknown metabolites individually accounted for less than 2.9% and 3.6% AR in the phenyl and carbonyl label treated soils, respectively, according to the applicant. The CA requested further information regarding increase in unknown metabolites towards the end of the studies. The applicant confirmed that despite increases in combined totals, no unidentified metabolites reached more than 3.6% AR.

In response to an information request from the CA, the applicant provided justification excluding 3-OH from the PECsoil calculations. The applicant states prolonged occurrence of anaerobic conditions (>90 days) are required for 3-OH to form in significant levels and that this is inconsistent with productive agriculture to assume that farmers will grow crops under conditions where prolonged presence of anaerobic conditions may be regularly expected. Furthermore, 3-OH exhibits rapid degradation (DT50 = 0.4 d) under aerobic conditions meaning, once aerobic conditions are re-established, the metabolite would degrade rapidly ensuring significant levels do not occur. This justification is accepted by the CA. 2,4-DBA was detected at quantities greater than determined in this anaerobic degradation study in the soil dissipation studies (CA.B.8.1.2.1). Therefore, PECsoil calculations have been undertaken for 2,4-DBA, using the results of the soil dissipation studies (as opposed to this anaerobic degradation study).

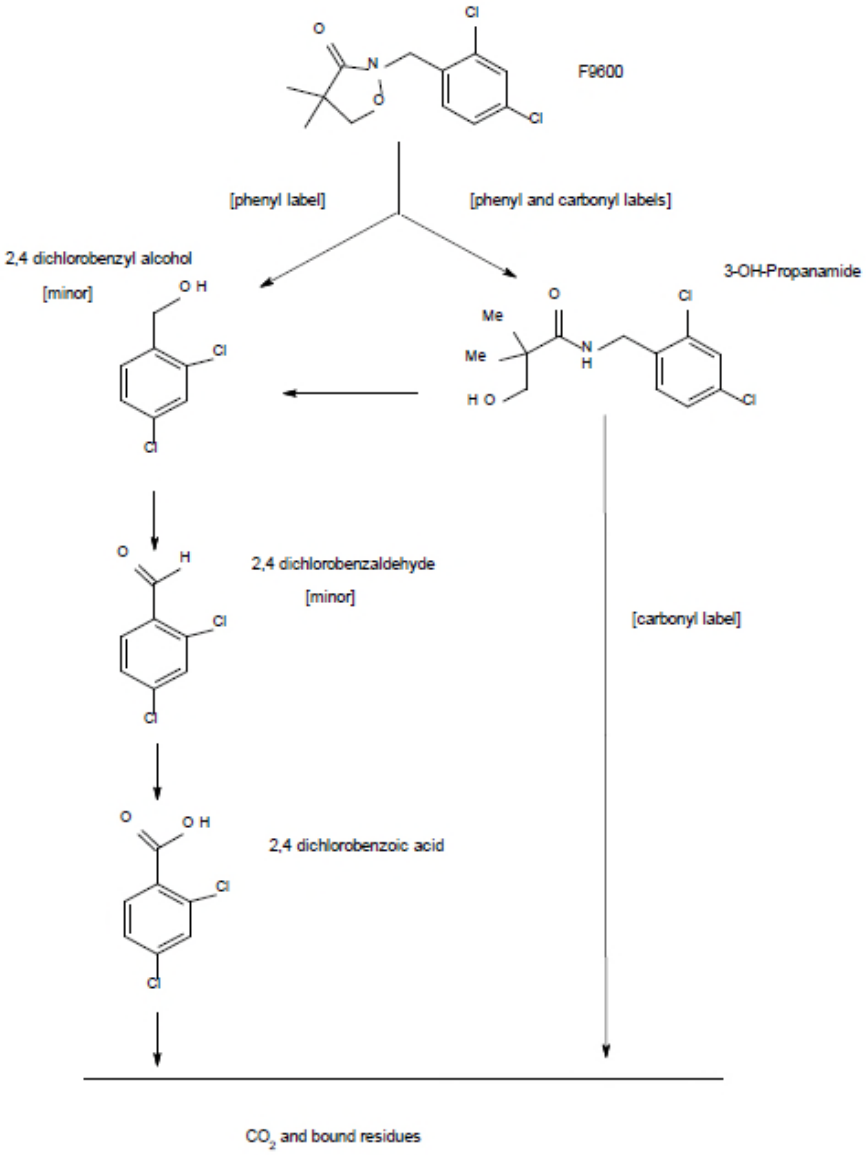
KINETIC ASSESSMENT

The kinetic assessment of the results is presented in section CA.B.8.1.1.5.1.

CONCLUSION

In two European and two US soils, [phenyl-U-^{14}C]- and [carbonyl-^{14}C]-bixlozone degraded to CO_2 (maximum 9.9 and 20.3% of applied radioactivity at the end of the aerobic phase (30 days) and anaerobic phase (120 days), respectively) and bound residues. The recoveries of applied radioactivity for the samples ranged from 90.0 to 105.3% for all soils. In the anaerobic phase of the study, the metabolite bixlozone-3-hydroxy-propanamide was detected as a major metabolite which was present at ≥ 10% applied radioactivity (maximum of 14.76% of applied radioactivity), 2,4-dichlorobenzoic acid was present at ≥ 5% at a single time-point (maximum individual measurement of 6.31% (mean 5.80 %) of applied radioactivity at day 120 and increasing at study end). 2,4-Dichlorobenzaldehyde was detected at a maximum of 2.4% of applied radioactivity and 2,4-dichlorobenzyl alcohol at a mean maximum concentration of 2.16% of applied radioactivity. All unknown metabolites individually accounted for less than 3.6% of applied radioactivity. The applicant's degradation pathway is summarised in Figure CA.B.**8.1.1.2.1-1**.

Figure CA.B.8.1.1.2.1-1.: Anaerobic degradation pathway



Overall, there were more metabolites identified for anaerobic degradation than for aerobic degradation of bixlozone. 2,4-dichlorobenzoic acid, 2,4-dichlorobenzyl alcohol and 2,4-dichlorobenzaldehyde were identified under aerobic conditions, whilst bixlozone-3-hydroxy-propanamide, 2,4-dichlorobenzoic acid, 2,4-dichlorobenzaldehyde and 2,4-dichlorobenzyl alcohol were identified under anaerobic conditions. Justification was provided and accepted excluding 3-OH from the PECsoil calculations.

CA.B.8.1.1.2.2. 3-OH-propanamide anaerobic route of degradation

Report:	KCA 7.1.2.1.4/01, Schwarzkopf, A., (2018a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600-3-OH-Propanamide Anaerobic Degradation in One Soil at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study No.: S17-04093, FMC Tracking No.: 2017EFT-ISX3564
Guidelines:	OECD 307 OPPTS 835.4200 SANCO/3029/99 rev.4
GLP:	Yes

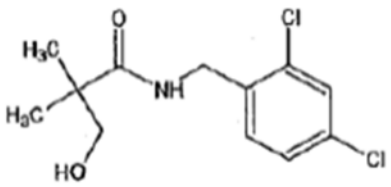
CA comments:	The CA noted the following minor deviations from the OECD guidelines, which are discussed in more detail in the main text: • Temperature deviation up to 22.83 °C for a single 9 hour period • No 30 day aerobic phase before anaerobic conditions (due to a short DT_{50} between 0.4-0.5 days determined in the aerobic degradation study) The CA does not consider these deviations to have significantly impacted upon the outcomes of the study; the method was concluded as being acceptable in the Vol. 3CA, B5. However, as justification was provided and accepted excluding 3-OH anaerobic degradation from the exposure calculations, the results of this study are not considered further in the DAR **This study is <u>not</u> relied upon.**

INTRODUCTION

The rate of degradation of bixlozone-3-OH-propanamide (hereby referred to as 3-OH), a metabolite of bixlozone, was investigated under anaerobic conditions at 20 ± 2 °C. The study was performed over a period of 120 days using one soil of European origin (St. Bauzille 12-060).

MATERIALS

Table CA.B.8.1.1.2.2-1: <u>Test Substance properties</u>

Test Item name:	bixlozone-3-OH-propanomide
Chemical Structure	
Chemical name:	*N*-[(2,4-dichlorophenyl)methyl]-3-hydroxy-2,2-dimethylpropanamide
CAS Number:	Not available
Purity analysed:	98.5 % w/w
Molecular weight:	276.2 g/mol
Storage conditions:	deep frozen (≤ -18°C), dark, dry

Soil

One field fresh European soil (St. Bauzille 12-060) was used for this study. The soil was sieved with a 2 mm sieve and stored at 4 °C prior to use (for a maximum of 28 days). The dry weight of the soil was determined prior to adjusting the water content. At least 5 g of the soil was dried overnight in a climate chamber at 105 °C. This determination was performed with duplicate samples. Water holding capacity (WHC) and biological activity were also determined at the test facility.

Table CA.B.8.1.1.2.2-2: Soil physiochemical properties table

Soil Characterisation	**St. Bauzille 12-00**
Sampling location	Site Fr1, St.Bauzille de la Silve, Herault, France
GPS Coordinates	43°3'33.2''N, 3°34'34.6''E
Pesticide Use History	No pesticides used in previous 5 years
Sampling Depth	0-10 cm
Collection Date	23 May 2017
Shipping Date	31 May 2017
Particle size distribution	
Sand (%)	18.3
Silt (%)	45.5
Clay (%)	36.2
Textural classification (USDA)	Silty clay loam
pH (0.01M $CaCl_2$)	7.26
% Organic matter	2.293
% Total organic carbon	1.33
CEC (meq/100g)	13.6
Bulk density, disturbed (g/L)	1212
Maximum water holding capacity %	42.66
% Moisture maintained in experiment	Flooded
Soil biomass (mg C/100g soil)	
At arrival	36.8
Aerobic Initial	77.1
Aerobic Final	36.8
Anaerobic Initial	60.2
Anaerobic Final	11.8

METHOD

Experimental conditions

All glass test vessels (300 mL) were filled with 100 g soil (dry substance) on 30 June 2017 and moisturised to 50 % WHC on 04 July 2017. The flasks were equilibrated for at least 10 days aerated in the dark at 20 °C.

The test was performed in the dark. The study overall mean temperature was 20.50 °C with a range from 20.02 – 22.83 °C. There was a short deviation of the temperature (about 9 hours up to 22.83 °C). The CA notes this is higher than the 20 ± 2 ˚C recommended in the OECD guidelines. However, due to it only being a minor exceedance of the temperature range and it only occurring for a short period of time, this is not expected to have significantly impacted upon the outcomes of the study.

During the applicant's aerobic degradation study (CA.B.8.1.1.4.2), a DT_{50} was established. The applicant states that since the test item is known to have a fast degradation rate under aerobic conditions, with a DT_{50} 0.4 - 0.5 days, the incubation phase under aerobic conditions prior to the anaerobic phase was not conducted. This is accepted by the CA. Directly after application, the treated soils (except the two replicates for sampling after 0 days) were flooded with approximately 120 mL deionized water and the anaerobic conditions were attained using an N_2 flow through system.

The soil system included flasks as follows:

- 22 flasks, treated with 3-OH. All were analysed.
- 36 untreated flasks, one blank and two concurrent recoveries (one recovery at approximately 5 % of applied amount and one at approximately 110 % of applied amount) per sampling interval. 27 were analysed (9 reserves).

- 12 flasks, treated with the same amount of solvent (acetonitrile/water (1/1, v/v)), were used for the determination of the biomass at the start and after the end of the study to assess any influence on biological activity, 8 were analysed (4 flasks as reserve).
- 6 additional flasks were not applied and not flooded with water (spare samples, not used).

The moisture content was checked prior to application and adjusted to 50 ± 5 % WHC.

Application was made on 10 July 2017. 16.1 µg of 3-OH per 100 g soil (dry weight) were applied in a volume of 400 µL per flask, equivalent to a field application of 0.40 kg a.i./ha assuming a transformation of 20 % from the parent bixlozone.

A stock solution (995 mg/L corrected for the purity of the test substance) was prepared by dissolving 10.1 mg of 3-OH in 10 mL acetonitrile. The application solution (40.25 mg/L) was prepared by diluting 0.809 mL of the stock solution with acetonitrile/water (1/1, v/v) to a final volume of 20 mL. 400 µL acetonitrile/water (1/1, v/v) were added to the flasks for the determination of the biomass. The application solution was applied dropwise to the soil and subsequently mixed by shaking the flask.

The application control was applied by spiking an aliquot of 400 µL of the application solution into 100 mL volumetric flasks and dilution to volume with acetonitrile/water (80/20,v/v). The application check at the 100 % level was performed before and after application. Concurrent recoveries at levels of 110 % (440 µL application solution) and the LOQ (20 µL application solution) were performed at every sampling date to demonstrate the extraction efficiency in the soil. To demonstrate the recovery in the water phase the decanted water was applied at a level of 55 % (220 µL application solution) and 2.5 % (10 µL application solution), calculated with a nominal water amount of 120 mL.

Sampling

Duplicate samples of each system were worked up at the following sampling time intervals: 0, 0.083 (2 h), 0.25 (6 h), 1, 2, 3, 7, 14, 30, 60 and 120 days after treatment.

Soil samples were extracted with acetonitrile (80 v/v) and water (20 v/v).

Directly after application two replicates were worked up immediately (0 day sampling). All other tests vessels were flooded with water and connected to a N_2 flow-through system to obtain anaerobic conditions. Water phases were stored in a freezer ≤ -18°C and were analysed a maximum of 60 days later.

Methods of analysis

The combined extracts and the water phases were analysed for 3-OH residues by high performance liquid chromatography coupled with mass spectrometry (LCMS/MS) in multiple reaction monitoring mode using 3-OH standards in solvent for calibration. Quantification was performed by using linear regression with additional correction for bracketing standards. Injections of sample extracts and water phases were interspersed with injections of standard solutions after maximum 5 samples to verify the detector response and to adjust the calculated concentration. See Vol 3CA, B5 for further information.

RESULTS

Microbial biomass in the soil decreased from 44.7 mg C/100 g under aerobic conditions to 18.8 mg C/100 g on establishment of anaerobic conditions, but then remained stable throughout the anaerobic incubation phase. Oxygen concentrations and redox potentials in the water phase ranged from 0.3 to 1.8 mg/L and 55-220 mV, respectively. The redox potential of the soil phase declined over the course of the study, reaching a negative potential 14 days after treatment and a value of -110 mV on day 120 (range -132.5 to 176.5 mV).

During the phase of method validation, the mean recoveries of 3-OH per level were between 93.9 % and 95.2 % with mean values of relative standard deviation per level between 0.9 % and 3.7 % for water and soil (see Vol 3 CA, B5). Therefore, the samples were within the mean OECD recommended recovery range of 70 % and 110 %.

Table CA.B.8.1.1.2.2-3: Physico-chemical state of test

DAT	Temperature in water [°C]	O_2 in water [mg/L]	Redox potential in water [mV]	Redox potential in sediment [mV]	pH in water	pH in sediment
0.083	21.0	0.6	176.5	169.0	8.6	8.1
0.25	20.0	0.7	136.5	146.5	8.5	7.9
1	20.8	1.1	120.5	140.5	8.3	7.9
2	21.1	0.3	203.0	176.5	8.5	8.1
3	21.2	0.3	166.0	156.5	8.4	8.0
7	21.4	1.8	148.0	35.5	8.1	7.5
14	21.2	0.5	123.0	-64.0	8.2	7.6
30	20.4	0.5	55.0	-125.5	8.1	7.9
60	20.3	0.7	132.5	-132.5	8.4	7.9
120	20.1	1.6	219.5	-109.5	7.8	7.9

The results of the sampling in the water phase, soil phase and total system are described in Table CA.B.**8.1.1.2.2-5**.

Table CA.B.8.1.1.2.2-4: Recovery of bixlozone-3-OH-propanamide from fortified control samples (as % applied)

Sampling interval (day)	Water		Soil	
	LOQ	22 × LOQ	LOQ	22 × LOQ
0	93.7	96.4	98.0	95.3
1	97.2	108.1	96.6	106.9
2	102.0	100.2	96.6	108.5
3	98.3	97.7	88.1	108.2
7	97.4	101.2	99.5	90.1
14	88.5	96.6	87.0	93.6
30	93.7	96.6	86.8	93.3
60	91.0	108.9	97.1	102.5
120	105.5	109.5	103.7	109.0

Table CA.B.8.1.1.2.2-5: Degradation of bixlozone-3-OH-propanamide in soil St. Bauzille 12-060 under anaerobic conditions (as % applied)

Sampling interval (day)	Water	Soil	Total system
0	n.a	95.8	95.8
0	n.a	92.7	92.7
Mean	**n.a**	**94.3**	**94.3**
0.083	52.4	33.7	86.2
0.083	53.5	31.0	84.6
Mean	**53.0**	**32.4**	**85.4**
0.25	44.1	31.8	75.8
0.25	35.5	38.8	74.4
Mean	**39.8**	**35.3**	**75.1**
1	37.6	25.9	63.5
1	31.0	29.2	60.2
Mean	**34.3**	**27.6**	**61.9**
2	41.7	26.6	68.3
2	33.4	27.6	61.0
Mean	**37.5**	**27.1**	**64.6**
3	33.2	28.7	61.9
3	27.2	29.9	57.1
Mean	**30.2**	**29.3**	**59.5**
7	24.3	33.3	56.6
7	22.9	38.1	61.0
Mean	**23.6**	**35.7**	**59.3**
14	12.4	31.0	43.4
14	15.2	32.4	47.6
Mean	**13.8**	**31.7**	**45.5**
30	11.8	37.6	49.4
30	10.8	36.0	48.3
Mean	**11.3**	**36.8**	**48.1**
60	16.2	32.1	47.4
60	15.1	32.4	48.3
Mean	**15.6**	**32.2**	**47.8**
120	2.4	17.0	19.4
120	2.1	21.7	23.9
Mean	**2.3**	**19.3**	**21.6**

n.a Not analysed. Time 0 samples were taken prior to flooding.

KINETIC EVALUATION

The kinetic evaluation is presented in section CA.B.8.1.1.5.2.

CONCLUSION

The anaerobic degradation of non-labelled 3-OH was investigated in one soil under laboratory conditions. The study was performed over 120 days, with a final total system mean quantity of 21.6% of the initial dose. The kinetic assessment is presented in section CA.B.8.1.1.5.2.

CA.B.8.1.1.2.3. 2,4-dichlorobenzoic acid anaerobic route of degradation

Report:	KCA 7.1.2.1.4/02, Schwarzkopf, A., (2018b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	2,4-Dichlorobenzoic Acid Anaerobic Degradation in One Soil at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study No.: S17-04094, FMC Tracking No.: 2017EFT-ISX3565
Guidelines:	OECD 307 OPPTS 835.4200 SANCO/3029/99 rev.4
GLP:	Yes

CA comments	The following minor deviations from the OECD guidelines were noted which are described in more detail in the text below: • Temperature deviation up to 22.83 °C for a single 9 hour period • No day zero figures for anaerobic phase These were not considered to have significantly impacted on the outcomes of the study; the method was concluded as being acceptable in the Vol. 3CA, B5. However, as 2,4-DBA formed in greater quantities in the soil dissipation studies, the results of this anaerobic degradation study have not been considered further in the DAR. **This study is <u>not</u> relied upon.**

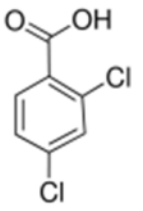
INTRODUCTION

The rate of degradation of 2,4-dichlorobenzoic acid (hereby referred to as 2,4-DBA), a metabolite of bixlozone, was investigated under anaerobic conditions at 20 °C ± 2 °C in the dark according to OECD 307 guidelines. The study was performed over a period of 4 days under aerobic conditions followed by 120 days under anaerobic conditions using one soil of European origin (St. Bauzille 12-060).

MATERIALS

Test Item

Table CA.B.8.1.1.2.3-1: <u>Test Item Properties</u>

Test Item Name	2,4-dichlorobenzoic acid
CAS number	50-84-0
Chemical Structure	
Purity analysed	99.9% w/w
Molecular weight	191.0 g/mol
Storage conditions	Ambient (≤ 30°C), dark, dry

Soil

Soil was collected from one European site which had no history of pesticide application in the previous 5 years. The soils were sieved to <2 mm and stored at 4 °C prior to use.

Table CA.B.8.1.1.2.3-2: Soil physiochemical properties table

Soil Characterisation	**St. Bauzille 12-00**
Sampling location	Site Fr1, St.Bauzille de la Silve, Herault, France
GPS coordinates	43°3’33.2’’N, 3°34’34.6’’E
Pesticide use history	No pesticides used for previous 5 years
Sampling depth	0-10 cm
Collection date	23 May 2017
Shipping date	31 May 2017
Particle size distribution	
Sand (%)	18.3
Silt (%)	45.5
Clay (%)	36.2
Textural classification (USDA)	Silty clay loam
pH (0.01M $CaCl_2$)	7.26
pH in water	7.86
% Organic matter	2.293
% Total organic carbon	1.33
Cation Excahange Capacity (meq/100g)	13.6
Bulk density, disturbed (g/L)	1212
Maximum water holding capacity %	42.66
% Moisture maintained in experiment	Flooded
Soil biomass (mg C/100g soil)	
At arrival	36.8
Aerobic initial	77.1
Aerobic final	36.8
Anaerobic initial	60.2
Anaerobic final	11.8

METHOD

Experimental conditions

One hundred grams (dry weight equivalent) soil samples were moistened to 50% maximum water holding capacity and incubated in glass test vessels, at 20 ± 2 °C, in the dark, for 13 days under aerobic conditions. During the aerobic phase the glass flasks were covered with a PU plug. During the anaerobic phase the flasks were equipped with a N_2 connection.

2,4-DBA in acetonitrile:water (1:1, v/v) was applied at a rate of 5.6 µg per test system, equivalent to a field application rate of 0.40 kg a.i./ha, assuming a transformation rate of 10% from bixlozone.

The stock solution (1009 mg/L corrected for the purity of the test substance) was prepared by dissolving 10.1 mg of 2,4-DBA in 10 mL acetonitrile. The application solution (14 mg/L) was prepared by diluting 0.278 mL of the stock solution with acetonitrile/water (1/1, v/v) to a final volume of 20 mL. To determine the biomass, 400 µL acetonitrile/water (1/1, v/v) was applied to the flasks. The

application solution was applied dropwise to the soil and subsequently mixed by shaking the flask. The application control was applied by spiking an aliquot of 400 µL of the application solution into 100 mL volumetric flasks and dilution to volume with acetonitrile/water (80/20, v/v). The application check at the 100 % level was performed before and after application. Concurrent recoveries at levels of 110 % (440 µL application solution) and the LOQ (20 µL application solution) were performed at every sampling date to demonstrate the extraction efficiency in the soil. To demonstrate the recovery in the water phase during the anaerobic part of the study, the decanted water was applied at a level of 55 % (220 µL application solution) and 2.5 % (10 µL application solution).

Test systems were incubated under aerobic conditions for 4 days in the dark at 20 ± 2 ºC, except for an increase of temperature to 22.83°C for approximately 9 hours. However, due to it only being a minor exceedance of the temperature range and it only occurring for a small period of time, this is not expected to have significantly impacted upon the outcomes of the study. The test systems were then flooded with water and connected to a nitrogen flow through system to establish anaerobic conditions. Test systems were incubated for a further 120 days under anaerobic conditions.

Sampling

Duplicate samples were removed for analysis after 0, 1, 2, 3 and 4 days of aerobic incubation and 1, 3, 7, 14, 31, 60 and 120 days after establishing anaerobic conditions. During the anaerobic incubation, redox potentials of the soil and water phases, and the oxygen concentration in the water phase, were measured at each sampling interval.

Methods of analysis

Water and soil phases were separated by decantation. Soils were extracted three times with acetonitrile: water (80:20, v/v, 1 × 200 mL, 2 × 100 mL) at ambient temperature and once with acetonitrile: water (80:20, v/v, 100 mL) in a microwave oven (55 ºC, 15 mins). Soil extracts were combined for analysis. Soil extracts and water phases were analysed for 2,4-DBA residues by LC-MS/MS (Soil: LOD 0.00056 mg/kg, LOQ 0.0028 mg/kg / water: LOD 0.00023 mg/L, LOQ 0.00115 mg/L). Soils were extracted immediately after sampling. Extracts were stored at ≤ -18 ºC and analysed within 1 day. Storage stability was verified by two concurrent recoveries in fortified control samples. See Vol 3CA, B5 for further information on the methods of analysis and fortification recovery results.

RESULTS

Table CA.B.8.1.1.2.3-3: Degradation of 2,4-dichlorobenzoic acid in St. Bauzille 12-060 soil under aerobic conditions (as % applied)

Sampling interval (day)	Soil
0	106.7
0	101.5
Mean	**104.1**
1	88.5
1	85.0
Mean	**86.8**
2	83.5
2	81.0
Mean	**82.3**
3	66.5
3	75.8
Mean	**71.2**
4	70.9
4	68.3
Mean	**69.6**

Table CA.B.8.1.1.2.3-4: Degradation of 2,4-dichlorobenzoic acid in St. Bauzille 12-060 soil under anaerobic conditions (as % applied)

Sampling interval (day)	Water	Soil	Total system
1	40.5	26.4	66.9
1	50.2	25.9	76.1
Mean	**45.3**	**26.2**	**71.5**
3	37.8	26.7	64.4
3	46.5	27.4	73.9
Mean	**42.1**	**27.0**	**69.2**
7	47.5	25.0	72.4
7	39.4	27.3	66.8
Mean	**43.4**	**26.2**	**69.6**
14	49.2	22.1	71.3
14	36.2	20.4	56.6
Mean	**42.7**	**21.3**	**63.9**
31	46.5	26.7	73.2
31	40.3	23.4	63.7
Mean	**43.4**	**25.0**	**68.5**
60	44.7	28.3	72.9
60	46.0	25.8	71.8
Mean	**45.4**	**27.0**	**72.4**
120	29.4	14.9	44.3
120	32.7	14.9	47.6
Mean	**31.1**	**14.9**	**45.9**

KINETIC ASSESSMENT

The kinetic assessment is presented in section CA.B.8.1.1.5.3.

CONCLUSION

The anaerobic degradation of non-labelled 2,4-DBA was investigated in one soil under laboratory conditions. The study was performed over 124 days, with a final total system mean quantity of 45.9% of the initial dose. The kinetic assessment is presented in section CA.B.8.1.1.5.3.

CA.B.8.1.1.3. ***Soil photolysis***

Report:	KCA 7.1.1.3, Tuffnail, W. (2016)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600: Phototransformation of [^{14}C]- F9600 on Soil Surfaces under Laboratory Conditions
Testing facility:	Quotient Bioresearch Ltd., UK
Document No:	Study no. FCC/02, FMC Tracking no. 2015EFT-ISX2045
Guidelines:	OECD Guidelines for the Testing of Chemicals. Draft Document. Phototransformation of Chemicals on Soil Surfaces (January 2002); US EPA OPPTS Guideline 835.2410 (November 2008) SETAC-Europe Procedures for Assessing the Environmental Fate and ecotoxicology of Pesticides Section 2 (1995)
GLP:	Yes (laboratory certified by UK National Authority)

CA comments	The mass balances of 11 of the replicate samples (8 irradiated samples and 3 dark control samples) dropped below the 90% AR OECD recommended recovery level. For the 3 dark control samples and 4 of the 8 irradiated samples, the CA does not consider this deviation to have significantly altered the outcomes of the study as the corresponding replicate recorded a value within the OECD recommended range. However, for the remaining four samples, these corresponded to two sampling events and so the CA has excluded these from the kinetic analysis as both replicates were outside of the OECD recommended range. **This study is relied upon.**

INTRODUCTION

[Carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone, were applied to thin layers (≤ 2 mm) of soil in order to study the soil photolysis of the new active substance bixlozone. The treated soils were continuously irradiated for up to 15 days. The irradiation intensity to the soil surface per day by artificial sunlight was approximately equivalent to 34 days of natural summer sunlight at latitude 30-50°N.

MATERIALS

Test Substance

Test substance	[carbonyl-^{14}C]-bixlozone [phenyl-U-^{14}C]-bixlozone
Lot/Batch no.	[carbonyl-^{14}C]-bixlozone: CFQ42476 [phenyl-U-^{14}C]-bixlozone: CFQ42475
Purity	[phenyl-U-^{14}C]-bixlozone: >97% (from HPLC) [carbonyl-^{14}C]-bixlozone: >97% (from HPLC)
CAS No.	81777-95-9
Specific activity	[carbonyl-^{14}C]-bixlozone: 59 mCi/mmol, 2.18 GBq/mmol [phenyl-U-^{14}C]-bixlozone: 60 mCi/mmol, 2.22 GBq/mmol

Soil

Fresh topsoil was collected from one European site (Leimersheim) on 2 February 2016 and from one US site (Madera) on 17 March 2016. The soils were transported under ambient conditions and on arrival they were stored at ca. 4 °C prior to the initiation of the experiment. Moist soil (pF 2) was sieved to 2 mm prior to use. A summary of the physical and chemical properties of the soils is provided in Table CA.B.**8.1.1.3-1**. The CA notes that the one of the soils used is a relatively sandy soil, which the OECD guideline points out generally should not be used. The guideline also recommends that the soil should be one that is used for soil aerobic degradation studies. The soil characteristics broadly match with soils used for aerobic degradation studies, with half of the

soils used containing generally ≥50 % particle size distributions in the 0.05 – 2 mm range used in these studies. Therefore, the CA considers the soil used for this study to be in line with those used in other soil degradation studies.

In addition, the moisture content was readjusted back to the field capacity value at every sampling point and the temperature was controlled to prevent drying out. The guideline suggests including an additional set of samples that is allowed to dry out before test substance application to measure phototransformation on irradiated dry soil surfaces. However, given the lack of degradation seen in moist soil, and the fact that the dark controls were also kept at this level, the CA considers that dry soil would not yield results changing the outcome of this study.

Table CA.B.8.1.1.3-1: Soil physiochemical properties

Soil Characterisation	2016/SOIL/02	2015/SOIL/39
Sampling location	Madera County USA	Leimersheim Germany
Particle size distribution		
Sand (%)	65	33
Silt (%)	18	36
Clay (%)	17	31
Textural classification (USDA)	Sandy loam	Clay loam
pH (0.01M $CaCl_2$)	6.2	7.3
% Organic matter	0.79	4.0
% Organic carbon†	0.46	2.3
CEC (meq/100g)	6.5	19.9
Bulk density, disturbed (g/cm^3)	1.09	1.07
% Moisture at pF2.0	16.0	37.7
% Moisture maintained in experiment	16.0	37.7
Soil biomass (µg C/g soil)	270	719

† % Organic carbon = % Organic matter / 1.724

STUDY DESIGN

A preliminary experiment was conducted over 7 days of continuous irradiation showed no significant degradation, but was not reported in detail.

50 µL (nominal) aliquots of [Carbonyl-^{14}C]-bixlozone and [phenyl-U-^{14}C]-bixlozone, were applied to thin layers (≤ 2 mm, 4 g dry weight) of soil slurry kept at field capacity (pF 2) in individual photolysis vessels. Forty eight vessels were prepared in total, with twenty four vessels used for the irradiated test and twenty four for control experiments in the dark. The experimental soils were continuously irradiated for up to 15 days using light from a xenon arc lamp (wavelengths <290 nm filtered out, similar spectrum to natural sunlight) and incubated at 20 ± 2°C. The daily averaged intensity was adjusted to 54.64 W/m² for the 290-400 nm range. Control soils were kept in the dark, also incubated at 20 ± 2 °C. The maximum incubation time of 15 days corresponded to approximately 32 - 34 days of natural summer sunlight at latitude 30-50°N. Air entering the incubation chamber was filtered to remove CO_2 and the vessels fitted with side arm traps of 2M sodium hydroxide to collect $^{14}CO_2$ and any volatile radioactivity formed.

Soils were sampled in duplicate from each soil type for each radio label immediately after treatment, and at 1, 4, 7, 10 and 15 days after treatment (DAT).

METHOD

Soil was extracted twice with acetonitrile and twice with acetonitrile/water (80:20 v/v). Additionally, the irradiated Madera soil treated with [carbonyl-^{14}C]-bixlozone was extracted once with acetonitrile/water/formic

acid (50:50:1 v/v/v, ca 10 mL) for the day 7, 10, and 15 samples and once with dichloromethane for the day 10 samples. Combined extracts were analysed by LSC (LOQ = 0.0047% AR) and analysed chromatographically by reverse phase HPLC (LOD = 0.25% AR; LOQ = 0.5% AR) at 30°C, using gradient elution of 0.1 % acetic acid in water and 0.1% acetic acid in methanol as the mobile phase, and a Zorbax RX-C8 250 x 4.6 mm column. Where possible, analysis was conducted on the day of sampling, otherwise, the samples were stored at < –15°C until analysis. The radioactivity in the sodium hydroxide trap solutions was confirmed as $^{14}CO_2$ by barium chloride precipitation. Radioactivity in extracted soil samples was quantified by combustion/LSC.

Soil samples were combusted using an automated sample oxidizer, with $^{14}CO_2$ trapped with Carbosorb E and Permfluor E+ scintillation cocktail.

Identity of the parent substance was made by co-chromatography of an internal reference standards and confirmed using LC/MS/MS at 40°C with an Agilent Zorbax RX-C8 column, 0.1 % acetic $acid_{(aq)}$ and 0.1 % acetic acid in methanol solvents. The ion transition monitored was 274-290 m/z.

RESULTS AND DISCUSSION

The total mass balance, distribution of radioactive residues, and the characterisation of the extractable residues are presented in Table CA.B.**8.1.1.3-2** and Table CA.B.**8.1.1.3-3** for samples treated with [carbonyl-^{14}C]-bixlozone and in

Table CA.B.8.1.1.3-4 and Table CA.B.**8.1.1.3-5** for samples treated with [phenyl-U-^{14}C]-bixlozone.

Table CA.B.8.1.1.3-2: ^{14}C distribution in irradiated soil (Madera, sandy loam) and dark control treated with [carbonyl-^{14}C]-bixlozone (LOD: 0.0047%)

% Applied radioactivity									
Time (day)	**Total extracted***	**Bixlozone**	**Unknown**				**NER¶**	**CO_2§**	**Mass Balance**
			Polar	**3**	**5**	**6**			
Irradiated									
0	100.42	97.32	ND	ND	1.35	1.76	0.18	NA	100.6
0	99.38	97.25	ND	ND	ND	2.13	0.21	NA	99.59
Mean	**99.90**	**97.29**	**ND**	**ND**	**0.68**	**1.95**	**0.20**	**NA**	**100.1**
1	94.04	91.11	ND	ND	0.96	1.97	1.79	0.12	95.95
1	93.61	91.66	ND	ND	ND	1.95	2.73	0.22	96.56
Mean	**93.83**	**91.39**	**ND**	**ND**	**0.48**	**1.96**	**2.26**	**0.17**	**96.26**
4	91.24	88.68	ND	ND	0.82	1.74	3.06	0.81	95.11
4	93.63	91.29	ND	ND	ND	2.34	3.17	0.11	96.91
Mean	**92.44**	**89.99**	**ND**	**ND**	**0.41**	**2.04**	**3.12**	**0.46**	**96.01**
7	83.35	78.83	ND	ND	0.67	1.70	2.93	1.20	87.48
7	90.55	87.78	ND	ND	ND	0.71	2.12	1.44	94.11
Mean	**86.95**	**83.31**	**ND**	**ND**	**0.34**	**1.21**	**2.53**	**1.32**	**90.80**
10	83.57	79.19	ND	ND	0.70	1.47	2.04	2.48	88.10
10	64.43	58.68	1.45	ND	0.91	0.96	3.64	4.21	72.29
Mean	**74.00**	**68.94**	**0.73**	**ND**	**0.81**	**1.22**	**2.84**	**3.35**	**80.20**
15	78.77	68.32	3.57	2.02	2.02	0.64	3.37	2.90	85.04
15	83.17	80.81	ND	ND	ND	ND	3.38	3.61	90.16
Mean	**80.97**	**74.57**	**1.79**	**1.01**	**1.01**	**0.32**	**3.38**	**3.26**	**87.60**
Dark control									
0	100.08	97.91	ND	ND	ND	2.17	0.26	NA	100.34
0	99.58	97.13	ND	ND	ND	2.45	0.26	NA	99.84
Mean	**99.83**	**97.52**	**ND**	**ND**	**ND**	**2.31**	**0.26**	**NA**	**100.09**
1	94.61	93.12	ND	0.57	ND	0.92	2.40	0.10	97.11
1	95.62	93.25	ND	ND	ND	2.37	2.78	0.18	98.59
Mean	**95.12**	**93.19**	**ND**	**0.29**	**ND**	**1.65**	**2.59**	**0.14**	**97.85**
4	95.15	94.17	ND	ND	ND	0.98	2.92	ND	98.07
4	96.27	94.28	ND	ND	ND	1.99	2.77	0.22	99.26
Mean	**95.71**	**94.23**	**ND**	**ND**	**ND**	**1.49**	**2.85**	**0.11**	**98.67**
7	95.60	92.68	ND	ND	1.00	1.91	3.01	0.19	98.80
7	95.97	93.70	ND	ND	0.62	1.64	3.35	0.24	99.56
Mean	**95.79**	**93.19**	**ND**	**ND**	**0.81**	**1.78**	**3.18**	**0.22**	**99.18**
10	93.06	89.84	ND	ND	1.68	1.55	3.80	0.18	97.04
10	92.49	90.53	ND	ND	ND	1.96	3.83	0.39	96.71
Mean	**92.78**	**90.19**	**ND**	**ND**	**0.84**	**1.76**	**3.82**	**0.29**	**96.88**
15	90.87	90.87	ND	ND	ND	ND	3.76	0.24	94.87
15	93.06	93.06	ND	ND	ND	ND	3.21	0.40	96.67
Mean	**91.97**	**91.97**	**ND**	**ND**	**ND**	**ND**	**3.49**	**0.32**	**95.77**

NA = not analysed; ND = not detected

* Extracts 1-4 combined (irradiated soil: 5 extracts for day 7 and 15 after treatment and 6 extracts for day 10)

¶ Non-extractable radioactivity from soil

§ Radioactivity in NaOH traps confirmed to be $^{14}CO_2$ by $BaCl_2$ analysis. Traps included to capture any volatile component contained < 0.04% of applied radioactivity

Table CA.B.8.1.1.3-3: ^{14}C distribution in irridated soil (Leimersheim, clay loam) and dark control treated with [carbonyl-^{14}C]-bixlozone (LOD: 0.0047%)

% Applied radioactivity									
Time (day)	**Total extracted***	**Bixlozone**	**Unknown**				**NER¶**	**CO_2§**	**Mass Balance**
			3	**4**	**5**	**6**			
Irradiated									
0	101.69	98.83	ND	ND	0.78	2.07	0.46	NA	102.15
0	103.64	100.84	ND	ND	0.77	2.03	0.68	NA	104.32
Mean	**102.67**	**99.84**	**ND**	**ND**	**0.78**	**2.05**	**0.57**	**NA**	**103.24**
1	98.46	96.26	ND	ND	0.79	1.41	1.28	0.13	99.87
1	100.22	98.01	ND	ND	0.74	1.47	1.38	0.06	101.66
Mean	**99.34**	**97.14**	**ND**	**ND**	**0.77**	**1.44**	**1.33**	**0.10**	**100.77**
4	97.77	95.82	ND	ND	0.66	1.28	2.73	1.18	101.68
4	94.32	91.78	ND	0.53	0.97	1.04	3.62	0.73	98.67
Mean	**96.05**	**93.80**	**ND**	**0.27**	**0.82**	**1.16**	**3.18**	**0.96**	**100.18**
7	93.23	92.08	ND	ND	ND	1.15	3.17	0.44	96.84
7	90.51	88.27	ND	ND	0.80	1.45	4.09	0.38	94.98
Mean	**91.87**	**90.18**	**ND**	**ND**	**0.40**	**1.30**	**3.63**	**0.41**	**95.91**
10	86.26	84.04	0.86	ND	0.63	0.72	5.06	1.61	92.93
10	94.71	91.58	ND	1.10	0.71	1.33	4.41	2.58	101.71
Mean	**90.49**	**87.81**	**0.43**	**0.55**	**0.67**	**1.03**	**4.74**	**2.10**	**97.32**
15	86.85	83.37	1.41	ND	1.13	0.96	8.24	1.07	96.16
15	80.23	76.19	1.53	1.03	0.70	0.78	6.58	3.84	90.65
Mean	**83.54**	**79.78**	**1.47**	**0.52**	**0.92**	**0.87**	**7.41**	**2.46**	**93.41**
Dark control									
0	104.10	101.88	ND	ND	0.70	1.51	0.45	NA	104.55
0	103.51	100.75	ND	ND	0.70	2.06	0.57	NA	104.08
Mean	**103.81**	**101.32**	**ND**	**ND**	**0.70**	**1.79**	**0.51**	**NA**	**104.32**
1	100.52	97.95	ND	ND	0.95	1.62	1.35	0.07	101.94
1	92.99	90.85	ND	ND	0.56	1.58	1.23	0.17	94.39
Mean	**96.76**	**94.40**	**ND**	**ND**	**0.76**	**1.60**	**1.29**	**0.12**	**98.17**
4	90.41	88.37	ND	ND	0.63	1.41	2.45	0.52	93.38
4	85.19	83.63	ND	ND	0.55	1.00	2.34	0.52	88.06
Mean	**87.80**	**86.00**	**ND**	**ND**	**0.59**	**1.21**	**2.40**	**0.52**	**90.72**
7	90.45	89.16	ND	ND	ND	1.29	2.73	0.83	94.01
7	92.77	90.93	ND	ND	0.58	1.26	2.68	0.78	96.23
Mean	**91.61**	**90.05**	**ND**	**ND**	**0.29**	**1.28**	**2.71**	**0.81**	**95.12**
10	82.77	80.99	ND	1.08	ND	0.70	3.41	1.48	87.66
10	85.49	83.94	ND	ND	0.44	1.09	3.55	1.25	90.29
Mean	**84.13**	**82.47**	**ND**	**0.54**	**0.22**	**0.90**	**3.48**	**1.37**	**88.98**
15	88.43	86.42	ND	ND	1.18	0.82	6.20	1.80	96.43
15	77.48	75.63	ND	ND	0.81	1.05	4.99	1.52	83.99
Mean	**82.96**	**81.03**	**ND**	**ND**	**1.00**	**0.94**	**5.60**	**1.66**	**90.21**

NA = not analysed; ND = not detected

* Extracts 1-4 combined

¶ Non-extractable radioactivity from soil

§ Radioactivity in NaOH traps confirmed to be $^{14}CO_2$ by $BaCl_2$ analysis. Traps included to capture any volatile component contained < 0.04% of applied radioactivity

Table CA.B.8.1.1.3-4: ^{14}C distribution in irridated soil (Madera, sandy loam) and dark control treated with [phenyl-^{14}C]-bixlozone (LOD: 0.0047%)

% Applied radioactivity										
Time (day)	**Total extrac-ted***	**Bixlozone**	**Unknown**					**NER¶**	**CO_2§**	**Mass Balance**
			1**	**3**	**4**	**5**	**6**			
Irradiated										
0	103.98	101.34	ND	ND	ND	0.85	1.79	0.09	NA	104.07
0	103.09	100.18	ND	ND	ND	0.76	2.14	0.08	NA	103.17
Mean	**103.54**	**100.76**	**ND**	**ND**	**ND**	**0.81**	**1.97**	**0.09**	**NA**	**103.62**
1	100.25	96.78	ND	ND	0.60	0.79	2.08	1.00	0.02	101.27
1	96.06	93.27	ND	ND	ND	0.87	1.90	1.18	ND	97.24
Mean	**98.16**	**95.03**	**ND**	**ND**	**0.30**	**0.83**	**1.99**	**1.09**	**0.01**	**99.26**
4	96.81	94.41	ND	ND	ND	0.70	1.70	2.20	0.07	99.08
4	91.54	88.22	1.22	ND	ND	0.83	1.28	2.04	0.12	93.70
Mean	**94.18**	**91.32**	**0.61**	**ND**	**ND**	**0.77**	**1.49**	**2.12**	**0.10**	**96.39**
7	91.74	89.93	ND	ND	ND	0.41	1.39	2.50	0.26	94.50
7	80.50	73.16	1.83	1.14	0.97	1.17	2.24	3.26	0.03	83.80
Mean	**86.12**	**81.55**	**0.92**	**0.57**	**0.49**	**0.79**	**1.82**	**2.88**	**0.15**	**89.15**
10	85.60	81.88	1.88	ND	ND	ND	1.83	2.40	0.43	88.43
10	90.17	84.73	2.38	ND	ND	0.77	2.29	3.24	0.75	94.16
Mean	**87.89**	**83.31**	**2.13**	**ND**	**ND**	**0.39**	**2.06**	**2.82**	**0.59**	**91.30**
15	83.13	78.73	2.29	ND	ND	0.62	1.49	5.20	1.17	89.50
15	79.28	73.77	2.90	1.28	ND	ND	1.32	4.99	1.17	85.44
Mean	**81.21**	**76.25**	**2.60**	**0.64**	**ND**	**0.31**	**1.41**	**5.10**	**1.17**	**87.47**
Dark control										
0	104.83	101.81	ND	ND	ND	1.10	1.92	0.17	NA	105.00
0	104.44	101.00	ND	ND	0.76	0.91	1.77	0.15	NA	104.59
Mean	**104.64**	**101.41**	**ND**	**ND**	**0.38**	**1.01**	**1.85**	**0.16**	**NA**	**104.80**
1	98.27	95.46	ND	ND	ND	0.71	2.1	2.07	0.01	100.35
1	99.77	97.13	ND	ND	ND	0.78	1.88	1.57	0.01	101.35
Mean	**99.02**	**96.30**	**ND**	**ND**	**ND**	**0.75**	**1.99**	**1.82**	**0.01**	**100.85**
4	95.37	92.31	ND	ND	ND	1.20	1.85	1.42	0.02	96.81
4	89.03	85.09	ND	ND	ND	1.91	2.04	1.60	0.03	90.66
Mean	**92.20**	**88.70**	**ND**	**ND**	**ND**	**1.56**	**1.95**	**1.51**	**0.03**	**93.74**
7	94.58	92.73	ND	ND	ND	ND	1.85	1.81	0.03	96.42
7	97.49	93.74	ND	0.72	ND	ND	3.04	2.14	0.03	99.66
Mean	**96.04**	**93.24**	**ND**	**0.36**	**ND**	**ND**	**2.45**	**1.98**	**0.03**	**98.04**
10	89.99	86.59	ND	ND	ND	0.79	2.61	1.62	0.09	91.70
10	89.84	87.11	ND	ND	ND	0.85	2.19	4.12	0.09	94.35
Mean	**89.92**	**86.85**	**ND**	**ND**	**ND**	**0.82**	**2.4**	**2.87**	**0.09**	**93.03**
15	90.31	87.81	ND	ND	ND	0.81	1.69	4.92	0.03	95.26
15	87.49	85.26	ND	ND	ND	ND	2.23	3.49	0.06	91.04
Mean	**88.90**	**86.54**	**ND**	**ND**	**ND**	**0.41**	**1.96**	**4.21**	**0.05**	**93.15**

NA = not analysed; ND = not detected

* Extracts 1-4 combined

** Tentatively identified as 2, 4-dichlorobenzoic acid

¶ Non-extractable radioactivity from soil

§ Radioactivity in NaOH traps confirmed to be $^{14}CO_2$ by $BaCl_2$ analysis. Traps included to capture any volatile component contained < 0.04% of applied radioactivity

Table CA.B.8.1.1.3-5: ^{14}C distribution in irradiated soil (Leimersheim, clay loam) and dark control treated with [phenyl-^{14}C]-bixlozone (LOD: 0.0047%)

% Applied radioactivity											
Time (day)	**Total extrac-ted***	**Bixlozone**	**Unknown**						**NER¶**	**CO_2§**	**Mass Balance**
			1**	**2**	**3**	**4**	**5**	**6**			
Irradiated											
0	99.53	96.60	ND	ND	ND	ND	0.86	2.08	0.42	NA	99.95
0	98.21	95.45	ND	ND	ND	ND	0.76	2.00	0.81	NA	99.02
Mean	**98.87**	**96.03**	**ND**	**ND**	**ND**	**ND**	**0.81**	**2.04**	**0.62**	**NA**	**99.49**
1	97.48	94.36	ND	ND	ND	ND	0.96	2.16	1.76	0.06	99.30
1	97.80	96.10	ND	ND	ND	ND	ND	1.70	1.89	0.03	99.72
Mean	**97.64**	**95.23**	**ND**	**ND**	**ND**	**ND**	**0.48**	**1.93**	**1.83**	**0.05**	**99.51**
4	94.50	91.82	ND	ND	ND	ND	1.02	1.66	3.90	0.40	98.80
4	93.15	90.64	ND	ND	ND	ND	0.74	1.77	4.62	0.28	98.05
Mean	**93.83**	**91.23**	**ND**	**ND**	**ND**	**ND**	**0.88**	**1.72**	**4.26**	**0.34**	**98.43**
7	96.28	94.14	ND	ND	ND	ND	0.73	1.41	2.83	0.17	99.28
7	88.65	86.30	ND	ND	ND	ND	1.06	1.29	5.23	0.53	94.41
Mean	**92.47**	**90.22**	**ND**	**ND**	**ND**	**ND**	**0.90**	**1.35**	**4.03**	**0.35**	**96.85**
10	95.55	90.43	2.45	ND	0.82	ND	0.75	1.11	3.60	0.89	100.04
10	92.31	88.68	1.02	ND	ND	ND	0.90	1.70	3.18	0.02	95.51
Mean	**93.93**	**89.56**	**1.74**	**ND**	**0.41**	**ND**	**0.83**	**1.41**	**3.39**	**0.46**	**97.78**
15	86.85	78.82	2.52	0.69	0.91	1.35	0.69	1.88	7.04	1.19	95.12
15	88.29	82.01	3.85	ND	1.17	ND	1.25	ND	7.40	0.98	96.69
Mean	**87.57**	**80.42**	**3.19**	**0.35**	**1.04**	**0.68**	**0.97**	**0.94**	**7.22**	**1.09**	**95.91**
Dark control											
0	100.98	98.28	ND	ND	ND	ND	0.72	1.98	0.47	NA	101.45
0	101.10	98.34	ND	ND	ND	ND	0.94	1.82	0.62	NA	101.72
Mean	**101.04**	**98.31**	**ND**	**ND**	**ND**	**ND**	**0.83**	**1.90**	**0.55**	**NA**	**101.59**
1	99.07	97.60	ND	ND	ND	ND	ND	1.47	1.65	0.05	100.77
1	97.93	95.55	ND	ND	ND	ND	0.46	1.92	1.92	0.06	99.91
Mean	**98.50**	**96.58**	**ND**	**ND**	**ND**	**ND**	**0.23**	**1.70**	**1.79**	**0.06**	**100.34**
4	97.40	93.95	0.95	ND	ND	ND	0.53	1.96	3.04	ND	100.44
4	96.17	94.76	ND	ND	ND	ND	0.58	0.83	3.79	0.16	100.12
Mean	**96.79**	**94.36**	**0.48**	**ND**	**ND**	**ND**	**0.56**	**1.40**	**3.42**	**0.08**	**100.28**
7	92.35	90.75	ND	ND	ND	ND	0.83	0.78	3.35	0.46	96.16
7	100.70	97.63	ND	ND	ND	ND	1.06	2.01	3.23	0.33	104.27
Mean	**96.53**	**94.19**	**ND**	**ND**	**ND**	**ND**	**0.95**	**1.40**	**3.29**	**0.40**	**100.22**
10	98.68	95.50	0.61	ND	ND	ND	1.16	1.39	3.10	0.70	102.48
10	96.69	94.03	ND	ND	ND	ND	1.38	1.28	3.23	0.18	100.11
Mean	**97.69**	**94.77**	**0.31**	**ND**	**ND**	**ND**	**1.27**	**1.34**	**3.17**	**0.44**	**101.30**
15	89.80	88.44	ND	ND	ND	ND	ND	1.36	3.88	0.31	93.99
15	90.63	84.82	3.05	ND	ND	ND	0.75	2.00	5.12	1.58	97.33
Mean	**90.22**	**86.63**	**1.53**	**ND**	**ND**	**ND**	**0.38**	**1.68**	**4.50**	**0.95**	**95.66**

NA = not analysed; ND = not detected

* Extracts 1-4 combined

** Tentatively identified as 2, 4-dichlorobenzoic acid

¶ Non-extractable radioactivity from soil

§ Radioactivity in NaOH traps confirmed to be $^{14}CO_2$ by $BaCl_2$ analysis. Traps included to capture any volatile component contained < 0.04% of applied radioactivity

The CA notes that recoveries for 8 irradiated and 3 non-irradiated samples were not in the OECD range of 90-110% AR; the recoveries <90% AR ranged from 72.29% AR to 89.50% AR. All 8 of the irradiated recoveries <90% AR occurred in the Madera soil samples, with 4 recoveries <90% each in the phenyl-labelled tests (Table CA.B.8.1.1.3-4) and the carbonyl-labelled tests (Table CA.B.8.1.1.3-2) respectively. The three non-irradiated samples with recoveries <90% AR occurred in the carbonyl-labelled Leimersheim soil samples (Table CA.B.8.1.1.3-3). The applicant justifies this deviation by noting that mass balances can be an issue with this type of study as the vessels are opened to air during moisture control, which means that the system is not completely closed throughout the study. The applicant also stated that, generally, there is at least a single replicate for all time points. The CA notes there were two occasions where both replicates recorded recoveries <90% AR: Madera. day 10, irradiated, carbonyl-label (88.10% and 72.29%) and Madera, day 15, irradiated, phenyl-label (89.50% and 85.44%). Regarding the 72.29% recovery specifically, the applicant states that although this recovery is ~18% lower than the other replicate, the aliquot variance was <0.5% and therefore this sample was considered acceptable.

For the irradiated samples where at least one replicate was within the OECD recommended range, the CA accepts the applicant's justification that this is unlikely to significantly affect the outcomes of the study. Furthermore, the CA notes the longest (and therefore most conservative as no major photolytic metabolites were observed) irradiated DT50 values were calculated for the Leimersheim soil where all irradiated samples were within the OECD acceptable range. For the two Madera sampling intervals where both replicates were <90% AR, the CA does not consider this data robust and so the CA has omitted the results from the kinetic evaluation (see kinetics paragraph below for further information).

Individual recoveries ranged between 83.99 − 105.0% AR in dark controls, with three individual recoveries below 90 % in the Leimersheim soil at 4, 10 and 15 DAT with the carbonyl label. The CA judges this acceptable due to the data from previous time points being within acceptable ranges, the other replicate from that time point being within the acceptable range and the parent data from this time point being in line with the previous time points.

Non-extracted radioactivity reached a maximum of 7.4% in light and of 5.6% AR in dark conditions.

The radioactivity in the sodium hydroxide trap solutions was confirmed as $^{14}CO_2$ by barium chloride precipitation. After 15 days of continuous radiation, 3.3 and 2.5% AR carbon dioxide was evolved from the soils treated with carbonyl labelled bixlozone and 1.2 and 1.1% AR from the soils treated with phenyl labelled bixlozone. In the dark control sample, < 1.7% AR carbon dioxide was evolved from any treatment. Only very minor amounts of radioactivity (< 0.04% AR in any individual sample) were observed in the traps to capture any other volatile component.

No degradates were observed > 5% AR in the two soils for either label in either irradiated or dark control samples. The largest degradate reached a maximum of 3.85% applied radioactivity (Leimersheim soil, phemyl label, day 15, replicate 2; the mean value of the two day 15 replicates was 3.19% AR) which was identified as 2,4-dichlorobenzoic acid. A number of minor degradates were also observed, none exceeding 3.57% of applied radioactivity. None of these degradates were identified by the applicant.

DT_{50} and DT_{90} values for the rate of photolysis of [^{14}C]-bixlozone in the two soils tested were determined following FOCUS degradation kinetics (2014). All calculations were performed using the software package CAKE 2.0 and are summarised in Table CA.B.8.1.1.3-6. All SFO visual fits were good. Photolysis of [^{14}C]-bixlozone was slow in soil with a mean half-life ($DegT_{50}$) of 36 days continuous irradiation equivalent to *ca*. 78 equivalent summer sunlight days (Madera soil) and 59 days continuous irradiation equivalent summer sunlight days at *ca*. 125 days (Leimersheim soil). The CA agrees with the applicant's fits for all radiolabels/soils except the irradiated carbonyl and phenyl labelled Madera soil, where the CA considers it appropriate to omit specific sampling interval results due to them recording mass balances outside the OECD recommended range. For these labels/soil, the CA has used KinGUI v2 with NLLS selected to perform the kinetic assessment. For the carbonyl label, all results except day 10 were modelled. For the phenyl label, all results except the day 15 were modelled. For both kinetic assessments, the day 0 mass balances were used as the initial sampling points and, for both runs, the SFO model was considered to provide good visual and statistical fits of the data and so no further kinetic models were tested.

The results of the kinetic assessment is presented below. The irradiated DT50 values have not been corrected to account for degradation in the dark control samples. The applicant's original irradiated Madera results are included for informational purposes but are greyed-out and italicised to indicate they are not considered further.

Table CA.B.8.1.1.3-6: Degradation rate of [^{14}C]-bixlozone on soil in irradiated and dark conditions

Soil	**Label**	**Conditions**	**Kinetic model**	**DT_{50} (days)**	**DT_{90} (days)**	**DT_{50} (days) 30-50°N**	**χ^2 (%)**	**t-test**	**Visual assessment**	**Agrees with applicant?**
Madera	Carbonyl	Irradiated	SFO	41	137	93	1.44	<0.001	Good	N
			SFO	*33*	*110*	*75*	*4.02*	*0.001*	*Good*	*n/a*
		dark	SFO	204	676	-	1.3	0.003	Good	Y
	Phenyl	irradiated	SFO	31	103	71	2.72	0.002	Good	N
			SFO	*38*	*128*	*81*	*2.24*	*<0.001*	*Good*	*n/a*
		dark	SFO	72	240	-	2.57	<0.001	Good	Y
Leimers-heim	Carbonyl	irradiated	SFO	50	166	108	1.01	<0.001	Good	Y
		dark	SFO	51	168	-	2.96	<0.001	Good	Y
	Phenyl	irradiated	SFO	67	221	142	1.31	<0.001	Good	Y
		dark	SFO	104	344	-	1.38	<0.001	Good	Y
Geometric mean (n=4)		irradiated	-			100	-			

Greyed, italicised text indicates the applicant's kinetic fit which has not been considered further

Figure CA.B.8.1.1.3-1: SFO kinetic fit for Madera Soil Carbonyl Label (Irradiated)

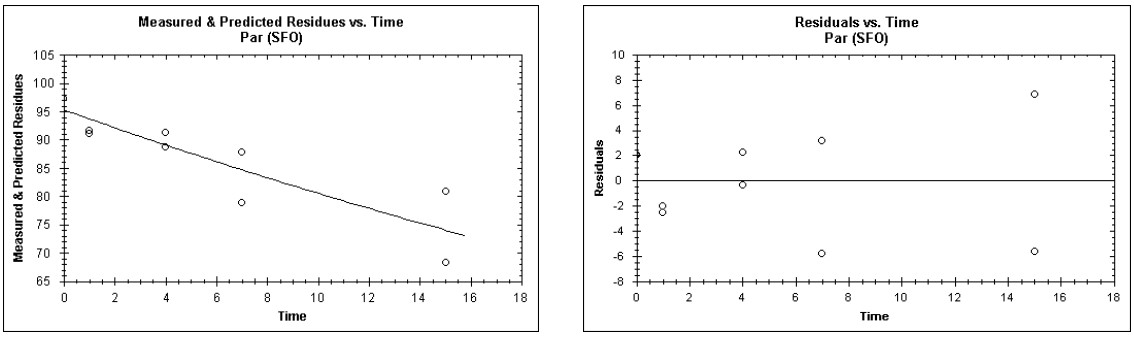


Figure CA.B.8.1.1.3-2: : SFO kinetic fit for Madera Soil, Carbonyl label (dark control)

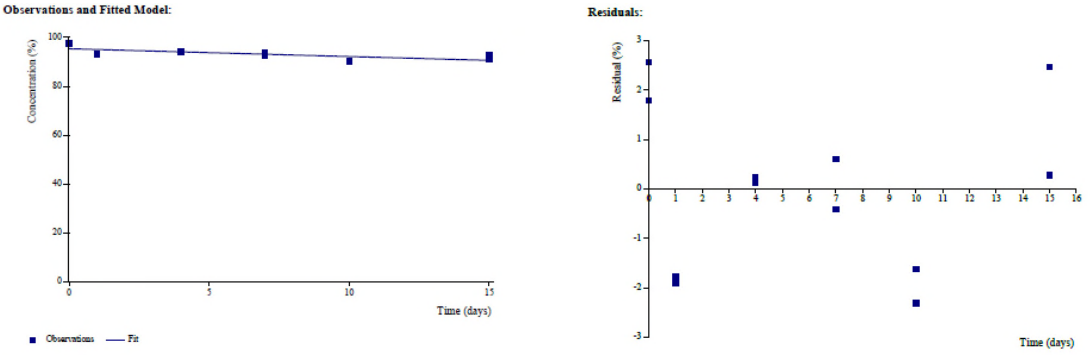


Figure CA.B.8.1.1.3-3: : SFO kinetic fit for Madera Soil, Phenyl label (irradiated)

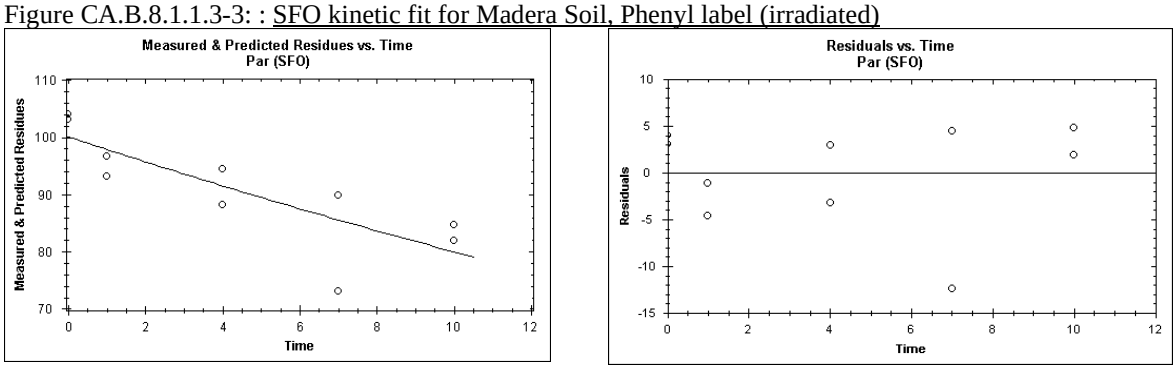


Figure CA.B.8.1.1.3-4: : SFO kinetic fit for Madera soil Phenyl label (dark control)

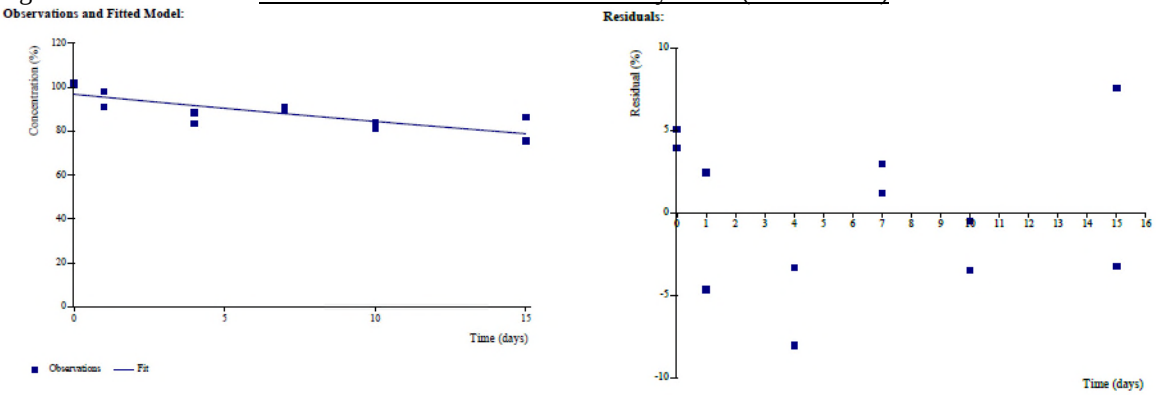


Figure CA.B.8.1.1.3-5: SFO kinetic fit for Leimershiem soil, Carbonyl label (irradiated)

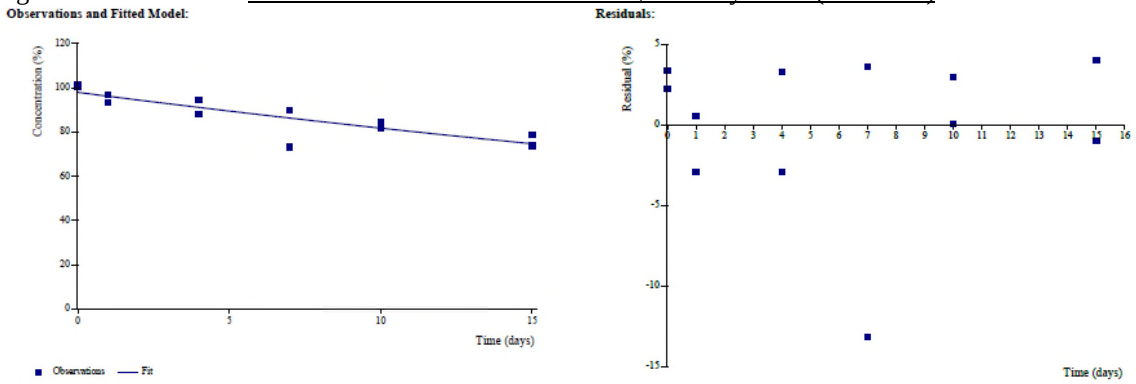


Figure CA.B.8.1.1.3-6: : SFO kinetic fit for Leimershiem soil, Carbonyl label (dark control)

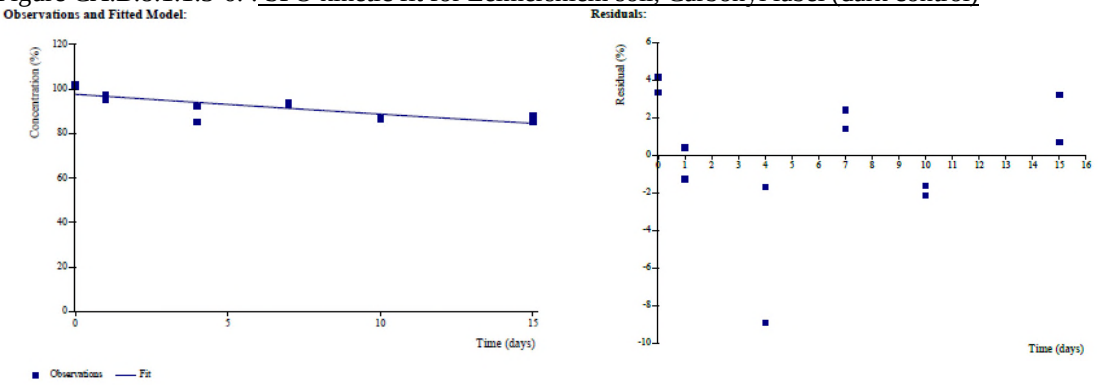


Figure CA.B.8.1.1.3-7: SFO kinetic fit Leimersheim soil, Phenyl label (irradiated)

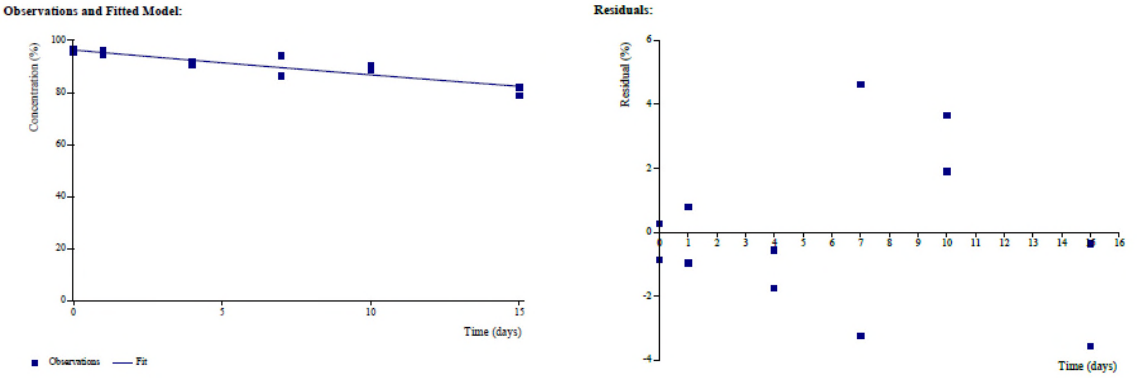
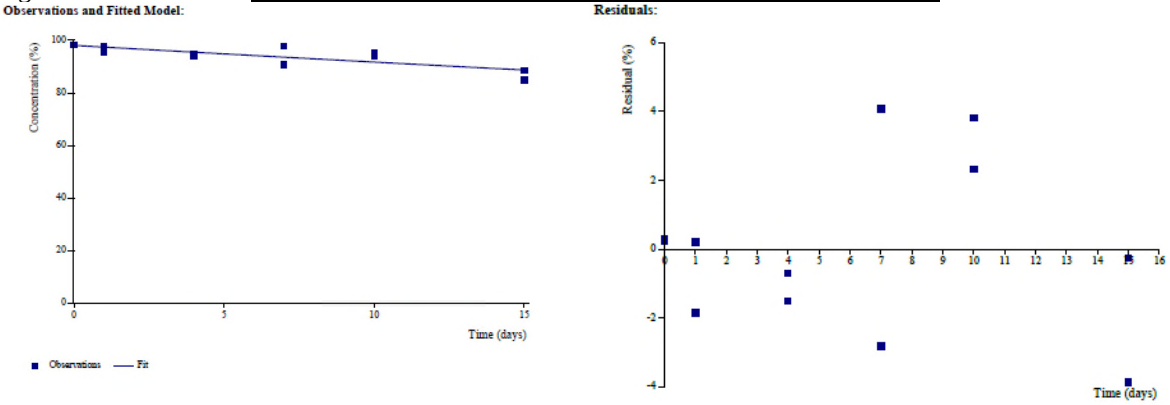


Figure CA.B.8.1.1.3-8: SFO kinetic fit for Leimersheim soil, Phenyl label (dark control)



CONCLUSION

[Carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone degraded slowly on soil surface under irradiated conditions (DT_{50} > 31 days of continuous degradation), with no degradates > 5% AR being observed in either irradiated or dark control samples. The largest degradate reached a maximum of 3.85 % AR in one replicate (mean of both replicates was 3.19% AR) of the irradiated samples after 15 days continuous irradiation and was tentatively identified as 2, 4-dichlorobenzoic acid. A number of minor degradates were also observed, none exceeding 3.57% of applied radioactivity. Degradation in the dark controls was slower over the incubation period, except for carbonyl labelled Leimersheim soil where it was almost identical. Therefore, soil photolysis is not expected to be a significant route of degradation in the field of bixlozone.

CA.B.8.1.1.4. *Rate of aerobic degradation*

CA.B.8.1.1.4.1. Rate of aerobic degradation of the active substance

Report:	KCA 7.1.1.1 Simmonds, R., (2015a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Route and Rate of Aerobic Degradation in Seven Soils at 20°C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/001; FMC Tracking no. 2013EFT-ISX1021
Guidelines:	OECD Guideline 307 (April 2002); US EPA OPPTS Guideline 835.4100 (October 2008)
Deviations from guideline:	None affecting the outcome of the study
GLP:	Yes (laboratory certified by UK National Authority)

Report:	KCA 7.1.2.1.1-02 Kong, L., (2017b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600: Normalisation of Laboratory DT50 for Temperature (20°C) and Moisture (pF 2.0)
Document No:	FMC Tracking no. 2017WHP-ISX3143
Guidelines:	FOCUS (2014)
Deviations	None
GLP:	No, modelling study

CA comments	Post the CA's evaluation, the applicant updated their kinetic assessment using the latest version of CAKE (v3.3). As a result, the applicant derived slightly different endpoints from their original submission. However, the CA's kinetic evaluation presented in the text below is still considered acceptable and so the applicant's revised modelling has not been considered further. **This study is relied upon.**

INTRODUCTION

This is the kinetic evaluation of the results of the Simmonds, R., 2015a laboratory bixlozone aerobic degradation study (section CA.B.8.1.1.1) conducted on 7 test soils.

The degradation rates were determined by the applicant for each soil with CAKE 2.0 software following FOCUS kinetic guidance. The CA has used Kingui v2 in order to run fitting for independent evaluation.

MATERIALS AND METHODS

All information on materials and methods are summarised at point section CA.B.8.1.1.1.

Data treatment and summary of endpoints

DT_{50} and DT_{90} values for the degradation of [^{14}C]-bixlozone in the seven soils tested were determined following FOCUS kinetics (2014)[1]. The replicates were fitted individually and were combined for the two radiolabels (i.e. 4 samples per time point); given the results seen were similar for the two radiolabels, this is considered acceptable. Initial mass (M0) was left as 'not fixed' in the kinetic fit. Initial mass (M0) was taken as total radioactivity (i.e. extracted parent and non-extracted radioactivity) multiplied by radiochemical purity (98.57 %). For the other time points the measured extracted parent value was used. The CA accepts this approach as a best estimate of the amount of parent substance dosed into the system in this case and that it was close to the measured value, given the lack of any metabolites and volatiles formed and the very low level of radioactivity

[1] FOCUS Kinetics, v1.1 (2014)30

present as non-extracted on day 0. Applicant calculations were performed using the numerical software package CAKE v 2.0, whereas the CA has used Kingui v2.0 in order to perform independent fitting. Data used for kinetic fitting by the CA is presented in Table CA.B.**8.1.1.4.1-1** with the data at 0 DAT corrected for radiochemical purity. A summary of the CA's validated rates of degradation of [^{14}C]-bixlozone in soil are presented in Table CA.B.**8.1.1.4.1-2**, with the full evaluation proceeding after.

Table CA.B.8.1.1.4.1-1: Data used by the CA for kinetic fitting

Soil	**Days After Treatment (DAT)**	**bixlozone (%AR)**			
		[phenyl-U-^{14}C]-bixlozone		**[carbonyl-^{14}C]-bixlozone**	
Lufa 6S	0	97.1	97.3	97.2	95.8
	7	94.9	94.1	92.8	93.0
	14	87.9	88.2	86.3	88.1
	30	80.0	81.8	78.1	81.3
	75	65.1	64.9	63.2	64.1
	120	50.4	53.0	52.5	49.7
Lufa 5M	0	96.8	96.5	98.9	97.0
	7	94.2	94.8	92.8	92.9
	14	88.9	88.4	86.9	88.4
	30	79.2	78.0	80.1	81.3
	75	65.5	64.8	60.5	60.9
	120	42.1	45.3	50.5	47.7
Lufa 2.2	0	97.1	97.3	97.3	96.7
	7	96.0	96.4	95.9	95.6
	14	90.8	93.7	94.3	92.8
	30	84.2	87.1	90.6	89.7
	75	81.2	81.8	83.2	80.3
	120	74.3	76.5	75.8	75.8
RefeSol 02-A	0	96.4	96.8	92.1	92.4
	7	88.0	86.6	77.1	80.8
	14	69.8	72.6	69.6	65.5
	30	96.2	97.6	91.0	90.8
	75	87.7	85.9	82.5	82.3
	120	71.3	73.3	66.7	65.9
CA-SL	0	96.8	96.4	97.0	97.3
	7	95.1	95.0	94.5	95.2
	14	91.8	87.1	94.0	91.5
	30	84.4	85.9	91.7	86.6
	75	70.9	67.9	70.2	70.7
	120	58.6	54.7	54.8	56.3
Iowa	0	96.7	96.3	96.6	96.5
	7	91.1	92.0	88.9	88.0
	14	81.5	80.1	83.5	82.7
	30	73.0	69.6	69.5	68.0
	75	43.0	41.6	45.7	44.0
	120	24.2	24.9	25.9	27.3
LAD-SCL-PF	0	96.2	96.1	95.9	101.5
	7	98.8	97.6	96.8	97.4
	14	93.6	96.0	95.4	95.3
	30	83.8	84.7	90.5	90.2
	75	75.5	75.5	73.4	72.7
	120	59.6	60.6	61.0	63.8

Table CA.B.8.1.1.4.1-2: Summary of degradation rate of [^{14}C]-bixlozone in soil under aerobic conditions

Soil	"Best-fit" kinetic model†	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	t-test	Visual fit
Lufa 6S	FOMC	136	868	0.8	n/a	Good
Lufa 5M	SFO	115	384	1.0	<0.001	Good
Lufa 2.2	FOMC	1000	>1000	0.6	n/a	Good
RefeSol 02-A	DFOP	358	>1000	1.0	0.0017 / 0.0067	Good
CA-SL	SFO	154	512	1.0	<0.001	Good
Iowa	SFO	64.1	213	1.2	<0.001	Good
LAD-SCL-PF	SFO	176	584	1.1	<0.001	Good
Soil	**"Modelling" Kinetic model**	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**t-test**	**Visual fit**
Lufa 6S	SFO	131	433	1.1	<0.001	Good
Lufa 5M	SFO	115	384	1.6	<0.001	Good
Lufa 2.2	SFO	330	>1000*	1.0	<0.001	Fair
RefeSol 02-A	SFO	225	749	2.6	<0.001	Good
CA-SL	SFO	154	512	1.0	<0.001	Good
Iowa	SFO	64.1	213	1.2	<0.001	Good
LAD-SCL-PF	SFO	176	584	1.1	<0.001	Good
Geometric mean (N=7)	**-**	**153**	**501**	**-**	**-**	**-**

*1000 days used for geomean calculation

CA FITTING FOR PERSISTENCE END POINTS

Lufa 6S CA fitting

Table CA.B.8.1.1.4.1-3: CA Kinetic fitting of data from bixlozone in Lufa 6S soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	130.23	432.62	1.104	k	5.32×10^{-3}	$<2 \times 10^{-16}$	-	-	Good
FOMC	**136.02**	**868.5**	**0.7994**	**α** **β**	**1.4402** **220.0444**	**-**	**2.424** **401.987**	**0.4562** **38.1014**	**Good**
DFOP	132.19	469.89	0.7843	k_1 k_2 g	0.05017 0.00477 0.0614	0.4582 0.0886 -	0.976 0.011 0.954	-0.8752 0.001913 -0.8313	Good

[a)]Best-fit model highlighted bold

The CA has selected the FOMC fitting as the best fit for Lufa 6S soil. This is due to the better statistical fit (lower chi^2 value) compared to SFO. FOMC fitting was then compared to DFOP, which did not improve the fit statistically due to the k_1 failing the t-test, alongside no visual improvement. Overall, the CA has selected FOMC due to the better statistical fit, although it is recognised that less reliance should be placed on this when degradation is extrapolated so far beyond study duration. It should be noted that any of the fits result in the same regulatory outcome (i.e. field dissipation studies triggered by DT_{50} >60d and DT_{90} >200d).

Figure CA.B.8.1.1.4.1-1: CA kinetic fitting and residuals for data from bixlozone in Lufa 6S soil. SFO fitting

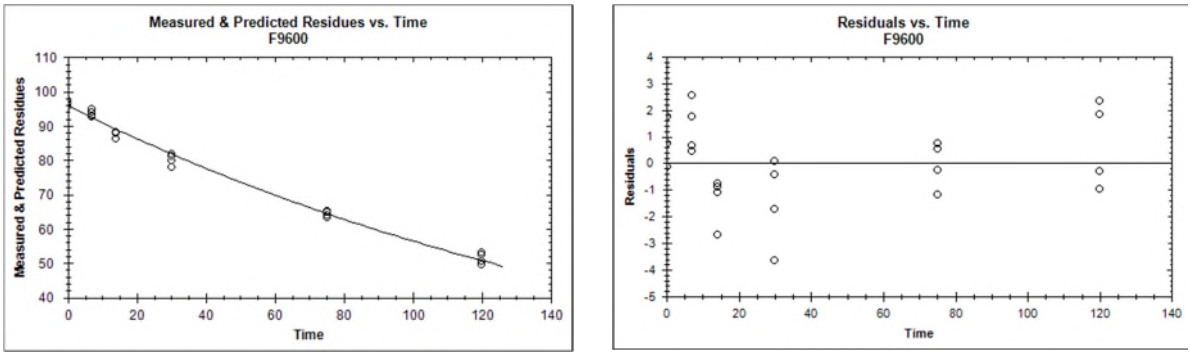


Figure CA.B.8.1.1.4.1-2: CA kinetic fitting and residuals for data from bixlozone in Lufa 6S soil. FOMC fitting

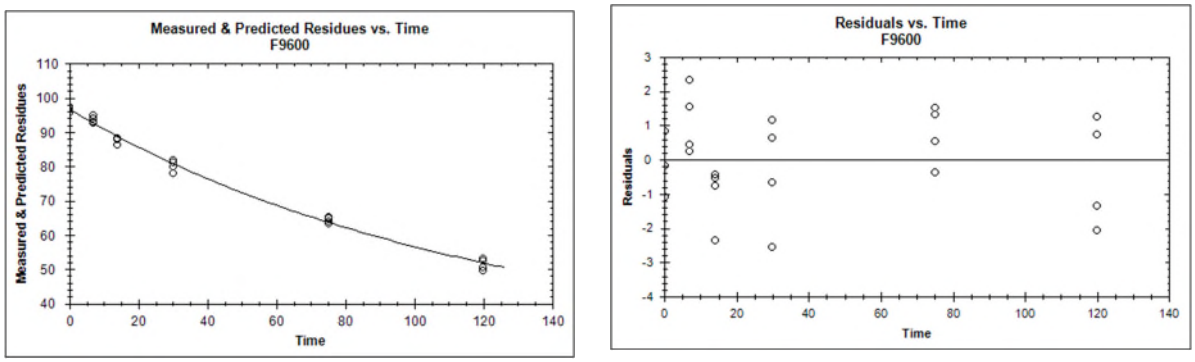
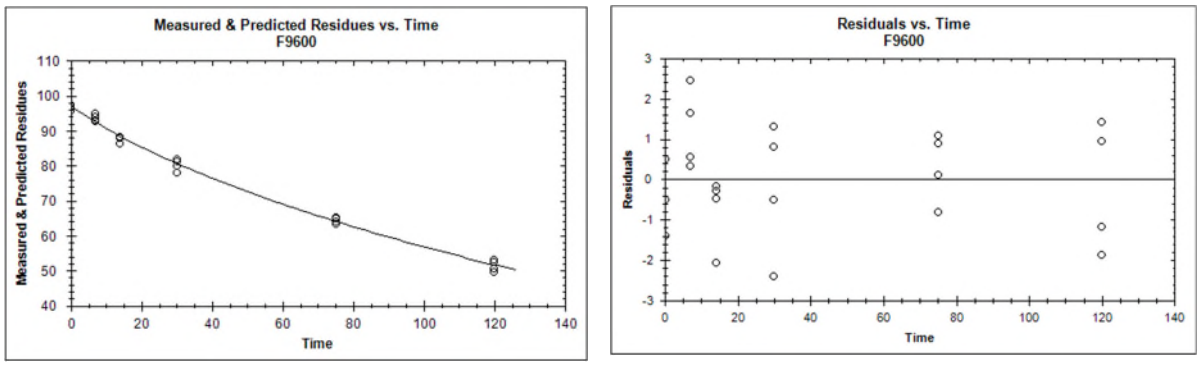


Figure CA.B.8.1.1.4.1-3: CA kinetic fitting and residuals for data from bixlozone in Lufa 6S soil. DFOP fitting



Lufa 5M CA fitting

Table CA.B.8.1.1.4.1-4: CA Kinetic fitting of data from bixlozone in Lufa 5M soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	**114.66**	**380.9**	**0.934**	**k**	**$6.045x10^{-3}$**	**$<2x10^{-16}$**	-	-	**Good**
FOMC	112.54	378.27	1.079	α β	68.73 $1.11x10^{4}$	-	642.38 104236	-504.9 $-8.2x10^{4}$	Good

[a)]Best-fit model highlighted bold

The CA has selected the SFO fitting as the best fit for Lufa 5M soil (noting that SFO and FOMC fits gave very similar results). This agrees with the applicant's choice of fit, however there are slight differences in the values calculated by the applicant and CA, with CA DT_{50} values being slightly lower.

Figure CA.B.8.1.1.4.1-4: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. SFO fitting

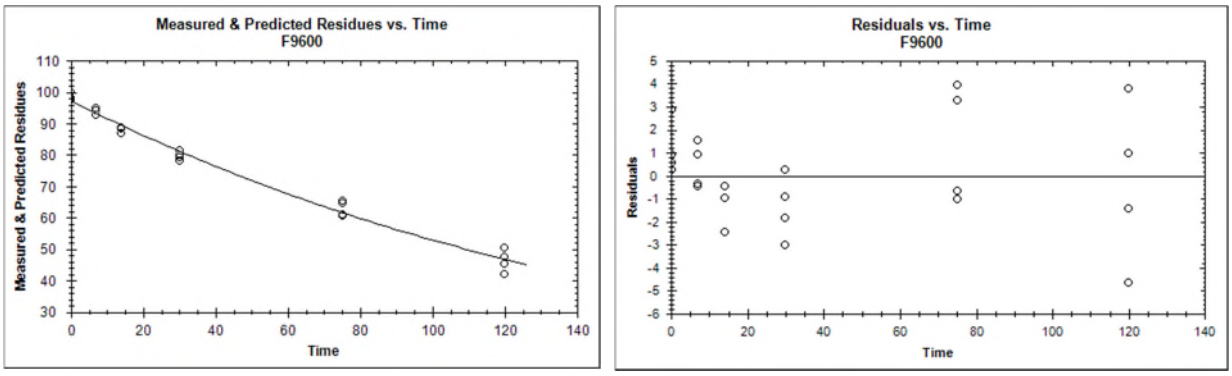
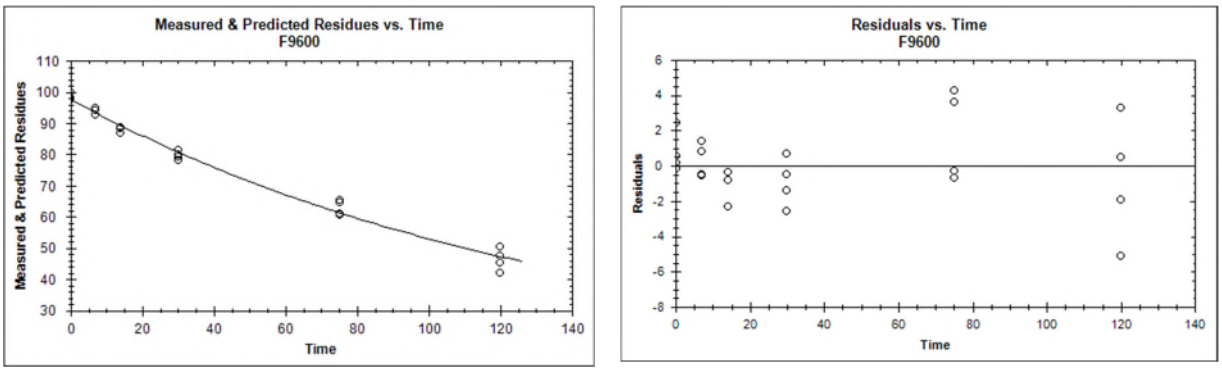


Figure CA.B8.1.1.4.1-5.: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. FOMC fitting



Lufa 2.2 CA fitting

Table CA.B.8.1.1.4-1: CA Kinetic fitting of data from bixlozone in Lufa 2.2 soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	328.63	1091.7	1.117	k	2.109×10^{-3}	1.45×10^{-15}	-	-	Fair
FOMC	**1000**	**>1000**	**0.6193**	**α** **β**	**0.2190** **55.7623**	**-**	**0.335** **104.306**	**0.1033** **7.2190**	**Good**
DFOP	432.38	>1000*	0.6679	k_1 k_2 g	0.0346528 1.401×10^{-3} 0.0836702	0.07846 0.0329 -	0.081 0.002 0.182	-0.0115268 4.956×10^{-4} -0.0145769	Good

[a)]Best-fit model highlighted bold

*DFOP result was not reported by Kingui, however, the CA ran the same data in CAKE and obtained a value of 1580 days

The CA has selected the FOMC fitting as the best fit for Lufa 2.2 soil. This is due to SFO fitting being visually and statistically worse than FOMC, and the fit not being improved with DFOP. DFOP fitting had a higher chi^2 value and no improvement on the visual fitting. The applicant selected DFOP, however, the CA notes the slightly higher chi^2. Overall, the CA has selected FOMC due to the better statistical fit, although it is recognised that less reliance should be placed on this when degradation is extrapolated so far beyond study duration. A DT_{50} value >1000 days was calculated by Kinguii, however, the CA has corrected it to 1000 days in line with the FOCUS guidance. It should be noted that any of the fits result in the same regulatory outcome (i.e. field dissipation studies triggered by DT_{50} >60d and DT_{90} >200d).

Figure CA.B.8.1.1.4.1-6: CA kinetic fitting and residuals for data from bixlozone in Lufa 2.2 soil. SFO Fitting

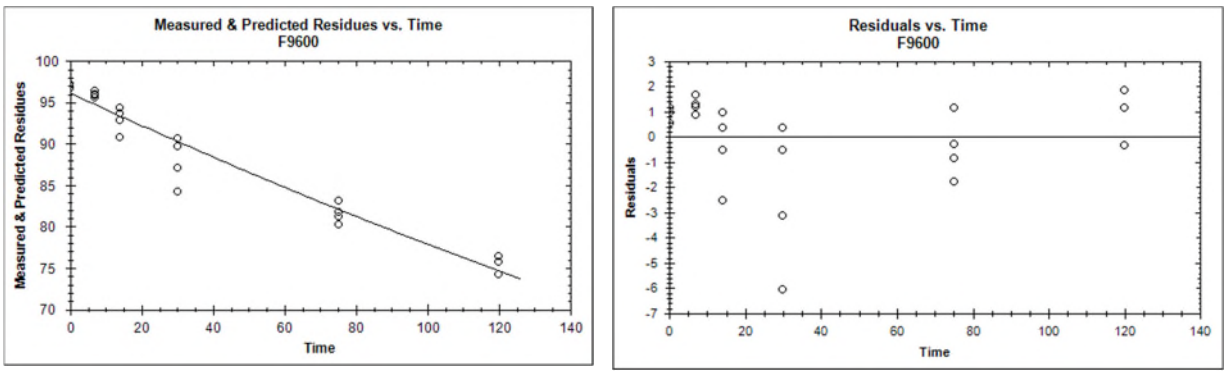


Figure CA.B.8.1.1.4.1-7: CA kinetic fitting and residuals for data from bixlozone in Lufa 2.2 soil. FOMC fitting

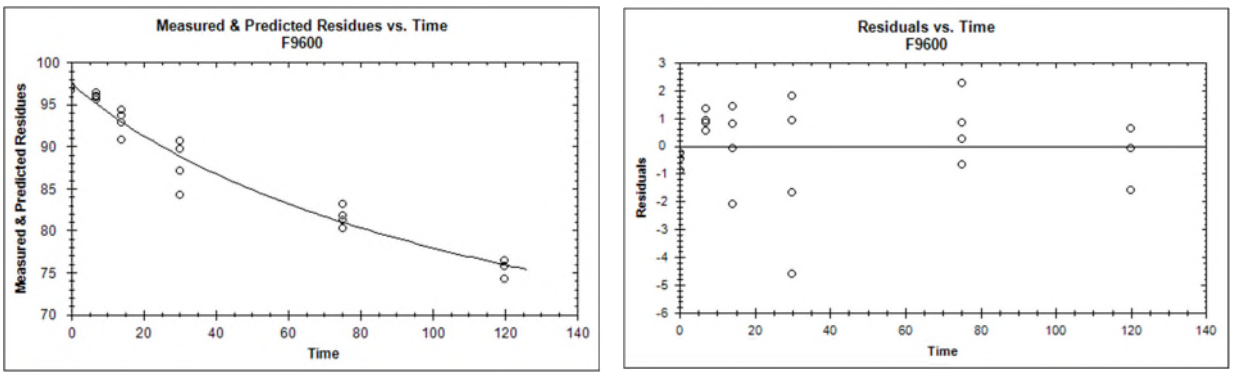
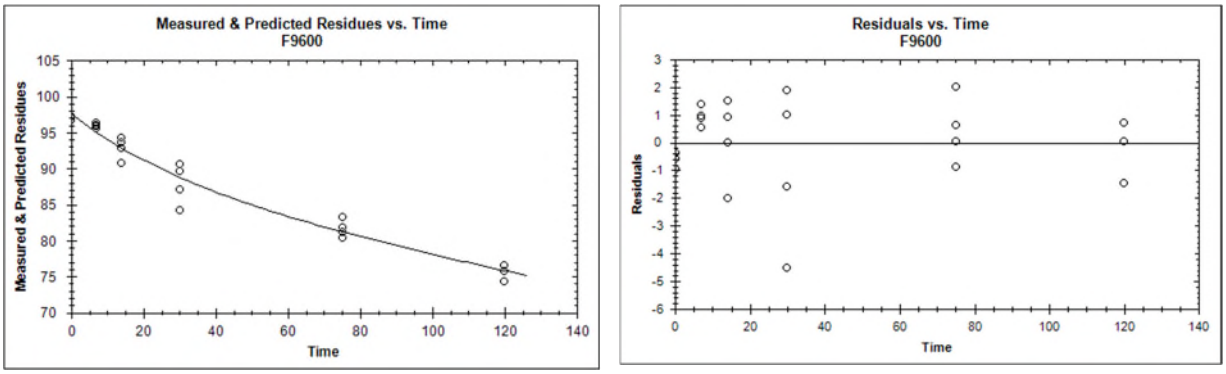


Figure CA.B.8.1.1.4.1-8: CA kinetic fitting and residuals for data from bixlozone in Lufa 2.2 soil. DFOP fitting



RefeSol 02-A CA fitting

Table CA.B.8.1.1.4.1-5: CA Kinetic fitting of data from bixlozone in RefeSol 02-A soil[a)]

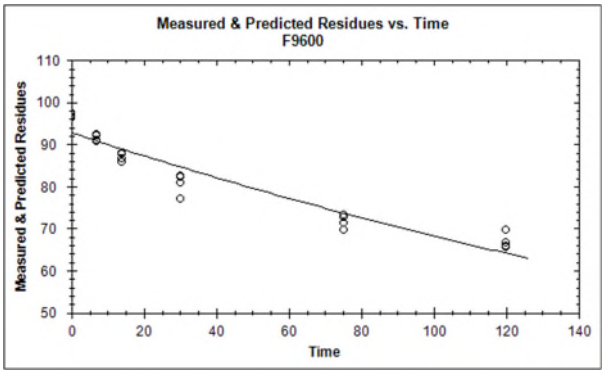
Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	224.92	747.16	2.684	k	3.082×10^{-3}	6.86×10^{-13}	-	-	Fair
FOMC	809.69	>1000	0.218	α β	0.179 17.22	-	0.21 23.93	0.1790 17.223	Good
DFOP	**361.37**	**>1000***	**0.1228**	**k_1** **k_2** **g**	**0.04164** **1.336×10^{-3}** **0.1897**	**0.00291** **0.00921** **-**	**0.068** **0.002** **0.281**	**0.0152** **3.156×10^{-4}** **0.001897**	**Good**

[a)]Best-fit model highlighted bold

*Kingui reported the DT_{90} value as '???', however, the CA ran the same data on CAKE and a DT_{90} of 1560 days was obtained.

The CA has selected the DFOP fitting as the best fit for Lufa 2.2 soil. This is due to SFO fitting being visually and statistically worse than FOMC, and marginally improved again when run with DFOP. DFOP fitting has a slightly better statistical fitting, however, there is no additional improvement on the visual fitting. The CA observes that there is very little difference between the FOMC and DFOP fits, both in terms of visual and statistical fit; for example the chi^2 values are both very low at 0.218 for FOMC and 0.1228 for DFOP. It should be noted that any of the fits result in the same regulatory outcome (i.e. field dissipation studies triggered by DT_{50} >60d and DT_{90} >200d). This agrees with the applicant's selection, however there are minor differences in DT_{50} value.

Figure CA.B.8.1.1.4.1-9: CA kinetic fitting and residuals for data from bixlozone in RefeSol 02-A soil. SFO fitting



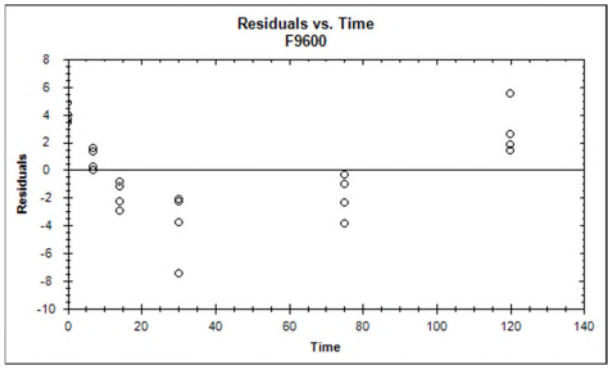
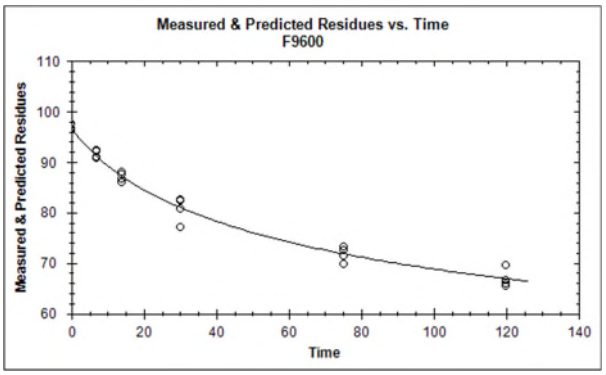


Figure CA.B.8.1.1.4.1-10: CA kinetic fitting and residuals for data from bixlozone in RefeSol 02-A soil. FOMC fitting



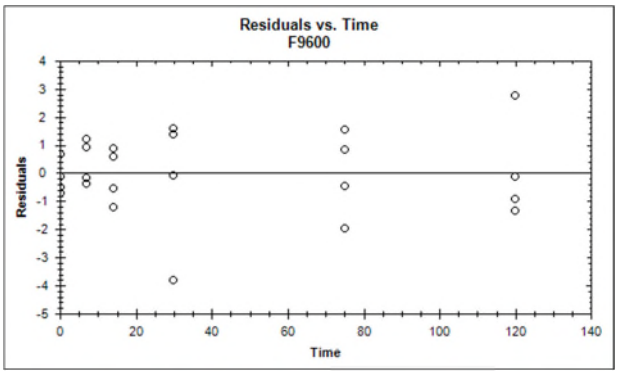
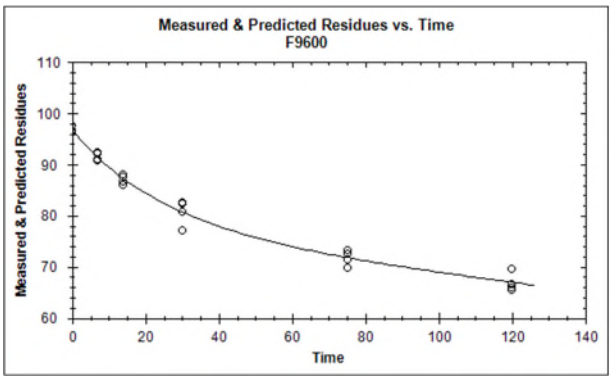
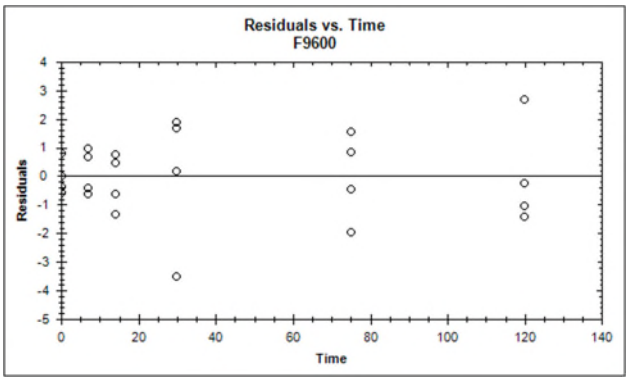


Figure CA.B.8.1.1.4.1-11: CA kinetic fitting and residuals for data from bixlozone in RefeSol 02-A soil. DFOP fitting





CA-SL CA fitting

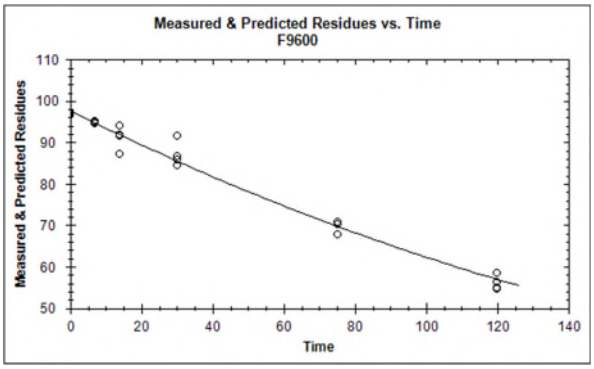
Table CA.B.8.1.1.4.1-6: CA Kinetic fitting of data from bixlozone in CA-SL soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	**153.5**	**509.91**	**0.885**	**k**	**$4.52x10^{-3}$**	**$<2x10^{-16}$**	**-**	**-**	**Good**
FOMC	148.75	497.81	1.083	α β	1.09 $2.331x10^{4}$	-	462.51 99123	-244 $-5.25x10^{4}$	Good

[a)]Best-fit model highlighted bold

Both the SFO and FOMC models give a similar visual fit to the data. The CA has selected the SFO fitting as the best fit for CA-SL soil. This is due to SFO fitting being statistically better than FOMC, with a lower chi^2 and p values >0.05. This agrees with the applicant's selection, although there are very minor differences in DT_{50} and DT_{90} values calculated by the CA and the applicant. The lower confidence interval being <0 is also noted for both the α and β parameters of the FOMC model. However, it is recognised that slow degradation can lead to poorer statistical fitting, and it is noted that either fitting produces the same regulatory outcome.

Figure CA.B.8.1.1.4.1-12: CA kinetic fitting and residuals for data from bixlozone in CA-SL soil. SFO fitting



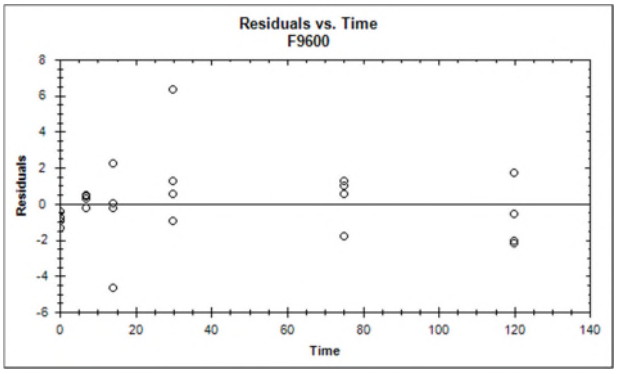
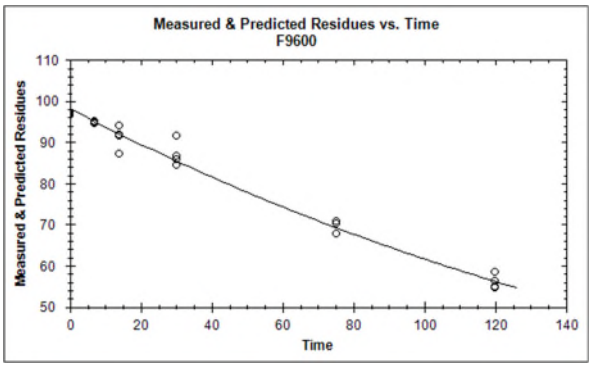
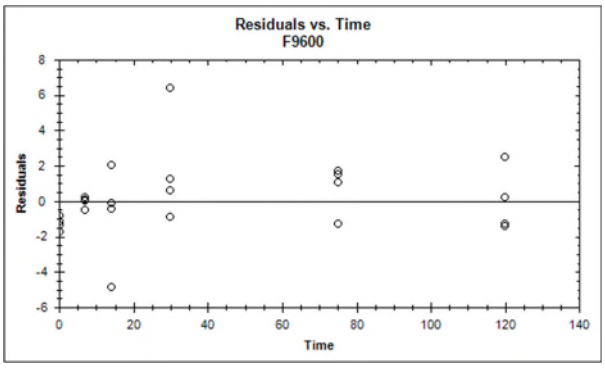


Figure CA.B.8.1.1.4.1-13: CA kinetic fitting and residuals for data from bixlozone in CA-SL soil. FOMC fitting





Iowa CA fitting

Table CA.B.8.1.1.4.1-7: CA Kinetic fitting of data from bixlozone in Iowa soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**63.95**	**212.44**	**0.744**	**k**	**0.01084**	**$2x10^{-16}$**	**-**	**-**	**Good**
FOMC	59.645	199.21	1.866	α β	149.5 $1.284x10^4$	0.279 0.280	641.55 55231.57	-342.5 $-2.95x10^4$	Good

[a)]Best-fit model highlighted bold

The CA has selected the SFO fitting as the best fit for Iowa soil. This is due to SFO fitting being statistically better than FOMC, with a lower chi^2 and p values >0.05. This agrees with the applicant's selection, however there are very minor differences between the DT_{50} and DT_{90} value calculated by the CA and the applicant and therefore CA data is presented. It is recognised that slow degradation can lead to poorer statistical fitting which may have resulted in the negative lower confidence intervals observed for the FOMC model, and it is noted that either fitting produces the same regulatory outcome. The FOMC model however marginally underpredicts degradation from day 30 onwards and consequently the residuals for the SFO model show more even scatter either side of the 0 line than the FOMC model. Consequently, based on the visual fit and the better statistical indicators, the CA accepted the SFO model as the best fit model.

Figure CA.B.8.1.1.4.1-14: CA kinetic fitting and residuals for data from bixlozone in Iowa soil. SFO fitting

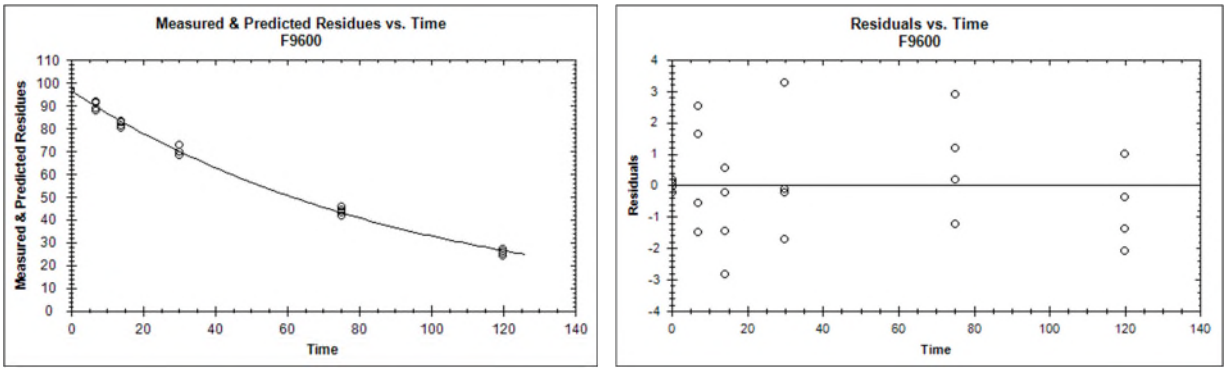
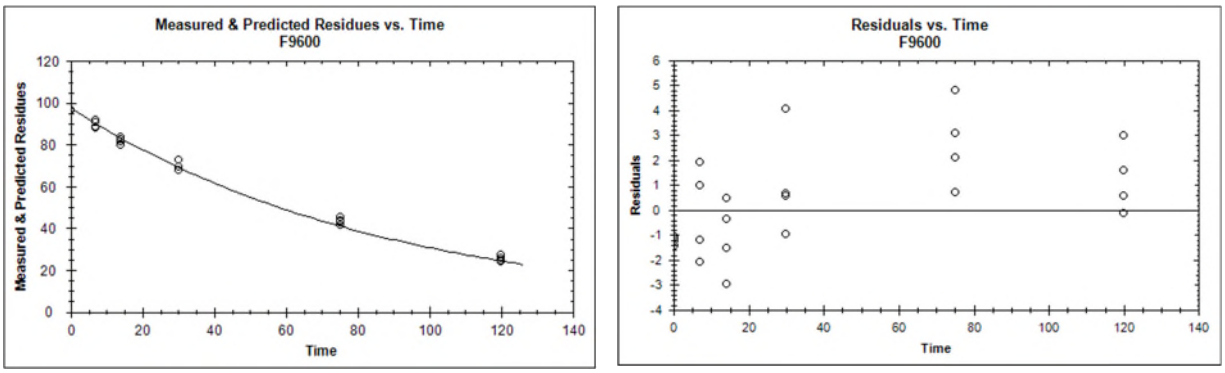


Figure CA.B8.1.1.4.1-15.: CA kinetic fitting and residuals for data from bixlozone in Iowa soil. FOMC fitting



LAD-SCL-PF CA fitting

Table CA.B.8.1.1.4.1-8: CA Kinetic fitting of data from bixlozone in LAD-SCL-PF soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**175.32**	**582.39**	**1.013**	**k**	**$3.954x10^{-3}$**	**$<2x10^{-16}$**	**-**	**-**	**Good**
FOMC	166.55	558.18	1.308	α β	91.13 $2.181x10^{4}$	-	531.5 127502.7	-0.03492 $-8.388x10^{4}$	Good

[a)]Best-fit model highlighted bold

The CA has selected the SFO fitting as the best fit for LAD-SCL-PF soil. This is due to SFO fitting being statistically better than FOMC, with a lower chi^2 and p value <0.05. This agrees with the applicant's selection, however there are very minor differences between the DT_{50} and DT_{90} value calculated by the CA and the applicant and therefore CA data is presented. It is recognised that slow degradation can lead to poorer statistical fitting which may have resulted in the negative lower confidence intervals observed for the FOMC model, and it is noted that either fitting produces the same regulatory outcome. The FOMC model however very marginally underpredicts degradation from day 30 onwards and consequently the residuals for the SFO model show more even scatter either side of the 0 line than the FOMC model. Consequently, based on the visual fit and the better statistical indicators, the CA accepted the SFO model as the best fit model.

Figure CA.B.8.1.1.4.1-16: CA kinetic fitting and residuals for data from bixlozone in LAD-SCL-PF. SFO fitting

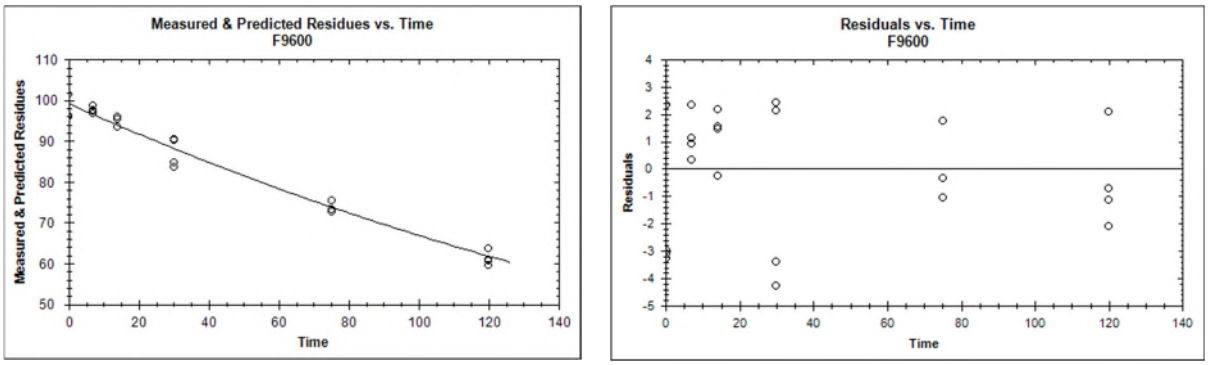
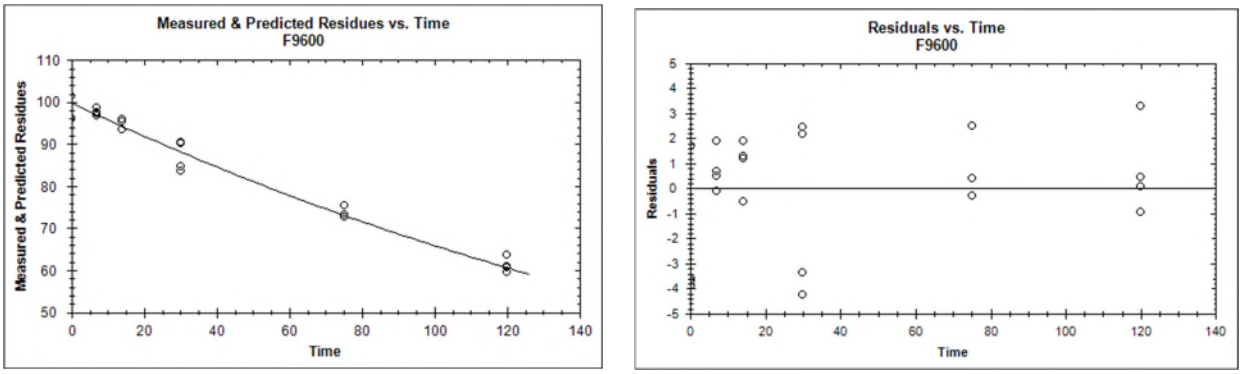


Figure CA.B.8.1.1.4.1-17: CA kinetic fitting and residuals for data from bixlozone in LAD-SCL-PF. FOMC fitting



The CA agrees with applicant fitting for Lufa 5M, RefeSol 02-A, CA-SL, Iowa, and LAD-SCL-PF soils, as the accepted fittings used the same model and there were only minor differences between DT_{50} values. For Lufa 6S and Lufa 2.2, the CA disagrees with the applicant. Therefore, in Table CA.B.**8.1.1.4.1-9**, accepted applicant values for soils Lufa 5M, RefeSol 02-A, CA-SL, Iowa and LAD-SCL-PF, and CA values for Lufa 6S and Lufa 2.2 are given as final persistence fittings for bixlozone. However, it is recognised that slow degradation can lead to more uncertain statistical fitting, and it is noted that in situations of marginal difference between kinetic models, the same regulatory outcome is reached.

Table CA.B.8.1.1.4.1-9: CA final persistence fits for aerobic laboratory degradation of bixlozone

Soil	Selected kinetic model	DT_{50} (days)	DT_{90} (days)	χ^2	t-test	Visual fit	Applicant fitting accepted?
Lufa 6S	FOMC	136	869	0.8	-	Good	N
Lufa 5M	SFO	115	384	1.0	<0.001	Good	Y
Lufa 2.2	FOMC	1000	>1000	0.6	-	Good	N
RefeSol 02-A	DFOP	358	>1000	1.0	0.0017/0.0067	Good	Y
CA-SL	SFO	154	512	1.0	<0.001	Good	Y
Iowa	SFO	64.1	213	1.2	<0.001	Good	Y
LAD-SCL-PF	SFO	176	584	1.1	<0.001	Good	Y

As the DT_{50} and DT_{90} are >60 days and >200 days respectively, this triggers requirement for field studies. This conclusion is reached regardless of the kinetic models selected for those soils in which the discussion above has indicated that the selection of kinetic model is a marginal decision.

CA fitting for modelling endpoints

All the SFO fitting from the persistence sections are acceptable for use in modelling according to the criteria in FOCUS kinetics guidance. CA and applicant values are very similar and therefore the CA has accepted the applicant's fits for modelling endpoints. The SFO fittings presented in the persistence section above. A summary of the accepted endpoints is given below.

DT_{50} values were also normalised to the reference soil moisture (pF 2.0) and temperature (20°C) by the applicant, following the "Generic Guidance for Tier 1 FOCUS Ground Water Assessments" (FOCUS, 2014) using a moisture exponent B of 0.7. The normalised rates of degradation of [^{14}C]-bixlozone in soil are summarised in Table CA.B.**8.1.1.4.1-10**.

Table CA.B.8.1.1.4.1-10: Degradation rate of [^{14}C]-bixlozone in soil under aerobic conditions normalised to 20°C and pF 2

bixlozone, Laboratory studies, aerobic conditions										
Soil name	**Soil type**	**pH $CaCl_2$**	**temp. °C**	**Soil moisture [% w/w]**		**DT_{50} (d)**	**DT_{90} (d)**	**DT_{50} (d) 20°C pF2/10kPa**	**χ^2 (%)**	**Kinetic model**
				study	**reference**					
Lufa 6S	Clay	6.9	20	26.5	31	131	433	117.4	1.1	SFO
Lufa 5M	Sandy loam	7.2	20	18.0	20.8	115	384	103.8	1.6	SFO
Lufa 2.2	Loamy sand	5.4	20	11.5	11.3	330	>1000	330	1.0	SFO
RefeSol 02-A	Silt loam	6.1	20	27.8	37	225	749	184.3	2.6	SFO
CA-SL	Loamy sand	6.9	20	11.5	13.4	154	512	138.3	1.0	SFO
Iowa	Silt loam	6.8	20	31.8	42.3	64.1	213	52.5	1.2	SFO
LAD-SCL-PF	Clay	8.0	20	29.7	40.9	176	584	140.7	1.1	SFO
Geometric mean (n=7)								134		
pH-dependency: y/n								N (see section CA.B.8.1.4)		

The worst case non-normalised laboratory best-fit DT_{50} for bixlozone was 1000 days. The normalised DT_{50} values ranged from 52.5 to 330 days with a geometric mean of 133.5 days.

CONCLUSIONS

In four European and three US soils, [^{14}C]-bixlozone degraded with best-fit DT_{50} values derived from a max of SFO, FOMC and DFOP kinetics in the range 64.1 days to >1000 days and normalised SFO DT_{50} values for use in exposure modelling in the range 52.5 to 330 days (geomean value of 133.5 days).

CA.B.8.1.1.4.2. Rate of aerobic 3-OH propanamide degradation

Report:	KCA 7.1.2.1.2-01 Göcer, M., (2016a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600-3-OH-Propanamide Aerobic Degradation in Three Soils at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01058, FMC Tracking no. 2016EFT-ISX2465
Guidelines:	FOCUS (2014) OECD Guideline 307 (April 2002); OPPTS Guideline 835.4200 (October 2008) SANCO/3029/99 rev.4
GLP:	Yes (laboratory certified by German National Authority)

CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

This is the kinetic evaluation of the results of the Göcer, M., 2016a laboratory 3-OH aerobic degradation study (section CA.B.8.1.1.1) conducted on 3 test soils.

The degradation rates were determined by the applicant for each soil with CAKE 2.0 software following FOCUS kinetic guidance. The CA has used Kingui v2 in order to run fitting for independent evaluation.

MATERIALS AND METHODS

All information on materials and methods are summarised at point section CA.B.8.1.1.1.2.

Data treatment and summary of endpoints

DT_{50} and DT_{90} values for the degradation of bixlozone-3-OH-propanamide in the three soils tested were determined following the recommendations of the FOCUS work group on degradation kinetics. Both replicates were included in the dataset plotted as separate data points and the initial value for mass was also allowed to be estimated by the model. All calculations were performed by the applicant using the numerical software package CAKE 3.2, which the CA has checked using kingui v2. It is noted the applicant used the raw data in mg/kg in their kinetic assessment whereas the CA used the percentages summarised in Table CA.B.**8.1.1.4.2-1**.

Table CA.B.8.1.1.4.2-1: Data used for kinetic fitting

	% Applied test substance		
Time (hours)	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
0	102.6	103.9	99.8
0	103.7	100.7	98.4
2	91.7	91.9	87.0
2	93.9	89.8	87.7
4	70.2	79.3	77.1
4	82.2	77.4	79.1
6	72.3	71.2	73.2
6	71.2	78.6	79.3
12	48.4	36.8	38.9
12	47.5	35.8	40.2
24	22.4	16.1	16.3
24	37.1	17.6	15.8
48	(4.4)	(3.6)	(4.7)
48	7.1	8.1	(4.9)

Values in brackets are <LOQ (5%); the measured values were used in the kinetic assessment

Lufa 2.1 soil fitting

The applicant ran all four kinetic models and concluded HS as the best-fit model, with SFO considered good enough for modelling endpoints. Following the FOCUS guidance, the CA ran SFO and FOMC initially. However, the CA does not consider the FOMC model to improve the visual and statistical fit of the data. Therefore, the CA considers the SFO fit to provide the best-fit model (for both persistence and modelling endpoints). As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil; the CA has also included the applicant's HS fit for information but this has not been validated by the CA and is not considered further.

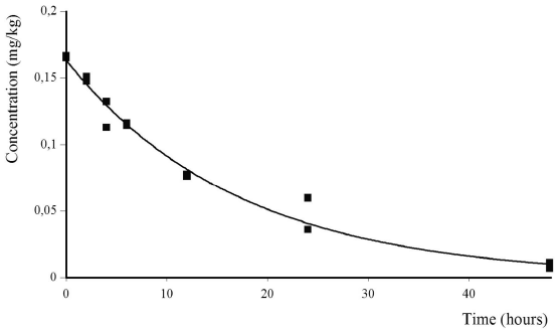
Table CA.B.8.1.1.4.2-2: Applicant's kinetic fitting of data from bixlozone-3-OH-Propanamide in Lufa 2.1 soil[a)]

Model	**DT_{50} (hours)**	**DT_{90} (hours)**	**χ^2 (%)**	**Estimated parameters**		**t-test**	**Upper 95% Confidence Interval**	**Lower 95% Confidence Interval**	**Visual fit**
SFO	**12.0**	**39.7**	**3.64**	**k**	**0.058**	**4.66E-9**	**-**	**-**	**Good**
FOMC	11.4	43.6	3.56	α β	5.745 88.49	-	20.12 338.4	-8.628 -156.8	Good
HS	11.6	41.8	3.43	k1 k2 tb	0.072 0.053 4.035	4.59E-4 4.06E-6 -	0.107 0.067 -	0.038 0.039 -	Good

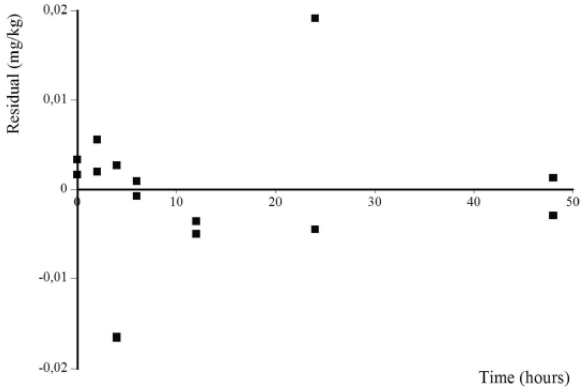
[a)]Best-fit model highlighted bold

Figure CA.B.8.1.1.4.2-1: Applicant's Lufa 2.1 3-OH kinetic fits

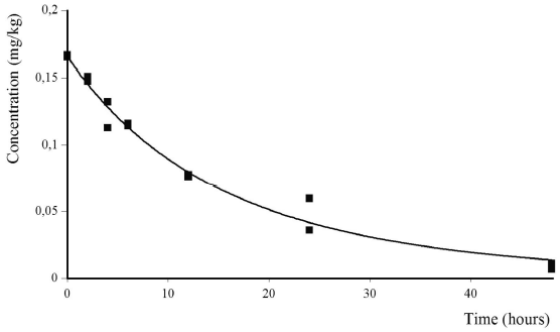
SFO



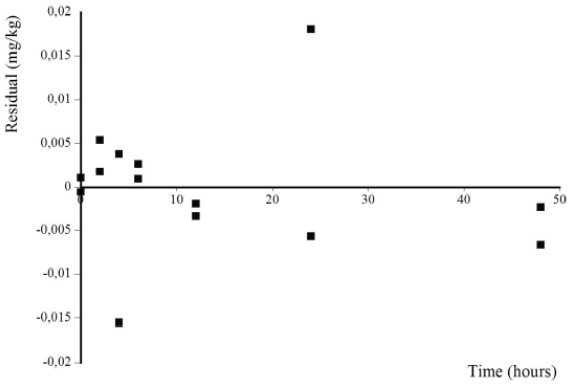
SFO



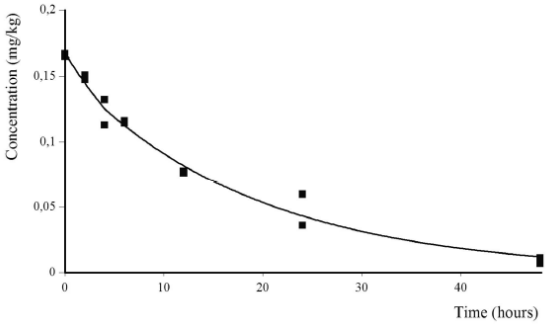
FOMC



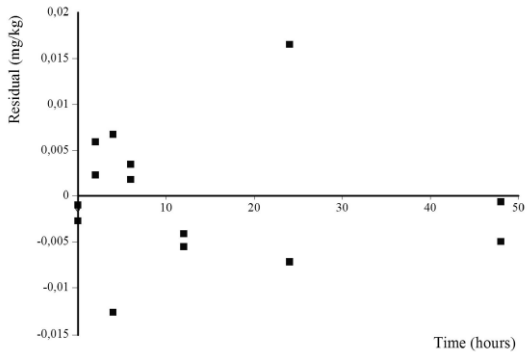
FOMC



HS



HS



Lufa 2.4 soil fitting

The applicant ran all four kinetic models and concluded SFO as the best-fit model. Following the FOCUS guidance, the CA ran SFO and FOMC initially. The CA agrees the FOMC model does not improve the visual and statistical fit of the data and so concurs SFO is the best-fit model. As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil.

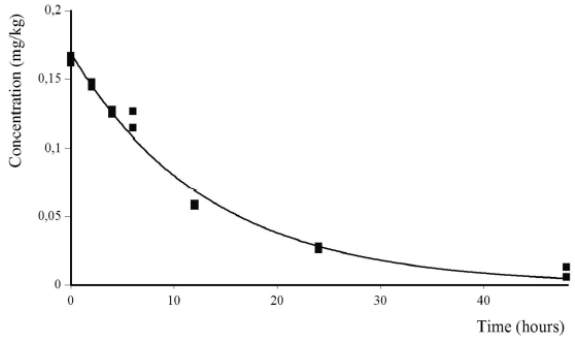
Table CA.B.8.1.1.4.2-3: Applicant's kinetic fitting of data from 3-OH in Lufa 2.4 soil[a)]

Model	DT_{50} (hours)	DT_{90} (hours)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**9.30**	**30.9**	**5.73**	**k**	**0.0746**	**2.96E-9**	**-**	**-**	**Good**
FOMC	7.58	25.6	6.22	α β	35.04 466.8	-	131 1.61E3	-23.26 -442.2	Good

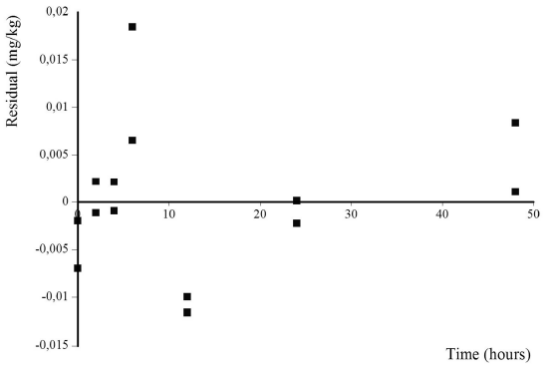
[a)]Best-fit model highlighted bold

Figure CA.B.8.1.1.4.2-2: Applicant's Lufa 2.4 3-OH kinetic fits

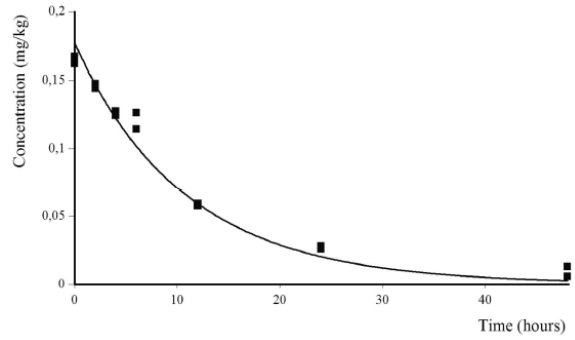
SFO



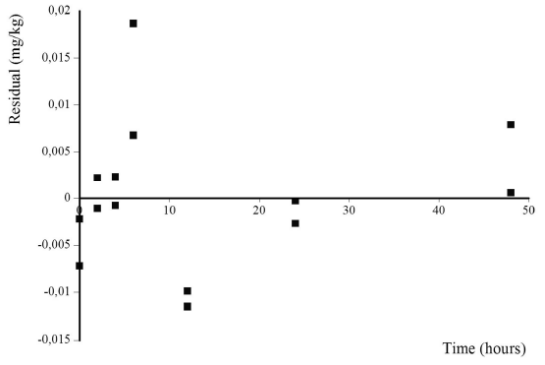
SFO



FOMC



FOMC



St. Bauzille 12-060 soil fitting

The applicant ran all four kinetic models and concluded HS as the best-fit model, with SFO good enough for modelling endpoints. Following the FOCUS guidance, the CA ran SFO and FOMC initially. However, the CA does not consider the FOMC model to improve the visual and statistical fit of the data. Therefore, the CA considers the SFO fit to provide the best-fit model (for both persistence and modelling endpoints). As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil; the CA has also included the applicant's HS fit for information but this has not been validated by the CA and is not considered further.

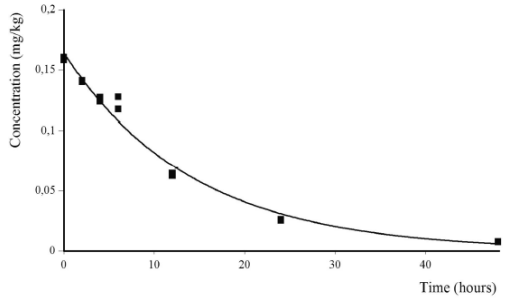
Table CA.B.8.1.1.4.2-4: Applicant's kinetic fitting of data from 3-OH in st. Bauzille 12-060 soil[a)]

Model	DT_{50} (hours)	DT_{90} (hours)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**9.96**	**33.1**	**5.89**	**k**	**0.070**	**3.00E-9**	**-**	**-**	**Good**
FOMC	7.91	26.5	6.41	α β	110.1 1.25E+3	-	161 1.93E+3	59.22 577.3	Good
HS	10.4	28.2	4.02	k1 k2 tb	0.050 0.090 6.046	9.29E-6 1.88E-5 -	0.065 0.119 -	0.036 0.061 -	Good

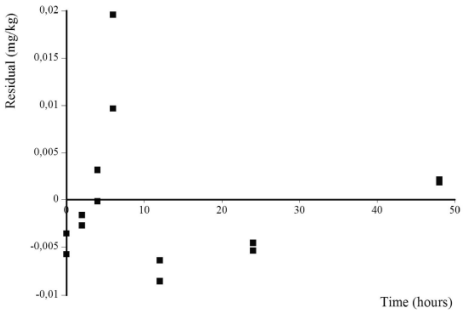
[a)]Best-fit model highlighted bold

Figure CA.B.8.1.1.4.2-3: Applicant's st. Bauzille 12-060 3-OH kinetic fits

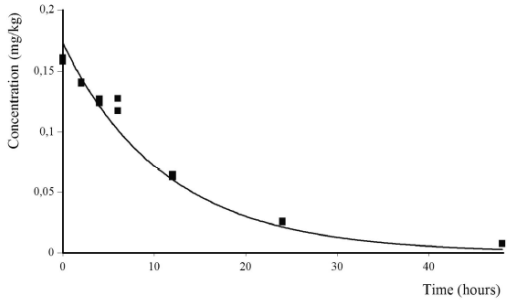
SFO



SFO



FOMC



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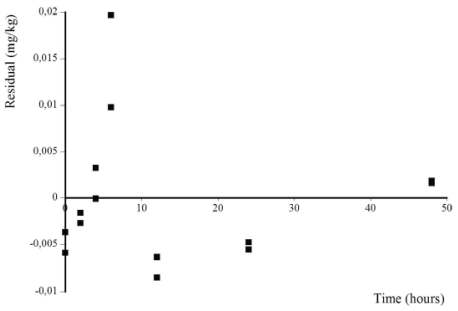
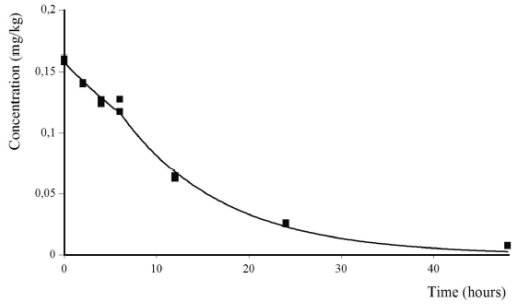
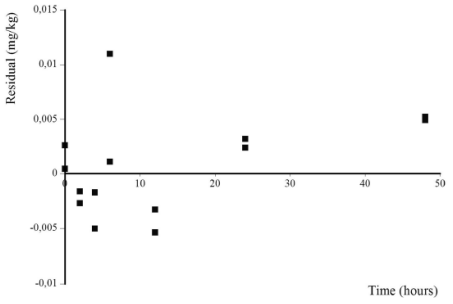


HS



HS



Rates of degradation of bixlozone-3-OH-propanamide in soil are summarised in Table CA.B.**8.1.1.4.2-5**. It is noted by the CA that all of the DT_{50} values are <0.5 days for each soil and so field soil dissipation studies are not triggered for this compound.

Table CA.B.8.1.1.4.2-5: Degradation rate of bixlozone-3-OH-propanamide in soil under aerobic conditions

Soil	Kinetic model†	DT_{50} (hours)	DT_{90} (hours)	χ^2 (%)	t-test	Visual fit
Lufa 2.1	SFO	12.0	39.7	3.64	< 0.001	Good
Lufa 2.4	SFO	9.3	30.9	5.73	< 0.001	Good
St. Bauzille 12-060	SFO	9.96	33.1	5.89	< 0.001	Good

† SFO = single first order

Using the data provided in the report the results were normalised to pF 2 and 20 °C using approaches outlined in the FOCUS Ground Water guidance[2], as shown in Table CA.B.**8.1.1.4.2-6**.

Table CA.B.8.1.1.4.2-6: Degradation rate of bixlozone-3-OH-propanamide in soil under aerobic conditions normalized to 20°C and pF 2.

bixlozone-3-OH-propanamide, Laboratory studies, aerobic conditions										
Soil name	Soil type	pH $CaCl_2$	temp. °C	Soil moisture [% w/w]		DT_{50} (h)	DT_{90} (h)	DT_{50} (h) 20°C pF2/10kPa	χ^2 (%)	Kinetic model
				study	reference					
Lufa 2.1	Loamy sand	4.84	20	15.8	14	12.0	39.7	12.0	3.64	SFO
Lufa 2.4	Loam	7.41	20	24.6	25	9.3	30.9	9.2	5.73	SFO
St. Bauzille 12-060	Silty clay	7.53	20	23.2	40	10.0	33.1	6.8	5.89	SFO
Geometric mean (n = 3)								9.1		

CONCLUSIONS

In three European soils incubated at 20ºC and 50% MWHC, non-labelled bixlozone-3-OH-propanamide degraded with normalised SFO DT_{50} values in the range 6.8 to 12.0 hours (geomean value = 9.1 hours).

[2] Generic Guidance for Tier 1 FOCUS Ground Water Assessments, v2.2, May 2014

CA.B.8.1.1.4.3. Rate of aerobic 2,4-dichlorobenzoic acid degradation

Report:	KCA 7.1.2.1.2-02 Göcer, M., (2016b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	2,4-Dichlorobenzoic Acid Aerobic Degradation in Three Soils at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01059, FMC Tracking no. 2016EFT-ISX2468
Guidelines:	FOCUS (2014) OECD Guideline 307 (April 2002); OPPTS Guideline 835.4200 (October 2008) SANCO/3029/99 rev.4
GLP:	Yes (laboratory certified by German National Authority)

CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

This is the kinetic evaluation of the results of the Göcer, M., 2016b laboratory 2,4-DBA aerobic degradation study (section CA.B.8.1.1.1.3) conducted on 3 test soils.

The degradation rates were determined by the applicant for each soil with CAKE 2.0 software following FOCUS kinetic guidance. The CA has used Kingui v2 in order to run fitting for independent evaluation.

MATERIALS AND METHODS

All information on materials and methods are summarised at point section CA.B.8.1.1.1.3.

Data treatment and summary of endpoints

DT_{50} and DT_{90} values for the degradation of 2,4-DBA in the three soils tested were determined following the recommendations of the FOCUS work group on degradation kinetics. Both replicates were included in the dataset plotted as separate data points and the initial value for mass was also allowed to be estimated by the model. All calculations were performed by the applicant using the numerical software package CAKE 3.2, which the CA has checked using Kingui v2. It is noted the applicant used the raw data in mg/kg in their kinetic assessment whereas the CA used the percentages summarised in Table CA.B.**8.1.1.4.3-1**.

Table CA.B.8.1.1.4.3-1: Data used for kinetic fitting

	% Applied test substance		
Time (days)	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
0	95.4	95.2	92.9
0	95.7	94.5	96.0
0.25	97.3	99.8	100.1
0.25	96.3	101.2	100.6
1	89.8	95.6	92.9
1	92.6	96.2	94.4
2	81.9	69.1	74.8
2	78.2	68.7	73.0
4	58.1	39.3	49.4
4	57.1	46.6	61.1
7	47.4	20.6	46.4
7	47.7	27.5	54.8

	% Applied test substance		
Time (days)	**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**
15	38.7	11.0	29.5
15	45.3	(2.5)	34.4
30	5.9	[0.5]	(2.6)
30	11.2	[0.5]	14.5
60	(4.8)	n.d.	5.5
60	5.2	n.d.	5.4

Values in brackets are <LOQ (5%); the measured values were used in the kinetic assessment. The values in square-brackets were <LOD (1%) and so values equivalent to ½ LOD (0.5%) have been used in the kinetic assessment.
n.d. = not determined

Lufa 2.1 soil fitting

The applicant ran all four kinetic models and concluded HS as the best-fit model, with SFO good enough for modelling endpoints. Following the FOCUS guidance, the CA ran SFO and FOMC initially. The CA agrees with the applicant that the FOMC model provided the slightly better visual fit (and lower χ^2) value and so proceeded to run DFOP and HS fits. The CA notes the resulting visual fits of the three biphasic models were very similar and calculated very similar DT50 values. Because the HS model resulted in the lowest lower χ^2 value and the t-test was passed for both rate constants, the CA accepts the applicant's conclusion that HS provided the best-fit model. The CA also agrees that SFO was good enough for modelling endpoints. As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil.

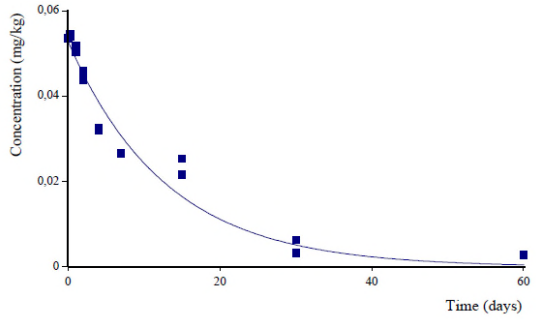
Table CA.B.8.1.1.4.3-2: Applicant's kinetic fitting of data from 2,4-dichlorobenzoic acid in Lufa 2.1 soil[a)]

Model	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**Estimated parameters**		**t-test**	**Upper Confidence Interval (95%)**	**Lower Confidence Interval (95%)**	**Visual fit**
SFO	8.94	29.7	9.11	k	0.0776	3.63E-8	-	-	Good
FOMC	7.35	43.7	8.11	α β	1.593 13.47	-	3.012 30.25	0.173 -3.308	Good
DFOP	7.37	38.6	7.79	k1 k2 g	0.377 0.050 0.303	0.096 0.001 -	0.968 0.080 -	-0.214 0.021 -	Good
HS	**7.52**	**38.7**	**6.80**	**k_1** **k_2** **tb**	**0.124** **0.052** **4.219**	**1.13E-6** **3.36E-5** **-**	**0.159** **0.071** **-**	**0.089** **0.032** **-**	**Good**

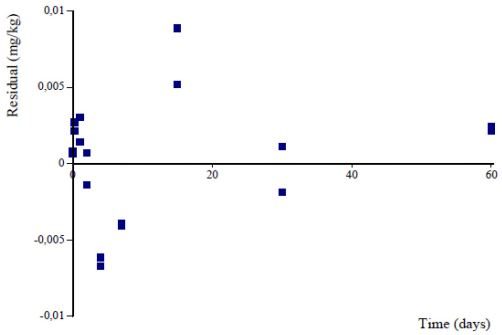
[a)]Best-fit model highlighted bold

Figure CA.B.: Applicant's Lufa 2.1 2,4-DBA kinetic fits

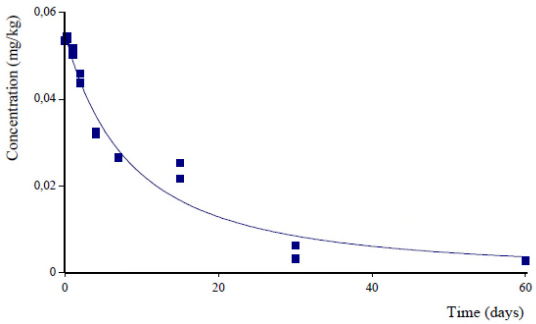
SFO



SFO



FOMC



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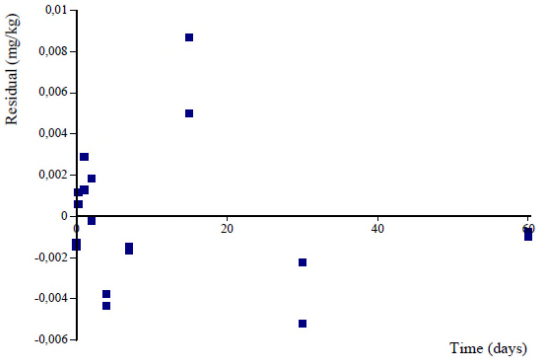
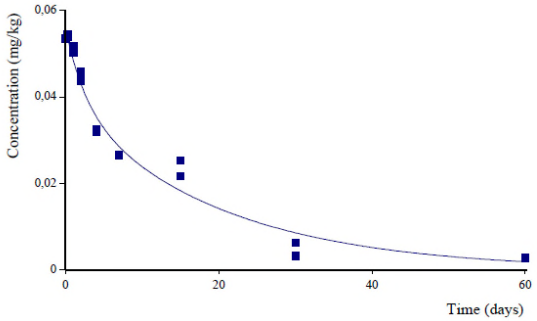
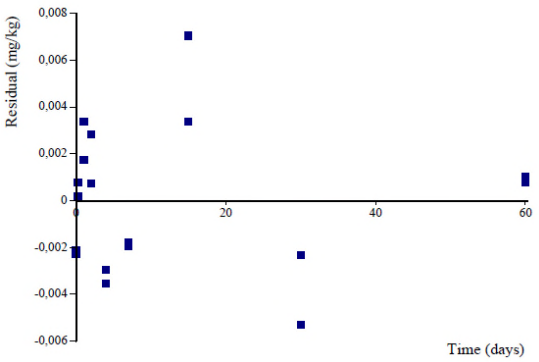


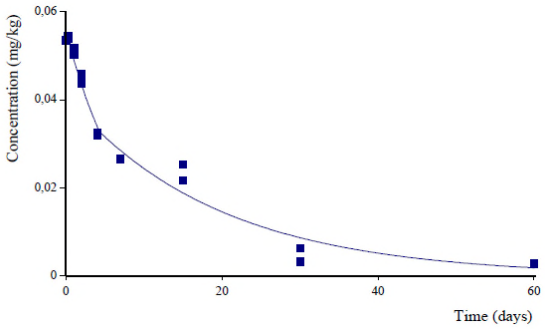
DFOP



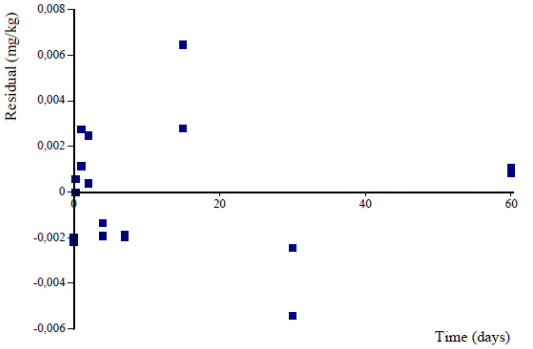
DFOP



HS



HS



Lufa 2.4 soil fitting

The applicant ran all four kinetic models and concluded SFO as the best-fit model. Following the FOCUS guidance, the CA ran SFO and FOMC initially. The CA agrees the FOMC model does not improve the visual and statistical fit of the data and so concurs SFO is the best-fit model. As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil.

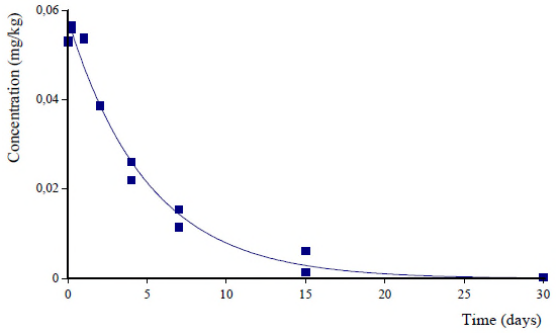
Table CA.B.8.1.1.4.3-3: CA Kinetic fitting of data from bixlozone-2,4-dichlorobenzoic acid in Lufa 2.4 soil[a)]

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**3.49**	**11.6**	**7.72**	**k**	**0.198**	**4.39E-9**	**-**	**-**	**Good**
FOMC	2.77	9.38	10.7	α β	45.49 180.6	-	nd	nd	Good

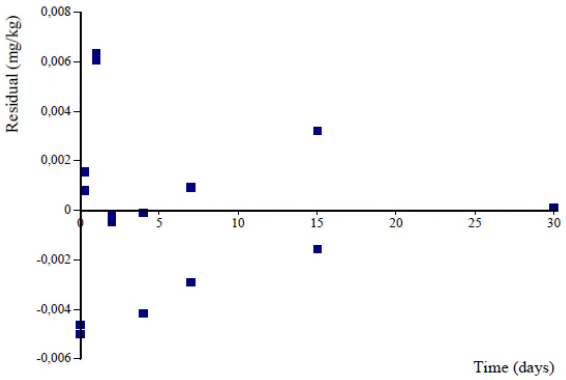
[a)]Best-fit model highlighted bold
nd – CAKE states the error could not be calculated because the covariance matrix could not be created

Figure CA.B.: Applicant's Lufa 2.4 2,4-DBAkinetic fits

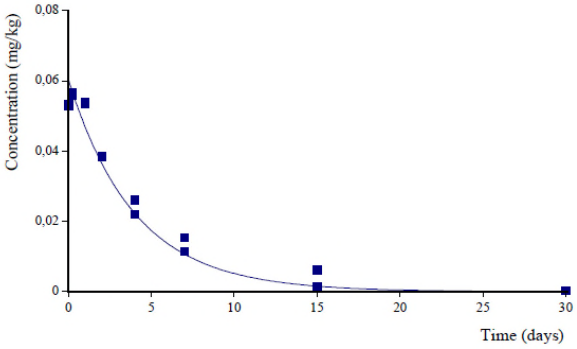
SFO



SFO



FOMC



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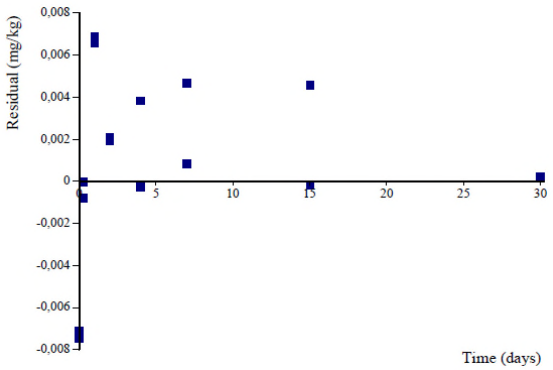


St. Bauzille 12-060 soil fitting

The applicant ran all four kinetic models and concluded HS as the best-fit model, with SFO good enough for modelling endpoints. Following the FOCUS guidance, the CA ran SFO and FOMC initially. The CA agrees with the applicant that the FOMC model provided the slightly better visual fit (and lower χ^2) value and so proceeded to run DFOP and HS fits. The CA notes the resulting visual fits of the three biphasic models were very similar and calculated very similar DT50 values. Because the HS model resulted in the lowest lower χ^2 value and the t-test was passed for both rate constants, the CA accepts the applicant's conclusion that HS provided the best-fit model. The CA also agrees that SFO was good enough for modelling endpoints. As the CA obtained very similar values to the applicant, the applicant's values are accepted for this soil.

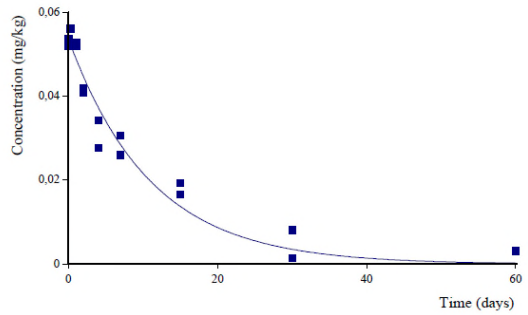
Table CA.B.8.1.1.4.3-4: Applicant's kinetic fitting of data from 2,4-DBA in St. Bauzille 12-060 soil[a)]

Model	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**Estimated parameters**		**t-test**	**Upper Confidence Interval (95%)**	**Lower Confidence Interval (95%)**	**Visual fit**
SFO	7.63	25.4	8.54	k	0.091	3.70E-8	-	-	Good
FOMC	6.36	38.1	7.05	α β	1.576 11.52	-	2.915 25.27	0.2379 -2.238	Good
DFOP	6.34	33.9	7.1	k1 k2 g	0.376 0.056 0.330	0.103 0.004 -	0.984 0.095 -	-0.233 0.018 -	Good
HS	**6.45**	**33.6**	**6.01**	**k1** **k2** **tb**	**0.137** **0.059** **3.995**	**1.03E-4** **3.21E-5** **-**	**0.196** **0.082** **-**	**0.078** **0.037** **-**	**Good**

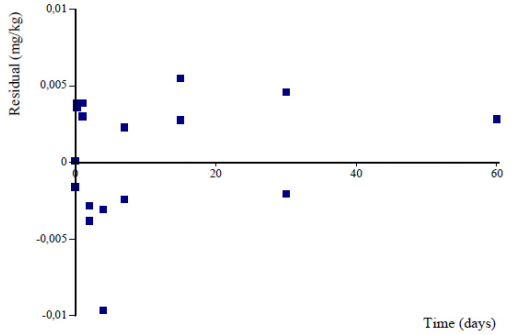
[a)]Best-fit model highlighted bold

Figure CA.B.: Applicant's St. Bauzille 12-060 2,4-DBA kinetic fits

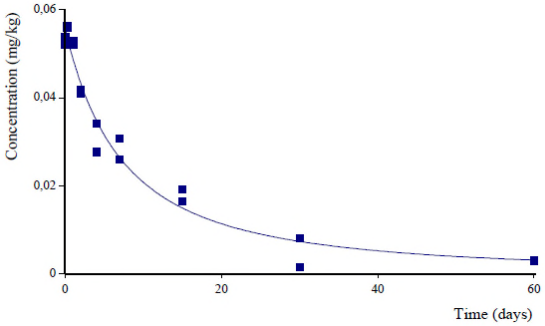
SFO



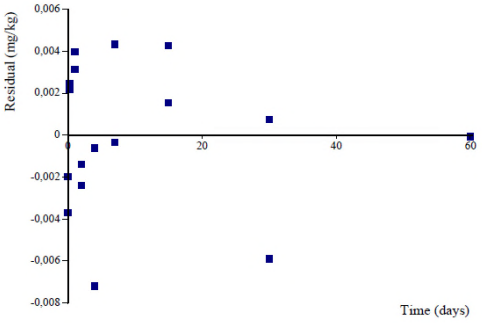
SFO



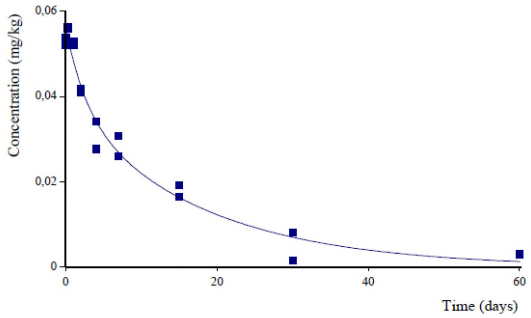
FOMC



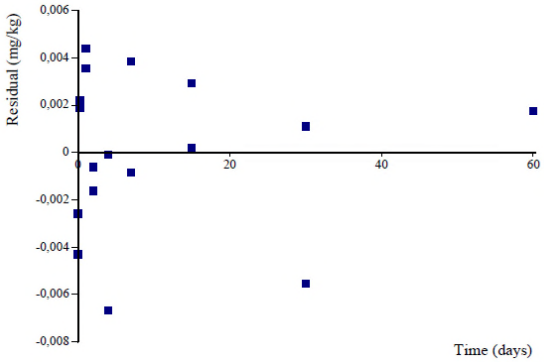
FOMC



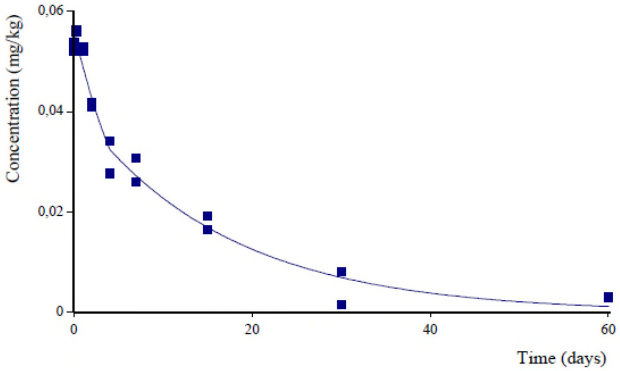
DFOP



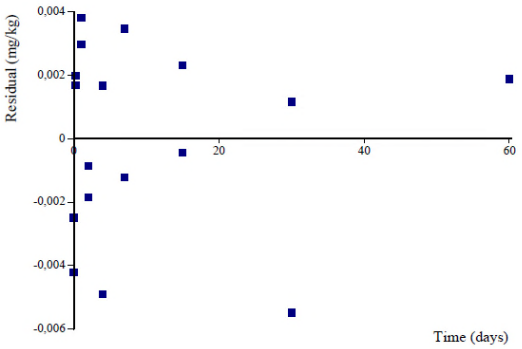
DFOP



HS



HS



The 2,4-DBA best-fit and non-normalised modelling endpoints are summarised in Table CA.B.**8.1.1.4.3-5**. It is noted by the CA that all of the best-fit DT_{50} values are <8 days for each soil and so soil dissipation studies are not triggered for this compound.

Table CA.B.8.1.1.4.3-5: Best-fit 2,4-DBA endpoints in soil under aerobic conditions

Soil	Kinetic model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	t-test	Visual fit	Agrees with applicant?
Best-fit endpoints							
Lufa 2.1	HS	7.5	38.8	6.79	k_1 <0.001 k_2 <0.001	Good	Y
Lufa 2.4	SFO	3.5	11.6	7.72	k < 0.001	Good	Y
St. Bauzille 12-060	HS	6.5	33.6	6.01	k_1 <0.001 k_2 <0.001	Good	Y
Modelling endpoints							
Lufa 2.1	SFO	8.9	29.7	9.11	k < 0.001	Good	Y
Lufa 2.4	SFO	3.5	11.6	7.72	k < 0.001	Good	Y
St. Bauzille 12-060	SFO	7.6	25.4	8.54	k < 0.001	Good	Y

For use in FOCUS modelling, the modelling endpoints were normalised to pF2 (FOCUS default value) and 20°C using the approach described in the FOCUS Ground Water guidance, as shown in Table CA.B.**8.1.1.4.3-6**.

Table CA.B.8.1.1.4.3-6: Degradation rate of 2,4-dichlorobenzoic acid in soil under aerobic conditions normalized to 20°C and pF2

2,4-dichlorobenzoic acid, laboratory studies, aerobic conditions											
Soil name	**Soil type**	**pH $CaCl_2$**	**temp. °C**	**Soil moisture [% w/w]**		**DT_{50} (d)**	**DT_{90} (d)**	**DT_{50} (d) 20°C pF2/10k Pa**	**χ^2 (%)**	**Kinetic model**	**Agrees with applicant?**
				study	**reference**						
Lufa 2.1	Loamy sand	4.84	20	15.8	14	8.9	29.7	8.9	9.11	SFO	Y
Lufa 2.4	Loam	7.41	20	24.6	25	3.5	11.6	3.5	7.72	SFO	Y
St. Bauzille 12-060	Silty loam	7.53	20	23.2	40	7.6	25.4	5.2	8.54	SFO	Y
Geometric mean (n = 3)								5.4			

CONCLUSION

In three European soils incubated at 20 ºC and 50% MWHC, non-labelled 2,4-dichlorobenzoic acid degraded with normalised SFO DT_{50} values in the range 3.5 to 8.9 days (geomean value = 5.4 days).

CA.B.8.1.1.5. *Rate of anaerobic degradation*

CA.B.8.1.1.5.1. Rate of anaerobic bixlozone degradation

Report:	KCA 7.1.1.2 Simmonds, R., (2015b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Route and Rate of Anaerobic Degradation in Four Soils at 20°C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/002, FMC Tracking no. 2013EFT-ISX1022
Guidelines:	FOCUS (2014) OECD Guideline 307 (April 2002); OPPTS Guideline 835.4200 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

CA comments:	For three of the four soils (except Lufa 6S), the applicant used incorrect data in their kinetic analysis due to a transcription error. Therefore, for these soils, the CA's kinetic evaluation is considered appropriate. **This study is relied upon.**

INTRODUCTION

Two European and two US soils (pH (0.01M $CaCl_2$) range 7.1 – 7.4) were treated with [phenyl-U-^{14}C]- and [carbonyl-^{14}C]-bixlozone at 1.09 µg/g dry weight. The soils were incubated at pF 2-2.5 in the dark at 20 ± 2°C for 30 days and then flooded with nitrogen purged de-ionised water to an approximate depth of 2 cm above the soil surface to establish anaerobic conditions which were maintained by a flow of nitrogen through the flasks for *ca* 120 days. Full study details are summarised in section CA.B.8.1.1.2.1. Kinetic assessments of the two major anaerobic metabolites bixlozone-3-OH-propanamide and 2,4-dichlorobenzoic acid are presented in sections CA.B.8.1.1.5.2 and CA.B.8.1.1.5.3 respectively. As such, no further consideration of the metabolites are considered in this kinetic assessment.

[^{14}C]-bixlozone degradation rates were determined by the applicant for each soil with CAKE v2.0 software following FOCUS kinetics guidance. The CA has used Kingui v2 in order to run fitting for independent evaluation.

METHOD

All information on materials and methods are summarised at section CA.B.8.1.1.2.1. The applicant has fitted data for parent only in their kinetic analysis, and will provide subsequent studies for kinetic analysis of metabolites. The CA has followed the same approach and only modelled parent data (see Table CA.B.**8.1.1.5.1-1**).

The degradation rates of bixlozone under anaerobic conditions were determined following the recommendations of the FOCUS work group on degradation kinetics. The results at the end of the aerobic phase were used as time zero for the anaerobic phase. The experimental data collected from both the phenyl and carbonyl labelled samples were treated as replicates and fitted individually during the kinetic analysis.

The CA notes that due to a transcription error, the applicant used incorrect data in their kinetic fittings for all soils except Lufa 6S. The applicant therefore provided an updated kinetic assessment of the data. However, this revised kinetic assessment was provided after the CA had conducted its kinetic assessment and so therefore has not been considered further.

Table CA.B.8.1.1.5.1-1: Data used for CA kinetic fitting (% applied radioactivity)

Lufa 6S			**CA-SL**		
Time (days)	**[phenyl-U-^{14}C]-bixlozone**	**[carbonyl-^{14}C]-bixlozone**	**Time (days)**	**[phenyl-U-^{14}C]-bixlozone**	**[carbonyl-^{14}C]-bixlozone**
0	83.71	80.60	**0**	88.73	87.76
0	80.64	82.05	**0**	89.23	87.80
3	81.49	81.76	**3**	92.04	91.44
3	81.79	83.18	**3**	89.90	93.55
14	81.37	80.47	**14**	85.04	85.56
14	78.75	81.20	**14**	83.75	86.04
45	75.08	77.77	**45**	83.93	83.22
45	78.77	77.19	**45**	84.55	83.27
90	69.65	68.19	**90**	83.78	80.83
90	70.30	65.17	**90**	83.71	81.50
120	50.54	46.73	**120**	84.11	78.85
120	47.35	50.25	**120**	81.02	81.13
Lufa 5M			**Iowa**		
Time (days)	**[phenyl-U-^{14}C]-bixlozone**	**[carbonyl-^{14}C]-bixlozone**	**Time (days)**	**[phenyl-U-^{14}C]-bixlozone**	**[carbonyl-^{14}C]-bixlozone**
0	78.63	81.51	**0**	76.16	73.78
0	80.30	81.83	**0**	84.66	76.66
3	81.88	81.29	**3**	77.05	72.76
3	82.36	82.52	**3**	78.01	78.22
14	78.89	79.72	**14**	74.61	73.14
14	78.63	78.39	**14**	77.22	72.88
45	79.15	73.38	**45**	72.48	70.14
45	73.61	77.10	**45**	72.63	72.35
90	71.49	68.43	**90**	64.64	71.66
90	70.42	68.53	**90**	69.71	70.92
120	71.21	72.73	**120**	64.45	63.10
120	67.23	72.56	**120**	64.78	67.30

RESULTS

All SFO visual fits were good with the exception of soil Lufa 6S. However the first order multi-compartment (FOMC) kinetic fit for Lufa 6S was no better, and hence SFO was retained as the most appropriate fit. The CA notes it is not clear why the Lufa 6S visual fit was not as good as for the other soils. The CA has checked the study summary (section CA.B.8.1.1.2.1) and there is nothing to indicate any artefacts of the study that would impact the study results which would require the omission any data. Therefore, the Lufa 6S SFO fit is accepted.

CA kinetic fitting for persistence endpoint of anaerobic degradation of bixlozone

Lufa 6S CA fitting

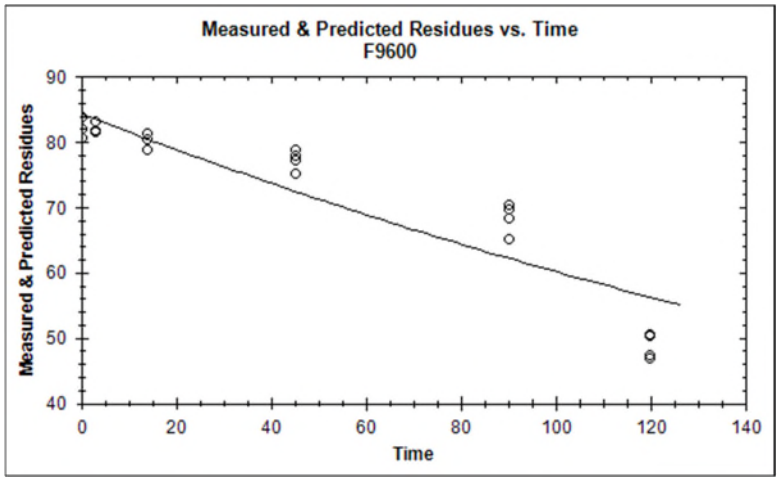
Table CA.B.8.1.1.5.1-2: CA Kinetic fitting of data from bixlozone in Lufa 6S soil

Model	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**Estimated parameters**		**t-test**	**Upper Confidence Interval (95%)**	**Lower Confidence Interval (95%)**	**Visual fit**
SFO	**204.98**	**680.91**	**4.967**	**k**	**$3.382x10^{-3}$**	**$5.35x10^{-10}$**	**-**	**-**	**Fair**
FOMC	175.68	587.09	5.848	α β	134.8 34090	n/a	832.70 210745.33	-563 -142600	Fair

Best fit model highlighted in bold

The CA has selected the SFO fitting as the best fit for Lufa 6S soil. This is due to the lower chi^2 value for SFO fitting, k passing the t-test and a slightly narrower residuals plot. FOMC does not significantly improve visual fit and confidence levels for α and β encompass 0 (with a very large range for β). The DT_{50} is uncertain for both fits as they are extrapolated far beyond the study duration. Therefore, the CA accepts the applicant's choice of SFO fitting, as the parameter values are very similar to those in the CA fitting.

Figure CA.B.8.1.1.5.1-1: CA kinetic fitting and residuals for data from bixlozone in Lufa 6S soil. SFO fitting and residuals



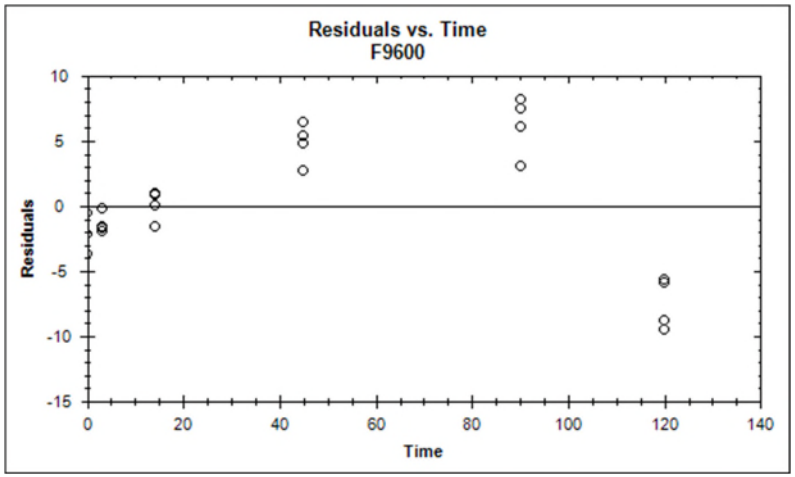
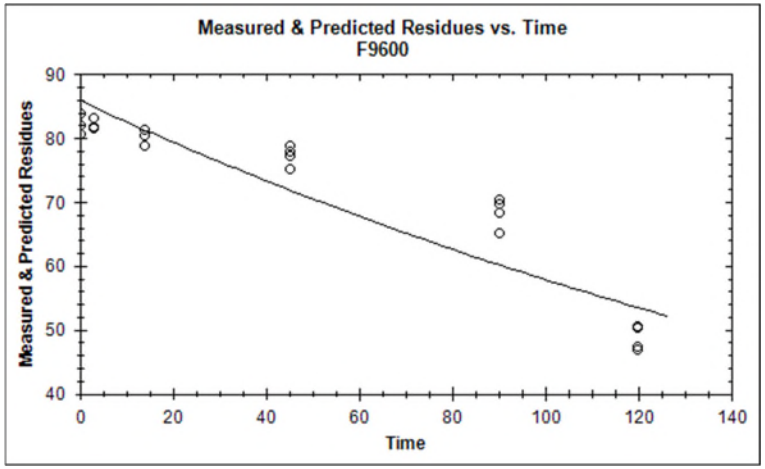
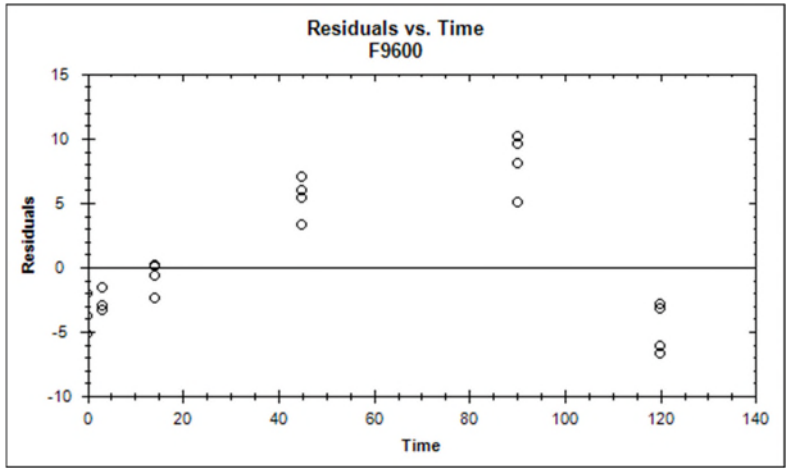


Figure CA.B.8.1.1.5.1-2: CA kinetic fitting and residuals for data from bixlozone in Lufa 6S soil. FOMC fitting and residuals





Lufa 5M CA fitting

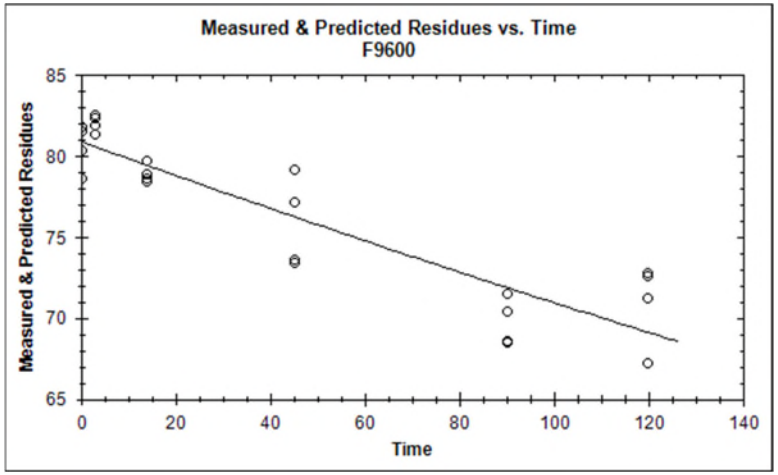
Table CA.B.8.1.1.5.1-3: CA Kinetic fitting of data from bixlozone in Lufa 5M soil

Model	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**Estimated parameters**		**t-test**	**Upper Confidence Interval (95%)**	**Lower Confidence Interval (95%)**	**Visual fit**
SFO	**528.08**	**>1000**	**1.388**	**k**	**0.001313**	**$1.15x10^{-9}$**	**-**	**-**	**Good**
FOMC	>1000	>1000	1.254	α β	0.12877 51.02225	n/a	0.238 131.049	0.01925 -29.0041	Good

Best fit model highlighted in bold

The CA has selected the SFO fitting as the best fit for Lufa 5M soil. This is due to k passing the t-test, low chi^2 and good visual fitting. FOMC did not improve the visual fitting and a large range in the confidence intervals, which also encompass 0, indicating it is not statistically reliable. It is recognised, however, that the DT_{50} of either fit is greater than the 60 day threshold triggering field studies, and is extrapolated beyond the study duration and is therefore uncertain. The CA SFO parameter values are slightly different to the applicant's (due to the applicant using incorrect data in their kinetic fit), and so the CA fitting should be used.

Figure CA.B.8.1.1.5.1-3: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. SFO fitting and residuals



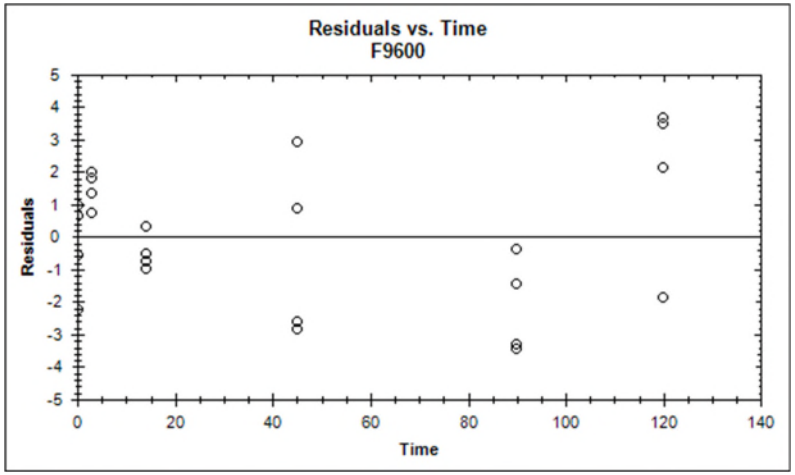
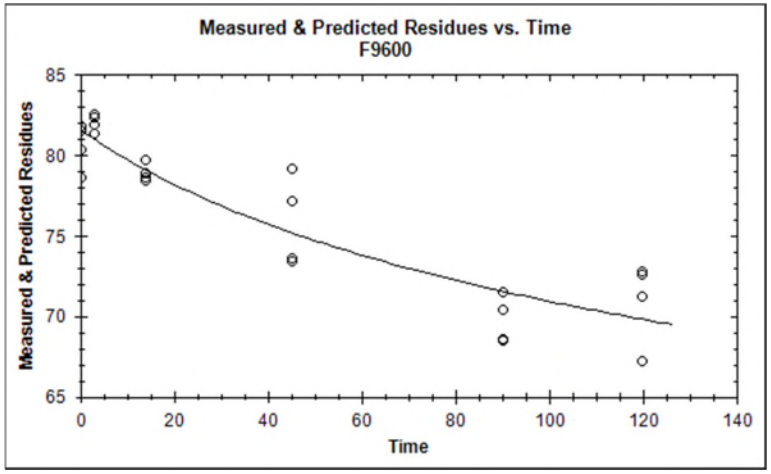
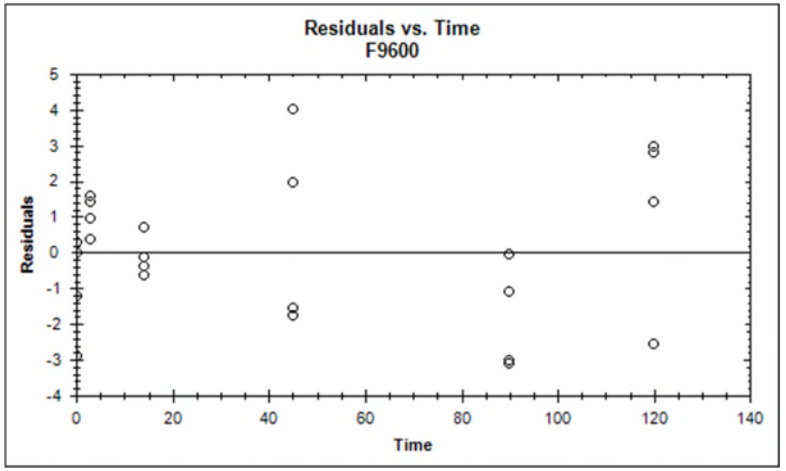


Figure CA.B.8.1.1.5.1-4: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. FOMC fitting and residuals





CA-SL CA fitting

Table CA.B.8.1.1.5.1-4: CA Kinetic fitting of data from bixlozone in CA-SL soil

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval (95%)	Lower Confidence Interval (95%)	Visual fit
SFO	**866.67**	**>1000**	**1.747**	**k**	**$7.998x10^{-4}$**	**$9.65x10^{-7}$**	**-**	**-**	**Fair**
FOMC	>10,000	>10,000	1.517	α β	0.0369 8.2993	n/a	0.058 22.481	0.01622 -5.88212	Fair

Best fit model highlighted in bold

The CA has selected the SFO fitting as the best fit for CA-SL soil. This is due to k passing the t-test and the fitting having a low chi^2, with a more even spread of residuals above and below the line. Additionally, there is no improvement of the visual fit between SFO and FOMC, with both models underpredicting the initial data points but acceptably fitting the latter data points. The CA also recognises that the DT_{50} of either fit is greater than the 60 day threshold triggering field studies and that the DT_{50} is much longer than the study duration, and therefore is uncertain. The CA SFO parameter values are slightly different to the applicant's (due to the applicant using incorrect data in their kinetic fit), and CA fitting should be used as the applicant did not use the raw data for bixlozone.

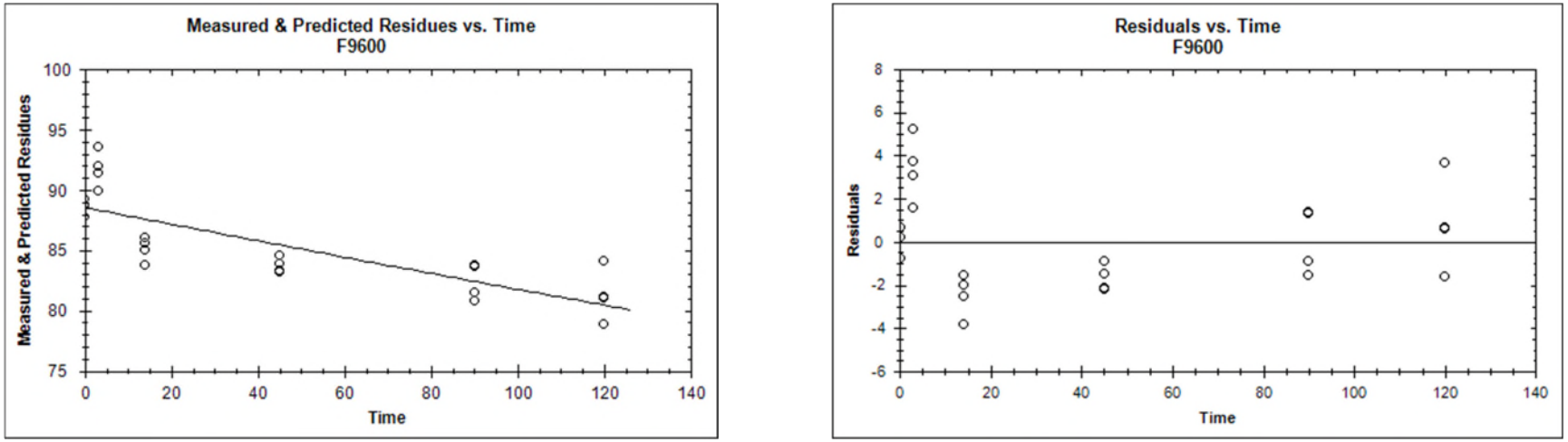
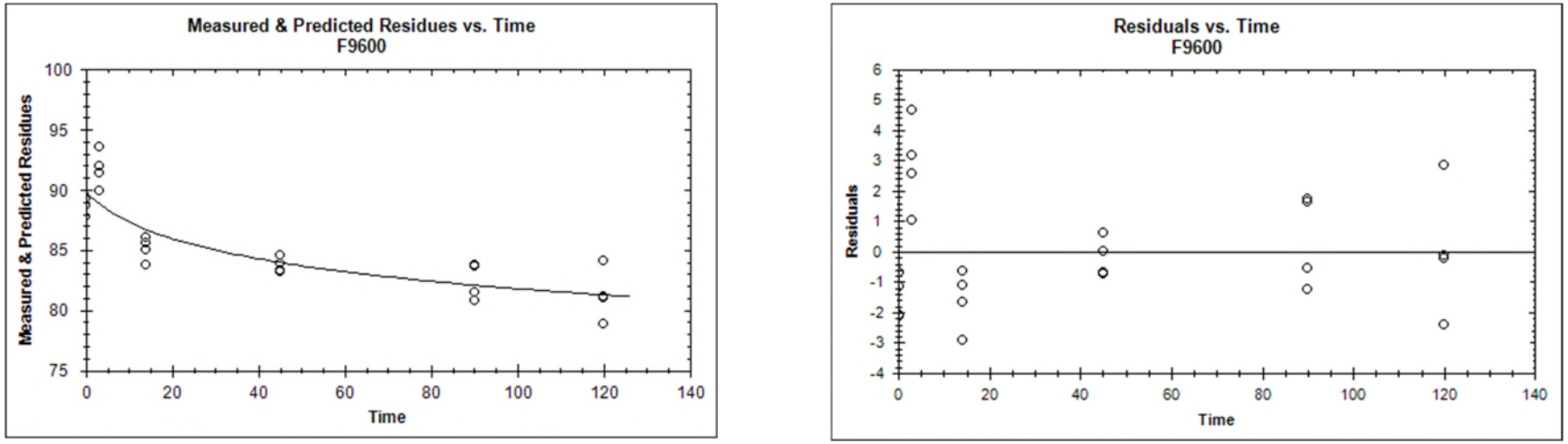


Figure CA.B.8.1.1.5.1-5: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. SFO Fitting and residuals

Figure CA.B.8.1.1.5.1-6: CA kinetic fitting and residuals for data from bixlozone in Lufa 5M soil. FOMC fitting and residuals



Iowa CA fitting

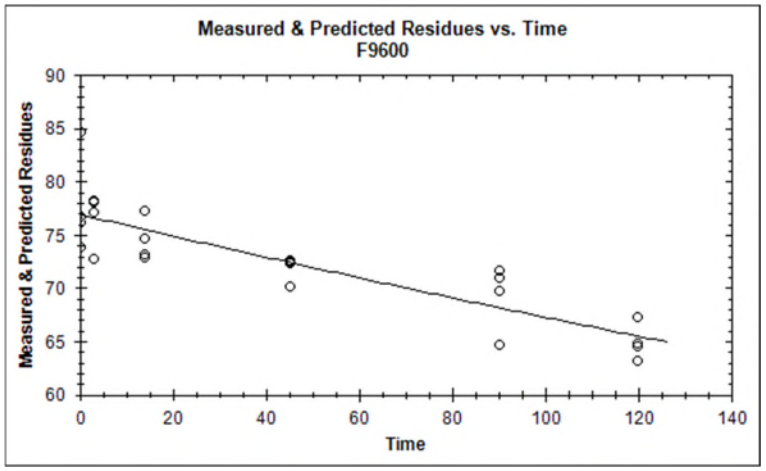
Table CA.B.8.1.1.5.1-5: CA Kinetic fitting of data from bixlozone in Iowa soil

Model	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	Estimated parameters		t-test	Upper Confidence Interval	Lower Confidence Interval	Visual fit
SFO	**516.46**	**>1000**	**0.847**	**k**	**$1.342x10^{-3}$**	**$4.29x10^{-8}$**	**-**	**-**	**Good**
FOMC	692.03	>1000	0.930	α β	0.990 68.23	n/a	19 14108.3	-17.02 -12740	Good

Best fit model highlighted in bold

The CA has selected the SFO fitting as the best fit for Iowa soil. This is due to the low chi^2 value and k passing the t-test. FOMC does not improve the visual fit and confidence levels for β and α also encompass 0, suggesting that values for these parameters are not statistically reliable. The CA notes that DT50 values for both models are extrapolated well beyond the end of the study and some uncertainty is therefore associated with these values. The CA SFO parameter values are slightly different to the applicant's (due to the applicant using incorrect data in their kinetic fit), and CA fitting should be used as the applicant did not use the raw data for bixlozone.

Figure CA.B.8.1.1.5.1-7: CA kinetic fitting and residuals for data from bixlozone in Iowa soil. SFO Fitting and residuals



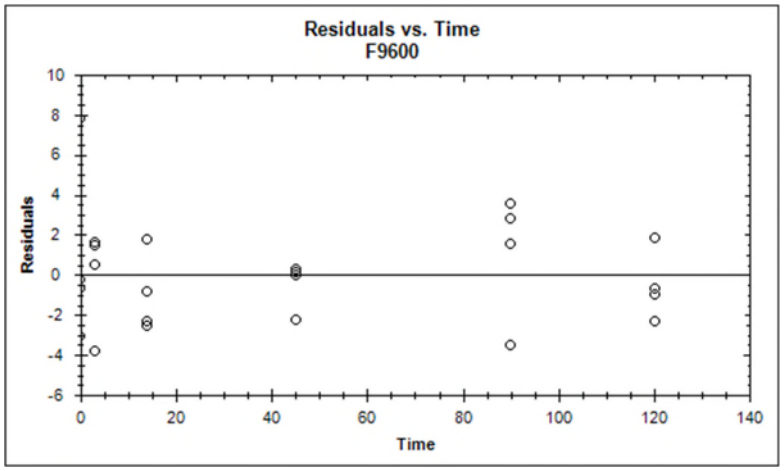
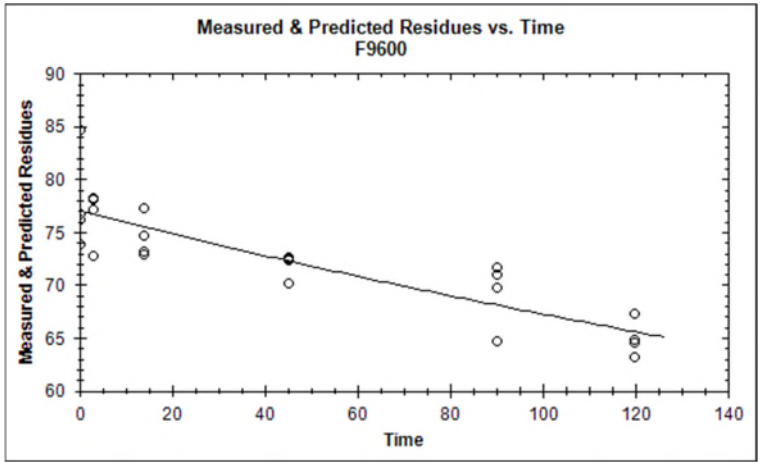
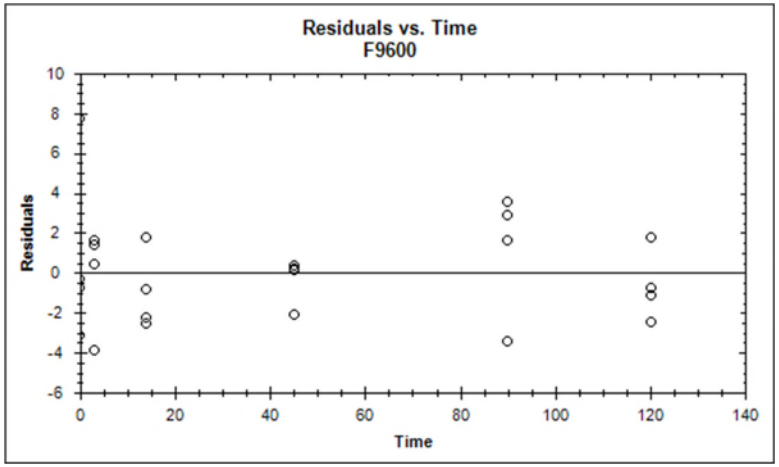


Figure CA.B.8.1.1.5.1-8: CA kinetic fitting and residuals for data from bixlozone in Iowa soil. FOMC fitting and residuals





CA modelling fitting for anaerobic degradation of bixlozone

As the accepted fits for the anaerobic degradation of bixlozone are all using SFO, they are also appropriate for use as modelling fits. A summary of the endpoints is given in Table CA.B.**8.1.1.5.1-6** below.

Table CA.B.8.1.1.5.1-6: Degradation rate of [^{14}C]-bixlozone in soil under anaerobic conditions

Soil	**Kinetic model†**	**DT_{50} (days)**	**DT_{90} (days)**	**χ^2 (%)**	**t-test**	**Agreed with applicant?**
Lufa 6S	SFO	206	685	5.78	<0.001	Y
Lufa 5M	SFO	528	>1000	1.39	<0.001	N (raw data)
CA-SL	SFO	867	>1000	1.75	<0.001	N (raw data)
Iowa	SFO	516	>1000	0.85	<0.001	N (raw data)
Geometric mean (n = 4)		470	1561	-		

† SFO = single first order

CA.B.8.1.1.5.2. Bixlozone-3-OH-Propanamide, Anaerobic Degradation in One Soil at 20ºC in the Dark

Report:	KCA 7.1.2.1.4/01, Schwarzkopf, A., (2018a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600-3-OH-Propanamide Anaerobic Degradation in One Soil at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study No.: S17-04093, FMC Tracking No.: 2017EFT-ISX3564
Guidelines:	OECD 307 OPPTS 835.4200 SANCO/3029/99 rev.4
GLP:	Yes

CA Comments:	The CA notes the applicant did not differentiate between modelling and best-fit endpoints in their kinetic assessment. Therefore, the modelling kinetic assessment has been undertaken by the CA. However, as justification was provided and accepted excluding 3-OH anaerobic degradation from the exposure calculations, the results of this study are not considered further in the DAR **This study is <u>not</u> relied upon.**

INTRODUCTION

This is the kinetic evaluation of the 3-OH-proanamide (hereby referred to as 3-OH) anaerobic degradation study, summarised in section CA.B.8.1.1.2.2.

METHOD

The applicant undertook a kinetic evaluation in order to derive persistence and modelling endpoints. Kinetic analysis was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*]. The applicant carried out the kinetic evaluation using CAKE v3.3 with IRLS as selected optimisation. For completeness, the CA has repeated the modelling using KinGUI v2 with NLLS selected. The goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test). The CA notes the applicant does not differentiate between modelling and best-fit endpoints in their assessment. Therefore, the modelling endpoint assessment is presented by the CA.

The CA notes the applicant has provided best-fit kinetic assessments for the soil phase, water phase and the total system. However, the CA considers it appropriate to only assess the total system degradation, therefore, only total system kinetics have been validated. The data used by the CA in the kinetics is summarised in Table CA.B.**8.1.1.5.2-1**.

Table CA.B.8.1.1.5.2-1: 3-OH total system anaerobic results

Sampling interval (day)	Total system (as % applied)
0	95.8
0	92.7
0.083	86.2
0.083	84.6
0.25	75.8
0.25	74.4
1	63.5
1	60.2
2	68.3
2	61.0
3	61.9
3	57.1
7	56.6
7	61.0
14	43.4
14	47.6
30	49.4
30	48.3
60	47.4
60	48.3
120	19.4
120	23.9

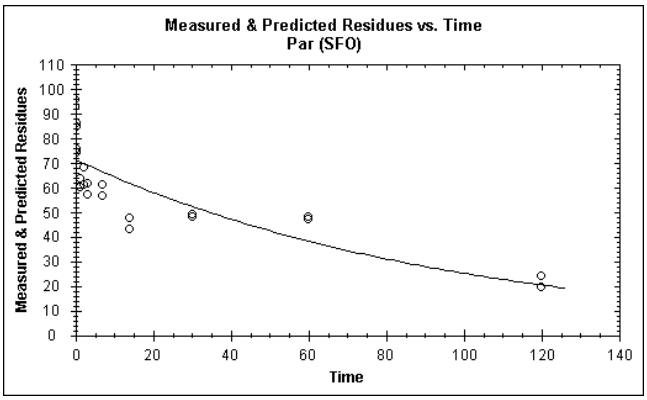
Modelling endpoints

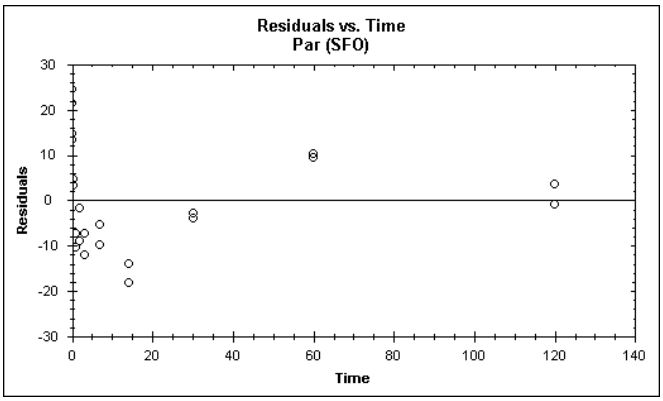
As mentioned above, the applicant did not differentiate between modelling and best-fit kinetics. Therefore, the modelling kinetic assessment has been undertaken by the CA. The CA ran SFO initially. It showed an acceptable visual fit and acceptable χ^2 and t-test values (see Figure CA.B.**8.1.1.5.2-1** and Table CA.B.**8.1.1.5.2-2**). Therefore, the SFO model was considered good enough to determine appropriate modelling endpoints.

Table CA.B.8.1.1.5.2-2: bixlozone-3-OH Propanamide Selected Modelling Endpoints

Model	Visual fit	χ^2 error [%]	Kinetic parameters	t-test	DT50 [d]	DT90 [d]
SFO	Acceptable	14.87	M(0): 71.37 K: 0.01049	1.58E-4	66.1	220

Figure CA.B.8.1.1.5.2-1: 3-OH SFO kinetic fit





Triggering/persistence endpoints

The applicant ran all kinetic models and concluded the HS model provided the best visual and statistical fit. The CA has repeated the applicant's modelling and obtained very similar results to the applicant, therefore, the applicant's kinetic results are accepted and are summarised in Table CA.B.**8.1.1.5.2-3** and Figure CA.B.**8.1.1.5.2-2**. The CA agrees with the applicant that the biphasic fits result in a better visual and statistical fit to the SFO fit. The CA notes there is very little difference between the DFOP and HS model kinetic fits, therefore, the applicant's conclusion of the HS model providing the best-fit is accepted.

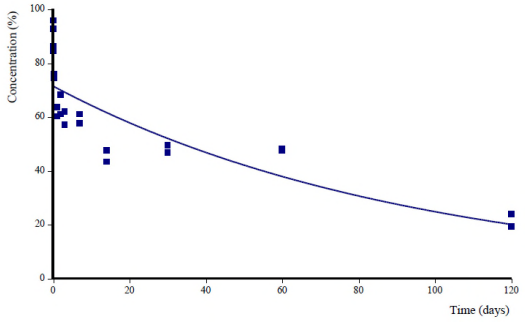
Table CA.B.8.1.1.5.2-3: Applicant's 3-OH best-fit kinetic results

Model		SFO	FOMC	DFOP	**HS**
Visual fit		Acceptable	Acceptable	Acceptable	**Acceptable**
DT_{50} (days)		65.6	17.1	36.0	**37.0**
DT_{90} (days)		218	>10,000	258	**257**
χ^2 error (%)		14.9	8.44	6.27	**6.33**
k ($days^{-1}$)	k or k_1	0.01057	n/a	3.391	**0.901**
	k_2	n/a	n/a	7.24E-3	**7.32E-3**
P value	k or k_1	1.53E-4	n/a	3.56E-3	**1.09E-3**
	k_2	n/a	n/a	7.33E-7	**5.96E-7**
g/tb		n/a	n/a	0.351	**0.473**
alpha		n/a	0.1278	n/a	**n/a**
beta		n/a	0.0758	n/a	**n/a**
95% CI (lower/upper)	alpha	n/a	0.089 / 0.166	n/a	**n/a**
	beta	n/a	-0.055 / 0.207	n/a	**n/a**

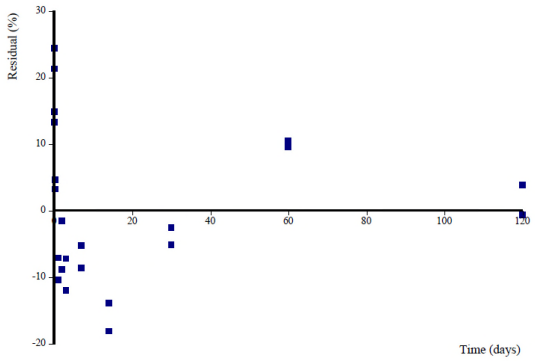
Best-fit model in bold

Figure CA.B.8.1.1.5.2-2: Applicant's 3-OH kinetic graphs

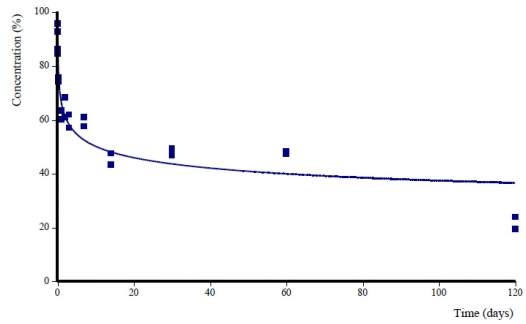
SFO:



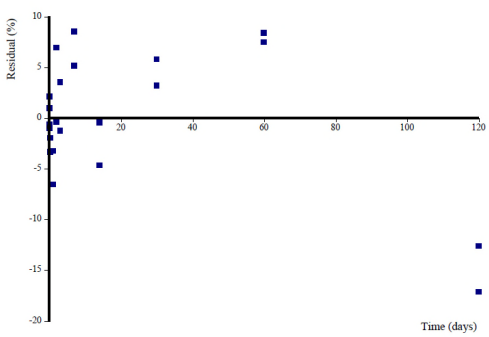
SFO:



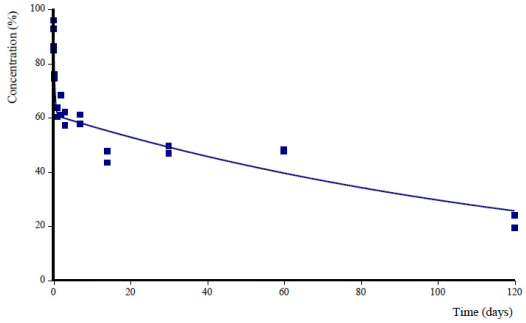
FOMC:



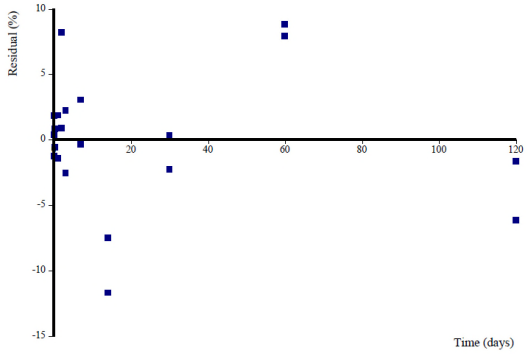
FOMC:



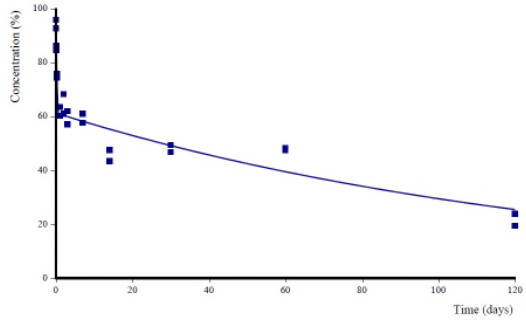
DFOP:



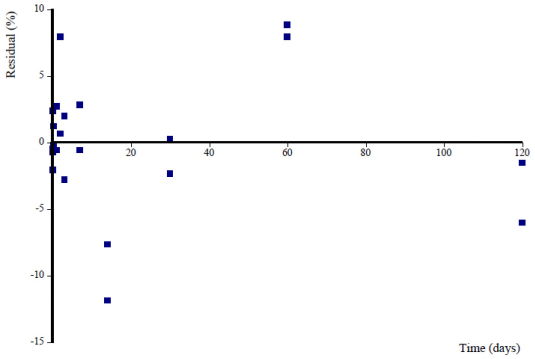
DFOP:



HS:



HS:



Conclusion

In one European soil, bixlozone-3-OH-propanamide degraded in soil incubated under anaerobic conditions with a modelling DT_{50}/DT_{90} of 66.1 / 220 days and a best-fit DT_{50}/DT_{90} of 37 / 257 days.

CA.B.8.1.1.5.3. 2,4-dichlorobenzoic acid Anaerobic Degradation in One Soil at 20ºC in the Dark

Report:	KCA 7.1.2.1.4/02, Schwarzkopf, A., (2018b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	2,4-Dichlorobenzoic Acid Anaerobic Degradation in One Soil at 20 °C in the Dark
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study No.: S17-04094, FMC Tracking No.: 2017EFT-ISX3565
Guidelines:	OECD 307 OPPTS 835.4200 SANCO/3029/99 rev.4
GLP:	Yes

CA Comments:	The CA notes the applicant did not differentiate between modelling and best-fit endpoints in their kinetic assessment. Therefore, the modelling kinetic assessment has been undertaken by the CA. However, as 2,4-DBA formed in greater quantities in the soil dissipation studies, the results of this anaerobic degradation study have not been considered further in the DAR. **This study is <u>not</u> relied upon.**

INTRODUCTION

This is the kinetic evaluation of the 2,4-dichlorobenzoic acid (hereby referred to as 2,4-DBA) anaerobic degradation study, summarised in section CA.B.8.1.1.2.3.

METHOD

The applicant undertook a kinetic evaluation in order to derive persistence and modelling endpoints. Kinetic analysis was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*]. The applicant carried out the kinetic evaluation using CAKE v3.3 with IRLS as selected optimisation. For completeness, the CA has repeated the modelling using KinGUI v2 with NLLS selected. The goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test). The CA notes the applicant does not differentiate between modelling and best-fit endpoints in their assessment. Therefore, the modelling endpoint assessment is presented by the CA.

The CA notes the applicant has provided best-fit kinetic assessments for the soil aerobic phase, soil anaerobic phase, water phase and the total system. However, the CA considers it appropriate to only assess the total system degradation, therefore, only total system kinetics have been validated. The CA notes the applicant's total system kinetics considered an initial time point corresponding to day 1 of the anaerobic phase. The CA considers the final time point of the aerobic phase to correspond to day 0 of the anaerobic phase and so has included these in the kinetic evaluation. The data used by the CA in the kinetics is summarised in .

Table CA.B.8.1.1.5.3-1: <u>2,4-DBA total system anaerobic results</u>

Sampling interval (day)	Total system
0	70.9
0	68.3
1	66.9
1	76.1
3	64.4
3	73.9
7	72.4
7	66.8
14	71.3

14	56.6
31 31	73.2 63.7
60 60	72.9 71.8
120 120	44.3 47.6

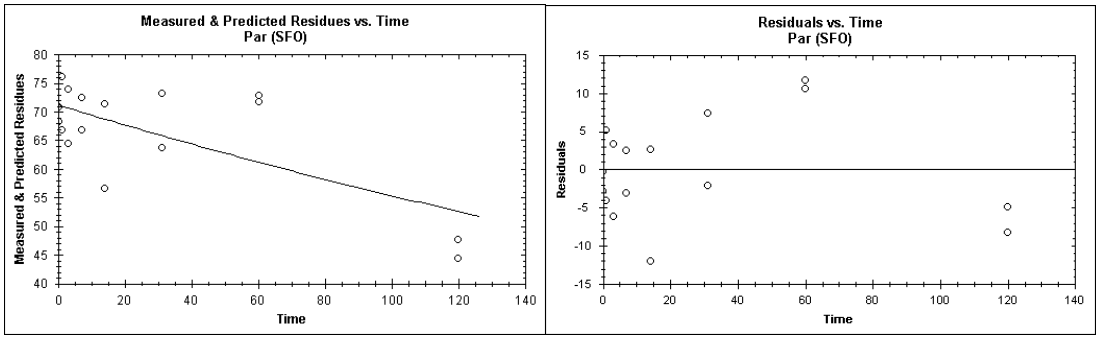
Modelling endpoints

The CA ran SFO model first. It showed an acceptable visual fit and acceptable χ^2 and t-test values (see Figure CA.B.**8.1.1.5.3-1** and Table CA.B.**8.1.1.5.3-2**). Therefore, the SFO model was considered good enough to determine appropriate modelling endpoints.

Table CA.B.8.1.1.5.3-2: 2,4-DBA modelling endpoints

Model	Visual fit	χ^2 error [%]	K	t-test	DT50 [d]	DT90 [d]
SFO	Acceptable	6.06	2.52E-3	2.85E-3	275	913

Figure CA.B.8.1.1.5.3-1: 2,4-DBA SFO kinetic fit



Triggering/persistence endpoints

The applicant ran all kinetic models and concluded the SFO model provided the best visual and statistical fit. The CA has repeated the SFO and FOMC modelling and obtained slightly longer DT50 values to the applicant, believed to be due to the CA using the final aerobic sampling result as a day 0 anaerobic value. The CA agrees with the applicant that the biphasic fit was no better than the SFO fit and so agrees SFO is the best-fit model. As the CA obtained slightly longer DT50 values, the CA's kinetic results are considered appropriate and are summarised in Table CA.B.**8.1.1.5.3-3** and Figure CA.B.**8.1.1.5.3-2**.

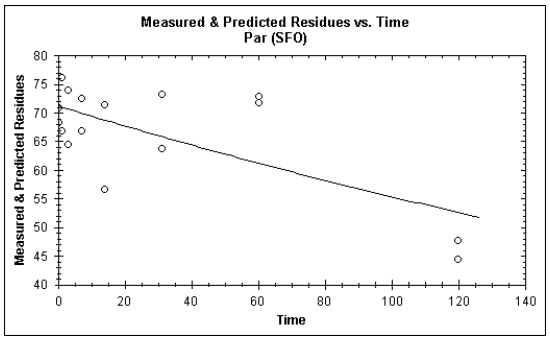
Table CA.B.8.1.1.5.3-3: CA's 2,4-DBA best-fit kinetic results

Model		**SFO**	FOMC
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**275**	275
DT_{90} (days)		**913**	914
χ^2 error (%)		**6.06**	6.46
k ($days^{-1}$)		**2.52E-3**	n/a
P value		**2.85E-3**	n/a
alpha		**n/a**	1.87E+3
beta		**n/a**	7.41E+5
95% CI (lower/upper)	alpha	**n/a**	7.03E+2 / 3.03E+3
	beta	**n/a**	7.41E+5 / 7.41E+5

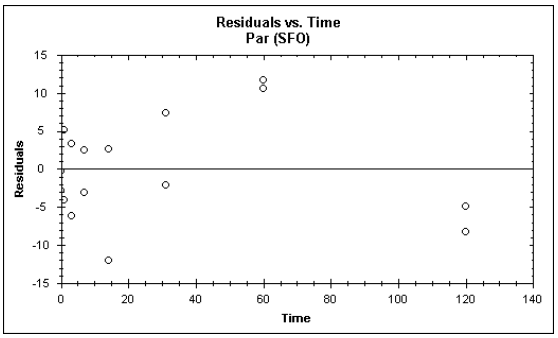
Best-fit model in bold

Figure CA.B.8.1.1.5.3-2: CA's 2,4-DBA kinetic graphs

SFO:



SFO:



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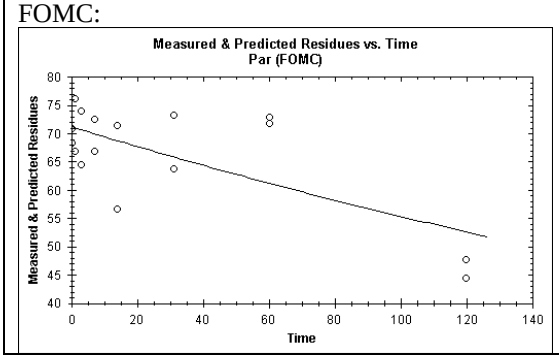
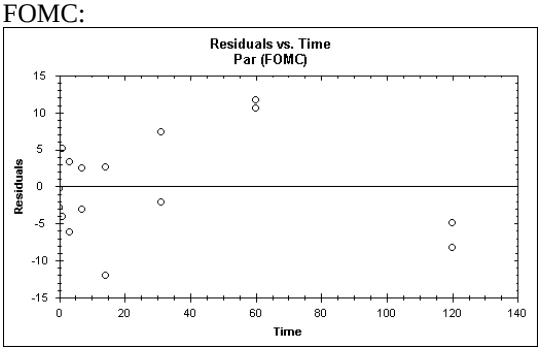


FOMC:



CONCLUSION

In one European soil, 2,4-DBA degraded slowly in soil incubated under anaerobic conditions with a DT_{50} value 275 days and DT_{90} value of 913 days.

CA.B.8.1.2. **Field studies**

CA.B.8.1.2.1. ***Soil dissipation studies***

CA.B.8.1.2.1.1. Soil dissipation in France and Italy

Report:	KCA 7.1.2.2.1-01 Gemrot, F. (2018a)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Soil Dissipation Study after One Application of F9600-4 SC or F9600-21 CS in Southern Europe (Southern France and Italy) - 2015 and 2017
Document No:	SGS Study Number: 15SGS088 FMC Tracking Number: 2015EFT-ISX1947
Guidelines:	-SETAC 1995 -SANCO/3029/99 rev. 4 -EPA (Oct 2008). Fate, Transport and Transformation Test Guidelines. -EFSA Guidance Document for evaluating laboratory and field dissipation studies to obtain DegT50 values of active substances of plant protection products and transformation products of these active substances in soil. EFSA Journal 2014;12(5): 3662. -ISO 10381-6:2009 (handling & storage soil for assessment of microbial biomass) -OECD Guidance Document for Conducting Pesticide Terrestrial Field Dissipation Studies (ENV/JM/MONO(2016)6, released 04 March 2016).
GLP:	Yes

CA Comments:	A number of samples at site FR01 and IT01 (see main body of text for more info) were kept at ambient temperature in the field rather than being kept cool prior to being frozen. However, as all samples were frozen with 12 hours of sampling, the CA does not consider this to have significantly impacted on the outcomes of the study. Furthermore, bixlozone residues >LOQ were detected at site IT02 in samples taken 3 days prior to application. The applicant has indicated these are likely due to lab contamination. This issue is further explored in the main body of text; the CA does not consider this issue to affect the validity of the study. **This study is relied upon.**

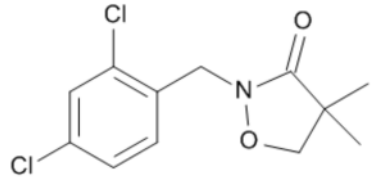
INTRODUCTION

A study was conducted to determine the dissipation and mobility of bixlozone residues in soil, following a single application of the formulated products, F9600-21 CS or F9600-4 SC, to bare soil. The trial sites were located in southern France and Italy.

Soil cores were collected to a depth of at least 30 cm prior to application, within three hours of application and then approximately 3, 7, 14, 30, 90, 180, 270 and 365 days after last application; one test site, IT01, samples were taken up to 736 days after application. Soil cores were cut into 0-10, 10-20, and 20-30 cm sections for analysis.

MATERIALS

Table CA.B.8.1.2.1.1-1: Test item

Common name:	Bixlozone
Chemical name ~(IUPAC):	2-(2,4-dichlorobenzyl)-4,4-dimethyl-1,2-oxazolidin-3-one)
Chemical structure:	
Cas number:	81777-95-9
Formulation name:	**F9600-4 SC**
Main uses:	Herbicide
Formulation type:	Suspension Concentrate (SC)
Nominal concentration:	36 % w/w
Nominal density:	1.11g/mL
Batch number:	PL15-0061
Actual concentration:	37.4 % w/w
Formulation name:	**F9600-21 CS**
Main uses:	Herbicide
Formulation type:	Capsule Suspension (CS)
Nominal concentration:	36.3 % w/w
Nominal density:	1.16 g/mL
Batch number:	PL15-0062
Actual concentration:	34.9 % w/w

Field data

The applicant states the selected sites presented no risk of erosion, flooding and were not too stony. The selected sites were loam or sandy loam and allowed 0.9 to 1.0 m depth sampling. Plots were not set up in the shadow of neighbouring trees and the slope was not more than 1.5%. The applicant states the sites had been in cultivation for several years but no soil disinfectant (dazomet or metam) was used within the last 3 years before trial set up. Each test site consisted of bare soil. The area was cleared mechanically of all vegetation prior to trial set up and thereafter kept free of weeds by application of herbicide (e.g. glyphosate) at recommended commercial rates. Bare soil was maintained on the plots and around during the whole study period. The pesticide use history has been provided by the applicant for the preceding three years; the CA has confirmed no structurally similar compounds were used. The CA also confirmed that 2,4-dichlorobenzoic acid (2,4-DBA) is a metabolite of propiconazole and penconazole as well, however, neither of these active substances had been applied at the test sites. Therefore, the CA considers the previous pesticide use to have not significantly impacted upon the outcomes of the study. Details of the location and physical properties of the sites is provided in Table CA.B.**8.1.2.1.1-2**. The pesticide use history is provided in Table CA.B.**8.1.2.1.1-3**, with the cultural treatments and maintenance pesticides in Table CA.B.**8.1.2.1.1-4** and Table CA.B.**8.1.2.1.1-5** respectively.

Table CA.B.8.1.2.1.1-2: Chemical and physical soil characterisation

Field Number	FR01			IT01			IT02		
Country	France			Italy			Italy		
Region	Aquitaine			Lombardia			Lombardia		
City	Brannens			Caleppio di Settala			Caleppio di Settala		
ZIP code	33124			20090			20090		
GPS coordinates	44 31 23.7 N, 00 08 27.5 E			45 26 16.55 N, 09 23 32 E			45 26 16.55 N, 09 23 32 E		
Soil depth	0-30 cm	30-45 cm	45-60 cm	0-30 cm	30-45 cm	45-60 cm	0-30 cm	30-45 cm	45-60 cm
pH water	5.9	6.7	7	6.7	6.9	7.1	6.7	6.9	7.1
Organic Matter (%)	1.44	0.48	0.34	2.61	1.85	0.62	2.61	1.85	0.62
Organic Carbon (%)	0.84	0.28	0.2	1.51	1.07	0.36	1.51	1.07	0.36
Microbial biomass (as % OC)	Start of study: 3.28 End of study: 2.61			Start of study: 2.11 End of study: 19.4			Start of study: 2.11[a)] End of study: 1.37		
CEC (meq/100 g)	5.2	3.8	3.5	14.7	13.6	11.8	14.7	13.6	11.8
Maximum WHC Bar 0.1	33.3	27.9	25.2	43.1	39.5	34.5	43.1	39.5	34.5
WHC at pF2	19.0	17.7	17.2	30.3	29.3	25.3	30.3	29.3	25.3
Soil Properties									
Sand 2.00-0.05 mm % w/w	54	53	52	42	39	38	42	39	38
Silt 0.05-0.002 mm % w/w	36	36	36	37	40	39	37	40	39
Clay <0.002 mm % w/w	10	11	12	21	21	23	21	21	23
USDA Textural Class	Sandy Loam	Sandy Loam	Sandy Loam	Loam	Loam	Loam	Loam	Loam	Loam
Sand 2.00-0.063 mm % w/w	48	46	46	28	29	28	28	29	28
Silt 0.063-0.002 mm % w/w	41	42	41	51	51	49	51	51	49
Clay <0.002 mm % w/w	11	12	13	21	20	51	21	20	23
UK Textural Class	Sandy Silt Loam	Sandy Silt Loam	Sandy Silt Loam	Clay Loam	Clay Loam	Clay Loam	Clay Loam	Clay Loam	Clay Loam

[a)] This value was not included in the study report, however, the applicant has confirmed that site IT02 is in the same field as IT01 and so the starting biomass can be read across. For this reason, and because the soil was shown to be microbially active at the end of the study, this is accepted by the CA.

Table CA.B.8.1.2.1.1-3: Field use history

Trial number	Field crop	Date	Pesticide applied		Field rate / ha
FR01	Maize	05/05/12	DUAL GOLD SAFENEUR	s-metolachlor + benoxacor	2.0 L
		0505/12	LAGON	isoxaflutole + aclonifen	1.0 L
	Maize	20/04/13	DUAL GOLD SAFENEUR	s-metolachlor + benoxacor	2.0 L
		20/04/13	LAGON	isoxaflutole + aclonifen	1.0 L
	Maize	11/04/14	DUAL GOLD SAFENEUR	s-metolachlor + benoxacor	2.0 L
		11/04/14	LAGON	isoxaflutole + aclonifen	1.0 L
IT01	Head cabbage	Sept/12	TREBON UP	Etonfeprox	0.4 L
		Sept/12	TRIBASIC DEL	Copper	3.0 L
		Oct/12	TREBON UP	Etonfeprox	0.4 L
		Oct/12	TRIBASIC DEL	Copper	3.0 L
	Tomato	05/2013	SENCOR WG	metribuzin	0.5 kg

Trial number	Field crop	Date	Pesticide applied		Field rate / ha
		06/2013	SENCOR WG	metribuzin	0.5 kg
		06/2013	DECIS JET	deltamethrin	0.6 L
		07/2013	CURZATE R WG	cimoxanyl + copper	1.2 kg
		07/2013	DECIS JET	deltamethrin	0.6 L
		07/2013	CURZATE R WG	cimoxanyl + copper	1.2 kg
	Spring wheat	06/2014	ARIANNE II	clopyralid+ fluroxypir + MCPA	3.5 L
IT02	Head cabbage	Sept/12	TREBON UP	Etonfeprox	0.4 L
		Sept/12	TRIBASIC DEL	Copper	3.0 L
		Oct/12	TREBON UP	Etonfeprox	0.4 L
		Oct/12	TRIBASIC DEL	Copper	3.0 L
	Bare Soil	05/2013	SENCOR WG	metribuzin	0.5 kg
		06/2013	SENCOR WG	metribuzin	0.5 kg
		06/2013	DECIS JET	deltamethrin	0.6 L
		07/2013	CURZATE R WG	cimoxanyl + copper	1.2 kg
		07/2013	DECIS JET	deltamethrin	0.6 L
		07/2013	CURZATE R WG	cimoxanyl + copper	1.2 kg
	Spring Wheat	06/2014	ARIANNE II	clopyralid	3.5 L
	Bare Soil	07/07/15	KLARO ULTRA	glyphosate	10.0 L
		07/07/15	ANTIGRAM GOLD	s-metolachlor	1.5 L
		08/08/15	KLARO ULTRA	glyphosate	10.0 L
		08/08/15	ANTIGRAM GOLD	s-metolachlor	1.5 L
		05/09/15	KLARO ULTRA	glyphosate	10.0 L
		05/09/15	ANTIGRAM GOLD	s-metolachlor	1.5 L
		23/10/15	KLARO ULTRA	glyphosate	8.0 L
		07/07/15	KLARO ULTRA	glyphosate	10.0 L

Table CA.B.8.1.2.1.1-4: Cultural treatments

Field No.	Date	Description
FR01	20/05/15	Rotary harrow
IT01	23/04/15	Ploughing
	25/04/15	Rotary Harrow
	29/06/15	Rotary Harrow
IT02	02/09/16	Ploughing (ripper)
	02/09/16	Rotary Harrow

Table CA.B.8.1.2.1.1-5: Maintenance pesticides

Field No.	**Application Date**	**Product**	**Active Substance**	**Application Rate (/ha)**	**Purpose**
IT01	07/07/15	KLARO ULTRA	glyphosate	10.0 L	Herbicide
	07/07/15	ANTIGRAM GOLD	s-metolachlor	1.5 L	Herbicide
	08/08/15	KLARO ULTRA	glyphosate	10.0 L	Herbicide
	08/08/15	ANTIGRAM GOLD	s-metolachlor	1.5 L	Herbicide
	05/09/15	KLARO ULTRA	glyphosate	10.0 L	Herbicide
	05/09/15	ANTIGRAM GOLD	s-metolachlor	1.5 L	Herbicide
	23/10/15	KLARO ULTRA	glyphosate	10.0 L	Herbicide
	08/03/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	05/04/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	22/04/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	04/05/16	OURAGAN	glyphosate	8.0 L	Herbicide
	19/05/16	OURAGAN	glyphosate	8.0 L	Herbicide
	01/06/16	OURAGAN	glyphosate	8.0 L	Herbicide
	16/06/16	OURAGAN	glyphosate	8.0 L	Herbicide

Field No.	Application Date	Product	Active Substance	Application Rate (/ha)	Purpose
	06/07/16	OURAGAN	glyphosate	8.0 L	Herbicide
	20/07/16	OURAGAN	glyphosate	8.0 L	Herbicide
	03/08/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	22/08/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	13/09/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	07/10/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	16/03/17	SILGLIF	glyphosate	8.0 L	Herbicide
	08/04/17	SILGLIF	glyphosate	8.0 L	Herbicide
	26/04/17	SILGLIF	glyphosate	8.0 L	Herbicide
	10/05/17	SILGLIF	glyphosate	8.0 L	Herbicide
	24/05/17	SILGLIF	glyphosate	8.0 L	Herbicide
IT02	08/03/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	05/04/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	22/04/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	04/05/16	OURAGAN	glyphosate	8.0 L	Herbicide
	19/05/16	OURAGAN	glyphosate	8.0 L	Herbicide
	01/06/16	OURAGAN	glyphosate	8.0 L	Herbicide
	16/06/16	OURAGAN	glyphosate	8.0 L	Herbicide
	06/07/16	OURAGAN	glyphosate	8.0 L	Herbicide
	20/07/16	OURAGAN	glyphosate	8.0 L	Herbicide
	03/08/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	22/08/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	13/09/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	07/10/16	KLARO ULTRA	glyphosate	8.0 L	Herbicide
	16/03/17	SILGLIF	glyphosate	8.0 L	Herbicide
	08/04/17	SILGLIF	glyphosate	8.0 L	Herbicide
	26/04/17	SILGLIF	glyphosate	8.0 L	Herbicide
	10/05/17	SILGLIF	glyphosate	8.0 L	Herbicide
	24/05/17	SILGLIF	glyphosate	8.0 L	Herbicide

EXPERIMENTAL DESIGN

Trials FR01 and IT01 consisted of one untreated plot (referred to as “U”) and 4 treated plots (referred to as “T”) that received one application of formulated product as follows:
- T1: bixlozone -4 SC applied to bare soil
- T2: bixlozone- 4 SC applied to bare soil and incorporated to a depth of 7 cm just after application
- T3: F9600-21 CS applied to bare soil
- T4: F9600-21 CS applied to bare soil and incorporated to a depth of 7 cm just after application

Trial IT02 consisted of one untreated plot and one treated plot:
- T2: bixlozone- 4 SC applied to bare soil and incorporated to a depth of 7 cm just after application

Each main plot was separated by a 10 meter buffer zone. Plots were divided into 3 sub-plots a, b, and c. Each sub-plot was separated from the next by a 5 meter buffer zone. Around the untreated control plot a 20 meter buffer zone separated it from other plots.

Table CA.B.8.1.2.1.1-6: Experimental design

Trial number	FR01	IT01	IT02
Plot ID (1)	T1, T2, T3, T4	T1, T2, T3, T4	T2
Subplot width (m)	3	3	3
Subplot length (m)	25	20	20
Number of subplots	3	3	3
Total Plot area (m²)	225	180	180
Slope (%)	1	0	0

Distance minimum between untreated and treated plots (m)	≥20	≥20	≥20

Weather stations were installed on site for the duration of the trial. Historical data was taken from weather stations 7 km from FR01 and 3 km from IT01 and IT02. The weather data from the study period was compared against the average minimum and maximum temperatures and rainfall over the preceding 5 years (see Figure CA.B**8.1.2.1.1-1** to Figure CA.B.**8.1.2.1.1-3**). As can be seen, the averages temperatures were broadly similar to the 5 year norm at each test site, with the exception potentially of the summer 2017, at IT02, where the average minimum temperatures were much higher than the norm. Large fluctuations in monthly rainfall is also observed at each test site, with some months significantly drier than the norm and other months significantly wetter.

Figure CA.B8.1.2.1.1-1.: FR01 comparison of study weather versus the preceding 5 year average

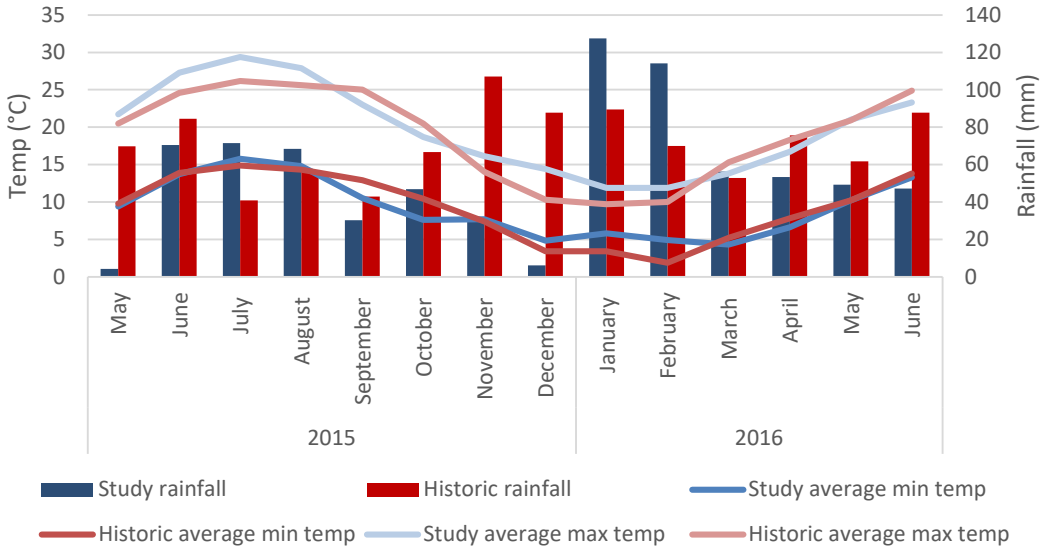


Figure CA.B.8.1.2.1.1-2: IT01 comparison of study weather versus the preceding 5 year average

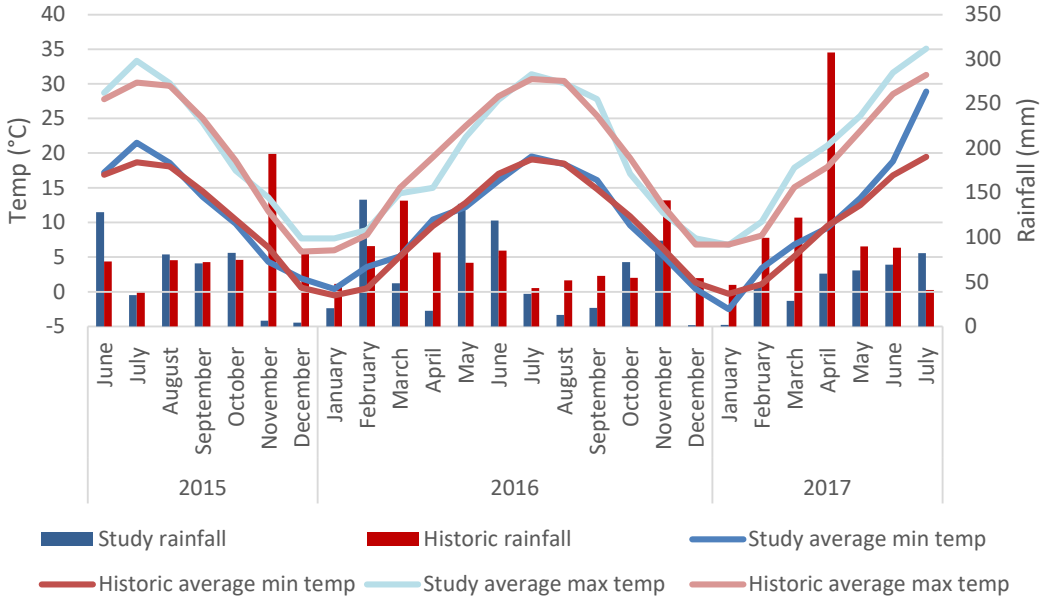
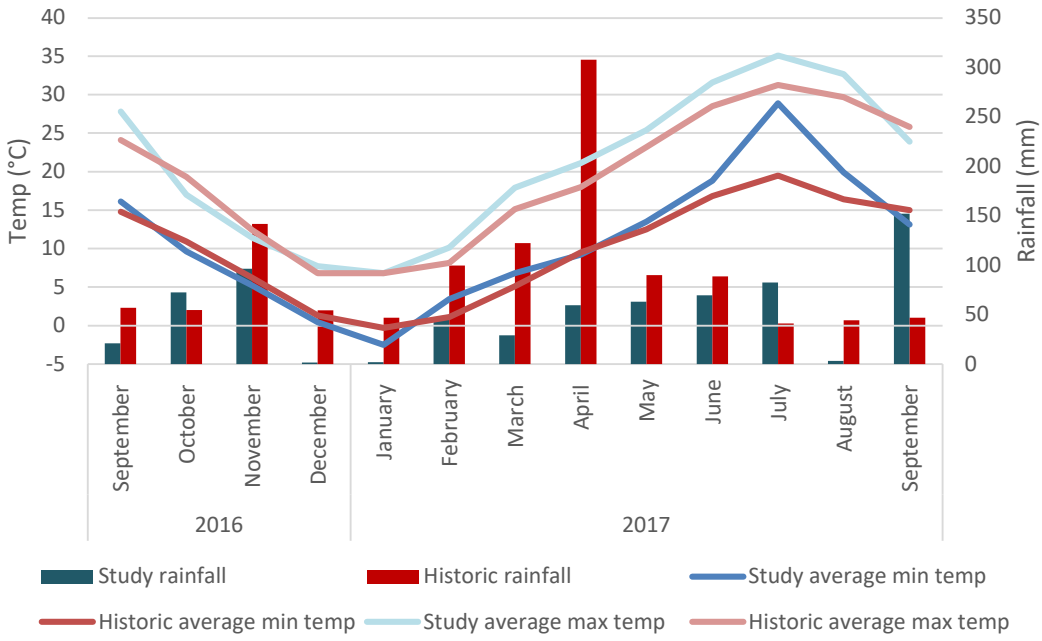


Figure CA.B.8.1.2.1.1-3: IT02 comparison of study weather versus the preceding 5 year average



Application of test item

The application equipment consisted of a boom sprayer. On each treated plot (T1, T2, T3 and T4) a single application was made. Calibrations of the spray equipment at the trial sites were accomplished by using the volume/time method for liquid applications. Before each application, the spray equipment and the sprayer speed were calibrated to deliver an average volume of spray mixture per unit time at a given pressure resulting in the desired spray volume per hectare.

The formulations F9600-4 SC and F9600-21 CS were only mixed with water. No adjuvant was added to the spray mixture. The target dose rate of active substance bixlozone was 375 g a.s./ha; this rate is the same as the maize use submitted in the CP dossier. The test items were applied as follows:

• On plot T1, F9600-4 SC at 0.9384 L/ha of formulated product (F.P.) on bare soil;
• On plot T2, F9600-4 SC at 0.9384 L F.P. on bare soil and then incorporated;
• On plot T3, F9600-21 CS at 0.8906 L F.P. on bare soil;
• On plot T4, F9600-21 CS at 0.8906 L F.P./ha on bare soil and then incorporated.

Trial IT02 consisted of one untreated plot and one treated plot where 0.9384 L FP bixlozone- 4 SC/ha was applied to the bare soil and incorporated.

Applications were made at a water volume of 200 or 350 litres per hectare of mixture according to the GAP.

Table CA.B.8.1.2.1.1-7: Application equipment data

Trial Number	Plot ID	Application No.	Sprayer Configuration	Nozzle Type	Number of Nozzles	Nozzle Spacing (cm)
FR01	T1, T2, T3, T4	A1	Boom	Flat fan	6	50
IT01	T1, T2, T3, T4	A1	Boom	Flat fan	6	50
IT02	T2	A1	Boom	Flat fan	6	50

Table CA.B.8.1.2.1.1-8: Application Data

Trial number	FR01				IT01				IT02
Plot ID	T1	T2	T3	T4	T1	T2	T3	T4	T2
Actual application date	05/06/2015	05/06/2015	05/06/2015	05/06/2015	30/06/2015	30/06/2015	30/06/2015	30/06/2015	05/09/2016
Target application rate (L FP/ha)	0.9384	0.9384	0.8906	0.8906	0.9384	0.9384	0.8906	0.8906	0.9384
Actual rate amount (L FP/ha)	0.943	0.951	0.907	0.883	0.941	0.933	0.895	0.893	0.946
Percent of deviation (%) (2)	0.5	1.3	1.8	-0.9	0.3	-0.6	0.5	-0.3	0.8
Nominal rate amount (g as/ha)	391.48	394.8	-	-	390.65	387.33	-	-	392.72
Spray volume applied (L/ha)	-	-	367.19	357.47	-	-	362.33	361.52	-
Treated area (m²)	200.9	202.7	203.6	198.2	351.1	347.8	351.7	351.1	352.8
Total spray mixture (mL)	225	225	225	225	180	180	180	180	180
Amount of test item added to spray mixture	6000	6000	6000	6000	7500	7500	7500	7500	7500
Spray mixture remaining (mL)	31.248	31.248	30.992	30.991	22.321	22.321	22.137	22.137	22.321
Spray mixture applied to plot area (mL)	1480	1440	1420	1540	1180	1240	1170	1180	1150

Conditions of application

The applications were carried out within two hours of mixing the spray solution and performed under conditions typical for the crop with either no wind or a light wind of less than 3 m/s.

Table CA.B.8.1.2.1.1-9: Environmental conditions at application

Trial No.	FR01				IT01				IT02
Actual Application Date	05/06/2015				30/06/2015				05/09/2016
Plot N°	T1	T2	T3	T4	T1	T2	T3	T4	T2
Time of Mixing	07:00	08:40	07:40	07:55	11:14	08:30	13:10	09:06	09:20
Time of Application	07:25	08:50*	07:45	08:20*	11:27	08:51*	13:19	09:24*	9:45*
Air Temp. (°C)	20.7				29.8	27.7	29.8	28.7	27.1
Wind Speed (m/s)	0				1.7	0.9	1.3	0.9	0.7
Wind Direction (Origin)	NAP				E				E
Relative Humidity (%)	81				29	37	24	30	60
Cloud Cover (%)	0				0				70
Soil Surface Moisture	Dry				Dry				Dry
Soil Temp. at	22.3				27.2	25.9	27.4	25.8	26.5

Trial No.	FR01	IT01				IT02
10 cm (°C)						
1st Rainfall after application within 3h and Amount (mm)	0	0				0

* Formulated product on T2 and T4 was incorporated in the first 7 cm soil top layer just after application

SAMPLING PROCEDURES

Soil microbial biomass determination

In the treated plot, soil (*ca* 0-30 cm depth) was taken from a minimum 10 different areas to obtain a bulk soil specimen of 4.0 kg minimum.

The applicant states the specimens were placed into polyethylene (PE) bags and loosely secured with rubber bands. Soil specimens were kept fresh in the field until they were stored at the field test site away from heat. The specimens were shipped within 72 hours after collection. Packaging was used to maintain a cool/fresh environment for the duration of transit, and cores were placed in freezers with a maximum of 8 h after beginning of sampling.

Spray mixture

Samples of 10 mL of homogenised spray mixture before and after application were taken on the application day at each test site. After collection in the field, they were transported with dry ice to be stored in freezers with 8 hours after end of application. Specimens were stored frozen in pre-labelled 50 mL glass jars and bagged with PE bags. Specimens were stored deep-frozen at target temperature below -18°C until shipment. Specimens were shipped frozen at a temperature - 18° C, by freezer truck to the analytical laboratory.

Petri-dishes

To confirm the amount of active substance applied, 10 petri-dishes, filled with 20 g of dried and sieved soil per sub plot, were placed in treated plots and opened just before application. They were closed and collected immediately after application. Once collected, specimens were stored cold with dry ice and placed in freezer storage within 8 hours after application. Specimens were stored deep-frozen at target temperature below -18°C until shipment to the analytical laboratory. Shipment occurred under frozen conditions at a temperature - 18° C, by freezer truck.

The applicant states some deviation occurred where the five petri-dishes taken for each sub-plot were incorrectly combined into a single sample for analysis. Therefore, there were 24 petri dish samples for analysis instead of 120. Because of this, the applicant states the application homogeneity cannot be checked within each sub-plot (however spray quality can be compared between sub plots). The CA does not consider this to have impacted upon the outcomes of the study.

Soil dissipation specimens

Zero contamination soil sampling equipment with acetate tubes were used for collection of soil cores. Only tubes full of soil were considered acceptable to provide specimens. One specimen comprised 10 tubes. Sampling holes were filled with uncontaminated soil to prevent wash off of treated soil to lower depths. A stick was used to indicate the location of each hole made in the sub plot used. Acetate tubes were closed using coloured caps to identify the top.

The cores were brought to field test site for freezing, being placed in horizontal position in order to avoid residue migration prior to shipment. The cores were placed in freezers with a maximum of 8 h

after beginning of sampling. The cores in their acetate tubes were put together into large heavy PE bags at each sampling date. Untreated and treated specimens were bagged separately.

Soil specimens were kept cool in the field until they were frozen (at a target temperature below -18°C) at the field test site until shipment to the analytical laboratory. The applicant states the time 0, 60 and 91 day samples at site FR01 were kept ambient in the field, instead of being stored cool, until storage at frozen conditions. Similarly, at site IT01, gel packs were not used for transportation of the time 0, 7, 367 and 542 day samples. However, because freezing took place within 12 hours of collection in every case, the CA does not consider this to have significantly impacted on the outcomes of the study.

Similarly, the CA notes the FR01 day 0, day 60 and day 91, soil specimens were stored at ambient temperature in the field for up to 9 hours before freezing. However, again the CA does not consider to have significantly impacted the study results.

ANALYTICAL METHOD

Soil specimens were analysed for bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide using analytical method CAM-0151/001; see Vol 3 CA, B5 for further information. Petri dish soil specimens were analysed for bixlozone only using the same method

Residues of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide were extracted from the soil by shaking with a QuEChERS salt mixture (EN method 15662) in a mixture of water and acetonitrile containing 0.1% formic acid. Following C-18 dispersive solid-phase extraction, an aliquot of the extract was diluted with water and acetic acid and analysed by high performance liquid chromatography with mass spectrometric detection (LC-MS/MS).

In soil, the LOQ for all analytes was set at 0.005 mg/kg and the LOD was set at 0.0015 mg/kg.

Spray solutions were analysed for levels of bixlozone. The solutions were diluted by a factor of 2500 with methanol. They were then further diluted by a factor of 100 with acetonitrile: water (25:75) containing 0.05% acetic acid. The diluted specimens were then analysed by LC-MS/MS, under the conditions used for soil specimens. The analytical batch contained one methanol control specimen, two methanol control specimens fortified at 0.3 mg/mL, two methanol control specimens fortified at 3.0 mg/kg and a reagent blank.

The laboratory reports specimens from the field were received frozen and in good condition. They were transferred to a freezer set to maintain a temperature of ≤-18°C, where they were kept at all times unless removed for preparation or analysis.

The validity of the analytical method was tested by determining the recoveries of fortified control samples. As shown in Table CA.B.**8.1.2.1.1-10**, the mean recoveries were within the OECD recommended range and so the analytical method is accepted by the CA.

Table CA.B.8.1.2.1.1-10: Recovery of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide from fortified control samples

Trial		**FR01**			**IT01**			**IT02**		
Fortification level (mg/kg)	**Recovery**	**Bixlozone**	**2,4-DBA**	**Bixlozone-3-OH-Prop**	**Bixlozone**	**2,4-DBA**	**Bixlozone-3-OH-Prop**	**Bixlozone**	**2,4-DBA**	**Bixlozone-3-OH-Prop**
0.005	Mean (%)	97	94	97	100	94	94	97	94	94
	RSD (%)	7.9	9.1	5.1	6.4	7.2	7.5	8.2	9.4	7.7
0.05	Mean (%)	99	96	97	102	94	96	99	96	99
	RSD (%)	12.0	11.2	6.6	8.0	7.3	7.0	6.8	6.9	5.3
Overall mean (%)		98	95	97	101	94	95	98	95	96
Overall RSD (%)		10.1	10.2	5.8	7.3	7.2	7.3	7.4	8.1	6.9

2,4-DBA = 2,4-dichlorobenzoic acid
bixlozone-3-OH-prop. = bixlozone-3-OH-propanamide

RESULTS

The residue results are presented in Table CA.B.**8.1.2.1.1-11** to Table CA.B.**8.1.2.1.1-15** below. For site FR01, bixlozone residues >LOQ were only detected in the 0-10 cm horizon, whereas, for sites IT01 and IT02, residues >LOQ were also detected in the 10-20 cm and 20-30 cm horizons as well.

The CA notes at site IT02, bixlozone residues were detected in the samples taken three days prior to application (Table CA.B.**8.1.2.1.1-15**). The CA queried these results with the applicant. The study director confirmed these results were not due to a wrong entry, instead sample contamination in the laboratory is suspected. The applicant also notes the untreated plot results were all <LOQ and the mean result of the samples three days prior to application was also <LOQ. The CA accepts the applicant's justification and does not consider this issue to affect the validity of the study. Furthermore, the -3 DAT results are not used in the kinetic evaluation and so there is no impact on the degradation rates derived.

The metabolite 2,4-dichlorobenze was detected in the 0 – 10 cm horizons (but not lower) at sites FR01 and IT01. The 2,4-DBA residues at each time point and sub-plot were compared against the time 0 bixlozone (0 - 30 cm) residue sub-plot values to determine the maximum occurrence. Although higher concentrations of 2,4-DBA were observed, the maximum occurrence of 2,4-DBA occurred at site IT01, sub-plot a, 14 DALA where the 0.0099 mg/kg residue value was equivalent to 16.98% of the time 0 bixlozone value (0.0583 mg/kg) on a mass basis; this is equivalent to 24.37% on a molar basis.

The metabolite 3-OH propenamide was not observed at levels above LOD in any soil sample over the 0-30 cm soil horizon at any test site.

Table CA.B.8.1.2.1.1-11: Residues of bixlozone and 2,4-dichlorobenzoic acid in FR01 trial plots T1 and T2

Sampling (DALA)	Sub-plot	bixlozone and 2,4-D residue in 0-30 cm horizons (mg/kg dwt)*											
		Treatment											
		T1 Bixlozone-4-SC, surface application						T2 Bixlozone-4-SC, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-8	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.2276	(0.0022)	<LOD	<LOD	<LOD	<LOD	0.2328	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1466	<LOD	<LOD	<LOD	<LOD	<LOD	0.2739	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1539	<LOD	<LOD	<LOD	<LOD	<LOD	0.2199	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1012	(0.0015)	<LOD	0.0102	<LOD	<LOD	0.2171	(0.0117)	<LOD	<LOD	<LOD	<LOD
	b	0.1446	<LOD	<LOD	0.0116	<LOD	<LOD	0.2491	(0.0025)	<LOD	<LOD	<LOD	<LOD
	c	0.1839	<LOD	<LOD	0.0159	<LOD	<LOD	0.1887	(0.0016)	<LOD	<LOD	<LOD	<LOD
6	a	0.1175	<LOD	(0.0022)	0.0234	<LOD	<LOD	0.2022	<LOD**	<LOD	<LOD	<LOD	<LOD
	b	0.1079	<LOD	<LOD	0.0211	<LOD	<LOD	0.1905	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1066	<LOD	<LOD	0.0187	<LOD	<LOD	0.1341	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.1276	(0.0018)	<LOD	(0.0041)	<LOD	<LOD	0.2323	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0794	(0.0027)	<LOD	(0.0033)	<LOD	<LOD	0.2834	(0.0015)	<LOD	<LOD	<LOD	<LOD
	c	0.0847	<LOD	<LOD	(0.0026)	<LOD	<LOD	0.3029	<LOD	<LOD	<LOD	<LOD	<LOD
28	a	0.0867	<LOD	<LOD	0.0057	<LOD	<LOD	0.1324	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0945	<LOD	<LOD	0.0065	<LOD	<LOD	0.1360	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1030	<LOD	<LOD	(0.0036)	<LOD	<LOD	0.1364	<LOD	<LOD	<LOD	<LOD	<LOD
60	a	0.0460	<LOD	<LOD	<LOD	<LOD	<LOD	0.0719	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0380	<LOD	<LOD	(0.0025)	<LOD	<LOD	0.0863	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0401	<LOD	<LOD	(0.0021)	<LOD	<LOD	0.0656	<LOD	<LOD	<LOD	<LOD	<LOD
91	a	0.0146	<LOD	<LOD	<LOD	<LOD	<LOD	0.0934	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0100	<LOD	<LOD	<LOD	<LOD	<LOD	0.0766	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0398	<LOD	<LOD	<LOD	<LOD	<LOD	0.0765	<LOD	<LOD	<LOD	<LOD	<LOD

179	a	0.0120	<LOD	<LOD	<LOD	<LOD	<LOD	0.0483	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0125	<LOD	<LOD	<LOD	<LOD	<LOD	0.0393	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0090	<LOD	<LOD	<LOD	<LOD	<LOD	0.0521	<LOD	<LOD	<LOD	<LOD	<LOD
285	a	0.0143	<LOD	<LOD	<LOD	<LOD	<LOD	0.0145	<LOD	<LOD	<LOD	<LOD	<LOD
	b	(0.0033)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0252	<LOD	<LOD	<LOD	<LOD	<LOD
	c	(0.0042)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0348	(0.0018)	<LOD	<LOD	<LOD	<LOD
369	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	0.0178	<LOD	<LOD	<LOD	<LOD	<LOD
	b	(0.0017)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0174	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	0.0124	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery
** This value is reported as 0.0008 mg/kg in the study report; the CA has corrected it to "<LOD"
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.1-12: Residues of bixlozone and 2,4-dichlorobenzoic acid in FR01 trial plots T3 and T4

Sampling (DALA)	**Sub-plot**	**bixlozone and 2,4-D residue in 0-30 cm horizons (mg/kg dwt)***											
		Treatment											
		T3 F9600-21 CS, surface application						**T4** F9600-21 CS, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-8	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.4778	0.0051	(0.0022)	<LOD	<LOD	<LOD	0.2144	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1690	(0.0024)	<LOD	<LOD	<LOD	<LOD	0.2545	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.2270	(0.0021)	<LOD	<LOD	<LOD	<LOD	0.1950	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1126	(0.0019)	<LOD	0.0156	<LOD	<LOD	0.2401	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0846	<LOD	<LOD	0.0147	<LOD	<LOD	0.1771	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1212	(0.0020)	<LOD	0.0228	<LOD	<LOD	0.2087	<LOD	<LOD	<LOD	<LOD	<LOD

6	a	0.1083	<LOD	<LOD	0.0181	<LOD	<LOD	0.1363	<LOD	<LOD	(0.0023)	<LOD	<LOD
	b	0.1672	<LOD	<LOD	0.0206	<LOD	<LOD	0.1883	<LOD	(0.0069)	<LOD	<LOD	<LOD
	c	0.0635	<LOD	<LOD	0.0165	<LOD	<LOD	0.1574	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.1000	<LOD	<LOD	(0.0038)	<LOD	<LOD	0.2381	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1100	<LOD	<LOD	(0.0040)	<LOD	<LOD	0.3105	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0894	<LOD	(0.0043)	(0.0030)	<LOD	<LOD	0.2355	<LOD	<LOD	<LOD	<LOD	<LOD
28	a	0.0888	<LOD	<LOD	0.0058	<LOD	<LOD	0.2454	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0854	<LOD	<LOD	0.0085	<LOD	<LOD	0.2548	<LOD	<LOD	(0.0015)	<LOD	<LOD
	c	0.0625	<LOD	<LOD	(0.0029)	<LOD	<LOD	0.2709	<LOD	<LOD	<LOD	<LOD	<LOD
60	a	0.0617	<LOD	<LOD	<LOD	<LOD	<LOD	0.1736	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0534	(0.0030)	<LOD	(0.0025)	<LOD	<LOD	0.1328	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0246	<LOD	<LOD	(0.0022)	<LOD	<LOD	0.1455	<LOD	<LOD	<LOD	<LOD	<LOD
91	a	0.0397	<LOD	<LOD	<LOD	<LOD	<LOD	0.0836	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0478	<LOD	<LOD	<LOD	<LOD	<LOD	0.1357	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0297	<LOD	<LOD	<LOD	<LOD	<LOD	0.1154	<LOD	<LOD	<LOD	<LOD	<LOD
179	a	0.0293	<LOD	<LOD	<LOD	<LOD	<LOD	0.0864	(0.0026)	<LOD	<LOD	<LOD	<LOD
	b	0.0188	<LOD	<LOD	<LOD	<LOD	<LOD	0.1199	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0165	<LOD	<LOD	<LOD	<LOD	<LOD	0.1405	(0.0044)	<LOD	<LOD	<LOD	<LOD
285	a	0.0328	<LOD	<LOD	<LOD	<LOD	<LOD	0.0640	<LOD	(0.0016)	<LOD	<LOD	<LOD
	b	0.0089	<LOD	<LOD	<LOD	<LOD	<LOD	0.0410	(0.0019)	<LOD	<LOD	<LOD	<LOD
	c	0.0066	(0.0023)	<LOD	<LOD	<LOD	<LOD	0.1559	(0.0023)	<LOD	<LOD	<LOD	<LOD
369	a	0.0094	<LOD	<LOD	<LOD	<LOD	<LOD	0.0869	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0180	<LOD	<LOD	<LOD	<LOD	<LOD	0.0959	(0.0048)	<LOD	<LOD	<LOD	<LOD
	c	0.0108	<LOD	<LOD	<LOD	<LOD	<LOD	0.0956	(0.0019)	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.1-13: Residues of bixlozone and 2,4-dichlorobenzoic acid in IT01 T1 and T2 trial plots

Sampling (DALA)	Sub-plot	bixlozone and 2,4-D residue in 0-30 cm horizons (mg/kg dwt)*											
		Treatment											
		T1 Bixlozone-4-SC, surface application						T2 Bixlozone-4-SC, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-1	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.0583	<LOD	<LOD	<LOD	<LOD	<LOD	0.1270	<LOD	(0.0017)	<LOD	<LOD	<LOD
	b	0.5065	0.0378	(0.0036)	(0.0017)	<LOD	<LOD	0.1272	<LOD	0.0077	<LOD	<LOD	<LOD
	c	0.2354	<LOD	(0.0040)	<LOD	<LOD	<LOD	0.0371	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.0723	<LOD	0.0062	(0.0020)	<LOD	<LOD	0.1229	0.0056	<LOD	<LOD	<LOD	<LOD
	b	0.2686	<LOD	(0.0024)	0.0075	<LOD	<LOD	0.1296	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.2857	<LOD	(0.0026)	0.0075	<LOD	<LOD	0.1159	<LOD	<LOD	<LOD	<LOD	<LOD
7	a	0.1314	<LOD	(0.0021)	0.0089	<LOD	<LOD	0.2234	<LOD	(0.0019)	(0.0015)	<LOD	<LOD
	b	0.3151	<LOD	(0.0016)	0.0161	<LOD	<LOD	0.1376	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1419	(0.0027)	<LOD	0.0093	<LOD	<LOD	0.0880	<LOD	<LOD	<LOD	<LOD	<LOD
14	a	0.1273	<LOD	<LOD	0.0099	<LOD	<LOD	0.1413	(0.0016)	0.0287	(0.0015)	<LOD	<LOD
	b	0.1487	0.0423	<LOD	0.0112	(0.0028)	<LOD	0.1561	0.0090	<LOD	(0.0016)	<LOD	<LOD
	c	0.1410	(0.0020)	<LOD	0.0109	<LOD	<LOD	0.1204	<LOD	<LOD	<LOD	<LOD	<LOD
30	a	0.0620	(0.0018)	<LOD	0.0078	<LOD	<LOD	0.1368	(0.0019)	<LOD	(0.0029)	<LOD	<LOD
	b	0.1475	0.0190	<LOD	0.0155	(0.0019)	<LOD	0.0828	<LOD	<LOD	(0.0016)	<LOD	<LOD
	c	0.1006	<LOD	<LOD	0.0194	<LOD	<LOD	0.1596	<LOD	<LOD	(0.0031)	<LOD	<LOD
58	a	0.0504	<LOD	<LOD	<LOD	<LOD	<LOD	0.1020	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0618	<LOD	<LOD	<LOD	<LOD	<LOD	0.2409	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0768	(0.0027)	<LOD	<LOD	<LOD	<LOD	0.1254	<LOD	<LOD	<LOD	<LOD	<LOD
91	a	0.0413	<LOD	<LOD	<LOD	<LOD	<LOD	0.0844	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0618	<LOD	<LOD	<LOD	<LOD	<LOD	0.1497	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0640	<LOD	<LOD	<LOD	<LOD	<LOD	0.0589	<LOD	<LOD	<LOD	<LOD	<LOD
174	a	0.0140	<LOD	<LOD	<LOD	<LOD	<LOD	0.1344	<LOD	<LOD	<LOD	<LOD	<LOD

	b	0.0256	<LOD	<LOD	<LOD	<LOD	<LOD	0.1227	(0.0034)	<LOD	<LOD	<LOD	<LOD
	c	0.0384	<LOD	<LOD	<LOD	<LOD	<LOD	0.1363	0.0068	<LOD	<LOD	<LOD	<LOD
	a	0.0097	(0.0015)	<LOD	<LOD	<LOD	<LOD	0.0673	(0.0031)	<LOD	<LOD	<LOD	<LOD
269	b	0.0197	(0.0027)	<LOD	<LOD	<LOD	<LOD	0.0451	0.0188	<LOD	<LOD	<LOD	<LOD
	c	0.0171	(0.0025)	<LOD	<LOD	<LOD	<LOD	0.0489	<LOD	<LOD	<LOD	<LOD	<LOD
	a	0.0078	<LOD	<LOD	<LOD	<LOD	<LOD	0.0294	0.0086	<LOD	<LOD	<LOD	<LOD
368	b	(0.0050)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0584	0.0168	<LOD	<LOD	<LOD	<LOD
	c	(0.0030)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0241	(0.0040)	<LOD	<LOD	<LOD	<LOD
	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0177	<LOD	<LOD	<LOD	<LOD	<LOD
454	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0170	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0275	<LOD	<LOD	<LOD	<LOD	<LOD
	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0074	<LOD	0.0130	<LOD	<LOD	<LOD
542	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0216	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0116	<LOD	(0.0031)	<LOD	<LOD	<LOD
	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0090	<LOD	<LOD	<LOD	<LOD	<LOD
640	b	n.a	n.a	n.a	n.a	n.a	n.a	(0.0035)	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0095	<LOD	<LOD	<LOD	<LOD	<LOD
	a	n.a	n.a	n.a	n.a	n.a	n.a	(0.0044)	<LOD	<LOD	<LOD	<LOD	<LOD
736	b	n.a	n.a	n.a	n.a	n.a	n.a	(0.0048)	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	(0.0048)	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery.
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.1-14: Residues of bixlozone and 2,4-dichlorobenzoic acid in IT01 T3 and T4 trial plots

Sampling (DALA)	Sub-plot	bixlozone and 2,4-D residue in 0-30 cm horizons (mg/kg dwt)*											
		Treatment											
		T3 F9600-21 CS, surface application						T4 F9600-21 CS, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-1	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.1526	<LOD	<LOD	<LOD	<LOD	<LOD	0.2692	0.0470	0.0286	<LOD	<LOD	<LOD
	b	0.3598	<LOD	(0.0046)	<LOD	<LOD	<LOD	0.1529	(0.0023)	0.0234	<LOD	<LOD	<LOD
	c	0.3650	<LOD	0.0087	<LOD	<LOD	<LOD	0.2407	<LOD	(0.0029)	<LOD	<LOD	<LOD
3	a	0.1398	<LOD	(0.0022)	0.0078	<LOD	<LOD	0.2425	<LOD	<LOD	(0.0015)	<LOD	<LOD
	b	0.1526	<LOD	(0.0030)	0.0115	<LOD	<LOD	0.1568	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1440	0.0078	0.0120	0.0081	<LOD	<LOD	0.1199	0.0195	<LOD	<LOD	<LOD	<LOD
7	a	0.0852	0.0108	(0.0041)	0.0099	(0.0018)	<LOD	0.1578	<LOD	(0.0037)	(0.0017)	<LOD	<LOD
	b	0.1547	<LOD	(0.0030)	0.0245	<LOD	<LOD	0.1350	0.0180	0.0065	<LOD	<LOD	<LOD
	c	0.1164	<LOD	(0.0021)	0.0163	<LOD	<LOD	0.1544	(0.0018)	(0.0018)	<LOD	<LOD	<LOD
14	a	0.1177	<LOD	(0.0016)	0.0195	<LOD	<LOD	0.1467	0.0121	(0.0017)	(0.0018)	<LOD	<LOD
	b	0.1229	<LOD	(0.0017)	0.0252	<LOD	<LOD	0.2935	<LOD	(0.0021)	<LOD	<LOD	<LOD
	c	0.0903	<LOD	(0.0018)	0.0168	<LOD	<LOD	0.2230	<LOD	<LOD	<LOD	<LOD	<LOD
30	a	0.0548	(0.0031)	<LOD	<LOD	0.0142	<LOD	0.1761	<LOD	<LOD	(0.0021)	<LOD	<LOD
	b	0.0434	<LOD	<LOD	<LOD	0.0156	<LOD	0.0604	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0634	<LOD	<LOD	<LOD	0.0157	<LOD	0.1624	<LOD	<LOD	<LOD	<LOD	<LOD
58	a	0.0491	(0.0047)	<LOD	<LOD	<LOD	<LOD	0.1560	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1107	0.0122	<LOD	<LOD	<LOD	<LOD	0.1400	0.0022	<LOD	<LOD	<LOD	<LOD
	c	0.0373	<LOD	<LOD	<LOD	<LOD	<LOD	0.1595	<LOD	<LOD	<LOD	<LOD	<LOD
91	a	0.0233	<LOD	<LOD	<LOD	<LOD	<LOD	0.1494	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0407	<LOD	<LOD	<LOD	<LOD	<LOD	0.1360	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0374	<LOD	<LOD	<LOD	<LOD	<LOD	0.0920	<LOD	<LOD	<LOD	<LOD	<LOD
174	a	0.0507	<LOD	<LOD	<LOD	<LOD	<LOD	0.1265	0.0257	<LOD	<LOD	<LOD	<LOD

	b	0.0228	<LOD	<LOD	<LOD	<LOD	<LOD	0.0876	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0207	<LOD	<LOD	<LOD	<LOD	<LOD	0.1020	<LOD	<LOD	<LOD	<LOD	<LOD
269	a	0.0075	<LOD	<LOD	<LOD	<LOD	<LOD	0.0734	0.0256	<LOD	<LOD	<LOD	<LOD
	b	0.0080	<LOD	<LOD	<LOD	<LOD	<LOD	0.1300	0.0284	<LOD	<LOD	<LOD	<LOD
	c	0.0208	0.0228	<LOD	<LOD	<LOD	<LOD	0.0973	0.0146	<LOD	<LOD	<LOD	<LOD
368	a	0.0116	(0.0020)	<LOD	<LOD	<LOD	<LOD	0.0625	0.0750	<LOD	<LOD	<LOD	<LOD
	b	(0.0042)	<LOD	<LOD	<LOD	<LOD	<LOD	0.1032	0.0073	<LOD	<LOD	<LOD	<LOD
	c	0.0080	<LOD	<LOD	<LOD	<LOD	<LOD	0.0798	(0.0049)	<LOD	<LOD	<LOD	<LOD
454	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0471	<LOD	<LOD	<LOD	<LOD	<LOD
	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0362	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0326	<LOD	<LOD	<LOD	<LOD	<LOD
542	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0537	0.0203	0.0054	<LOD	<LOD	<LOD
	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0361	0.0211	0.0064	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0778	0.0201	<LOD	<LOD	<LOD	<LOD
640	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0233	0.0055	<LOD	<LOD	<LOD	<LOD
	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0495	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0276	<LOD	<LOD	<LOD	<LOD	<LOD
736	a	n.a	n.a	n.a	n.a	n.a	n.a	0.0200	<LOD	<LOD	<LOD	<LOD	<LOD
	b	n.a	n.a	n.a	n.a	n.a	n.a	0.0190	<LOD	<LOD	<LOD	<LOD	<LOD
	c	n.a	n.a	n.a	n.a	n.a	n.a	0.0090	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery.
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.1-15: Residues of bixlozone in IT02 T2 trial plot

Sampling (DALA)	Sub-plot	bixlozone and 2,4-D residue in 0-30 cm horizons (mg/kg dwt)*					
		Treatment					
		T2 Bixlozone-4-SC, surface application					
		Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30
-3	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	(0.0019)	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0061	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.6716	0.0072	0.0217	<LOD	<LOD	<LOD
	b	0.5901	0.0274	0.0233	<LOD	<LOD	<LOD
	c	0.5868	0.0165	0.0264	<LOD	<LOD	<LOD
3	a	0.4496	0.0123	0.0095	(0.0033)	<LOD	<LOD
	b	0.2972	0.0130	0.0881	<LOD	<LOD	<LOD
	c	0.3669	0.0567	0.0696	<LOD	<LOD	<LOD
7	a	0.2434	0.0385	(0.0041)	(0.0034)	<LOD	<LOD
	b	0.2102	0.0289	0.0815	<LOD	<LOD	<LOD
	c	0.2936	0.0210	0.0058	(0.0019)	<LOD	<LOD
14	a	0.2484	<LOD	0.0075	(0.0027)	<LOD	<LOD
	b	0.1880	<LOD	(0.0043)	(0.0015)	<LOD	<LOD
	c	0.2559	0.0056	(0.0042)	<LOD	<LOD	<LOD
30	a	0.1037	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1329	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1147	(0.0029)	<LOD	<LOD	<LOD	<LOD
60	a	0.0298	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0760	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1834	<LOD	<LOD	<LOD	<LOD	<LOD
88	a	0.0705	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1281	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0783	<LOD	<LOD	<LOD	<LOD	<LOD
186**	a	0.0769	0.0168	<LOD	<LOD	<LOD	<LOD
	b	0.0771	0.0088	0.0094	<LOD	<LOD	<LOD
	c	0.0574	0.0142	0.0097	<LOD	<LOD	<LOD
263**	a	0.0069	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0170	0.0021	<LOD	<LOD	<LOD	<LOD
	c	0.0312	0.0064	0.0072	<LOD	<LOD	<LOD
360	a	(0.0026)	<LOD	<LOD	<LOD	<LOD	<LOD
	b	(0.0021)	<LOD	<LOD	<LOD	<LOD	<LOD
	c	(0.0020)	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery.
**The 30-40 and 40-50 cm soil layers were also analysed for 186 and 263 DALA. No residues of bixlozone above LOD were detected.
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

KINETICS

The kinetic assessment is presented in section 165CA.B.8.1.2.3.

CONCLUSION

In field dissipation trials in southern France and Italy, bixlozone residues were predominantly confined to the 0-10 cm soil layer, but on occasion were observed at lower layers down to 30 cm. Metabolite, 2,4-dichlorobenzoic acid forms in the 0-10 cm surface layer and was generally at higher concentrations in plots without incorporation, but was not observed in deeper soil layers. The maximum 2,4-dichlorobenzoic acid residue detected corresponded to 24.37% of parent (on a molar basis) and therefore is to be classed as a major soil metabolite. Bixlozone-3-OH-propanamide was not observed at levels above LOD in any soil sample over the 0-30 cm soil horizon at any of the trial sites.

CA.B.8.1.2.1.2. Soil dissipation in Germany and France

Report:	KCA 7.1.2.2.1-02 Gemrot, F. (2018b)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Soil Dissipation Study After One Application of F9600-4 SC or F9600-21 CS in Northern Europe (Germany) and Southern Europe (Southern France) - 2015 and 2016
Document No:	SGS Study Number: 15SGS111 FMC Tracking Number: 2015EFT-ISX2156
Guidelines:	-SETAC 1995 -SANCO/3029/99 rev. 4 -EPA (Oct 2008). Fate, Transport and Transformation Test Guidelines. -EFSA Guidance Document for evaluating laboratory and field dissipation studies to obtain DegT50 values of active substances of plant protection products and transformation products of these active substances in soil. EFSA Journal 2014;12(5): 3662. -ISO 10381-6:2009 (handling & storage soil for assessment of microbial biomass) -OECD Guidance Document for Conducting Pesticide Terrestrial Field Dissipation Studies (ENV/JM/MONO(2016)6, released 04 March 2016).
GLP:	Yes

CA Comments:	The following deviations from the guidelines were noted by the CA and are explored in more detail in the main body of text: • Pesticide use history was not submitted for 2012 for site FR02 • Some sampling delays because of weather limitations • One instance where freezing dipped below the prescribed -18ºC • A prolonged instance of soil moisture and temperatures not recorded because of probe failure. Data collected instead from neighbouring weather station. • Period between extraction and analysis overran to 7 days. Higher than the suggested 6 days proven by procedural recovery tests. However, the CA does not consider these issues to have significantly affected the outcomes of the study. **This study is relied upon.**

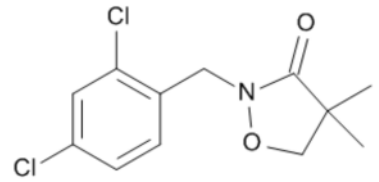
INTRODUCTION

Field studies were conducted in Germany and southern France to determine the dissipation and mobility of residues of bixlozone and its major metabolites in soil, following a single application of the formulated product (F9600-4 SC or F9600-21 CS) to bare soil. At both trial sites, each formulation was applied in autumn 2015 to the bare soil of two trial plots, then incorporated immediately after application in one of the treated plots. Bare soil was maintained throughout the trial period by application of herbicide (glyphosate and/or flumioxazin).

Soil cores were collected to a depth of at least 30 cm prior to application, within three hours of application and then approximately 3, 7, 14, 30, 90, 180, 270 and 365 days after last application. Soil cores were cut into 0-10, 10-20, and 20-30 cm sections for analysis.

MATERIALS

Table CA.B.8.1.2.1.2-1: Test Item

Common name:	Bixlozone
Chemical name ~(IUPAC)	2-(2,4-dichlorobenzyl)-4,4-dimethyl-1,2-oxazolidin-3-one)
Structure:	
CAS number:	81777-95-9
Formulation name:	F9600-4 SC
Main uses:	Herbicide
Formulation type:	Suspension concentrate (SC)
Nominal concentration:	36 % w/w
Nominal density:	1.11g/mL
Batch number:	PL15-0061
Actual concentration:	37.4 % w/w
Formulation Name:	F9600-21 CS
Main uses:	Herbicide
Formulation type:	Capsule suspension (CS)
Nominal concentration:	36.3 % w/w
Nominal density:	1.16 g/mL
Batch number:	PL15-0062
Actual concentration:	34.9 % w/w

Field Data

The applicant states the trial areas were mechanically cleared of all vegetation prior to trial set up and kept free of weeds thereafter by application of herbicide (e.g. glyphosate) at recommended commercial rates. The soil surface was rolled prior to plot set up. The surface of the plots was raked and hoed manually or with a small rotary cultivator at 5-10 cm depth to produce a fine level tilth. The soil was cultivated as fine seed bed before application to get a fine crumb structure. The trial sites did not receive application of maintenance pesticides except for herbicide maintenance. Only glyphosate and/or flumioxazin used at the registered rate were applied. Each trial consisted of 5 plots; 1 untreated control plot (plot U) and 4 treated plots labelled T1 to T4 (bare soil).

The applicant states the selected sites presented no risk of erosion or flooding and were not too stony. The selected sites were loamy sand and sandy loam and allowed sampling to a 0.9 to 1.0 m depth. Plots were not set up in the shadow of neighbouring trees and the sites were not on a slope. The sites have been in cultivation for several years but no soil disinfectant (e.g. dazomet or metam) had been used within the last three years before trial set up. For site GE01, pesticide use history has been provided by the applicant for the preceding three years. However, for site FR02, the applicant has only provided pesticide use history for the preceding two years in the study report, although the applicant does state clomazone was not used in the preceding three years. Of the pesticide use history provided, the CA's chemistry specialist has confirmed no structurally similar compounds were used. Therefore, for site GE01, the CA considers the previous pesticide use to have not significantly impacted upon the outcomes of the study. For site FR02, the pesticides used in the preceding two years were not structurally similar to bixlozone. In response to the CA's request for 2012 FR02 pesticide use history, the applicant confirmed that the test site was the same as site FR01 where pesticide use history was provided. As reported in section CA.B.8.1.2.1.1, Table CA.B.**8.1.2.1.1-5**, the 2012 pesticide use was the same as reported for 2013 and 2014; therefore, the pesticides used were not structurally similar to bixlozone.

Details of the location and physical properties of the sites is provided in
Table CA.B.**8.1.2.1.2-2**. The pesticide use history is provided in Table CA.B.8.1.2.1.2-3, with the cultural treatments and maintenance pesticides in Table CA.B.**8.1.2.1.2-4** and Table CA.B.**8.1.2.1.2-5** respectively.

Table CA.B.8.1.2.1.2-2: Chemical and physical soil characterisation

Field Number	GE01			FR02		
Country	Germany			France		
Region	Lower Saxony			Aquitaine		
City	Emstek			Brannens		
Zip code	49685			33120		
GPS coordinates	N52º52'36" E8º8'47"			N44º31'25" E0º8'26"		
Soil depth	0-30cm	30-45cm	45-60cm	0-30cm	30-45cm	45-60cm
pH water	5.9	6.3	6.4	5.1	5	4.9
Organic Matter (% w/w)	2.14	0.45	0.14	1.07	0.59	0.31
Organic Carbon (% w/w)	1.24	0.26	0.08	0.62	0.34	0.18
CEC (meq/100 g)	6	2.9	2.5	3.9	4	4.9
Maximum WHC (% w/w)	29.6	25.3	23.3	26.9	25.3	27.1
WHC at pF2 (% w/w)	19.5	17.8	01.2	17.9	17.2	16.4
Soil microbial biomass as % organic carbon	Start of study: 1.71 End of study: 1.13			Start of study: 2.10 End of study: 2.48		
Soil properties						
Sand 2.00-0.05 mm % w/w	86	85	84	67	60	56
Silt 0.05-0.002 mm % w/w	9	11	11	27	30	29
Clay < 0.002 mm % w/w	5	4	5	6	10	15
USDA Textural class	Loamy Sand			Sandy Loam		
Sand 2.00-0.063 mm % w/w	84	84	82	62	57	51
Silt 0.063-0.002 mm % w/w	12	12	13	31	33	35
Clay <0.002 mm % w/w	4	4	5	7	10	14
UK Textural class	Loamy Sand			Sandy Loam		

Table CA.B.8.1.2.1.2-3: Pesticide use history

Trial number	**Field crop**	**Date**	**Pesticide applied**		**Field rate / ha**
GE01	Rye	31/10/12	BACARA	diflufenican, flurtamone	1.0 L
		26/04/13	CAPALO	epoxiconazole, fenpropimorph, metrafenone	2.0 L
		26/04/13	CCC 720	chlormequat	1.0 L
		26/04/13	TALIUS	proquinazid	0.1 L
		26/04/13	MODDUS	trinexapac	0.2 L
		28/05/13	AMISTAR OPTI	azoxystrobin, chlorothalonil	2.2 L
		28/05/13	MATADOR	tebuconazole, triadimenol	0.5 L
	Triticale	01/11/13	BACARA	diflufenican, flurtamone	1.0 L
		01/04/14	ALTO 240 EC	Cyproconazole	0.5 L
		01/04/14	CCC 720	chlormequat	0.5 L
		23/04/14	MATADOR	tebuconazole, triadimenol	0.2 L
		23/04/14	INPUT CLASSIC	Prothioconazol, spiroxamine	1.1 L

Trial number	**Field crop**	**Date**	**Pesticide applied**		**Field rate / ha**
		23/04/14	CCC 720	chlormequat	0.4 L
		26/05/14	AMISTAR OPTI	azoxystrobin, chlorothalonil	1.6 L
		26/05/14	SEGURIS	Isopyrazam, epoxiconazole	0.4 L
	Rye	29/10/14	BACARA	diflufenican, flurtamone	1.0 L
		10/04/15	CAPALO	epoxiconazole, fenpropimorph, metrafenone	2.0 L
		10/04/15	CCC 720	chlormequat	1.0 L
		10/04/15	MODDUS	trinexapac	0.2 L
		10/04/15	TALIUS	proquinazid	0.1 L
FR02	Maize	20/04/13	DUAL GOLD SAFENEUR	S-metolachlor + benoxacor	2.0 L
		20/04/13	LAGON	Isoxaflutole + aclonifen	1.0 L
	Maize	11/04/14	DUAL GOLD SAFENEUR	S-metolachlor + benoxacor	2.0 L
		11/04/14	LAGON	Isoxaflutole + aclonifen	1.0 L

Table CA.B.8.1.2.1.2-4: Cultural treatments

Field No.	Date	Description
GE01	02/10/15	Plough with packer
	05/10/15	Rotary harrow and roller
FR02	15/10/15	Harrowing

Table CA.B.8.1.2.1.2-5: Maintenance pesticides

Field No.	Application Date	Product	Active Substance	Application Rate (/ha)	Purpose
GE01	11/05/16	ROUND UP PowerFlex	glyphosate 480 g/L	3.5 L	Herbicide
	23/06/16	ROUND UP PowerFlex	glyphosate 480 g/L	3.5 L	Herbicide
	13/07/16	VOROX F	Flumioxazin 500 g/L	1.2 kg	Herbicide
FR02	15/04/15	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	10/05/15	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	01/06/15	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide
	20/06/15	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide
	10/07/15	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide
	05/08/15	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	15/09/15	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	02/11/15	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	05/03/16	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	04/04/16	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	10/05/16	ROUNDUP	glyphosate 360 g/L	6.0 L	Herbicide
	01/06/16	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide
	17/06/16	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide
	05/07/16	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide

	25/07/16	BASTA F1	glufosinate-ammonium 150 g/L	5.0 L	Herbicide

EXPERIMENTAL DESIGN

Each trial consisted of 5 plots comprising of 1 untreated control plot (U, minimum 3 × 10 m) and 4 treated plots (T1 to T4, minimum 3 × 20 m), with each plot divided into 3 subplots (a, b and c). Each main plot was separated by a 10 meter buffer zone. Plots were divided into 3 sub-plots a, b, and c. Each sub-plot was separated from the next by a 5 meter buffer zone. Around the untreated control plot a 20 meter buffer zone separated it from other plots.

Table CA.B.8.1.2.1.2-6: Trial site data

Trial Number	Plot ID	Plot Width (m)	Plot Length (m)	Plot Area (m^2)	Slope (%)	Distance minimum between untreated and treated plots (m)
GE01	T1, T2, T3, T4	3.0	90.0	270.0	0	≥20
FR02	T1, T2, T3, T4	3.0	63.0	189.0	0	≥20

Daily records were collected by use of a weather station installed on site. Historical weather data were collected from institutional, permanent weather recording stations situated 7 (FR02) and 49 (GE01) km from the field sites. The weather data from the study period was compared against the average minimum and maximum temperatures and rainfall over the preceding 5 years (see Figure CA.B.**8.1.2.1.2-1** to Figure CA.B.**8.1.2.1.2-2**). As can be seen, the averages temperatures were broadly similar to the 5 year norm at each test site, with the exception potentially of November and December, at GE01, where the average minimum temperatures were much higher than the norm. Large fluctuations in monthly rainfall is also observed at each test site, with some months significantly drier than the norm and other months significantly wetter.

Figure CA.B.8.1.2.1.2-1: GE01 comparison of study weather versus the preceding 5 year average

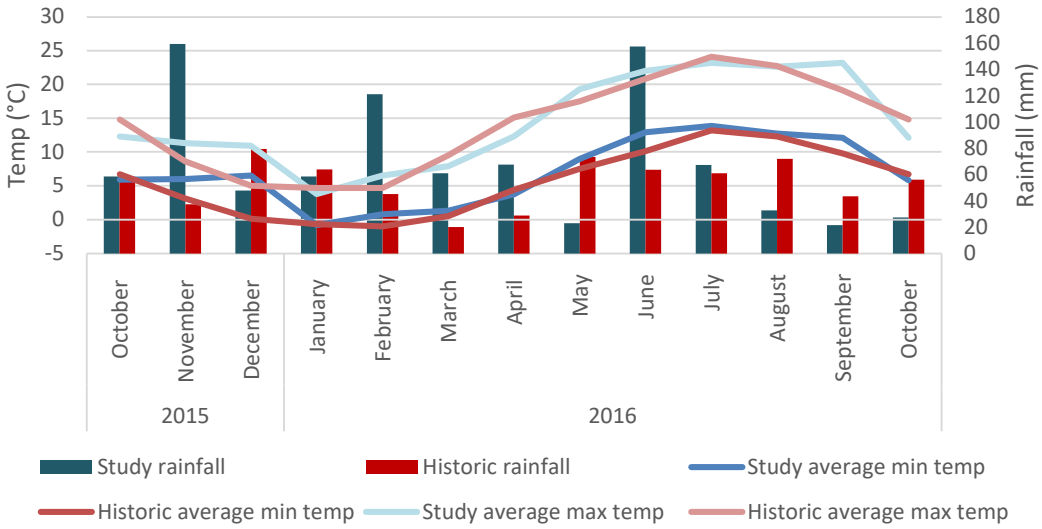
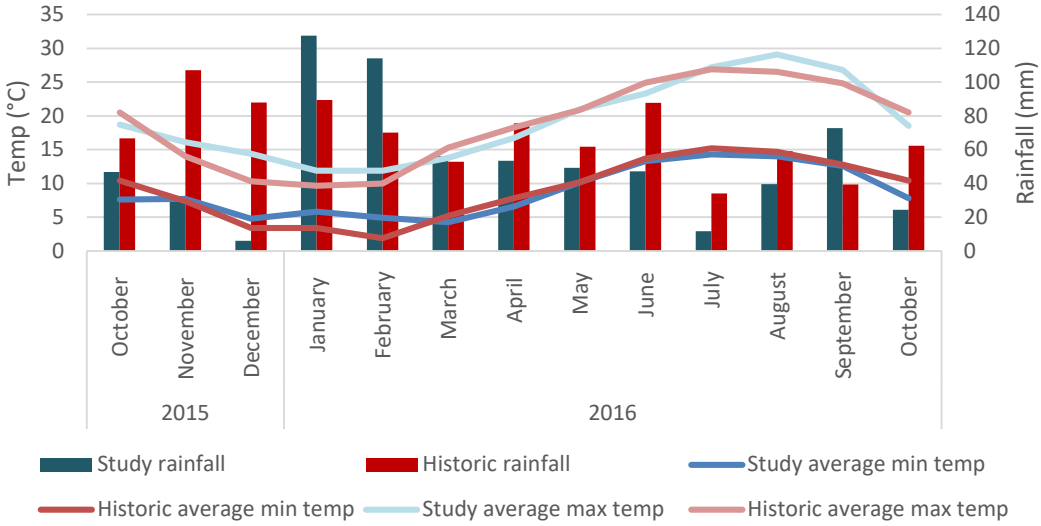


Figure CA.B.8.1.2.1.2-2: FR02 comparison of study weather versus the preceding 5 year average



Application of Test item

The application equipment consisted of a backpack boom sprayer with 6 flat fan nozzles; each was spaced 50cm apart. On each treated plot T1, T2, T3 and T4, a single application was made.

Calibrations of the spray equipment at the trial sites were accomplished by using the volume/time method for liquid applications. Before each application, the spray equipment and the sprayer speed were calibrated to deliver an average volume of spray mixture per unit time at a given pressure resulting in the desired spray volume per hectare.

The formulations F9600-4 SC and F9600-21 CS were only mixed with water. No adjuvant was added to the spray mixture. The target dose rate of active substance bixlozone was 250 g a.s./ha. The CA notes this target application rate is less than that applied in study Gemrot, 2018a (section CA.B.8.1.2.1.1), however, it is in line with the proposed winter wheat/barley (200 g/ha) and oilseed rape (300 g/ha) application rates in the applicant's CP submission. Therefore, the CA accepts the target application rates administered.

The test items were applied as follows:

- T1: F9600-4 SC at 0.63 L formulated product/ha to bare soil
- T2: F9600-4 SC at 0.63 L formulated product/ha to bare soil then incorporated to a depth of 7 cm just after application
- T3: F9600-21 CS at 0.594 L formulated product/ha to bare soil
- T4: F9600-21 CS at 0.594 L formulated product/ha to bare soil then incorporated to a depth of 7 cm just after application

Applications were made at a target water volume of 200 litres (actual volumes 198.5 to 207.4 L) per hectare of spray mixture. The application data is summarised in Table CA.B.**8.1.2.1.2-7**.

Table CA.B.8.1.2.1.2-7: Application data

Trial Number	GE01				FR02			
Plot ID	T1	T2	T3	T4	T1	T2	T3	T4
Actual application date	06/10/2015	06/10/2015	06/10/2015	06/10/2015	20/10/2015	20/10/2015	20/10/2015	20/10/2015
Target application rate (L FP/ha)	0.63	0.63	0.594	0.594	0.63	0.63	0.594	0.594
Actual rate amount (L FP/ha)	0.625	0.632	0.59	0.601	0.654	0.634	0.6	0.598
Percent of deviation (%) (1)	-0.8	0.3	-0.7	1.2	3.8	0.6	1	0.7
Nominal rate amount (g as/ha)	259.463	262.368	238.856	243.309	271.502	263.199	242.904	242.094
Spray volume applied	198.5	200.7	198.5	202.2	207.4	201.1	202.1	201.1
Treated area (m^2)	270	270	270	270	189	189	189	189
Total spray mixture (mL)	6500	6500	6500	6500	5000	5000	5000	5000
Amount of test item added to spray mixture (g)	22.727	22.727	22.394	22.394	17.488	17.485	17.232	17.236
Spray mixture remaining (mL)	1140	1080	1140	1040	1080	1200	1180	1200
Spray mixture applied to plot area (mL)	5360	5420	5360	5460	3920	3800	3820	3800

Conditions of application

The applications were carried out within two hours of mixing the spray solution and performed under conditions typical for the crop with either no wind or a light wind of less than 3 m/s (see Table CA.B.8.1.2.1.2-8).

Table CA.B.8.1.2.1.2-8: Environmental conditions at application

Trial No.	GE01				FR02			
Actual application date	06/10/2015				20/10/2015			
Plot N°	T1	T2	T3	T4	T1	T2	T3	T4
Time of Mixing	13:30	14:05	14:40	15:05	08:30	09:00	09:25	09:50
Time of Application	13:40	14:10	14:45	15:10	08:35	09:10	09:35	10:00
Air temperature (°c)	14.3	14.3	14.3	14.8	7.4			
Wind Speed (m/s)	2	1.4	1.3	1.1	0			
Wind direction (origin)	SE				NAP			
Relative humidity (%)	98				97			
Cloud cover (%)	100				20			
Soil surface moisture	Moist				Moist			
Soil Temp. At 10 cm (°C)	12.9	12.9	13.1	13.2	9.6			
1st Rainfall after application within 24h and amount (mm)	0.4 at 22:20				0			

SAMPLING PROCEDURES

Soil microbial biomass determination

Before the application and at the end of the study (365 ±7 days after application), soil microbial biomass determination was done at each test site. A representative soil specimen of 4.0 kg minimum weight was obtained from at least 10 different areas – about 0-30 cm depth - and randomly collected at each trial site from the treated plot and then combined and mixed to one composite soil sample for

biomass determination. Soil specimens were shipped at ambient temperature to the laboratory for analysis under GLP.

Spray mixture

Samples of 10 mL of homogenised spray mixture, before and after application, were taken with a pipette or a syringe on the application day at each test site. After collection in the field, they were stored with dry ice and then stored in freezers within 8 hours after end of application. Specimens were stored and shipped to the analytical laboratory at below -18°C.

Petri-dishes

15 Petri-dishes filled with 20g of dried and sieved soil per sub-plot were placed in each treated replicate and opened just before application. They were closed and collected immediately after application. After collection in the field, they were stored with dry ice then stored in freezers within 8 hours after end of application. Specimens were stored deep frozen at target temperature below -18°C until shipment to the analytical laboratory. Shipment occurred under frozen conditions at a temperature of -18°C, by freezer truck.

Soil dissipation specimens

Zero contamination soil sampling equipment with acetate tubes were used for collection of soil cores. Only tubes full of soil were considered acceptable to provide specimens. One specimen comprised 10 tubes. Sampling holes were re-filled after sampling with uncontaminated soil to prevent the wash off of treated soil to lower depths. A stick was used to indicate the location of each sampling hole in the sub-plot. Acetate tubes were closed using coloured caps to identify the top and bottom. The cores were brought to the field test site for freezing, being placed in horizontal position to avoid residue migration prior to shipment. The cores were placed in freezers within a maximum of 12h hours after sampling.

The CA notes that the temperature of the freezer was above the stated -18ºC during the storage of the German day 183 specimens (max -12.7°C) for a period of *ca* 15 hours. The applicant states this was due to the high volume of sample material. Given the deviation in temperature was relatively minor and did not continue for a prolonged period of time, the CA does not believe this to have had a significant impact on the study outcomes.

The CA notes soil sample collection was delayed at the day 90 and day 180 FR02 sampling events because of large amounts of rainfall limiting access to field sites (samples collected day 125 and 188 respectively). Furthermore, the FR02 day 270 sampling occasion could only be collected on day 290 due to mechanical auger failure. Also, the depth of a number of core samples collected were also impacted by water saturation at depths >50 cm. However, because revised sampling occasions were undertaken relatively close to the intended sampling occasion and the cores recorded depths of at least 0 – 30 cm (only one sample recorded a residue value >LOQ below 10 cm (see results section below)), the CA considers that these issues will not have significantly impacted on the outcomes of the study.

ANALYTICAL METHOD

Soil specimens were analysed for bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide using analytical method CAM-0151/001; see Vol 3 CA, B5 for further information. Petri dish soil specimens were analysed for bixlozone only using the same method.

Residues of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide were extracted from the soil by shaking with a QuEChERS salt mixture (EN method 15662) in a mixture of water and acetonitrile containing 0.1% formic acid. Following C-18 dispersive solid-phase extraction, an aliquot of the extract was diluted with water and acetic acid and analysed by high performance liquid chromatography with mass spectrometric detection (LC-MS/MS). Spray solutions were analysed for levels of bixlozone.

The solutions were diluted by a factor of 2500 with methanol. They were then further diluted by a factor of 100 with acetonitrile:water (25:75) containing 0.05% acetic acid. The diluted specimens were then analysed by LC-MS/MS, under the conditions used for soil specimens. The analytical batch

contained one methanol control specimen, two methanol control specimens fortified at 0.3 mg/mL, two methanol control specimens fortified at 3.0 mg/kg and a reagent blank.

All field specimens and procedural recoveries were analysed using the quantitation transition for each analyte.

The validity of the analytical method was tested by determining the recoveries of fortified control samples. As shown in Table CA.B.**8.1.2.1.2-9**, the mean recoveries were within the OECD recommended range and so the analytical method is accepted by the CA.

Each analytical batch contained at least one untreated field specimen, an untreated field specimen fortified at LOQ level (0.005 mg/kg), an untreated field specimen fortified at 10 × LOQ level (0.05 mg/kg) and a reagent blank. The LOD was set at 0.0015 mg/kg in soil.

The CA notes that the maximum period between extraction and analysis was up to 7 days, however residues of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanomide in soil extracts were only shown to be stable when stored at 4°C for up to 6 days. The applicant states the procedural recovery data demonstrates the stability over 7 days; the CA agrees with the applicant and does not consider this to have had a significant impact on the study outcomes.

Table CA.B.8.1.2.1.2-9: Recovery of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide from fortified control samples

Trial		**15SGS111 GE01**			**15SGS111 FR02**		
Fortification level (mg/kg)	**Recovery**	**Bixlozone**	**2,4-DBA**	**Bixlozone-3-OH-Prop**	**Bixlozone**	**2,4-DBA**	**Bixlozone-3-OH-Prop**
0.005	Mean (%)	93	89	90	103	93	94
	RSD (%)	11.6	6.6	8.5	10.4	6.4	7.1
0.05	Mean (%)	98	94	96	101	95	97
	RSD (%)	9.8	6.5	7.5	10.0	7.1	8.4
0.5	Mean (%)	98	95	96	106	95	99
	RSD (%)	7.4	6.4	6.1	9.7	6.1	5.9
Overall mean (%)		96	92	93	103	94	96
Overall RSD (%)		10.6	6.8	8.6	10.1	6.6	7.5

2,4-DBA = 2,4-dichlorobenzoic acid
bixlozone-3-OH-prop. = bixlozone-3-OH-propanamide

RESULTS

The 0 – 10 cm horizon residue results are presented in Table CA.B.**8.1.2.1.2-10** to Table CA.B.**8.1.2.1.2-13** below. For site GE01, residues were predominately confined to the 0 – 10 cm horizon, with sporadic residues detected in deeper horizons. For site FR02, residues were only detected in the 0-10 cm horizon.

No metabolite residue values >LOQ were detected at either test site.

Table CA.B.8.1.2.1.2-10: Residues of bixlozone in GE01 trial plots T1 and T2

Sampling (DALA)	Sub-plot	bixlozone and 2,4-DBA residue in 0-30 cm horizons (mg/kg dwt)*											
		Treatment											
		T1 Bixlozone-4-SC, surface application						T2 Bixlozone-4-SC, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-1	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.1259	<LOD	<LOD	<LOD	<LOD	<LOD	0.0737	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1137	<LOD	<LOD	<LOD	<LOD	<LOD	0.0434	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1372	<LOD	<LOD	<LOD	<LOD	<LOD	0.2406	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1805	<LOD	<LOD	<LOD	<LOD	<LOD	0.0475	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0929	<LOD	<LOD	<LOD	<LOD	<LOD	0.1519	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1362	<LOD	<LOD	<LOD	<LOD	<LOD	0.0737	<LOD	<LOD	<LOD	<LOD	<LOD
7	a	0.1812	(0.0023)	<LOD	<LOD	<LOD	<LOD	0.1531	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1494	(0.0020)	(0.0030)	<LOD	<LOD	<LOD	0.1582	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1477	<LOD	<LOD	<LOD	<LOD	<LOD	0.1350	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.3660	<LOD	<LOD	<LOD	<LOD	<LOD	0.0548	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1634	<LOD	<LOD	<LOD	<LOD	<LOD	0.2836	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0968	<LOD	<LOD	<LOD	<LOD	<LOD	0.0882	<LOD	<LOD	<LOD	<LOD	<LOD
29	a	0.1801	<LOD	<LOD	(0.0034)	<LOD	<LOD	0.1031	<LOD	<LOD	(0.0023)	<LOD	<LOD
	b	0.1772	<LOD	<LOD	(0.0031)	<LOD	<LOD	0.1652	<LOD	<LOD	(0.0031)	<LOD	<LOD
	c	0.1750	<LOD	<LOD	(0.0027	<LOD	<LOD	0.1155	<LOD	<LOD	(0.0024)	<LOD	<LOD
58	a	0.1155	<LOD	<LOD	(0.0018)	<LOD	<LOD	0.1629	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1611	(0.0019)	<LOD	<LOD	<LOD	<LOD	0.1196	0.0081	<LOD	(0.0015)	<LOD	<LOD
	c	0.1558	0.0096	<LOD	(0.0015)	<LOD	<LOD	0.0843	<LOD	<LOD	<LOD	<LOD	<LOD
92	a	0.1087	(0.0017)	<LOD	<LOD	<LOD	<LOD	0.1049	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1643	<LOD	<LOD	<LOD	<LOD	<LOD	0.0918	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1011	<LOD	<LOD	<LOD	<LOD	<LOD	0.1714	<LOD	<LOD	<LOD	<LOD	<LOD

183	a	0.0874	<LOD	<LOD	<LOD	<LOD	<LOD	0.1072	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0951	<LOD	<LOD	<LOD	<LOD	<LOD	0.0737	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0796	<LOD	<LOD	<LOD	<LOD	<LOD	0.0819	<LOD	<LOD	<LOD	<LOD	<LOD
267	a	0.0639	<LOD	(0.0017)	<LOD	<LOD	<LOD	0.0309	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0622	<LOD	<LOD	<LOD	<LOD	<LOD	0.0440	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0451	<LOD	<LOD	<LOD	<LOD	<LOD	0.0181	<LOD	<LOD	<LOD	<LOD	<LOD
359	a	0.0162	(0.0023)	<LOD	<LOD	<LOD	<LOD	0.0113	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0181	<LOD	<LOD	<LOD	<LOD	<LOD	0.0231	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0183	(0.0032)	<LOD	<LOD	<LOD	<LOD	0.0099	(0.0018)	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery.
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.2-11: Residues of bixlozone in GE01 trial plots T3 and T4

Sampling (DALA)	**Sub-plot**	**bixlozone and 2,4-DBA residue in 0-30 cm horizons (mg/kg dwt)***											
		Treatment											
		T3 F9600-21 CS, surface application						**T4** F9600-21 CS, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-1	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.1468	<LOD	<LOD	<LOD	<LOD	<LOD	0.1299	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1416	(0.0025)	0.0053	<LOD	<LOD	<LOD	0.1338	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0563	<LOD	<LOD	<LOD	<LOD	<LOD	0.1812	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1881	<LOD	<LOD	<LOD	<LOD	<LOD	0.0852	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0718	<LOD	<LOD	<LOD	<LOD	<LOD	0.1125	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1471	<LOD	<LOD	<LOD	<LOD	<LOD	0.1307	<LOD	<LOD	<LOD	<LOD	<LOD
7	a	0.2560	<LOD	<LOD	<LOD	<LOD	<LOD	0.0821	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1469	<LOD	<LOD	<LOD	<LOD	<LOD	0.0792	<LOD	<LOD	<LOD	<LOD	<LOD

	c	0.1527	<LOD	<LOD	<LOD	<LOD	<LOD	0.1336	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.1388	<LOD	<LOD	<LOD	<LOD	<LOD	0.1920	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0908	<LOD	<LOD	<LOD	<LOD	<LOD	0.2569	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.3547	<LOD	<LOD	<LOD	<LOD	<LOD	0.0790	<LOD	<LOD	<LOD	<LOD	<LOD
29	a	0.1855	<LOD	<LOD	(0.0024)	<LOD	<LOD	0.0858	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1626	<LOD	<LOD	(0.0021)	<LOD	<LOD	0.1451	<LOD	<LOD	(0.0020)	<LOD	<LOD
	c	0.1610	(0.0019)	<LOD	<LOD	<LOD	<LOD	0.0765	<LOD	<LOD	(0.0017)	<LOD	<LOD
58	a	0.1156	(0.0020)	<LOD	<LOD	<LOD	<LOD	0.1000	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1569	<LOD	<LOD	<LOD	<LOD	<LOD	0.1186	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0761	<LOD	<LOD	<LOD	<LOD	<LOD	0.1052	<LOD	<LOD	<LOD	<LOD	<LOD
92	a	0.1219	<LOD	<LOD	<LOD	<LOD	<LOD	0.0871	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1054	<LOD	(0.0017)	<LOD	<LOD	<LOD	0.1380	(0.0023)	<LOD	<LOD	<LOD	<LOD
	c	0.1026	<LOD	<LOD	<LOD	<LOD	<LOD	0.0933	<LOD	<LOD	<LOD	<LOD	<LOD
183	a	0.1156	<LOD	<LOD	<LOD	<LOD	<LOD	0.1802	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0993	<LOD	<LOD	<LOD	<LOD	<LOD	0.1325	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0791	<LOD	<LOD	<LOD	<LOD	<LOD	0.0736	<LOD	<LOD	<LOD	<LOD	<LOD
267	a	0.0726	<LOD	<LOD	<LOD	<LOD	<LOD	0.0663	(0.0016)	<LOD	<LOD	0.0052	<LOD
	b	0.0564	<LOD	<LOD	<LOD	<LOD	<LOD	0.0605	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0620	<LOD	<LOD	<LOD	<LOD	<LOD	0.0431	<LOD	<LOD	<LOD	<LOD	<LOD
359	a	0.0306	0.0064	(0.0045)	<LOD	<LOD	<LOD	0.0400	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0254	(0.0040)	<LOD	<LOD	<LOD	<LOD	0.0365	(0.0020)	<LOD	<LOD	<LOD	<LOD
	c	0.0191	(0.0031)	<LOD	<LOD	<LOD	<LOD	0.0358	(0.0034)	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

Table CA.B.8.1.2.1.2-12: Residues of bixlozone in FR02 trial plots T1 and T2

Sampling (DALA)	Sub-plot	bixlozone and 2,4-DBA residue in 0-30 cm horizons (mg/kg dwt)*											
		Treatment											
		T1 Bixlozone-4-SC, surface application						T2 Bixlozone-4-SC, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-5	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.1437	<LOD	<LOD	<LOD	<LOD	<LOD	0.1464	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1392	<LOD	<LOD	<LOD	<LOD	<LOD	0.1132	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1435	<LOD	<LOD	<LOD	<LOD	<LOD	0.1169	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1141	<LOD	(0.0015)	<LOD	<LOD	<LOD	0.1520	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1131	<LOD	<LOD	<LOD	<LOD	<LOD	0.1202	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1174	<LOD	(0.0017)	<LOD	<LOD	<LOD	0.1221	<LOD	<LOD	<LOD	<LOD	<LOD
6	a	0.1109	<LOD	<LOD	(0.0021)	<LOD	<LOD	0.1279	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1250	<LOD	<LOD	(0.0022)	<LOD	<LOD	0.1029	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1082	<LOD	<LOD	(0.0017)	<LOD	<LOD	0.1291	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.0934	<LOD	<LOD	(0.0023)	<LOD	<LOD	0.1100	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1229	<LOD	<LOD	(0.0023)	<LOD	<LOD	0.1267	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1268	<LOD	<LOD	(0.0017)	<LOD	<LOD	0.1213	<LOD	<LOD	<LOD	<LOD	<LOD
30	a	0.0867	<LOD	<LOD	(0.0035)	<LOD	<LOD	0.1173	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0756	<LOD	<LOD	(0.0023)	<LOD	<LOD	0.1257	<LOD	<LOD	(0.0018)	<LOD	<LOD
	c	0.1007	<LOD	<LOD	(0.0030)	<LOD	<LOD	0.1080	<LOD	<LOD	<LOD	<LOD	<LOD
58	a	0.0603	<LOD	<LOD	<LOD	<LOD	<LOD	0.1205	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0600	<LOD	<LOD	<LOD	<LOD	<LOD	0.1140	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0664	(0.0021)	<LOD	<LOD	<LOD	<LOD	0.0655	<LOD	<LOD	<LOD	<LOD	<LOD
125	a	0.0370	<LOD	<LOD	<LOD	<LOD	<LOD	0.1032	<LOD	<LOD	<LOD	<LOD	<LOD
	b	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	0.0387	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0302	<LOD	<LOD	<LOD	<LOD	<LOD	0.0510	<LOD	<LOD	<LOD	<LOD	<LOD
188	a	0.0127	<LOD	<LOD	<LOD	<LOD	<LOD	0.0326	<LOD	<LOD	<LOD	<LOD	<LOD

	b	0.0185	<LOD	<LOD	<LOD	<LOD	<LOD	0.0397	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0180	<LOD	<LOD	<LOD	<LOD	<LOD	0.0419	<LOD	<LOD	<LOD	<LOD	<LOD
290	a	(0.0015)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0076	<LOD	<LOD	<LOD	<LOD	<LOD
	b	(0.0019)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0108	<LOD	<LOD	<LOD	<LOD	<LOD
	c	(0.0030)	<LOD	<LOD	<LOD	<LOD	<LOD	(0.0036)	<LOD	<LOD	<LOD	<LOD	<LOD
372	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	(0.0024)	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	(0.0031)	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	(0.0025)	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)
n.a. Not available. The sample was not collected as the plot was saturated with water following heavy rainfall.

Table CA.B.8.1.2.1.2-13: Residues of bixlozone in FR02 trial plots T3 and T4

Sampling (DALA)	**Sub-plot**	**bixlozone and 2,4-DBA residue in 0-30 cm horizons (mg/kg dwt)***											
		Treatment											
		T3 F9600-21 CS, surface application						**T4** F9600-21 CS, soil incorporated					
		Bixlozone			2,4-DBA			Bixlozone			2,4-DBA		
		0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30	0-10	10-20	20-30
-5	a	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	b	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	c	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
0	a	0.1568	<LOD	<LOD	<LOD	<LOD	<LOD	0.1253	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1258	<LOD	<LOD	<LOD	<LOD	<LOD	0.1207	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1518	<LOD	<LOD	<LOD	<LOD	<LOD	0.0993	<LOD	<LOD	<LOD	<LOD	<LOD
3	a	0.1477	<LOD	<LOD	<LOD	<LOD	<LOD	0.1427	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1073	<LOD	<LOD	<LOD	<LOD	<LOD	0.0932	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.2110	<LOD	<LOD	<LOD	<LOD	<LOD	0.1327	<LOD	<LOD	<LOD	<LOD	<LOD
6	a	0.0973	<LOD	<LOD	<LOD	<LOD	<LOD	0.1457	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0800	<LOD	<LOD	<LOD	<LOD	<LOD	0.1276	<LOD	<LOD	<LOD	<LOD	<LOD

	c	0.1287	<LOD	<LOD	<LOD	<LOD	<LOD	0.1019	<LOD	<LOD	<LOD	<LOD	<LOD
13	a	0.1360	<LOD	<LOD	(0.0027)	<LOD	<LOD	0.1434	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0960	<LOD	<LOD	(0.0025)	<LOD	<LOD	0.1303	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.1118	<LOD	<LOD	(0.0019)	<LOD	<LOD	0.1458	<LOD	(0.0033)	<LOD	<LOD	<LOD
30	a	0.1143	<LOD	<LOD	(0.0022)	<LOD	<LOD	0.1334	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.1250	<LOD	<LOD	(0.0024)	<LOD	<LOD	0.1301	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0946	<LOD	<LOD	(0.0017)	<LOD	<LOD	0.1473	<LOD	<LOD	<LOD	<LOD	<LOD
58	a	0.0822	<LOD	<LOD	<LOD	<LOD	<LOD	0.1533	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0585	<LOD	<LOD	<LOD	<LOD	<LOD	0.0778	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0567	<LOD	<LOD	<LOD	<LOD	<LOD	0.0738	<LOD	<LOD	<LOD	<LOD	<LOD
125	a	0.0396	<LOD	<LOD	<LOD	<LOD	<LOD	0.1217	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0436	<LOD	<LOD	<LOD	<LOD	<LOD	0.0960	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0530	<LOD	<LOD	<LOD	<LOD	<LOD	0.1251	<LOD	<LOD	<LOD	<LOD	<LOD
188	a	0.0385	<LOD	<LOD	<LOD	<LOD	<LOD	0.1297	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0205	<LOD	<LOD	<LOD	<LOD	<LOD	0.0325	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0380	<LOD	<LOD	<LOD	<LOD	<LOD	0.0406	<LOD	<LOD	<LOD	<LOD	<LOD
290	a	0.0078	<LOD	<LOD	<LOD	<LOD	<LOD	0.0270	<LOD	<LOD	<LOD	<LOD	<LOD
	b	0.0088	<LOD	<LOD	<LOD	<LOD	<LOD	0.0259	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0137	<LOD	<LOD	<LOD	<LOD	<LOD	0.0585	<LOD	<LOD	<LOD	<LOD	<LOD
372	a	(0.0034)	<LOD	<LOD	<LOD	<LOD	<LOD	0.0467	(0.0033)	<LOD	<LOD	<LOD	<LOD
	b	0.0078	<LOD	<LOD	<LOD	<LOD	<LOD	0.0280	<LOD	<LOD	<LOD	<LOD	<LOD
	c	0.0129	<LOD	<LOD	<LOD	<LOD	<LOD	0.0327	<LOD	<LOD	<LOD	<LOD	<LOD

* Values corrected for mean procedural recovery
DALA = Days after last application
<LOD = below limit of detection (0.0015 mg/kg)
Values in brackets are below LOQ (0.0050 mg/kg) but above LOD (0.0015 mg/kg)

KINETICS

The kinetic assessment is presented in section CA.B.8.1.2.3.

CONCLUSION

Bixlozone steadily declined over 365 days in trial plots in Germany and France. Residues of bixlozone were confined to the 0-10 cm soil layer. Metabolites bixlozone-3-OH-propanamide and 2,4-dichlorobenzoic acid were below quantifiable levels throughout the trial period.

CA.B.8.1.2.1.3. Soil dissipation in Germany and the UK

Report:	KCA 7.1.2.2.1/03, Gezahegne, W. (2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Field soil dissipation study with one bare soil application of F9600-4 SC at two sites in North EU (Germany and UK) in 2016 – 2017
Testing Facility:	Eurofins Agroscience Services GmbH Carl-Goerdeler-Weg 5 D-21684 Stade Germany
Document No:	S16-02441
Guidelines:	-SETAC 1995 -SANCO/3029/99 rev. 4 -EPA (Oct 2008). Fate, Transport and Transformation Test Guidelines. -EFSA Guidance Document for evaluating laboratory and field dissipation studies to obtain DegT50 values of active substances of plant protection products and transformation products of these active substances in soil. EFSA Journal 2014;12(5): 3662. -ISO 10381-6:2009 (handling & storage soil for assessment of microbial biomass) -OECD Guidance Document for Conducting Pesticide Terrestrial Field Dissipation Studies (ENV/JM/MONO(2016)6, released 04 March 2016).
GLP:	Yes

CA Comments:	The following deviations from the guidelines were noted by the CA and are explored in more detail in the main body of text: • Some spilling of samples; reported but declared invalid. • Temperature during one shipment increased to -15ºC above the recommended 18ºC • sensor failures led to some water and soil data being taken from substitute weather stations. However, the CA does not consider these issues to have significantly affected the outcomes of the study. **This study is relied upon.**

INTRODUCTION

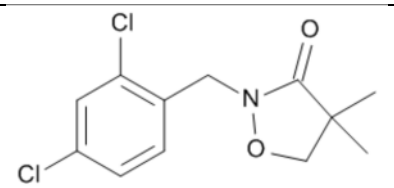
A study was carried out to determine bixlozone derived residue levels in soil after one spring application of bixlozone at sites in Germany and the UK in 2016 – 2017.

Soil cores were collected to a depth of at least 30 cm prior to application, within three hours of application and then approximately 3, 7, 14, 30, 60, 90, 180, 270 and 365 days after last application. Soil cores were cut into 0-10, 10-20, and 20-30 cm sections for analysis.

MATERIALS

Table CA.B.8.1.2.1.3-1: Test item

Common Name:	Bixlozone
Chemical Name ~(IUPAC)	2-(2,4-dichlorobenzyl)-4,4-dimethyl-1,2-oxazolidin-3-one)

Structure:	
CAS Number:	81777-95-9
Formulation Name:	F9600-4 SC
Main Uses:	Herbicide
Formulation Type:	Suspension concentrate (SC)
Nominal concentration:	36 % w/w
Nominal Density:	1.10g/mL
Batch Number:	PL15-0138
Actual Concentration:	36.4 % w/w

Field Data

The soil field trials were carried out at two locations in Europe (Germany and UK) in 2016 – 2017. The applicant states the field sites had ease of access, were not in areas prone to flooding and erosion, with relatively level ground (<1.5%) and were not too stony. No pasture, trees or vines had been cultivated in the last three years prior to application. Plots were not set-up in the headland or in the shadow of trees. All sites generally allowed year round irrigation (except under freezing conditions) and had homogenous soil over the whole site.

The pesticide use history has been provided by the applicant for the preceding three years; the CA's chemistry specialist has confirmed no structurally similar compounds were used. The CA's chemistry specialist also confirmed that 2,4-dichlorobenzoic acid (2,4-DBA) is a metabolite of propiconazole and penconazole as well, however, neither of these active substances had been applied at the test sites. Therefore, the CA considers the previous pesticide use to have not significantly impacted upon the outcomes of the study. Prior to the application of the test item the soil was cultivated as fine and firm seed bed. When necessary weed control was carried out with glyphosate. Weed cover was kept ≤ 10%.

Details of the location and physical properties of the sites is provided in Table CA.B.**8.1.2.1.3**-**2**. The field use history is provided in Table CA.B.**8.1.2.1.3**-**3**.

Table CA.B.8.1.2.1.3-2: Chemical and physical soil characterisation

Trial Number		GE02			UK01		
Country		Germany			UK		
Location		Burweg, Lower Saxony			Melbourne, Derbyshire		
GPS coordinates		53.620011 N, 9.275866 E			52.817318 N, 1.394447 W		
Previous crop in 2015		Potatoes			Fallow		
Field status in 2016		Fallow			Fallow		
Depth (cm)		0-30	30-60	60-100	0-30	30-60	60-100
Particle size distribution	Sand (%)	81.5	76.6	67.2	34.6	34.4	25.3
	Silt (%)	14.3	16.7	18.3	45.9	44.4	49.2
	Clay (%)	4.3	6.8	14.5	19.5	20.3	25.6
Textural Classification (USDA)		Loamy sand	Sandy loam	Sandy loam	Loam	Loam	Loam
pH ($CaCL_2$)		6.09	5.07	4.09	7.10	7.35	7.55
pH (H_2O)		5.19	4.73	4.40	7.11	7.24	7.33
Organic carbon (% w/w)		4.0	1.2	<0.5	2.1	0.88	0.57
Cation exchange capacity (meg/100g)		9.0	4.4	5.3	9.0	7.4	9.2
Soil bulk density (g/L)		1620	1980	2050	1530	1810	1870
Water holding capacity (%w/w)	WHC_{max}	28.4			24.4		
	pF 2.0	22.4			19.2		
	pF 4.0	8.7			11.2		
Microbial biomass carbon (mg		39.2 (0-20 cm)			63.3 (0-20 cm)		

C/100 g)		

Table CA.B.8.1.2.1.3-3: Field use history

Site	**Both sites**			
Year	**Fertilizer name**	**Rate**	**Unit**	**Composition**
2016 (until start of trial	None	N/A	N/A	N/A
Site	**GE02**		**UK01**	
Year	**Previous crop**	**Active ingredients**	**Previous crop**	**Active ingredients**
2013	Maize	Terbuthylazin, Pethoxamid, Dimethenamid-P, Topramezone, Mesotrione, Bromoxynil, Iodosulfuron, Foramsulfuron	Spring barley	Glyphosate
2014	Maize	Terbuthylazin, Pethoxamid, Dimethenamid-P, Topramezone, Mesotrione, Bromoxynil, Iodosulfuron, Foramsulfuron	Fallow	Glyphosate
2015	Maize (until end of May)	Terbuthylazin, Pethoxamid, Mesotrione, Nicoslufuron	Fallow	Glyphosate
	Potatoes (from June)	Mono and dipotassium phosphite, Potassium phosphates, Fluazinam, Difeconazole, Mandipropamid, Fluopicolide, Propamocarb HCL, Mancozeb, Dimethomorph, Cypoxanil, Metribuzin, Prosulfocarb, Rimsulfuron, Cyazofamid, Deltametrin, Cymoxanil, Metalaxyl M, Deiquat		
2016 (until start of trial)	Fallow	None	Fallow	Glyphosate

EXPERIMENTAL DESIGN

At both German and UK field sites the study area was divided into three plots; an untreated control plot (3 x 5 m), and two treated plots (plot 2, plot 3); each divided into 3 sub-plots (each 3 x.32 m). A minimum 2 m buffer zone separated each sub-plot with a >10 m buffer separating the treated plots from the untreated control plot.

Daily values of air temperature, soil temperature (at approx. 10 cm and 30 cm depth), volumetric soil moisture (at approx. 10 cm and 30 cm depth), wind speed, solar radiation, air humidity as well as rainfall were recorded by onsite weather stations. The air and soil temperature were recorded as daily minimum, maximum and average values. The rain as well as the solar radiation was recorded as sum per day, the volumetric soil moisture as average percent water content and all other data as daily mean values. The long term averages (1981 – 2010 for GE02 and 1971 – 2000 for UK01) were obtained from a weather station 21 km from GE02 and 9.37 km from UK01.

The CA notes that the GE02 onsite weather station failed on numerous occasions from December 2016 to February 2017, summarised in Table CA.B.**8.1.2.1.3-4**. As such, for these time periods, weather data was taken from a substitute weather station located 1.05 km from the test site. The CA notes this is marginally greater than the 1 km limit for tailored studies stated in the EFSA DegT50 guidance. However, because it was only marginally >1 km and the number of days affected was <1 month, the CA is of the opinion that the use of this weather station is unlikely to significantly impact upon the outcomes of the study

Similarly, the soil moisture station also failed at the GE02 test site during the study period. As such, the applicant collected soil moisture data from a substitute station located 100 m from the test site. Because the substitute station was located very close to the test site, the CA does not consider this to have impacted upon the study results.

Table CA.B.8.1.2.1.3-4: Dates when weather and soil data were obtained from the substitute stations

Substitute weather station dates in operation	Substitute soil station dates in operation
• Temperature, relative humidity, wind speed, radiation and rainfall data: - 23/12/16 – 29/12/16 - 03/01/17 – 04/01/17 - 07/01/17 – 12/01/17 - 15/01/17 – 15/01/17 - 20/01/17 – 26/01/17 - 31/01/17 – 01/02/17 • Relative humidity data: 05/02/17 – 27/04/17	• Soil temperature (10 cm): 15/12/16 – 10/03/17 • Soil temperature (30 cm): 10/03/17 – 11/05/17 • Soil moisture (10 cm): - 01/12/16 – 15/12/16 - 03/01/17 – 07/01/17 - 30/01/17 – 14/02/17 - 19/03/17 – 28/03/17 • Soil moisture (30 cm): - 31/01/17 – 18/02/19 - 10/03-17 – 30/03/17

The CA has compared the average temperatures and average precipitation for the study period against the long term average (see Figure CA.B**8.1.2.1.3-1** and Figure CA.B.**8.1.2.1.3-2**). As can be seen, the GE02 average temperature was broadly similar to the historical average. The average temperature at UK01 was slightly warmer for the study duration compared to the average. At both test sites, large fluctuations in monthly rainfall is observed, with some months significantly drier than the norm and other months significantly wetter; such as, the months August to November 2016 at site GE02 were significantly drier than average.

Figure CA.B8.1.2.1.3-1.: GE02 comparison of study weather versus the historical average

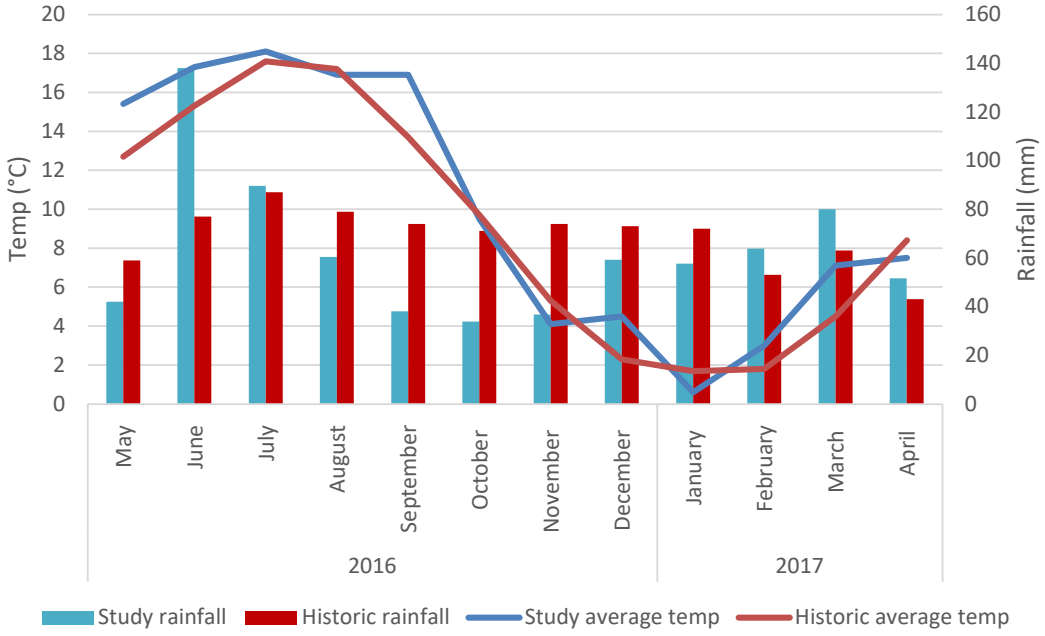
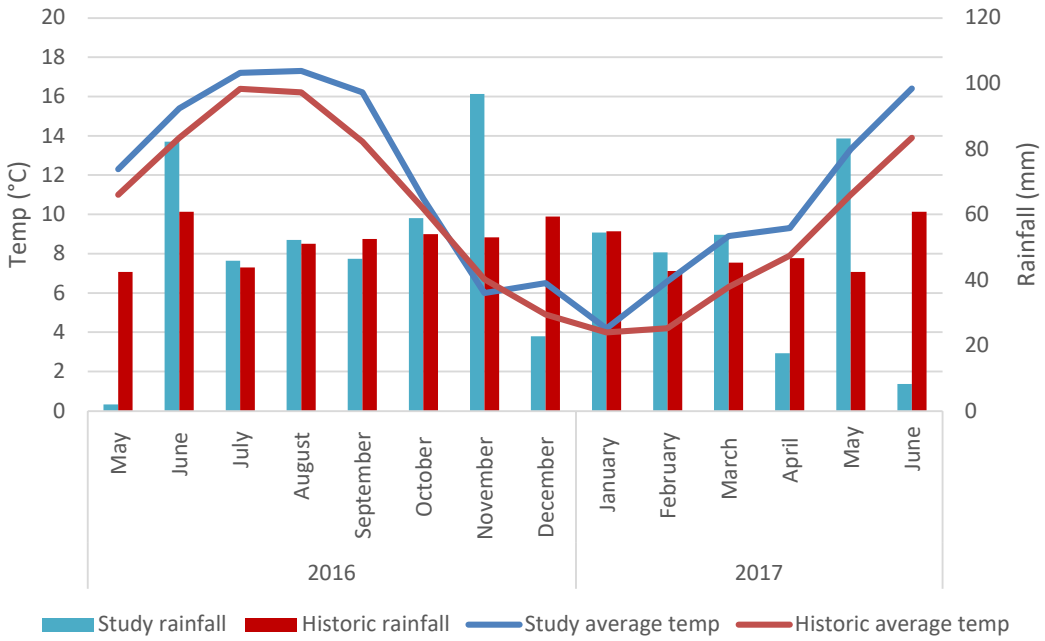


Figure CA.B.8.1.2.1.3-2: UK01 comparison of study weather versus the historical average



Application of Test Item

F9600-4 SC was applied to plots 2 and 3 on 18 May 2016 (Germany) and 28 May 2016 (UK) with a calibrated boom sprayer to bare soil at a target dose rate of 250 g/ha. The CA notes this target application rate is less than that applied in study Gemrot, 2018a (section CA.B.8.1.2.1.1), however, it is in line with the proposed winter wheat/barley (200 g/ha) and oilseed rape (300 g/ha) application rates in the applicant's CP submission. Therefore, the CA accepts the target application rates administered. Application was performed at a temperature below 25°C. The actual applied amount was calculated by measuring the remaining spray solution after application. Spray broth homogeneity was checked by the applicant visually after filling an aliquot of each spray mixture into a transparent vessel before application. The applicant states at all sites the spray tolerance (±10 %) was met. For plot 2 only, immediately after application the test item was mechanically incorporated into the soil to a soil depth of approximately 10 cm depth via a suitable tool (rotary harrow or power harrow). After incorporation, plot 2 was flat rolled.

Table CA.B.8.1.2.1.3-5: Application data

	GE02			UK01		
Treatment No.	**Plot 2**			**Plot 2**		
	SP1	**SP2**	**SP3**	**SP1**	**SP2**	**SP3**
Application date	18 May 2016	18 May 2016	18 May 2016	28 May 2016	28 May 2016	28 May 2016
Type of application	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil
Application volume actual (L/ha)	396	403	397	400	388	410
Deviation rate (%)	-0.91	+0.63	-0.68	+ 0.00	- 3.13	+ 2.60
g ai/ha applied [a]	247.7	251.6	248.3	250.0	242.2	256.5
mL product /ha applied	625.57	635.29	627.02	631.31	611.61	647.72
Air temperature (°C)	18.9	19.1	22.1	21.5		
Wind speed (m/s) (*behind wind	0.5*	0.5*	0.5*	0		

	GE02			UK01		
Treatment No.	**Plot 2**			**Plot 2**		
	SP1	**SP2**	**SP3**	**SP1**	**SP2**	**SP3**
shield)						
Wind direction	W			N/A		
Actual relative air humidity (%)	54.3	52.7	44.8	47.1		
Cloud cover (%)	70			50		
Ground cover (%)	Bare soil			N/A		
Wetness of soil surface	Moist			Dry		
Temperature of soil (°C) (10 cm)	13.9	14.2	14.2	20.2		
Treatment No.	**Plot 3**			**Plot 3**		
	SP1	**SP2**	**SP3**	**SP1**	**SP2**	**SP3**
Application date	18 May 2016	18 May 2016	18 May 2016	28 May 2016	28 May 2016	28 May 2016
Type of application	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil	overall to bare soil
Application volume actual (L/ha)	384	396	401	410	402	418
Deviation rate (%)	-4.04	-0.96	+0.23	+ 2.60	+ 0.52	+ 4.43
g ai/ha applied [a]	239.9	247.6	250.6	256.5	251.3	261.1
mL product /ha applied	605.81	625.25	632.76	647.72	634.59	659.28
Air temperature (°C)	16.6	17.3	18.9	22.8		
Wind speed (m/s) (*behind wind shield)	2	1-2	0.5*	0.6		
Wind direction	W			S		
Actual relative air humidity (%)	62.8	64.8	54.8	40.3		
Cloud cover (%)	10			80		
Ground cover (%)	Bare soil			N/A		
Wetness of soil surface	Moist			Dry		
Temperature of soil (°C) (10 cm)	12.1	12.3	12.6	18.9		

[a] based on nominal content of a.i. ; N/A: not applicable ; SP: Sub-Plot

SAMPLING PROCEDURES

Soil residue samples from 0 – 30 cm depth were taken by using a manual whilst soil cores from 0 – 50 cm depth were taken by using a hydraulic corer. Incomplete (less than 95% filled) or damaged cores (0 – 50 cm) were rejected and discarded. Cores not filled completely (between 95 – 100%) were filled up with crushed aluminium foil and capped.

After each sampling the holes were backfilled with untreated soil. Resampling was avoided by clearly marking already sampled areas. The remaining holes after sampling of the cores were filled with untreated soil or sand. Samples were kept out of the sunlight and untreated and treated samples were kept separate by an adequate space at all times. After collection soil cores were bagged, labelled and deep frozen less than 6 hours after collection.

In order to verify application results, 30 petri dishes (10.8 cm inner diameter) each filled with 50 ± 0.2 g of sieved (mesh size: 2 mm) top soil (0 – 10 cm) were placed across the treated plots (5 per subplot) right before the application. The soil had been taken from the surroundings of the treated plots prior to application. The soil was evenly distributed and levelled over the surface of the petri dishes. The petri dishes were covered until the time of application to prevent any potential losses due to wind. Immediately after application, the petri dishes were closed with a lid, sealed with adhesive tape and double wrapped in polyethylene bags to prevent any potential losses due to breaking. The samples were stored immediately in a mobile freezer and under deep frozen conditions on-site within 5 hours after the end of the application. The positions of petri dishes were marked to avoid resampling these places and recorded in the raw data. Soil characterisation, soil bulk density, water holding capacity, soil biomass and residue samples from the control plot were taken 1 day before application (DBA).

For site GE02, soil residues samples were taken from 0 – 30 cm depth immediately after application and at 3, 7 and 14 days after application (DAA). At 28 and 58 DAA soil residue samples were taken from 0 – 50 cm depth. At 90, 177, 272 and 358 DAA soil residue samples were taken from 0 – 100 cm depth.

For site UK01, soil residues samples were taken from 0 – 30 cm depth immediately after application and at 3, 6 and 15 DAA. At 30 and 58 DAA soil residue samples were taken from 0 – 50 cm depth. At 87, 185, 263 and 371 DAA soil residue samples were taken from 0 – 100 cm depth.

Soil Preparation

Upon receipt the soil samples were stored deep-frozen (≤ -18 °C). The CA notes that during shipment of the GE02 0, 15, 58, 87 DAA samples and 371 samples, the temperature increased to max of -14 °C (1.5 hours); the max period of time with a temperature >-18 °C was 13.5 hours (-15 °C). Similarly, for site UK01, the 0 DAA samples recorded temperatures of -16 °C for 5.5 hours during shipment.

Furthermore, the UK01 185 DAA treatment samples were stored at the analytical laboratory at -7 °C for 28 hours due to technical problem with the freezer.

To address these deviations, the applicant conducted an additional storage stability test over 51 hours and a temperature of -7 °C; the results are shown in Table CA.B.**8.1.2.1.3-6**. As can be seen, the analytes were shown to be stable over these conditions. Therefore, CA considers the deviation in temperature to not have impacted on the study outcomes.

Table CA.B.8.1.2.1.3-6: Short time storage stability test

Analyte	Matrix	Storage Period (hours)	Recovery in Stored Samples				Recovery in freshly fortified samples	
			single values	Mean uncorrected	RSD	Mean corrected	Single Values	Mean
			(%)	(%)	(%)	(%)	(%)	(%)
Bixlozone	Soil	0	93	96	4.5	100	NA	NA
			101					
			94					
		51	92	90	-	110	77	82
			88				87	
2,4-Dichlorobenzoic acid	Soil	0	94	94	1.1	100	NA	NA
			95					
			93					
		51	82	78	-	104	74	75
			75				75	
bixlozone-3-OH-Propanamide	Soil	0	102	97	4.3	100	NA	NA
			96					
			94					
		51	94	99	-	103	95	97
			104				99	

Cores were cut in layers (0-10 cm, 10-20 cm, 20-30 cm, 30-40 cm, 40-50 cm, 50-60 cm, 60-70 cm, 70-80 cm, 80-90 cm, 90-100 cm) in the deep frozen state and homogenised (only 0-10 cm, 10-20 cm, 20-30 cm; for some samples 30-40 cm and 40-50 cm additionally) by milling and sieving with dry ice. Two aliquots of at least 400 g frozen homogenised soil were taken and stored deep frozen. All samples were stored deep-frozen (≤ 18 °C) except samples mentioned above.

ANALYTICAL METHOD

Soil specimens were analysed for bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide using analytical method CAM-0151/001; see Vol 3 CA, B5 for further information. Petri dish soil specimens were analysed for bixlozone only using the same method. Soil specimens were extracted within 496 days of sampling.

Soil samples were extracted by weighing 5 g of soil 50 mL centrifuge tubes; 10 mL water of water was added and extraction with 10 mL acetonitrile containing 0.1% formic acid.. Following C-18 dispersive solid-phase extraction, aliquots of the extract were analysed by LC-MS/MS (LOD 0.0015 mg/kg / LOQ 0.005 mg/kg).

For each analytical set of specimen analysis, the method's applicability in terms of accuracy and repeatability was assessed by the applicant by fortification of control (untreated) test portions of the respective matrix and subsequent determination of the procedural recoveries upon applying the test method(s). Fortifications were performed at the level of 0.005 mg/kg, 0.05 mg/kg and 0.25 mg/kg, covering the range of the highest residues found in specimens. The fortification results are shown in Table CA.B.**8.1.2.1.3-7**; all results are within the OECD recommended range and so are accepted by the CA.

Table CA.B.8.1.2.1.3-7: Recovery of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide from fortified control samples

Fortification level (mg/kg)	Recovery	Bixlozone	2,4-DBA	bixlozone-3-OH-Prop
0.0050	Mean (%)	99	99	103
	RSD (%)	7.6	8.0	5.5
0.050	Mean (%)	96	90	99
	RSD (%)	6.6	9.4	7.0
0.25	Mean (%)	88*	-	-
	RSD (%)	-	-	-
Overall mean (%)		97	95	101
Overall RSD (%)		7.3	9.8	6.5

2,4-DBA = 2,4-dichlorobenzoic acid
bixlozone-3-OH-prop. = bixlozone-3-OH-propanamide
*Single sample only

RESULTS

The soil residue results from sites GE02 and UK01 are presented in Table CA.B.**8.1.2.1.3-8** to Table CA.B.**8.1.2.1.3-11** below.

At site GE02, soil residues of bixlozone were predominately confined to the top 10 cm horizon. Residues of 2,4-DBA were detected throughout the study and in all soil horizons. In residue terms, the maximum 2,4-DBA residue occurred at site GE02 (plot 2 – incorporated (Table CA.B.**8.1.2.1.3-8**)), 272 DAA, sub-plot 1, where the summed residue of 0 – 30 cm horizons equalled 0.0838 mg/kg. This is equivalent to 49.7% of bixlozone on a mass basis, when compared to the bixlozone 0 DAA, sub-plot 1 residue (0.1685 mg/kg). However, in percentage terms, the maximum occurrence of 2,4-DBA occurred at site GE02 plot 3 (bare soil (

Table CA.B.**8.1.2.1.3-9**)), where the summed 272 DAA, sub-plot 3 residue (0.0758 mg/kg) equates to 69.4% of bixlozone on a mass basis (when compared to the bixlozone, 0 DAA, sub-plot 3 residue (0.1093 mg/kg)). This is equivalent to 99.53% on a molar basis. Due to the quantities observed, the CA considers it appropriate for 2,4-DBA to be considered in the terrestrial exposure calculations and risk assessment. The CA notes the applicant also analysed the 272 DAA 30 – 50 cm horizons as well; no residues of 2,4-DBA were detected below 30 cm.

The metabolite 3-OH was detected at concentrations >LOD in only one sample; plot 3, 28 DAA, 10 – 20 cm horizon (0.0087 mg/kg). When compared to the bixlozone, plot 3, sub-plot 1, 0 DAA samples (0.1251 mg/kg), this equates to 6.95% of parent substance on a mass basis. This is equivalent to 6.90% on a molar basis. For soil dissipation studies, the data requirements state that metabolites <5% need not be considered further. However, as 3-OH was not detected in the laboratory aerobic degradation study and was only detected in one sample here (with the other two sub-plots recording residues <LOD), the CA does not consider it necessary for 3-OH to be included in the terrestrial exposure calculations or risk assessment. It is noted 3-OH was detected in the laboratory anaerobic degradation study (section CA.B.8.1.1.2.1), which could potentially explain the single residue detected at GE02 Plot 3, however, justification was submitted and accepted excluding the need to include 3-OH in the terrestrial exposure calculations and risk assessment.

At UK01, soil residues of bixlozone were predominately confined to the top 10 cm horizon. Residues of 2,4-DBA were detected throughout the study and to a depth of 30 cm. The maximum residue of 2,4-DBA detected was 0.0226 mg/kg in plot 3, sub-plot 2, 6 DAA, 0 – 10 cm depth. Furthermore, a mean residue of 0.0081 mg/kg was also detected in the 20 – 30 cm horizon for this sampling occasion. Therefore, this equates to a total residue of 0.0307 mg/kg over 0 – 30 cm. When compared to the bixlozone, plot 3, sub-plot 2, 0 DAA sample (0.1469 mg/kg), this equates to 20.9% of parent substance on a mass basis (29.99% molar basis).

Table CA.B.8.1.2.1.3-8: Residues of bixlozone and 2,4-DBA in GE02 Plot 2 (incorporated)

Sampling (DAA)	Sub-plot	Residues (mg/kg dwt) in GE02 Plot 2					
		Bixlozone			2,4-DBA		
		0-10 cm	10-20 cm	20-30 cm	0-10 cm	10-20 cm	20-30 cm
0	1	0.1685	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1215	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1197	<LOD	<LOD	<LOD	<LOD	<LOD
3	1	0.1185	<LOD	<LOD	0.0048	0.0087	0.0042
	2	0.0635	<LOD	<LOD	<LOD	0.0034	0.0199*
	3	0.1104	<LOD	<LOD	<LOD	<LOD	<LOD
7	1	0.1086	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0926	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1060	<LOD	<LOD	<LOD	<LOD	<LOD
14	1	0.0568	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0817	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0666	<LOD	<LOD	<LOD	<LOD	<LOD
28	1	0.0495	<LOD	<LOD	<LOD	0.0046	0.0026
	2	0.0795	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0826	<LOD	<LOD	<LOD	<LOD	<LOD
58	1	0.0641	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0797	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0643	<LOD	<LOD	<LOD	<LOD	<LOD
90	1	0.0337	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0467	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0262	<LOD	<LOD	<LOD	<LOD	<LOD
177	1	0.0605	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0418	<LOD	<LOD	<LOD	0.0058	<LOD
	3	0.0460	<LOD	<LOD	<LOD	<LOD	<LOD
272	1	0.0657	<LOD	<LOD	0.0220	0.0154	0.0464
	2	0.0432	<LOD	<LOD	0.0071	0.0090	0.0072
	3	0.0583	<LOD	<LOD	0.0044	<LOD	<LOD
358	1	0.0348	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0372	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0203	<LOD	<LOD	<LOD	<LOD	<LOD

DAA = Days after application
Plot 2: bixlozone-4-SC applied to bare soil then incorporated
<LOD = below limit of detection (0.0015 mg/kg)

*Mean of 2 replicates

Table CA.B.8.1.2.1.3-9: Residues of bixlozone, 2,4-DBA and 3-OH in GE02 Plot 3 (soil surface)

Sampling (DAA)	Sub-plot	Residues (mg/kg dwt) in GE02 Plot 3								
		Bixlozone			2,4-DBA			3-OH		
		0-10 cm	10-20 cm	20-30 cm	0-10 cm	10-20 cm	20-30 cm	0-10 cm	10-20 cm	20-30 cm
0	1	0.1251	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1469	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1093	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
3	1	0.1383	<LOD	0.0017	0.0051	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1021	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1150	<LOD	0.0079*	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
7	1	0.1217	<LOD	<LOD	0.0016	0.0083	<LOD	<LOD	<LOD	<LOD
	2	0.0987	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1205	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
14	1	0.0679	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0843	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0678	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
28	1	0.0739	<LOD	<LOD	0.0026	<LOD	<LOD	<LOD	0.0087	<LOD
	2	0.0492	<LOD	<LOD	<LOD	0.0056	<LOD	<LOD	<LOD	<LOD
	3	0.0696	<LOD	<LOD	0.0039	<LOD	0.0047	<LOD	<LOD	<LOD
58	1	0.0423	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0420	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0523	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
90	1	0.0527	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0314	0.0022	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0396	0.0019	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
177	1	0.0357	<LOD	<LOD	<LOD	0.0026	<LOD	<LOD	<LOD	<LOD
	2	0.0260	<LOD	<LOD	<LOD	0.0024	<LOD	<LOD	<LOD	<LOD
	3	0.0330	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
272	1	0.0402	0.0024	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0264	0.0019	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0268	<LOD	<LOD	<LOD	0.0513	0.0245	<LOD	<LOD	<LOD
358	1	0.0185	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0123	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0190	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD

DAA = Days after application
Plot 3: bixlozone-4-SC applied to bare soil
<LOD = below limit of detection (0.0015 mg/kg)
*Mean of 2 replicates

Table CA.B.8.1.2.1.3-10: Residues of bixlozone and 2,4-DBA in UK01 Plot 2 (incorporated)

Sampling (DAA)	Sub-plot	Residues (mg/kg dwt) in UK01 Plot 2					
		Bixlozone			2,4-DBA		
		0-10 cm	10-20 cm	20-30 cm	0-10 cm	10-20 cm	20-30 cm
0	1	0.0867	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1035	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1673	<LOD	<LOD	<LOD	<LOD	<LOD
3	1	0.1456	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1488	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0966	<LOD	<LOD	<LOD	<LOD	<LOD
6	1	0.1075	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1079	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0957	<LOD	<LOD	<LOD	<LOD	<LOD
15	1	0.0890	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0944	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1010	<LOD	<LOD	<LOD	<LOD	<LOD
30	1	0.0770	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0887	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0728	<LOD	<LOD	<LOD	<LOD	<LOD
58	1	0.1045	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0599	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0650	<LOD	<LOD	<LOD	<LOD	<LOD
87	1	0.0578	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0586	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0796	<LOD	<LOD	<LOD	<LOD	<LOD
185	1	0.0410	<LOD	<LOD	<LOD	<LOD	0.0022
	2	0.0568	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0390	<LOD	<LOD	<LOD	<LOD	<LOD
263	1	0.0407	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0448	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0462	<LOD	<LOD	<LOD	<LOD	<LOD
371	1	0.0372	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0345	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0489	<LOD	<LOD	<LOD	<LOD	<LOD

DAA = Days after application
Plot 2: bixlozone-4-SC applied to bare soil then incorporated
<LOD = below limit of detection (0.0015 mg/kg)

Table CA.B.8.1.2.1.3-11: Residues of bixlozone and 2,4-DBA in UK01 Plot 3 (soil surface)

Sampling (DAA)	Sub-plot	Residues (mg/kg dwt) in UK01 Plot 3					
		Bixlozone			2,4-DBA		
		0-10 cm	10-20 cm	20-30 cm	0-10 cm	10-20 cm	20-30 cm
0	1	0.1237	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.1409	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1840	<LOD	<LOD	<LOD	<LOD	<LOD
3	1	0.1161	<LOD	<LOD	0.0137	<LOD	<LOD
	2	0.0700	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1013	<LOD	<LOD	<LOD	<LOD	<LOD
6	1	0.1383	<LOD	<LOD	0.0037	<LOD	0.0017
	2	0.0857	<LOD	<LOD	0.0226	<LOD	0.0081*
	3	0.1279	<LOD	<LOD	<LOD	<LOD	<LOD
15	1	0.0695	<LOD	<LOD	0.0018	<LOD	<LOD
	2	0.0596	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0925	<LOD	<LOD	<LOD	<LOD	<LOD
30	1	0.0677	0.0023	<LOD	<LOD	<LOD	<LOD
	2	0.0791	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.1104	0.0027	<LOD	<LOD	<LOD	<LOD
58	1	0.0664	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0701	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0682	0.0021	<LOD	<LOD	<LOD	<LOD
87	1	0.0508	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0636	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0668	<LOD	<LOD	<LOD	<LOD	<LOD
185	1	0.0325	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0229	<LOD	<LOD	<LOD	<LOD	<LOD
	3	0.0270	<LOD	<LOD	<LOD	<LOD	<LOD
263	1	0.0263	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0188	0.0017	<LOD	<LOD	<LOD	<LOD
	3	0.0274	<LOD	<LOD	<LOD	<LOD	<LOD
371	1	0.0173	<LOD	<LOD	<LOD	<LOD	<LOD
	2	0.0219	0.0020	<LOD	<LOD	<LOD	<LOD
	3	0.0269	<LOD	<LOD	<LOD	<LOD	<LOD

DAA = Days after application
Plot 3: bixlozone-4-SC applied to bare soil
<LOD = below limit of detection (0.0015 mg/kg)
*Mean of 2 replicates

KINETICS

The kinetic evaluation is presented in section CA.B.8.1.2.3.

CONCLUSION

Bixlozone residues declined steadily in trial plots in Germany and the UK. Residues of bixlozone were predominantly confined to the 0-10 cm soil layer. Metabolite, 2,4-dichlorobenzoic acid was observed at both trial sites in soil samples up to 30 cm depth and at a maximum occurrence of 69.4% on a mass basis and 99.53% on a molar basis. Metabolite 3-OH propanamide was detected in only one sample at a level of 6.95% (on a mass basis) and does not require consideration in the terrestrial exposure calculations.

CA.B.8.1.2.2. ***Storage Stability***

Report:	KCA 7.1.2.2.1/04, Rawle, N. (2017)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Storage stability study of F9600 and its metabolites (2,4-dichlorobenzoic acid and F9600-3-OH-propanamide) in soil samples stored under frozen conditions
Testing facility:	CEM Analytical Services Ltd. (CEMAS), UK
Document No:	Study no. CEMS-7213, FMC Tracking No. 2015RES-ISX2038
Guidelines:	OECD 506 (2007) EC Guideline 1607/VI/97 rev. 2, appendix H 7032/VI/95 rev. 5 US EPA Guidelines, OPPTS 860.1380 (1996)
GLP:	Yes.

CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

The aim of the study was to evaluate the stability of bixlozone and its metabolites (2,4- dichlorobenzoic acid and bixlozone-3-OH-propanamide) in soil following frozen storage for up to 24 months.

MATERIALS

Reference Items

Table CA.B.8.1.2.2-1 **:** Reference Item bixlozone

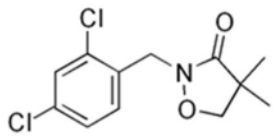
Identity:	Bixlozone
Chemical name (IUPAC):	2-(2,4-dichlorobenzyl)-4,4-dimethyl-1,2-oxazolidin-3-one
CAS number:	81777-95-9
Chemical formula:	C12H13Cl2NO2
Molecular weight:	274 g/mol
Structure:	
Purity (%):	99.8
Physical Description	White Solid
Storage:	Room Temperature

Table CA.B.8.1.2.2-2: Reference Item 2,4-Dichlorobenzoic acid

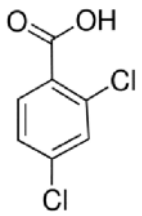
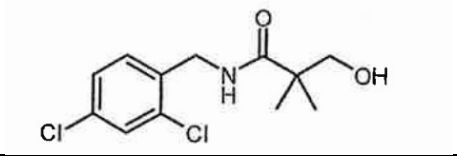
Identity:	2,4-Dichlorobenzoic acid
Chemical name (IUPAC):	2,4-Dichlorobenzoic acid
CAS number:	50-84-0
Chemical formula:	C7H4Cl2O2
Molecular weight:	191 g/mol
Structure:	
Purity (%):	99.7
Physical Description	White Solid
Storage:	Room Temperature

Table CA.B.8.1.2.2-3: Reference Item bixlozone-3-OH-Propanamide

Identity:	bixlozone-3-OH-Propanamide
Chemical name (IUPAC):	N-[(2,4-dichlorophenyl)methlyl]-3-hydroxy-2,2dimethylpropanamide
CAS number:	Not assigned
Chemical formula:	C12H15Cl2NO2
Molecular weight:	276 g/mol
Structure:	
Purity (%):	98.5
Physical Description	White Solid
Storage:	Room Temperature

METHOD

Test System

The storage stability study was carried out on LUFA 2.4 soil (see Table CA.B.**8.1.2.2-4**). No pesticides had been used at the sampling location in the previous 5 years.

Table CA.B.8.1.2.2-4: Soil Data

Soil No.	2.4
Source	Lufa Speyer, Germany
Sampling date	02/02/2015
organic carbon % C	2.03
Nitrogen in % N	0.22
pH (0.01 M CaCl2)	7.3
cation exchange capacity (meq/ 100g)	33
Particle Size (mm) distribution according to USDA (%)	
Clay (<0.002)	25.8
Silt (0.002-0.05)	41.1
Sand (0.05-2)	33.2
Soil Type	loam
maximum water holding capacity (g/100g)	43.8
weight per volume (g/1000ml)	1265

The specimens were weighed into 50 mL centrifuge tubes and then placed in a freezer set to maintain a temperature of <-18ºC. Specimens were stored under these frozen conditions at all times except when removed for analysis. Specimens were not radiolabelled.

Storage Setup

Aliquots of the specimens (5 g) were weighed out and placed in separate, individually-labelled 50 mL centrifuge tubes. The stored fortification specimens were fortified at the beginning of the study with a fortification solution containing bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3OH-propanamide to achieve the fortification level of 0.05 mg/kg (10 x LOQ (0.005 mg/kg)). Each specimen was left to stand for at least five minutes after fortification to allow the spiking solution to soak into the matrix before proceeding with the extraction or frozen storage.

Six additional spare sets of fortified specimens were prepared at the start of the study to allow for any extra time points or repeat analysis.

All unfortified specimens (used for controls and procedural recoveries) as well as the fortified specimens were stored in a freezer set to maintain a specimen temperature of <-18ºC. The specimens remained frozen throughout the storage unless removed for analysis.

Analytical Procedures

Residues of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide were extracted from the soil specimens by the addition of HPLC water followed by acetonitrile containing 0.1% formic acid. After mixing after each addition, a sachet of QuEChERS salt mixture (EN 15662) was added to each tube. The specimens were mixed on a reciprocating shaker and then centrifuged. 1 mL of the acetonitrile layer was measured and added to a dispersive solid phase extraction (SPE) mixture containing 150 mg of anhydrous magnesium sulphate and 50 mg of endcapped C18 sorbent. Following mixing and centrifuging,

250 µL of supernatant was transferred to a vial and diluted with 750 µL of HPLC water with 0.05% acetic acid. The final determination of the residues was carried out by HPLC-MS/MS.

Significant enhancement or suppression was observed during the method validation study and therefore matrix-matched standards have been used for the quantification of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide during this study. See Vol 3 CA, B5 for further information.

Analysis of Stored Specimens

At the initial time point, the analysis consisted of the following specimens of each matrix, plus a reagent blank:

- 1 unfortified control specimen.
- 2 control specimens that were freshly fortified at 0.05 mg/kg immediately prior to extraction to be used for procedural recoveries.
- 3 specimens that were fortified at 0.05 mg/kg immediately prior to extraction to be used for day-0 storage.

At the 3, 6, 12, 18 and 24 months storage time points, the analysis consisted of the following specimens of each matrix, plus a reagent blank:

- 1 unfortified control specimen.
- 2 control specimens that were freshly fortified at 0.05 mg/kg immediately prior to extraction to be used for procedural recoveries.
- 3 specimens that were fortified at 0.05 mg/kg and stored under frozen conditions for 3, 6, 12, 18 or 24 months.

One unfortified control specimen was analysed at each time-point to demonstrate that no residues of bixlozone, 2,4-dichlorobenzoic acid or bixlozone-3-OH-propanamide were present above 30% of the limit of quantification (LOQ). No residues were detected in the control specimens, as confirmed by the chromatograms.

Table CA.B.8.1.2.2-5: Stability of bixlozone in Soil Stored at <-18ºC

Storage time		Mean residue (mg/kg)		Mean recovery (%)		
Months (nominal)	**Days (actual)**	**Uncorrected (A)**	**Corrected (B)**	**Procedural**	**Uncorrected residue (C)**	**Corrected residue (D)**
0	0	0.0503	0.0513	98	100†	100†
3	90	0.0513	0.0507	101	102	99
6	179	0.0489	0.0440	111	97	86
12	360	0.0455	0.0494	92	90	96
18	545	0.0427	0.0533	80	85	104
24	733	0.0492	0.0464	106	98	90

† = nominally 100%
A = Measured residue
B = Measured residue corrected for mean procedural recovery
C = Percentage of 0 day using uncorrected residues (A / 0 time A) × 100%
D = Percentage of 0 day using corrected residues (B / 0 time B) × 100%

Table CA.B.8.1.2.2-6: Stability of 2,4-Dichlorobenzoic acid in soil Stored at <-18ºC

Storage time		Mean residue (mg/kg)		Mean recovery (%)		
Months (nominal)	**Days (actual)**	**Uncorrected (A)**	**Corrected (B)**	**Procedural**	**Uncorrected residue (C)**	**Corrected residue (D)**
0	0	0.0478	0.0514	93	100†	100†
3	90	0.0460	0.0470	98	96	91
6	179	0.0450	0.0464	97	94	90
12	360	0.0437	0.0460	95	91	89
18	545	0.0472	0.0555	85	99	108
24	733	0.0471	0.0495	95	98	96

† = nominally 100%
A = Measured residue
B = Measured residue corrected for mean procedural recovery
C = Percentage of 0 day using uncorrected residues (A / 0 time A) × 100%
D = Percentage of 0 day using corrected residues (B / 0 time B) × 100%

Table CA.B.8.1.2.2-7: Stability of bixlozone-3-OH-Propanamide in Soil Stored at <-18ºC

Storage time		Mean residue (mg/kg)		Mean recovery (%)		
Months (nominal)	**Days (actual)**	**Uncorrected (A)**	**Corrected (B)**	**Procedural**	**Uncorrected residue (C)**	**Corrected residue (D)**
0	0	0.0479	0.0520	92	100†	100†
3	90	0.0468	0.0483	97	98	93
6	179	0.0470	0.0443	106	98	85
12	360	0.0452	0.0480	94	94	92
18	545	0.0550	0.0491	112	115	94
24	733	0.0474	0.0474	100	99	91

† = nominally 100%
A = Measured residue
B = Measured residue corrected for mean procedural recovery
C = Percentage of 0 day using uncorrected residues (A / 0 time A) × 100%
D = Percentage of 0 day using corrected residues (B / 0 time B) × 100%

CONCLUSION

Residues of bixlozone, 2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide showed no significant decrease (≤15% as compared to the zero-time value) in soil when stored deep frozen at <-18°C for up to 24 months.

CA.B.8.1.2.3. *Soil dissipation kinetics*

Report:	KCA 7.1.2.2.1/05, Montesano, V., Jarvis, T., Sneath, H. (2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Normalisation of the field dissipation data for F9600-4 SC or F9600-21 CS from four locations in Europe and the determination of the normalised field DT_{50} values
Testing facility:	Exponent International Ltd, UK
Document No:	Report no.; 1508442.UK0-9677, FMC Tracking no.; 2018EFT-ISX4194
Guidelines:	EFSA Journal 2014; 12(5): 3662 FOCUS Kinetics (2014)
GLP:	No

Report:	KCA 7.1.2.2.1/06, Sneath, H., Tallentire, E. (2020)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Field dissipation kinetics for F9600 and its metabolite 2,4-dichlorobenzoic acid from four locations in Europe
Testing facility:	Exponent International Ltd, UK
Document No:	Report no.; 1508442.UK0-6455, FMC Report no.; FMC-54089
Guidelines:	FOCUS Kinetics (2014)
GLP:	No

CA comments	The CA's comments are presented in the main body of text. No significant issues were identified. **This study is relied upon.**

INTRODUCTION

The field dissipation of bixlozone was investigated in three studies, Gemrot (2018 a, b) and Gezahegne (2018), at seven field sites in Europe using F9600-4 SC and/or F9600-21 CS formulations (see section CA.B.8.1.2.1). At all of the trial sites, the formulation was sprayed onto bare soil. In some plots the formulation was incorporated into the first few centimetres of the top soil immediately after application, to minimise surface dissipation processes. Residues of bixlozone, 2,4-dichlorobenzoic acid (2,4-DBA) and 3-OH propanamide (3-OH) were detected; however, as 3-OH was only detected in a single sample and at concentrations <10%, this result has not been considered further. The trial sites, treatments and formulations are summarised in Table CA.B.**8.1.2.3-1**.

Table CA.B.8.1.2.3-1: Description of the field dissipation studies

Study	Country	Code	Plot	Type of application	Formulation
15SGS088 (Gemrot, 2018a)	France	FR01	T1	Bare soil	F9600-4 SC
			T2	Incorporated	F9600-4 SC
			T3	Bare soil	F9600-21 CS
			T4	Incorporated	F9600-21 CS
	Italy	IT01	T1	Bare soil	F9600-4 SC
			T2	Incorporated	F9600-4 SC
			T3	Bare soil	F9600-21 CS
			T4	Incorporated	F9600-21 CS
	Italy	IT02	T2	Incorporated	F9600-4 SC
15SGS111 (Gemrot, 2018b)	Germany	GE01	T1	Bare soil	F9600-4 SC
			T2	Incorporated	F9600-4 SC
			T3	Bare soil	F9600-21 CS
			T4	Incorporated	F9600-21 CS
	France	FR02	T1	Bare soil	F9600-4 SC

Study	Country	Code	Plot	Type of application	Formulation
			T2	Incorporated	F9600-4 SC
			T3	Bare soil	F9600-21 CS
			T4	Incorporated	F9600-21 CS
S16-02441 (Gezahegne, 2018)	Germany	GE02	2	Incorporated	F9600-4 SC
			3	Bare soil	F9600-4 SC
	UK	UK01	2	Incorporated	F9600-4 SC
			3	Bare soil	F9600-4 SC

The applicant has supplied normalised and non-normalised kinetics for each field site. The applicant's normalised kinetics are to produce appropriate endpoints for the groundwater and higher-tier drainflow calculations and the non-normalised kinetics are to produce appropriate endpoints for the PECsoil calculations and for comparison against accumulation and persistence triggers. For the PECsoil calculations and accumulation comparison, non-normalised DissT50 (or DegT50 for 'modern'/'tailored' studies) values are appropriate. However, for the persistence assessment (as per the SANCO guidance), non-normalised DegT50 values are appropriate because for persistence assessments the DT50 should refer to degradation. For the trial sites where incorporation occurred, the non-normalised endpoint will be a DegT50 value and so is appropriate for comparison against the persistence criteria and for the PECsoil and accumulation trigger comparison. However, for the trials where incorporation did not occur, the CA considers it appropriate to determine DissT50 values (by using all of the residue data) for the PECsoil and accumulation trigger assessment and DegT50 values (by excluding the samples undertaken prior to 10 mm rainfall as per the EFSA DegT50 guidance) for the persistence assessment. This approach is further justified by evidence of rapid early decline before 10 mm rainfall in some of the trails. The CA notes the applicant has not made this differentiation in their assessment of the non-normalised data. Therefore, the CA has performed the non-normalised DegT50 calculations for the trials where incorporation did not occur.

The CA has handled the field study data in a similar, but not identical, manner to the applicant. The CA considers it appropriate to apply slightly different data handling criteria to the top horizon (0 – 10 cm) than the 10 – 20 cm and 20 – 30 cm horizons; the applicant has applied the same criteria to each horizon. Replicates were considered separately (i.e. data for each sub-plots (a, b and c) were considered separately for kinetic fitting). Time 0 residues detected in soil horizons deeper than 10 cm were added to the 0 – 10 cm horizon (unless the test item was incorporated upon application).

Measured values between LOD (0.0015 mg/kg dry weight) and LOQ (<0.0050 mg/kg dry weight) were set to the measured value. With the exception of 0 DAT values, the last sampling interval before the first detectable amount was set to ½ LOD. Values <LOD after a detectable value were also set to ½ LOD. If a measured value was detected in either the 10 – 20 cm or the 20 – 30 cm horizon and adjacent horizon value was <LOD, this too was also set to ½ LOD. All subsequent samples <LOD were omitted unless later samples >LOD were reported. Residues from each horizon were summed. The applicant generally did not set the residues to ½ LOD, in the lower horizons, in the preceding or succeeding time points when residues >LOD were detected. Therefore, the CA generally obtained slightly higher summed residues to the applicant and so, for completeness, repeated the kinetic modelling. However, in the majority of cases, the resulting differences in kinetic fits were sufficiently minor that the applicant's fits could still be accepted.

Additionally, for the metabolite 2,4-dichlorobenzoic acid (2,4-DBA), the initial amount at 0 DAT were set to zero, the last sampling interval before the first detectable amount were set to ½ LOD, and any prior non-detects were omitted. Total measured residues of 2,4-dichlorobenzoic acid were converted to parent equivalents for kinetic fitting using molecular weight correction factor of 1.435 (bixlozone; 274.15 g/mol, 2,4-dichlorobenzoic acid; 191.01 g/mol).

The sum of the bixlozone residues and, where relevant, the 2,4-dichlorobenzoic acid residues in each plot at each sampling interval are presented in Table CA.B.**8.1.2.3-5** to Table CA.B.**8.1.2.3-25**. The applicant states for trials plots in which 2,4-dichlorobenzoic acid was below quantifiable levels throughout the trial period, concentrations of the metabolite are too low to enable robust kinetics for the metabolite in these plots, and the 2,4-dichlorobenzoic acid residues are not considered further in this report. This is accepted by the CA. Furthermore, the CA notes there were some instances of 2,4-DBA greater than LOQ, however, these were often sporadic or where a clear decline phase could not be observed and so a robust kinetic evaluation could not be

undertaken. Further information is provided in the relevant kinetic evaluation section, summarised in Table CA.B.**8.1.2.3-2**.

Table CA.B.8.1.2.3-2.: Summary of kinetic evaluation undertaken for each field site

Field site	Plot	Compounds detected in study	Compounds included in kinetic evaluation	Link to kinetics
FR01	T1	Bixlozone and 2,4-DBA	Bixlozone and 2,4-DBA	Section CA.B.8.1.2.3.1
	T2	Bixlozone	Bixlozone	Section CA.B.8.1.2.3.2
	T3	Bixlozone and 2,4-DBA	Bixlozone and 2,4-DBA	Section CA.B.8.1.2.3.3
	T4	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.4
IT01	T1	Bixlozone and 2,4-DBA	Bixlozone and 2,4-DBA	Section CA.B.8.1.2.3.5
	T2	Bixlozone and 2,4-DBA	bixlozone	Section CA.B.8.1.2.3.6
	T3	Bixlozone and 2,4-DBA	Bixlozone and 2,4-DBA	Section CA.B.8.1.2.3.7
	T4	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.8
IT02	T2	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.9
GE01	T1	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.10
	T2	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.11
	T3	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.12
	T4	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.13
FR02	T1	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.14
	T2	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.15
	T3	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.16
	T4	Bixlozone	Bixlozone	Section CA.B.8.1.2.3.17
GE02	2	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.18
	3	Bixlozone, 2,4-DBA and 3-OH	Bixlozone	Section CA.B.8.1.2.3.19
UK01	2	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.20
	3	Bixlozone and 2,4-DBA	Bixlozone	Section CA.B.8.1.2.3.21

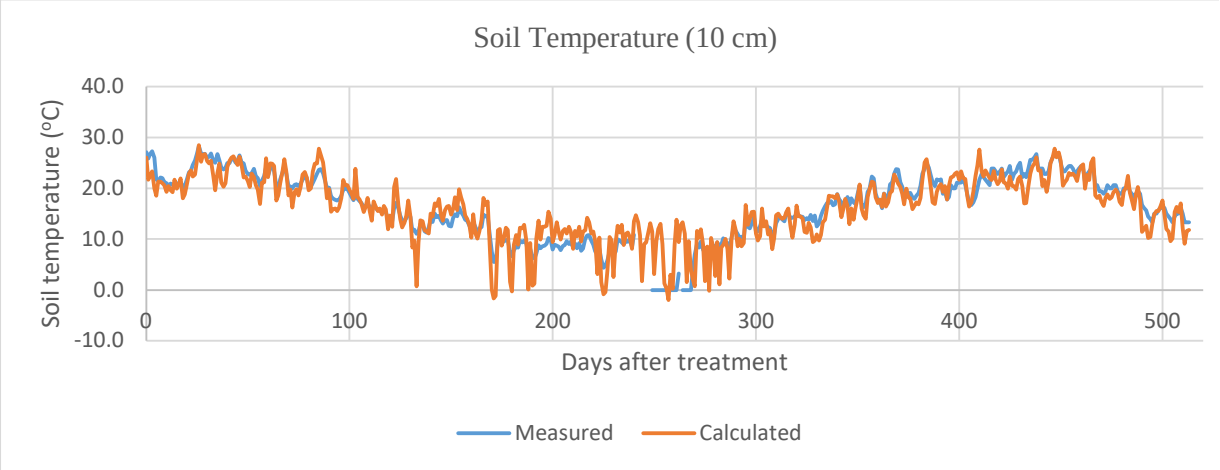
Using the weather data and soil analysis data, the applicant has normalised the field data to 20°C and pF2 using the time step normalsiation approach. For a number of the test sites, the applicant has used the program PERSIST (version 1.0) to calculate the daily soil moisture and temperature values, in order to normalise the days. The applicant notes daily soil temperature and soil moisture data at 10 cm depth were provided for the sites located at FR01, IT01 and IT02. However, for the French site, the applicant states the volumetric soil moisture values were above 100% in some cases, and hence were not considered as robust. The applicant further states there was no evidence that the Italian site used other methodology, therefore, for both sites the calculated values using PERSIST were considered. To further justify the use of PERSIST, the applicant compared the calculated soil temperatures with the recorded values, noting they showed good alignment (see Figure CA.B.**8.1.2.3-1**). Therefore, the CA accepts the use of PERSIST for these trial sites.

Similarly, for sites GE01 and FR02, the applicant notes although daily soil moisture and temperature values were provided for each site, the French site measured the moisture content using a tensiometer with the values having been informed in cBar,. Therefore, the applicant did not consider this data as robust and so used PERSIST to calculate daily soil moisture and temperature values instead. For consistency, the applicant also used PERSIST for site GE01. Again, to further justify the use of PERSIST, the applicant compared the calculated soil temperatures with the recorded values, noting they showed good alignment (see Figure CA.B.**8.1.2.3-1**). The applicant also compared the soil moisture values for site GE01, noting they too showed good alignment. Therefore, the CA accepts the use of PERSIST for these trial sites.

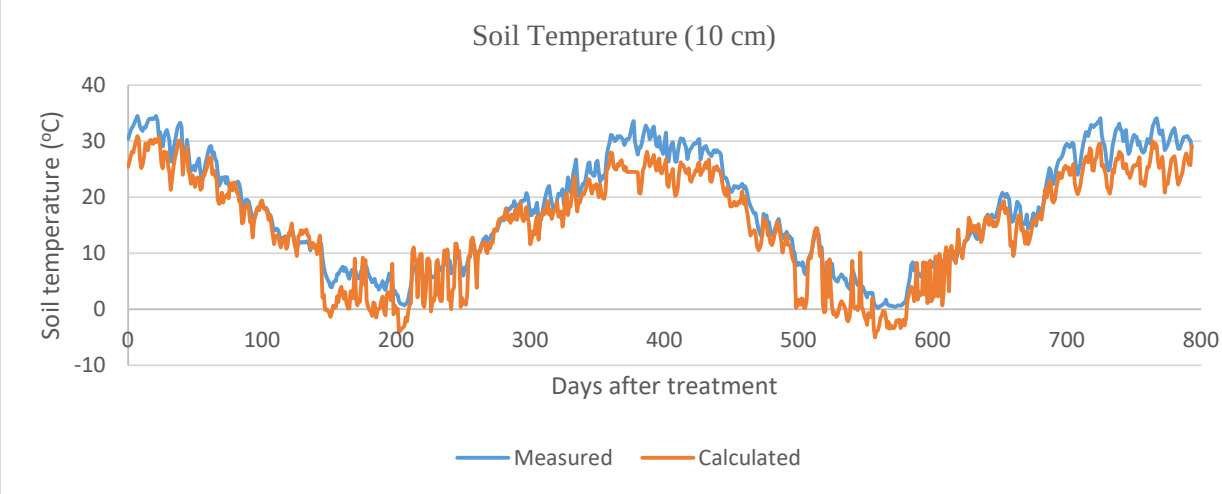
For sites GE02 and UK01, no concerns were raised with the reported daily soil moisture and temperature values and so the applicant used these for the timestep normalisation. This too is accepted by the CA.

Figure CA.B.8.1.2.3-1: Comparison of calculated and recorded soil temperatures and calculated and recorded soil moistures (site GE01 only)

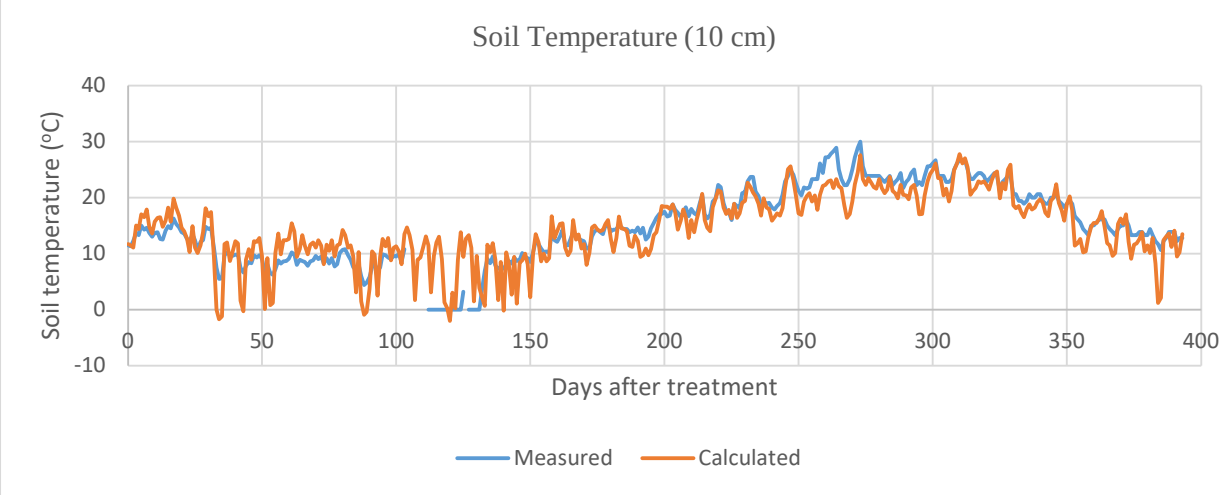
Site FR01



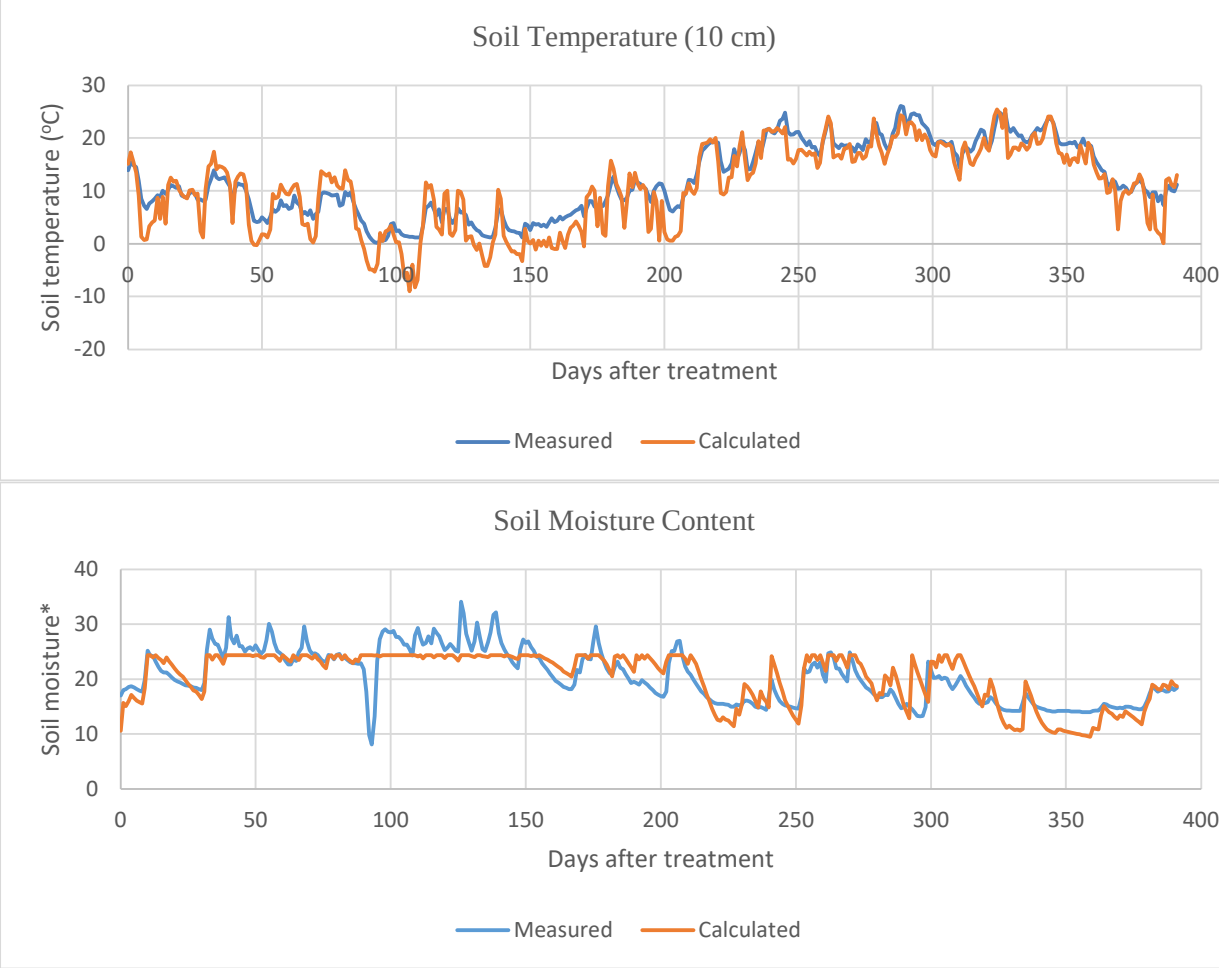
Site IT01 and IT02



Site FR02



Site GE01



*Measured: %v/v / Calculated: %w/w

The site parameters used in PERSIST are summarised in Table CA.B.**8.1.2.3-3**. The latitudes and altitudes were obtained by the applicant using Google Earth. However, as shown in Table CA.B.**8.1.2.3-3**, the CA obtained slightly different values when the CA entered the GPS coordinates, provided in the full study reports, into Google Earth (version 9.3.116.1). However, the differences had a negligible impact on the final corrected day values and so the applicant's normalised days are accepted. A Q10 of 2.58, a bulk density of 1.5 g/cm^3 and soil moisture and temperature values for the top 10 cm horizon were modelled.

Table CA.B.8.1.2.3-3: Site details for PERSIST

Site	Applicant's values		CA's values		WHC at pF2 (%w/w)	Soil type (USDA)
	Latitude	Altitude	Latitude	Altitude		
FR01	44.522	~ 45 m	44.523	29	19.0	Sandy loam
IT01	45.45	~ 105 m	45.438	97	30.3	Loam
IT02	45.45	~ 105 m	45.438	97	30.3	Loam
GE01	52.834	~55 m	52.877	49	19.5	Loamy sand
FR02	44.522	~45 m	44.524	29	17.9	Sandy loam

For sites GE02 and UK01, the measured soil temperature and moisture content at each site were used to determine the day length normalisation. The water holding capacity at pF2 (w/w%) was reported for both sites on a gravimetric basis. However, as the soil moisture values were reported as volumetric values, the applicant has used the default volumetric values from the FOCUS (2014) groundwater report as the reference moisture values. This is accepted by the CA. The site details are summarised in Table CA.B.**8.1.2.3-4**. It is noted the soil temperature at 4 and 5 DALA were not recorded at site GE02. The applicant has instead used 14.3°C, which is

the mean value of all the reported soil temperatures for May 2016 (the month of application). This approach is accepted by the CA. The CA obtained similar normalised days as to the applicant (see Table CA.B.**8.1.2.3-4**) and so the applicant's values are accepted.

Table CA.B.8.1.2.3-4: Normalisation details for sites GE02 and UK01

Site	Soil type (USDA)	Water Holding Capacity (w/w %) at pF2 from study	Default Soil Moisture at pF 2 (w/w %) (FOCUS, 2014)	Default Soil Moisture at pF 2 (v/v %) (FOCUS, 2014)	Applicant's final sampling occasion corrected day	CA's final sampling occasion corrected day
GE02	Loamy sand	22.4	14	20	180.2	180.9
UK01	Loam	19.2	25	34	148.3	150.0

The applicant performed the kinetic assessment using CAKE v3.2 (normalised) and v3.3 (non-normalised) with IRLS selected. For completeness, the CA has validated the modelling using KinGUII v2.1 with NLLS selected; the CA also used KinGUII v2.1(with NLLS selected) to perform the non-normalised persistence kinetic assessment for the field trials where incorporation did not occur. The kinetic assessment has been performed in line with FOCUS (2014) and EFSA DegT50 guidance. For the modelling endpoint assessment of the bare soil trials, the results prior to 10 mm rainfall have been omitted from the kinetic fits to account for the potential effect of surface processes.

Table CA.B.8.1.2.3-5: Total residues of bixlozone and 2,4-DBA in soil (mg/kg) – FR01, Plot T1 (F9600-4 SC, bare soil)

Formulation			Plot T1 (bare soil) / F9600-4 SC							
Actual sampling (DALA)	Normalised day	Sub-plot	Bixlozone residue (mg/kg dwt)				2,4-dichlorobenzoic acid residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues	0-10	10-30	Sum of residues	bixlozone equiv.*
0[a)]	0[a)]	a	0.2298	0	0	0.2298	0	0	0	0
		b	0.1466	0	0	0.1466	0	0	0	0
		c	0.1539	0	0	0.1539	0	0	0	0
3[a)]	2.5[a)]	a	0.1012	0.0015	0.0008	0.1035	0.0102	0	0.0102	0.0146
		b	0.1446	0	0	0.1446	0.0116	0	0.0116	0.0166
		c	0.1839	0	0	0.1839	0.0159	0	0.0159	0.0228
6	4.5	a	0.1175	0.0008	0.0022	0.1205	0.0234	0	0.0234	0.0336
		b	0.1079	0.0008	0	0.1087	0.0211	0	0.0211	0.0303
		c	0.1066	0	0	0.1066	0.0187	0	0.0187	0.0268
13	11.9	a	0.1276	0.0018	0.0008	0.1302	0.0041	0	0.0041	0.0059
		b	0.0794	0.0027	0.0008	0.0829	0.0033	0	0.0033	0.0047
		c	0.0847	0	0	0.0847	0.0026	0	0.0026	0.0037
28	26.5	a	0.0867	0.0008	0	0.0875	0.0057	0	0.0057	0.0082
		b	0.0945	0.0008	0	0.0953	0.0065	0	0.0065	0.0093
		c	0.1030	0	0	0.1030	0.0036	0	0.0036	0.0052
60	64.0	a	0.0460	0	0	0.0460	0.0008	0	0.0008	0.0011
		b	0.0380	0	0	0.0380	0.0025	0	0.0025	0.0036
		c	0.0401	0	0	0.0401	0.0021	0	0.0021	0.0030
91	98.0	a	0.0146	0	0	0.0146	0.0008	0	0.0008	0.0011
		b	0.0100	0	0	0.0100	0.0008	0	0.0008	0.0011
		c	0.0398	0	0	0.0398	0.0008	0	0.0008	0.0011
179	147.8	a	0.0120	0	0	0.0120	0	0	0	0
		b	0.0125	0	0	0.0125	0	0	0	0
		c	0.0090	0	0	0.0090	0	0	0	0
285	183.6	a	0.0143	0	0	0.0143	0	0	0	0
		b	0.0033	0	0	0.0033	0	0	0	0
		c	0.0042	0	0	0.0042	0	0	0	0
369	234.1	a	0.0008	0	0	0.0008	0	0	0	0
		b	0.0017	0	0	0.0017	0	0	0	0
		c	0.0008	0	0	0.0008	0	0	0	0

*2,4-dichlorobenzoic acid residues as bixlozone equivalents using molecular weight correction factor of 1.435 (bixlozone; 274.15 g/mol, 2,4-dichlorobenzoic acid; 191.01 g/mol).

[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-6: Total residues of bixlozone in soil (mg/kg) – FR01, Plot T2 (F9600-4 SC, incorporated)

Formulation			Plot T2 (incorporated) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Sub-plot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.2328	0	0	0.2328
		b	0.2739	0	0	0.2739
		c	0.2199	0	0	0.2199
3	2.5	a	0.2171	0.0117	0.0008	0.2296
		b	0.2491	0.0025	0.0008	0.2524
		c	0.1887	0.0016	0.0008	0.1911
6	4.5	a	0.2022	0.0008	0	0.2030
		b	0.1905	0.0008	0	0.1913
		c	0.1341	0.0008	0	0.1349
13	11.9	a	0.2323	0	0	0.2323
		b	0.2834	0.0015	0.0008	0.2857
		c	0.3029	0	0	0.3029
28	26.5	a	0.1324	0	0	0.1324
		b	0.1360	0.0008	0	0.1368
		c	0.1364	0	0	0.1364
60	64.0	a	0.0719	0	0	0.0719
		b	0.0863	0	0	0.0863
		c	0.0656	0	0	0.0656
91	98.0	a	0.0934	0	0	0.0934
		b	0.0766	0	0	0.0766
		c	0.0765	0	0	0.0765
179	147.8	a	0.0483	0	0	0.0483
		b	0.0393	0	0	0.0393
		c	0.0521	0.0008	0	0.0529
285	183.6	a	0.0145	0	0	0.0145
		b	0.0252	0	0	0.0252
		c	0.0348	0.0018[a)]	0.0008	0.0374
369	234.1	a	0.0178	0	0	0.0178
		b	0.0174	0	0	0.0174
		c	0.0124	0.0008	0	0.0132

[a)] The CA notes the applicant had this value in the 20 – 30 cm horizon, however, the CA has checked the study report and confirmed it is in the 10 – 20 cm horizon.

Table CA.B.8.1.2.3-7: Total residues of bixlozone and 2,4-dichlorobenzoic acid in soil (mg/kg) – FR01, Plot T3 (F9600-21 CS, bare soil)

Formulation			Plot T3 (bare soil) / F9600-21 CS							
Actual sampling (DALA)	Normalised day	Sub-plot	Bixlozone residue (mg/kg dwt)				2,4-dichlorobenzoic acid residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues	0-10	10-30	Sum of residues	bixlozone equiv.*
0[a)]	0[a)]	a	0.4851	0	0	0.4851	0	0	0	0
		b	0.1714	0	0	0.1714	0	0	0	0
		c	0.2291	0	0	0.2291	0	0	0	0
3[a)]	2.5[a)]	a	0.1126	0.0019[b)]	0.0008	0.1153	0.0156	0	0.0156	0.0224
		b	0.0846	0	0	0.0846	0.0147	0	0.0147	0.0211
		c	0.1212	0.0020	0.0008	0.1240	0.0228	0	0.0228	0.0327
6	4.5	a	0.1083	0.0008	0	0.1091	0.0181	0	0.0181	0.0260
		b	0.1672	0	0	0.1672	0.0206	0	0.0206	0.0296
		c	0.0635	0.0008	0.0008	0.0651	0.0165	0	0.0165	0.0237
13	11.9	a	0.1000	0	0	0.1000	0.0038	0	0.0038	0.0055
		b	0.1100	0	0	0.1100	0.0040	0	0.0040	0.0057
		c	0.0894	0.0008	0.0043	0.0945	0.0030	0	0.0030	0.0043
28	26.5	a	0.0888	0	0	0.0888	0.0058	0	0.0058	0.0083
		b	0.0854	0.0008	0	0.0862	0.0085	0	0.0085	0.0122
		c	0.0625	0	0.0008	0.0633	0.0029	0	0.0029	0.0042
60	64.0	a	0.0617	0	0	0.0617	0.0008	0	0.0008	0.0011
		b	0.0534	0.0030	0.0008	0.0572	0.0025	0	0.0025	0.0036
		c	0.0246	0	0	0.0246	0.0022	0	0.0022	0.0032
91	98.0	a	0.0397	0	0	0.0397	0	0	0	0
		b	0.0478	0.0008	0	0.0486	0.0008	0	0.0008	0.0011
		c	0.0297	0	0	0.0297	0.0008	0	0.0008	0.0011
179	147.8	a	0.0293	0	0	0.0293	0	0	0	0
		b	0.0188	0	0	0.0188	0	0	0	0
		c	0.0165	0.0008	0	0.0173	0	0	0	0
285	183.6	a	0.0328	0	0	0.0328	0	0	0	0
		b	0.0089	0	0	0.0089	0	0	0	0
		c	0.0066	0.0023	0.0008	0.0097	0	0	0	0
369	234.1	a	0.0094	0	0	0.0094	0	0	0	0
		b	0.0180	0	0	0.0180	0	0	0	0
		c	0.0108	0.0008	0	0.0116	0	0	0	0

*2,4-dichlorobenzoic acid residues as bixlozone equivalents using molecular weight correction factor of 1.435 (bixlozone; 274.15 g/mol, 2,4-dichlorobenzoic acid; 191.01 g/mol).

a) Sampling occasions prior to 10 mm rainfall

b) The CA notes the applicant had this value as 0.0033 mg/kg. The CA has checked the study report and has confirmed the correct value is 0.0019. The CA has also updated the summed value accordingly.

Table CA.B.8.1.2.3-8: Total residues of bixlozone in soil (mg/kg) – FR01, Plot T4 (F9600-21 CS, incorporated)

Formulation			Plot T4 (incorporated) / F9600-21 CS			
Actual sampling (DALA)	Normalised day	Sub-plot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.2144	0	0	0.2144
		b	0.2545	0	0	0.2545
		c	0.1950	0	0	0.1950
3	2.5	a	0.2401	0	0	0.2401
		b	0.1771	0	0.0008	0.1779
		c	0.2087	0	0	0.2087
6	4.5	a	0.1363	0	0	0.1363
		b	0.1883	0.0008	0.0069	0.1960
		c	0.1574	0	0	0.1574
13	11.9	a	0.2381	0	0	0.2381
		b	0.3105	0	0.0008	0.3113
		c	0.2355	0	0	0.2355
28	26.5	a	0.2454	0	0	0.2454
		b	0.2548	0	0	0.2548
		c	0.2709	0	0	0.2709
60	64.0	a	0.1736	0	0	0.1736
		b	0.1328	0	0	0.1328
		c	0.1455	0	0	0.1455
91	98.0	a	0.0836	0.0008	0	0.0844
		b	0.1357	0	0	0.1357
		c	0.1154	0.0008	0	0.1162
179	147.8	a	0.0864	0.0026	0.0008	0.0898
		b	0.1199	0.0008	0	0.1207
		c	0.1405	0.0044	0.0008	0.1457
285	183.6	a	0.0640	0.0008	0.0016	0.0664
		b	0.0410	0.0019	0.0008	0.0437
		c	0.1559	0.0023	0.0008	0.1590
369	234.1	a	0.0869	0	0.0008	0.0877
		b	0.0959	0.0048	0.0008	0.1015
		c	0.0956	0.0019	0.0008	0.0983

Table CA.B.8.1.2.3-9: Total residues of bixlozone and 2,4-dichlorobenzoic acid in soil (mg/kg) – IT01, Plot T1 (F9600-4 SC, bare soil)

Formulation			Plot T1 (bare soil) / bixlozone4 SC								
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)				2,4dichlorobenzoic acid residue (mg/kg dwt)				
			10	10-20	20-30	Sum of residues	10	10-20	20-30	Sum of residues	bixlozone equiv.*
0[a)]	0[a)]	a	0.0583	0	0	0.0583	0	0	0	0	0
		b	0.5496	0	0	0.5496	0	0	0	0	0
		c	0.2394	0	0	0.2394	0	0	0	0	0
3[a)]	3.5[a)]	a	0.0723	0.0008	0.0062	0.0793	0.0020	0	0	0.0020	0.0029
		b	0.2686	0.0008	0.0024	0.2718	0.0075	0	0	0.0075	0.0108
		c	0.2857	0.0008	0.0026	0.2891	0.0075	0	0	0.0075	0.0108
7[a)]	9.1[a)]	a	0.1314	0.0008	0.0021[b)]	0.1343	0.0089	0	0	0.0089	0.0128
		b	0.3151	0.0008	0.0016	0.3175	0.0161	0.0008	0	0.0169	0.0243
		c	0.1419	0.0027	0.0008	0.1454	0.0093	0	0	0.0093	0.0133
14[a)]	18.1[a)]	a	0.1273	0.0008	0.0008	0.1289	0.0099	0	0	0.0099	0.0142
		b	0.1487	0.0423	0.0008	0.1918	0.0112	0.0028[c)]	0.0008	0.0148	0.0212
		c	0.1410	0.0020	0.0008	0.1438	0.0109	0	0	0.0109	0.0156
30	43.1	a	0.0620	0.0018	0.0008	0.0646	0.0078	0	0	0.0078	0.0112
		b	0.1475	0.0190	0.0008	0.1673	0.0155	0.0019[d)]	0.0008	0.0182	0.0261
		c	0.1006	0.0008	0	0.1014	0.0194	0	0	0.0194	0.0278
58	85.2	a	0.0504	0.0008	0	0.0512	0.0008	0	0	0.0008	0.0011
		b	0.0618	0.0008	0	0.0626	0.0008	0.0008	0	0.0016	0.0023
		c	0.0768	0.0027	0.0008	0.0803	0.0008	0	0	0.0008	0.0011
91	120.4	a	0.0413	0	0	0.0413	0	0	0	0	0
		b	0.0618	0	0	0.0618	0	0	0	0	0
		c	0.0640	0.0008	0	0.0648	0	0	0	0	0
174	153.9	a	0.0140	0.0008	0	0.0148	0	0	0	0	0
		b	0.0256	0.0008	0	0.0264	0	0	0	0	0
		c	0.0384	0.0008	0	0.0392	0	0	0	0	0
269	174.7	a	0.0097	0.0015	0.0008	0.0120	0	0	0	0	0
		b	0.0197	0.0027	0.0008	0.0232	0	0	0	0	0
		c	0.0171	0.0025	0.0008	0.0204	0	0	0	0	0
368	260.3	a	0.0078	0.0008	0	0.0086	0	0	0	0	0
		b	0.0050	0.0008	0	0.0058	0	0	0	0	0
		c	0.0030	0.0008	0	0.0038	0	0	0	0	0

*2,4-dichlorobenzoic acid residues as bixlozone equivalents using molecular weight correction factor of 1.435 (bixlozone; 274.15 g/mol, 2,4-dichlorobenzoic acid; 191.01 g/mol).

a) Sampling occasions prior to 10 mm rainfall

b) The CA notes the applicant had this value as 0.0033 mg/kg. The CA has checked the study report and has confirmed the correct value is 0.0021 mg/kg. The CA has also updated the summed value accordingly.

c) The CA notes the applicant had this value as 0 mg/kg. The CA has checked the study report and has confirmed the correct value is 0.0028 mg/kg. The CA has also updated the summed value accordingly.

d) The CA notes the applicant had this value as 0 mg/kg. The CA has checked the study report and has confirmed the correct value is 0.0019 mg/kg. The CA has also updated the summed value accordingly.

Table CA.B.8.1.2.3-10: Total residues of bixlozone in soil (mg/kg) – IT01, Plot T2 (F9600-4 SC, incorporated)

Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)				2,4dichlorobenzoic acid residue (mg/kg dwt)				
			0-10	10-20	20-30	Sum of residues	0-10	10-20	20-30	Sum of residues	bixlozone equiv.*
0	0	a	0.1287	0	0	0.1287	0	0	0	0	0
		b	0.1349	0	0	0.1349	0	0	0	0	0
		c	0.0371	0	0	0.0371	0	0	0	0	0
3	3.5	a	0.1229	0.0056	0.0008	0.1293	0.0008	0	0	0.0008	0.0011
		b	0.1296	0	0	0.1296	0	0	0	0	0
		c	0.1159	0	0	0.1159	0	0	0	0	0
7	9.1	a	0.2234	0.0008	0.0019	0.2261	0.0015	0	0	0.0015	0.0022
		b	0.1376	0.0008	0	0.1384	0.0008	0	0	0.0008	0.0011
		c	0.0880	0	0	0.0880	0	0	0	0	0
14	18.1	a	0.1413	0.0016	0.0287	0.1716	0.0015	0	0	0.0015	0.0022
		b	0.1561	0.0090	0.0008	0.1659	0.0016	0	0	0.0016	0.0023
		c	0.1204	0	0	0.1204	0.0008	0	0	0.0008	0.0011
30	43.1	a	0.1368	0.0019	0.0008	0.1395	0.0029	0	0	0.0029	0.0042
		b	0.0828	0.0008	0	0.0836	0.0016	0	0	0.0016	0.0023
		c	0.1596	0	0	0.1596	0.0031	0	0	0.0031	0.0044
58	85.2	a	0.1020	0	0	0.1020	0.0008	0	0	0.0008	0.0011
		b	0.2409	0	0	0.2409	0.0008	0	0	0.0008	0.0011
		c	0.1254	0	0	0.1254	0.0008	0	0	0.0008	0.0011
91	120.4	a	0.0844	0	0	0.0844	0	0	0	0	0
		b	0.1497	0.0008	0	0.1505	0	0	0	0	0
		c	0.0589	0.0008	0	0.0597	0	0	0	0	0
174	153.9	a	0.1344	0	0	0.1344	0	0	0	0	0
		b	0.1227	0.0034	0.0008	0.1269	0	0	0	0	0
		c	0.1363	0.0068	0.0008	0.1439	0	0	0	0	0
269	174.7	a	0.0673	0.0031	0.0008	0.0712	0	0	0	0	0
		b	0.0451	0.0188	0.0008	0.0647	0	0	0	0	0
		c	0.0489	0.0008	0	0.0497	0	0	0	0	0
368	260.3	a	0.0294	0.0086	0.0008	0.0388	0	0	0	0	0
		b	0.0584	0.0168	0.0008	0.0760	0	0	0	0	0
		c	0.0241	0.0040	0.0008	0.0289	0	0	0	0	0
454	343.4	a	0.0177	0.0008	0.0008	0.0193	0	0	0	0	0
		b	0.0170	0.0008	0	0.0178	0	0	0	0	0
		c	0.0275	0.0008	0.0008	0.0291	0	0	0	0	0
542	373.8	a	0.0074	0.0008	0.0130	0.0212	0	0	0	0	0
		b	0.0216	0	0	0.0216	0	0	0	0	0
		c	0.0116	0.0008	0.0031	0.0155	0	0	0	0	0
640	395.8	a	0.0090	0	0.0008	0.0098	0	0	0	0	0
		b	0.0035	0	0	0.0035	0	0	0	0	0
		c	0.0095	0	0.0008	0.0103	0	0	0	0	0
736	483.3	a	0.0044	0	0	0.0044	0	0	0	0	0
		b	0.0048	0	0	0.0048	0	0	0	0	0
		c	0.0048	0	0	0.0048	0	0	0	0	0

Table CA.B.8.1.2.3-11: Total residues of bixlozone and 2,4-dichlorobenzoic acid in soil (mg/kg) – IT01, Plot T3 (F9600-21 CS, bare soil)

Formulation			Plot T3 (bare soil) / F9600-21 CS								
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)				2,4dichlorobenzoic acid residue (mg/kg dwt)				
			10	10-20	20-30	Sum of residues	10	10-20	20-30	Sum of residues	bixlozone equiv.*
0[a)]	0[a)]	a	0.1526	0	0	0.1526	0	0	0	0	0
		b	0.3644	0	0	0.3644	0	0	0	0	0
		c	0.3737	0	0	0.3737	0	0	0	0	0
3[a)]	3.5[a)]	a	0.1398	0.0008	0.0022	0.1428	0.0078	0.0008	0	0.0086	0.0123
		b	0.1526	0.0008	0.0030	0.1564	0.0115	0	0	0.0115	0.0165
		c	0.1440	0.0078	0.0120	0.1638	0.0081	0	0	0.0081	0.0116
7[a)]	9.1[a)]	a	0.0852	0.0108	0.0041	0.1001	0.0099	0.0018[b)]	0.0008	0.0125	0.0179
		b	0.1547	0.0008	0.0030	0.1585	0.0245	0	0	0.0245	0.0352
		c	0.1164	0.0008	0.0021	0.1193	0.0163	0	0	0.0163	0.0234
14[a)]	18.1[a)]	a	0.1177	0.0008	0.0016	0.1201	0.0195	0.0008	0	0.0203	0.0291
		b	0.1229	0.0008	0.0017	0.1254	0.0252	0.0008	0	0.0260	0.0373
		c	0.0903	0.0008	0.0018	0.0929	0.0168	0.0008	0	0.0176	0.0253
30	43.1	a	0.0548	0.0031	0.0008	0.0587	0.0008	0.0142[c)]	0.0008	0.0158	0.0227
		b	0.0434	0.0008	0.0008	0.0450	0.0008	0.0156[c)]	0.0008	0.0172	0.0247
		c	0.0634	0.0008	0.0008	0.0650	0.0008	0.0157[c)]	0.0008	0.0173	0.0248
58	85.2	a	0.0491	0.0047	0.0008	0.0546	0	0.0008	0	0.0008	0.0011
		b	0.1107	0.0122	0.0008	0.1237	0	0.0008	0	0.0008	0.0011
		c	0.0373	0	0	0.0373	0	0.0008	0	0.0008	0.0011
91	120.4	a	0.0233	0.0008	0	0.0241	0	0	0	0	0
		b	0.0407	0.0008	0	0.0415	0	0	0	0	0
		c	0.0374	0	0	0.0374	0	0	0	0	0
174	153.9	a	0.0507	0	0	0.0507	0	0	0	0	0
		b	0.0228	0	0	0.0228	0	0	0	0	0
		c	0.0207	0.0008	0	0.0215	0	0	0	0	0
269	174.7	a	0.0075	0.0008	0	0.0083	0	0	0	0	0
		b	0.0080	0	0	0.0080	0	0	0	0	0
		c	0.0208	0.0228	0.0008	0.0444	0	0	0	0	0
368	260.3	a	0.0116	0.0020	0.0008	0.0144	0	0	0	0	0
		b	0.0042	0	0	0.0042	0	0	0	0	0
		c	0.0080	0.0008	0	0.0088	0	0	0	0	0

*2,4-dichlorobenzoic acid residues as bixlozone equivalents using molecular weight correction factor of 1.435 (bixlozone; 274.15 g/mol, 2,4-dichlorobenzoic acid; 191.01 g/mol).

a) Sampling occasions prior to 10 mm rainfall

b) The CA notes the applicant had this value as 0 mg/kg. The CA has checked the study report and has confirmed the correct value is 0.0018 mg/kg. The CA has also updated the summed value accordingly.

c) The CA notes the applicant had these values in the 0 – 10 cm horizon, however, the CA has checked the study report and confirmed it is in the 10 – 20 cm horizon.

Table CA.B.8.1.2.3-12: Total residues of bixlozone in soil (mg/kg) –IT01, Plot T4 (F9600-21 CS, incorporated)

Formulation			Plot T4 (incorporated) / F9600-21 CS			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.3448	0	0	0.3448
		b	0.1786	0	0	0.1786
		c	0.2436	0	0	0.2436
3	3.5	a	0.2425	0.0008	0.0008	0.2441
		b	0.1568	0.0008	0.0008	0.1584
		c	0.1199	0.0195	0.0008	0.1402
7	9.1	a	0.1578	0.0008	0.0037	0.1623
		b	0.1350	0.0180	0.0065	0.1595
		c	0.1544	0.0018	0.0018	0.1580
14	18.1	a	0.1467	0.0121	0.0017	0.1605
		b	0.2935	0.0008	0.0021	0.2964
		c	0.2230	0.0008	0.0008	0.2246
30	43.1	a	0.1761	0	0	0.1761
		b	0.0604	0.0008	0	0.0612
		c	0.1624	0	0	0.1624
58	85.2	a	0.1560	0	0	0.1560
		b	0.1400	0.0022	0.0008	0.1430
		c	0.1595	0	0	0.1595
91	120.4	a	0.1494	0.0008	0	0.1502
		b	0.1360	0.0008	0	0.1368
		c	0.0920	0	0	0.0920
174	153.9	a	0.1265	0.0257	0.0008	0.1530
		b	0.0876	0.0008	0	0.0884
		c	0.1020	0.0008	0	0.1028
269	174.7	a	0.0734	0.0256	0.0008	0.0998
		b	0.1300	0.0284	0.0008	0.1592
		c	0.0973	0.0146	0.0008	0.1127
368	260.3	a	0.0625	0.0750	0.0008	0.1383
		b	0.1032	0.0073	0.0008	0.1113
		c	0.0798	0.0049	0.0008	0.0855
454	343.4	a	0.0471	0.0008	0.0008	0.0487
		b	0.0362	0.0008	0.0008	0.0378
		c	0.0326	0.0008	0	0.0334
542	373.8	a	0.0537	0.0203	0.0054	0.0794
		b	0.0361	0.0211	0.0064	0.0636
		c	0.0778	0.0201	0	0.0979
640	395.8	a	0.0233	0.0055	0.0008	0.0296
		b	0.0495	0.0008	0	0.0503
		c	0.0276	0.0008	0	0.0284
736	483.3	a	0.0200	0.0008	0	0.0208
		b	0.0190	0	0	0.0190
		c	0.0090	0	0	0.0090

Table CA.B.8.1.2.3-13: Total residues of bixlozone in soil (mg/kg) – IT02, Plot T2 (F9600-4 SC, incorporated)

Formulation			Plot T2 (incorporated) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Sub-plot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.6716	0.0072	0.0217	0.7005
		b	0.5901	0.0274	0.0233	0.6408
		c	0.5868	0.0165	0.0264	0.6297
3	2.6	a	0.4496	0.0123	0.0095	0.4714
		b	0.2972	0.0130	0.0881	0.3983
		c	0.3669	0.0567	0.0696	0.4932
7	6.3	a	0.2434	0.0385	0.0041	0.2860
		b	0.2102	0.0289	0.0815	0.3206
		c	0.2936	0.0210	0.0058	0.3204
14	12.2	a	0.2484	0.0008	0.0075	0.2567
		b	0.1880	0.0008	0.0043	0.1931
		c	0.2559	0.0056	0.0042	0.2657
30	22.3	a	0.1037	0	0.0008	0.1045
		b	0.1329	0	0.0008	0.1337
		c	0.1147	0.0029	0.0008	0.1184
60	36.3	a	0.0298	0	0	0.0298
		b	0.0760	0	0	0.0760
		c	0.1834	0.0008	0	0.1842
88	44.8	a	0.0705	0.0008	0	0.0713
		b	0.1281	0.0008	0.0008	0.1297
		c	0.0783	0.0008	0.0008	0.0799
186	58.8	a	0.0769	0.0168	0.0008	0.0945
		b	0.0771	0.0088	0.0094	0.0953
		c	0.0574	0.0142	0.0097	0.0813
263	108.2	a	0.0069	0.0008	0	0.0077
		b	0.0170	0.0021	0.0008	0.0199
		c	0.0312	0.0064	0.0072	0.0448
360	233.1	a	0.0026	0	0	0.0026
		b	0.0021	0.0008	0	0.0029
		c	0.0020	0.0008	0.0008	0.0036

Table CA.B.8.1.2.3-14: Total residues of bixlozone in soil (mg/kg) – GE01, Plot T1 (bixlozone-4SC, bare soil)

Formulation			Plot T1 (bare soil) / bixlozone4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0[a)]	0[a)]	a	0.1259	0	0	0.1259
		b	0.1137	0	0	0.1137
		c	0.1372	0	0	0.1372
3[a)]	1.6[a)]	a	0.1805	0.0008	0	0.1813
		b	0.0929	0	0	0.0929
		c	0.1362	0	0	0.1362
7	2.7	a	0.1812	0.0023	0.0008	0.1843
		b	0.1494	0.0020	0.0030	0.1544
		c	0.1477	0	0	0.1477
13	4.0	a	0.3660	0.0008	0	0.3668
		b	0.1634	0	0	0.1634
		c	0.0968	0	0	0.0968
29	9.7	a	0.1801	0	0	0.1801
		b	0.1772	0.0008	0	0.1780
		c	0.1750	0.0008	0	0.1758
58	20.7	a	0.1155	0.0008	0	0.1163
		b	0.1611	0.0019	0.0008	0.1638
		c	0.1558	0.0096	0.0008	0.1662
92	31.6	a	0.1087	0.0017	0.0008	0.1112
		b	0.1643	0.0008	0	0.1651
		c	0.1011	0.0008	0	0.1019
183	47.1	a	0.0874	0.0008	0.0008	0.0890
		b	0.0951	0	0	0.0951
		c	0.0796	0	0	0.0796
267	96.9	a	0.0639	0.0008	0.0017	0.0664
		b	0.0622	0	0	0.0622
		c	0.0451	0.0008	0	0.0459
359	170.6	a	0.0162	0.0023	0.0008	0.0193
		b	0.0181	0	0	0.0181
		c	0.0183	0.0032	0.0008	0.0223

[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-15: Total residues of bixlozone in soil (mg/kg) – GE01, Plot T2 (F9600-4 SC, incorporated)

Formulation			**Plot T2 (incorporated) / bixlozone4 SC**			
Actual sampling (DALA)	**Normalised day**	**Subplot**	**Bixlozone residue (mg/kg dwt)**			
			0-10	**10-20**	**20-30**	**Sum of residues**
0	0	a	0.0737	0	0	0.0737
		b	0.0434	0	0	0.0434
		c	0.2406	0	0	0.2406
3	1.6	a	0.0475	0	0	0.0475
		b	0.1519	0	0	0.1519
		c	0.0737	0	0	0.0737
7	2.7	a	0.1531	0	0	0.1531
		b	0.1582	0	0	0.1582
		c	0.1350	0	0	0.1350
13	4.0	a	0.0548	0	0	0.0548
		b	0.2836	0	0	0.2836
		c	0.0882	0	0	0.0882
29	9.7	a	0.1031	0	0	0.1031
		b	0.1652	0.0008	0	0.1660
		c	0.1155	0	0	0.1155
58	20.7	a	0.1629	0	0	0.1629
		b	0.1196	0.0081	0.0008	0.1285
		c	0.0843	0	0	0.0843
92	31.6	a	0.1049	0	0	0.1049
		b	0.0918	0.0008	0	0.0926
		c	0.1714	0	0	0.1714
183	47.1	a	0.1072	0	0	0.1072
		b	0.0737	0	0	0.0737
		c	0.0819	0	0	0.0819
267	96.9	a	0.0309	0	0	0.0309
		b	0.0440	0	0	0.0440
		c	0.0181	0.0008	0	0.0189
359	170.6	a	0.0113	0	0	0.0113
		b	0.0231	0	0	0.0231
		c	0.0099	0.0018	0.0008	0.0125

Table CA.B.8.1.2.3-16**:** Total residues of bixlozone in soil (mg/kg) – GE01, Plot T3 (F9600-21 CS, bare soil)

Formulation			**Plot T3 (bare soil) / F9600-21 CS**			
Actual sampling (DALA)	**Normalised day**	**Subplot**	**Bixlozone residue (mg/kg dwt)**			
			0-10	**10-20**	**20-30**	**Sum of residues**
0[a)]	0[a)]	a	0.1468	0	0	0.1468
		b	0.1494	0	0	0.1494
		c	0.0563	0	0	0.0563
3[a)]	1.6[a)]	a	0.1881	0	0	0.1881
		b	0.0718	0	0	0.0718
		c	0.1471	0	0	0.1471
7	2.7	a	0.2560	0	0	0.2560
		b	0.1469	0	0	0.1469
		c	0.1527	0	0	0.1527
13	4.0	a	0.1388	0	0	0.1388
		b	0.0908	0	0	0.0908
		c	0.3547	0.0008	0	0.3555
29	9.7	a	0.1855	0.0008	0	0.1863
		b	0.1626	0	0	0.1626
		c	0.1610	0.0019	0.0008	0.1637
58	20.7	a	0.1156	0.0020	0.0008	0.1184
		b	0.1569	0	0.0008	0.1577
		c	0.0761	0.0008	0	0.0769
92	31.6	a	0.1219	0.0008	0	0.1227
		b	0.1054	0.0008	0.0017	0.1079
		c	0.1026	0	0	0.1026
183	47.1	a	0.1156	0	0	0.1156
		b	0.0993	0	0.0008	0.1001
		c	0.0791	0	0	0.0791
267	96.9	a	0.0726	0.0008	0.0008	0.0742
		b	0.0564	0.0008	0	0.0572
		c	0.0620	0.0008	0	0.0628
359	170.6	a	0.0306	0.0064	0.0045	0.0415
		b	0.0254	0.0040	0.0008	0.0302
		c	0.0191	0.0031	0.0008	0.0230

[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-17**:** Total residues of bixlozone in soil (mg/kg) – GE01, Plot T4 (F9600-21 CS, incorporated)

Formulation			**Plot T4 (bare soil) / F9600-21 CS**			
Actual sampling (DALA)	**Normalised day**	**Subplot**	**Bixlozone residue (mg/kg dwt)**			
			0-10	**10-20**	**20-30**	**Sum of residues**
0	0	a	0.1299	0	0	0.1299
		b	0.1338	0	0	0.1338
		c	0.1812	0	0	0.1812
3	1.6	a	0.0852	0	0	0.0852
		b	0.1125	0	0	0.1125
		c	0.1307	0	0	0.1307
7	2.7	a	0.0821	0	0	0.0821
		b	0.0792	0	0	0.0792
		c	0.1336	0	0	0.1336
13	4.0	a	0.1920	0	0	0.1920
		b	0.2569	0	0	0.2569
		c	0.0790	0	0	0.0790
29	9.7	a	0.0858	0	0	0.0858
		b	0.1451	0	0	0.1451
		c	0.0765	0	0	0.0765
58	20.7	a	0.1000	0	0	0.1000
		b	0.1186	0.0008	0	0.1194
		c	0.1052	0	0	0.1052
92	31.6	a	0.0871	0	0	0.0871
		b	0.1380	0.0023	0.0008	0.1411
		c	0.0933	0	0	0.0933
183	47.1	a	0.1802	0.0008	0	0.1810
		b	0.1325	0.0008	0	0.1333
		c	0.0736	0	0	0.0736
267	96.9	a	0.0663	0.0016	0.0008	0.0687
		b	0.0605	0.0008	0	0.0613
		c	0.0431	0.0008	0	0.0439
359	170.6	a	0.0400	0.0008	0	0.0408
		b	0.0365	0.0020	0.0008	0.0393
		c	0.0358	0.0034	0.0008	0.0400

Table CA.B.8.1.2.3-18: Total residues of bixlozone in soil (mg/kg) – FR02, Plot T1 (F9600-4 SC, bare soil)

Formulation			Plot T1 (bare soil) / bixlozone4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0[a)]	0[a)]	a	0.1437	0	0	0.1437
		b	0.1392	0	0	0.1392
		c	0.1435	0	0	0.1435
3[a)]	1.3[a)]	a	0.1141	0.0008	0.0015	0.1164
		b	0.1131	0	0	0.1131
		c	0.1174	0.0008	0.0017	0.1199
6[a)]	3.3[a)]	a	0.1109	0	0.0008	0.1117
		b	0.1250	0	0	0.1250
		c	0.1082	0	0.0008	0.1090
13	7.9	a	0.0934	0	0	0.0934
		b	0.1229	0	0	0.1229
		c	0.1268	0	0	0.1268
30	17.0	a	0.0867	0	0	0.0867
		b	0.0756	0	0	0.0756
		c	0.1007	0.0008	0	0.1015
58	26.0	a	0.0603	0	0	0.0603
		b	0.0600	0	0	0.0600
		c	0.0664	0.0021	0.0008	0.0693
125	49.5	a	0.0370	0	0	0.0370
		b	n/a	n/a	n/a	n/a
		c	0.0302	0.0008	0	0.0310
188	76.6	a	0.0127	0	0	0.0127
		b	0.0185	0	0	0.0185
		c	0.0180	0	0	0.0180
290	154.0	a	0.0015	0	0	0.0015
		b	0.0019	0	0	0.0019
		c	0.0030	0	0	0.0030
372	221.1	a	0.0008	0	0	0.0008
		b	0.0008	0	0	0.0008
		c	0.0008	0	0	0.0008

n/a: sample not collected due to waterlogging
[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-19: Total residues of bixlozone in soil (mg/kg) – FR02, Plot T2 (F9600-4 SC, incorporated)

Formulation			Plot T2 (incorporated) / bixlozone4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.1464	0	0	0.1464
		b	0.1132	0	0	0.1132
		c	0.1169	0	0	0.1169
3	1.3	a	0.1520	0	0	0.1520
		b	0.1202	0	0	0.1202
		c	0.1221	0	0	0.1221
6	3.3	a	0.1279	0	0	0.1279
		b	0.1029	0	0	0.1029
		c	0.1291	0	0	0.1291
13	7.9	a	0.1100	0	0	0.1100
		b	0.1267	0	0	0.1267
		c	0.1213	0	0	0.1213
30	17.0	a	0.1173	0	0	0.1173
		b	0.1257	0	0	0.1257
		c	0.1080	0	0	0.1080
58	26.0	a	0.1205	0	0	0.1205
		b	0.1140	0	0	0.1140
		c	0.0655	0	0	0.0655
125	49.5	a	0.1032	0	0	0.1032
		b	0.0387	0	0	0.0387
		c	0.0510	0	0	0.0510
188	76.6	a	0.0326	0	0	0.0326
		b	0.0397	0	0	0.0397
		c	0.0419	0	0	0.0419
290	154.0	a	0.0076	0	0	0.0076
		b	0.0108	0	0	0.0108
		c	0.0036	0	0	0.0036
372	221.1	a	0.0024	0	0	0.0024
		b	0.0031	0	0	0.0031
		c	0.0025	0	0	0.0025

Table CA.B.8.1.2.3-20: Total residues of bixlozone in soil (mg/kg) – FR02, Plot T3 (F9600-21 CS, bare soil)

Formulation			Plot T3 (bare soil) / F9600-21 CS			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0[a)]	0[a)]	a	0.1568	0	0	0.1568
		b	0.1258	0	0	0.1258
		c	0.1518	0	0	0.1518
3[a)]	1.3[a)]	a	0.1477	0	0	0.1477
		b	0.1073	0	0	0.1073
		c	0.2110	0	0	0.2110
6[a)]	3.3[a)]	a	0.0973	0	0	0.0973
		b	0.0800	0	0	0.0800
		c	0.1287	0	0	0.1287
13	7.9	a	0.1360	0	0	0.1360
		b	0.0960	0	0	0.0960
		c	0.1118	0	0	0.1118
30	17.0	a	0.1143	0	0	0.1143
		b	0.1250	0	0	0.1250
		c	0.0946	0	0	0.0946
58	26.0	a	0.0822	0	0	0.0822
		b	0.0585	0	0	0.0585
		c	0.0567	0	0	0.0567
125	49.5	a	0.0396	0	0	0.0396
		b	0.0436	0	0	0.0436
		c	0.0530	0	0	0.0530
188	76.6	a	0.0385	0	0	0.0385
		b	0.0205	0	0	0.0205
		c	0.0380	0	0	0.0380
290	154.0	a	0.0078	0	0	0.0078
		b	0.0088	0	0	0.0088
		c	0.0137	0	0	0.0137
372	221.1	a	0.0034	0	0	0.0034
		b	0.0078	0	0	0.0078
		c	0.0129	0	0	0.0129

[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-21: Total residues of bixlozone in soil (mg/kg) – FR02, Plot T4 (F9600-21 CS, incorporated)

Formulation			Plot T4 (incorporated) / F9600-21 CS)			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.1253	0	0	0.1253
		b	0.1207	0	0	0.1207
		c	0.0993	0	0	0.0993
3	1.3	a	0.1427	0	0	0.1427
		b	0.0932	0	0	0.0932
		c	0.1327	0	0	0.1327
6	3.3	a	0.1457	0	0	0.1457
		b	0.1276	0	0	0.1276
		c	0.1019	0	0.0008	0.1027
13	7.9	a	0.1434	0	0	0.1434
		b	0.1303	0	0	0.1303
		c	0.1458	0.0008	0.0033	0.1499
30	17.0	a	0.1334	0	0	0.1334
		b	0.1301	0	0	0.1301
		c	0.1473	0	0.0008	0.1481
58	26.0	a	0.1533	0	0	0.1533
		b	0.0778	0	0	0.0778
		c	0.0738	0	0	0.0738
125	49.5	a	0.1217	0	0	0.1217
		b	0.0960	0	0	0.0960
		c	0.1251	0	0	0.1251
188	76.6	a	0.1297	0	0	0.1297
		b	0.0325	0	0	0.0325
		c	0.0406	0	0	0.0406
290	154.0	a	0.0270	0.0008	0	0.0278
		b	0.0259	0	0	0.0259
		c	0.0585	0	0	0.0585
372	221.1	a	0.0467	0.0033	0.0008	0.0508
		b	0.0280	0	0	0.0280
		c	0.0327	0	0	0.0327

Table CA.B.8.1.2.3-22: Total residues of bixlozone in soil (mg/kg) – GE02, Plot 2 (F9600-4 SC, incorporated)

Formulation			Plot 2 (incorporated) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.1685	0	0	0.1685
		b	0.1215	0	0	0.1215
		c	0.1197	0	0	0.1197
3	1.6	a	0.1185	0	0	0.1185
		b	0.0635	0	0	0.0635
		c	0.1104	0	0	0.1104
7	3.9	a	0.1086	0	0	0.1086
		b	0.0926	0	0	0.0926
		c	0.1060	0	0	0.1060
14	8.4	a	0.0568	0	0	0.0568
		b	0.0817	0	0	0.0817
		c	0.0666	0	0	0.0666
28	21.4	a	0.0495	0	0	0.0495
		b	0.0795	0	0	0.0795
		c	0.0826	0	0	0.0826
58	46.5	a	0.0641	0	0	0.0641
		b	0.0797	0	0	0.0797
		c	0.0643	0	0	0.0643
90	76.4	a	0.0337	0	0	0.0337
		b	0.0467	0	0	0.0467
		c	0.0262	0	0	0.0262
177	132.6	a	0.0605	0	0	0.0605
		b	0.0418	0	0	0.0418
		c	0.0460	0	0	0.0460
272	153.4	a	0.0657	0	0	0.0657
		b	0.0432	0	0	0.0432
		c	0.0583	0	0	0.0583
358	180.2	a	0.0348	0	0	0.0348
		b	0.0372	0	0	0.0372
		c	0.0203	0	0	0.0203

Table CA.B.8.1.2.3-23: Total residues of bixlozone in soil (mg/kg) – GE02, Plot 3 (F9600-4 SC, bare soil)

Formulation			Plot 3 (bare soil) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0[a)]	0[a)]	a	0.1251	0	0	0.1251
		b	0.1469	0	0	0.1469
		c	0.1093	0	0	0.1093
3[a)]	1.6[a)]	a	0.1383	0.0008	0.0017	0.1408
		b	0.1021	0	0	0.1021
		c	0.1150	0.0008	0.0079	0.1237
7	3.9	a	0.1217	0	0.0008	0.1225
		b	0.0987	0	0	0.0987
		c	0.1205	0	0.0008	0.1213
14	8.4	a	0.0679	0	0	0.0679
		b	0.0843	0	0	0.0843
		c	0.0678	0	0	0.0678
28	21.4	a	0.0739	0	0	0.0739
		b	0.0492	0	0	0.0492
		c	0.0696	0	0	0.0696
58	46.5	a	0.0423	0	0	0.0423
		b	0.0420	0.0008	0	0.0428
		c	0.0523	0.0008	0	0.0531
90	76.4	a	0.0527	0	0	0.0527
		b	0.0314	0.0022	0.0008	0.0344
		c	0.0396	0.0019	0.0008	0.0423
177	132.6	a	0.0357	0.0008	0	0.0365
		b	0.0260	0.0008	0	0.0268
		c	0.0330	0.0008	0	0.0338
272	153.4	a	0.0402	0.0024	0.0008	0.0434
		b	0.0264	0.0019	0.0008	0.0291
		c	0.0268	0	0	0.0268
358	180.2	a	0.0185	0.0008	0	0.0193
		b	0.0123	0.0008	0	0.0131
		c	0.0190	0	0	0.0190

[a)] Sampling occasions prior to 10 mm rainfall

Table CA.B.8.1.2.3-24: Total residues of bixlozone in soil (mg/kg) – UK01, Plot 2 (F9600-4 SC, incorporated)

Formulation			Plot 2 (incorporated) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0	0	a	0.0867	0	0	0.0867
		b	0.1035	0	0	0.1035
		c	0.1673	0	0	0.1673
3	1.7	a	0.1456	0	0	0.1456
		b	0.1488	0	0	0.1488
		c	0.0966	0	0	0.0966
6	2.8	a	0.1075	0	0	0.1075
		b	0.1079	0	0	0.1079
		c	0.0957	0	0	0.0957
15	9.6	a	0.0890	0	0	0.0890
		b	0.0944	0	0	0.0944
		c	0.1010	0	0	0.1010
30	19.6	a	0.0770	0	0	0.0770
		b	0.0887	0	0	0.0887
		c	0.0728	0	0	0.0728
58	40.6	a	0.1045	0	0	0.1045
		b	0.0599	0	0	0.0599
		c	0.0650	0	0	0.0650
87	60.0	a	0.0578	0	0	0.0578
		b	0.0586	0	0	0.0586
		c	0.0796	0	0	0.0796
185	98.0	a	0.0410	0	0	0.0410
		b	0.0568	0	0	0.0568
		c	0.0390	0	0	0.0390
263	113.2	a	0.0407	0	0	0.0407
		b	0.0448	0	0	0.0448
		c	0.0462	0	0	0.0462
371	148.3	a	0.0372	0	0	0.0372
		b	0.0345	0	0	0.0345
		c	0.0489	0	0	0.0489

Table CA.B.8.1.2.3-25: Total residues of bixlozone in soil (mg/kg) – Trial UK01, Plot 3 (F9600-4 SC, bare soil)

Formulation			Plot 3 (bare soil) / F9600-4 SC			
Actual sampling (DALA)	Normalised day	Subplot	Bixlozone residue (mg/kg dwt)			
			0-10	10-20	20-30	Sum of residues
0[a)]	0[a)]	a	0.1237	0	0	0.1237
		b	0.1409	0	0	0.1409
		c	0.1840	0	0	0.1840
3[a)]	1.7[a)]	a	0.1161	0	0	0.1161
		b	0.0700	0	0	0.0700
		c	0.1013	0	0	0.1013
6[a)]	2.8[a)]	a	0.1383	0	0	0.1383
		b	0.0857	0	0	0.0857
		c	0.1279	0	0	0.1279
15	9.6	a	0.0695	0.0008	0	0.0703
		b	0.0596	0	0	0.0596
		c	0.0925	0.0008	0	0.0933
30	19.6	a	0.0677	0.0023	0.0008	0.0708
		b	0.0791	0	0	0.0791
		c	0.1104	0.0027	0.0008	0.1139
58	40.6	a	0.0664	0.0008	0	0.0672
		b	0.0701	0	0	0.0701
		c	0.0682	0.0021	0.0008	0.0711
87	60.0	a	0.0508	0	0	0.0508
		b	0.0636	0	0	0.0636
		c	0.0668	0.0008	0	0.0676
185	98.0	a	0.0325	0	0	0.0325
		b	0.0229	0.0008	0	0.0237
		c	0.0270	0	0	0.0270
263	113.2	a	0.0263	0	0	0.0263
		b	0.0188	0.0017	0.0008	0.0213
		c	0.0274	0	0	0.0274
371	148.3	a	0.0173	0	0	0.0173
		b	0.0219	0.0020	0.0008	0.0247
		c	0.0269	0	0	0.0269

[a)] Sampling occasions prior to 10 mm rainfall

CA.B.8.1.2.3.1. FR01 T1

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted DFOP provided the best visual fit of the models and had the lowest χ^2 value, however, k1 failed the t-test. Nevertheless, due to the DFOP fit providing the best fit of the initial rapid decline phase, the applicant concludes the DFOP model best-fits the data. The applicant then re-ran the DFOP parent fit with the metabolite data modelled in sequence (both SFO and DFOP models were ran for the metabolite). The applicant concludes the DFOP – SFO fit was visually and statistically acceptable, therefore, these endpoints were selected as the best-fit for this test system.

To validate the applicant's modelling, the CA ran SFO – SFO and DFOP – SFO fits of the non-normalised data. The CA concurs with the applicant that the parent DFOP fit best describes the data. As this assessment is determining the best-fit kinetics, the CA accepts the applicant's argument to disregard the k1 t-test result; furthermore, the CA notes the k1 t-test value in the CA's simulations (with NLLS selected) was 0.06 and so is below the 0.1 threshold.

The CA notes χ^2 values >20% are obtained for the metabolite simulations. However, the CA considers this to be due to the dataset and not the kinetic model selected. As such, the CA agrees with the applicant in selecting SFO as the best-fit model for the metabolite data. As the CA's values were similar to the applicant's (DT50 differences of <2 days), the applicant's values are accepted on this occasion.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.1-1** and the graphical outputs in Figure CA.B.**8.1.2.3.1-1** and Figure CA.B.**8.1.2.3.1-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.1-4**.

Table CA.B.8.1.2.3.1-1: Summary of FR01 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**							**CA's modelling**			
Pathway		**Bixlozone only**			**Bixlozone → 2,4-DBA**				**Bixlozone → 2,4-DBA**			
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**
Model		SFO	FOMC	DFOP	**DFOP**	**SFO**	DFOP	DFOP	SFO	SFO	DFOP	SFO
Visual fit		Good	Good	Good	**Good**	**Acceptable**	Good	Acceptable	Acceptable	Poor	Good	Acceptable
DT_{50} (days)		30.8	21.4	21.4	**19.6**	**2.77**	19.7	2.78	29.7	3.29	21.5	3.05
DT_{90} (days)		102	157	118	**125**	**9.22**	126	9.25	98.7	10.9	119	10.1
DT_{90}/3.32 (days)		n/a	47.1	35.5	**37.7**	**n/a**	45.7	2.78	n/a	n/a	35.8	n/a
DT_{50} (days) - Slow phase		n/a	n/a	41.8	**45.3**	**n/a**	13.8	2.79	n/a	n/a	42.1	n/a
χ^2 error (%)		14.4	12.9	8.84	**9.13**	**35.5**	9.14	44.7	14.3	86.9	8.90	44.7
k ($days^{-1}$)	k or k_1	0.02251	n/a	0.3704	**0.2813**	**0.2499**	0.281	0.2491	0.0233	0.2110	0.3526	0.2276
	k_2	n/a	n/a	0.0166	**0.0153**	**n/a**	0.01518	0.249	n/a	n/a	0.0165	n/a
P value	k or k_1	9.83E-007	n/a	0.1422	**0.05238**	**0.03518**	n.d.	n.d.	2.85E-11	0.268	0.0568	0.1289
	k_2	n/a	n/a	2.14E-004	**6.19E-005**	**n/a**	1.49E-11	n.d.	n/a	n/a	2.29E-7	n/a
g		n/a	n/a	0.2869	**0.3265**	**n/a**	0.3274	0.5442	n/a	n/a	0.2884	n/a
alpha		n/a	1.219	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a	n/a
beta		n/a	27.87	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a	n/a
95% CI (lower/upper)	alpha	n/a	0.01924 / 2.418	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a	n/a
	beta	n/a	-17.28 / 73.03	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a	n/a

n.d – Not determined: parameter could not be calculated by the CAKE model.
Best-fit model shown in bold

Figure CA.B.8.1.2.3.1-1: Applicant’s FR01 T1 parent-only kinetic fits

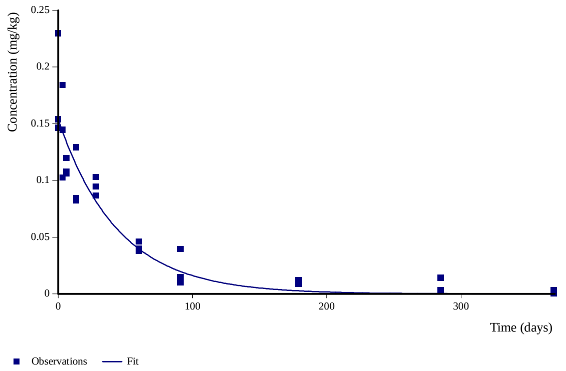
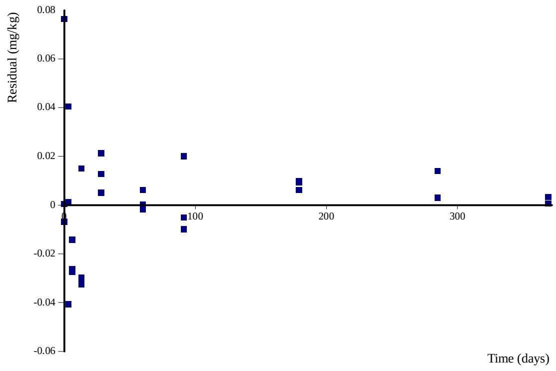
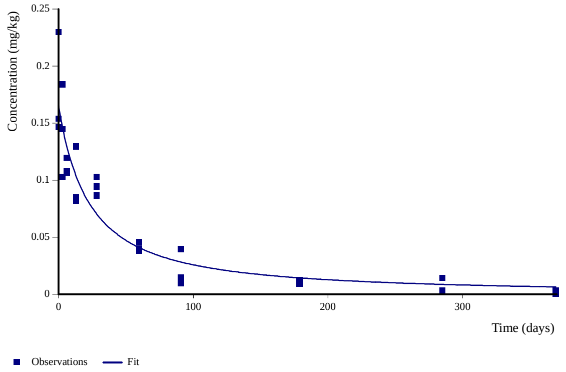
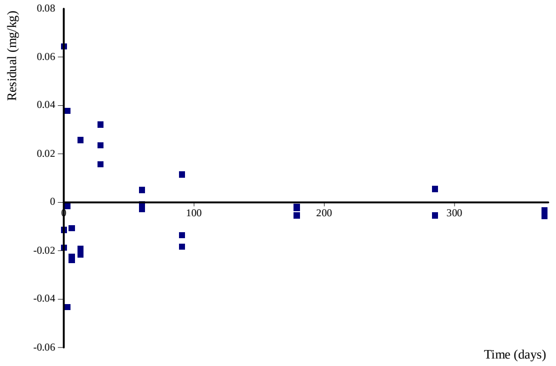
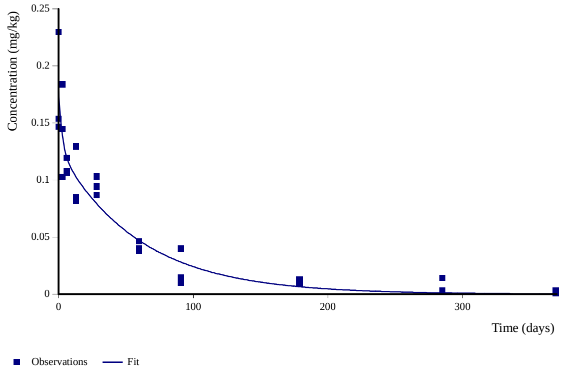
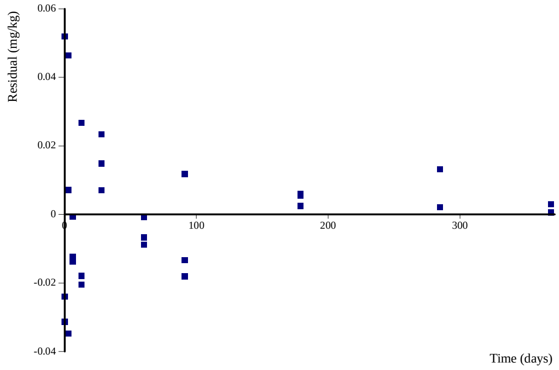
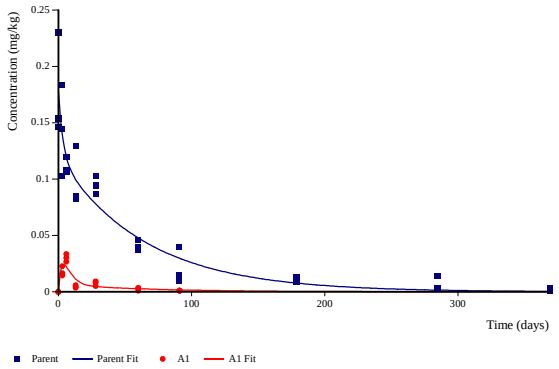
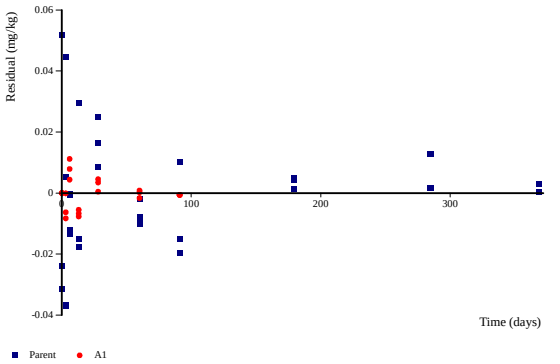
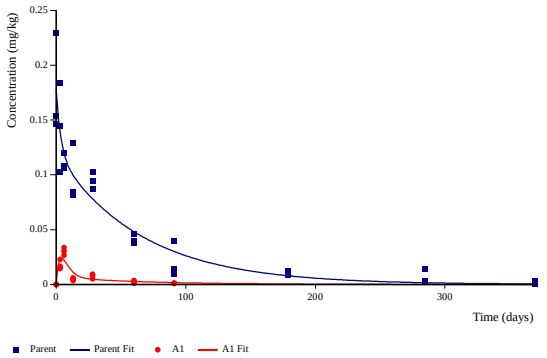
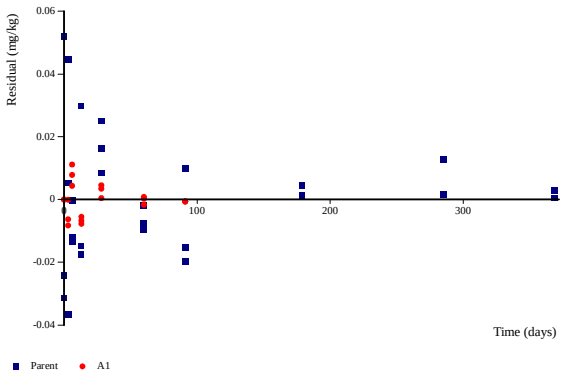
Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Figure CA.B.8.1.2.3.1-2: Applicant's FR01 T1 parent and metabolite kinetic fits

Fitted Model	**Residuals**
DFOP-SFO	DFOP-SFO
	
DFOP-DFOP	DFOP-DFOP
	

Persistence endpoints

As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a very good visual and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.1-2** and Figure CA.B.**8.1.2.3.1-3**.

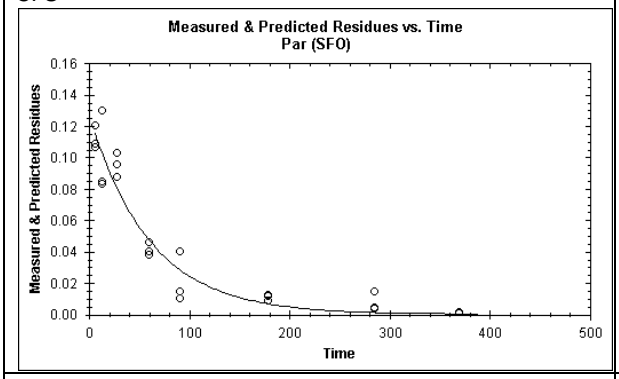
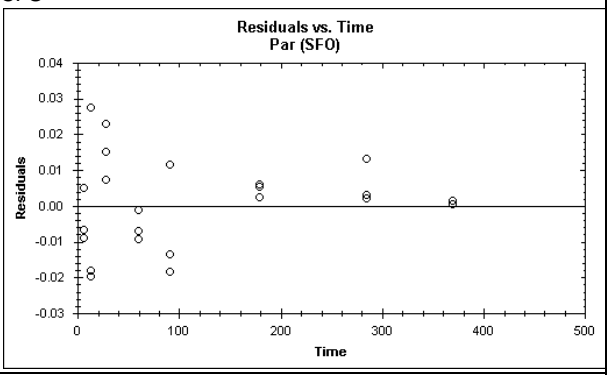
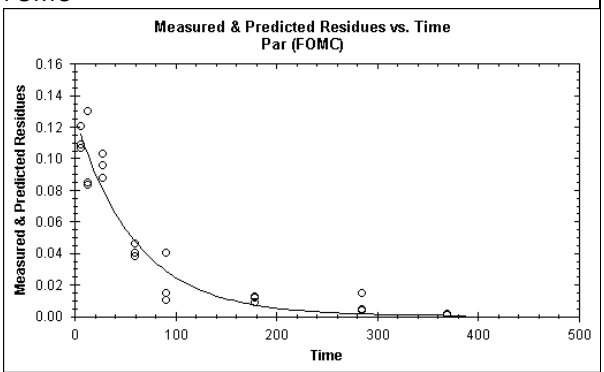
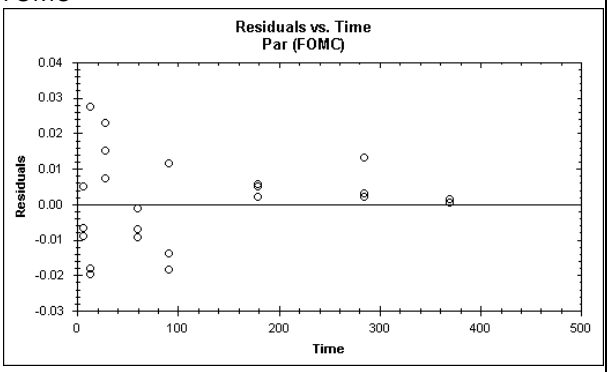
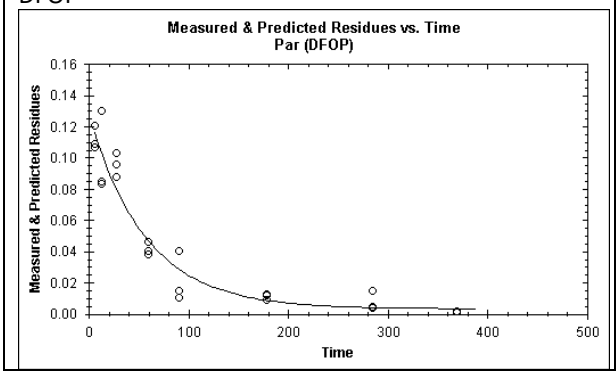
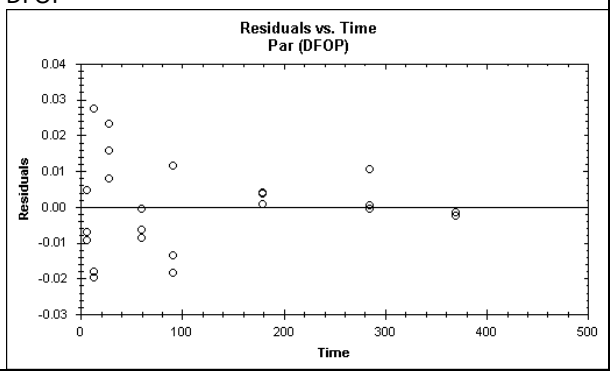
The CA has not considered 2,4-DBA in the persistence kinetic evaluation as formation occurred prior to 10 mm rainfall.

Table CA.B.8.1.2.3.1-2: Summary of FR01 T1 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Very good**	Very good	Very good
DT_{50} (days)		**41.9**	41.7	40.7
DT_{90} (days)		**139**	141	145
DT_{90}/3.32 (days)		**n/a**	42.3	43.6
DT_{50} (days) - Slow phase		**n/a**	n/a	39.3
χ^2 error (%)		**11.5**	12.3	12.9
k ($days^{-1}$)	k or k_1	**0.01656**	n/a	2.220E-14
	k_2	**n/a**	n/a	1.762E-2
P value	k or k_1	**3.05E-8**	n/a	0.5000
	k_2	**n/a**	n/a	0.0055
g		**n/a**	n/a	2.353E-2
alpha		**n/a**	6.093E+1	n/a
beta		**n/a**	3.647E-3	n/a
95% CI (lower/upper)	alpha	**n/a**	-2.347E+3 / 2.468E+3	n/a
	beta	**n/a**	-1.422E+5 / 1.495E+5	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.1-3: CA's FR01 T1 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). As >5% 2,4-DBA was formed prior to 10 mm rainfall, it has not been considered in the kinetic assessment. The applicant concluded the SFO fit provided a very good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.1-3** and the graphical outputs in Figure CA.B.**8.1.2.3.1-4**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.1-4**.

Table CA.B.8.1.2.3.1-3: Summary of FR01 T1 modelling endpoint kinetic modelling

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Very good**	Very good
DT_{50} (days)		**43.5**	43.5
DT_{90} (days)		**145**	145
DT_{50} (days) - Slow phase		**n/a**	43.5
χ^2 error (%)		**9.12**	10.5
k ($days^{-1}$)	k or k_1	**0.01593**	0.2647
	k_2	**n/a**	0.0159
P value	k or k_1	**2.05E-9**	Not determined
	k_2	**n/a**	4.38E-9
g		**n/a**	1.23E-7

Selected model shown in bold

Figure CA.B.8.1.2.3.1-4: Applicant's FR01 T1 parent-only modelling kinetic fits

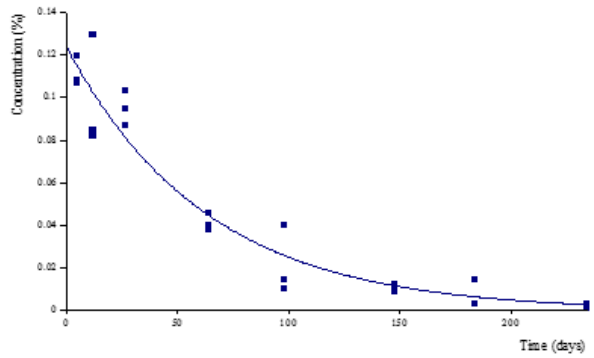
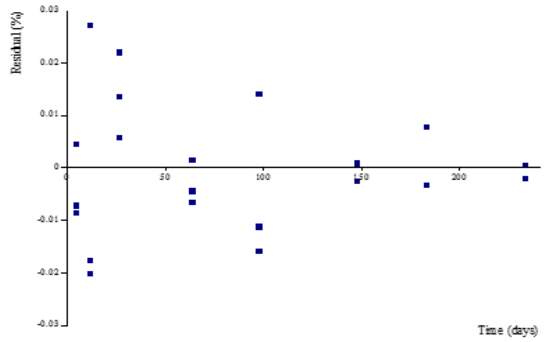
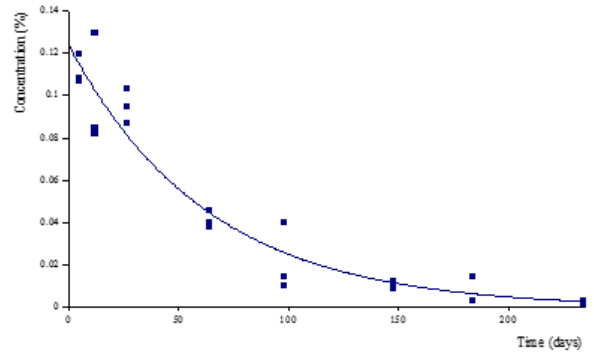
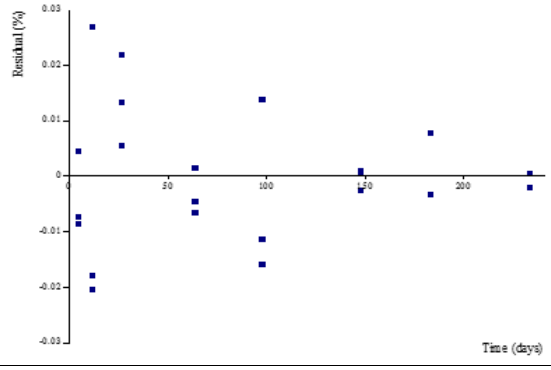
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.1-4: Summary of FR01 T1 modelling, persistence and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	19.6	125
2,4-DBA	SFO	2.77	9.22
Persistence endpoints			
Bixlozone	SFO	41.9	139
Modelling endpoints			
Bixlozone	SFO	43.5	145

CA.B.8.1.2.3.2. FR01 T2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared (no metabolites were detected in this trial). The applicant noted SFO provided a good visual and statistical fit of the data, whereas, the biphasic models were statistically unacceptable. Therefore, the applicant concludes SFO as the best-fit model.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs and is able to replicate the applicant's results. The CA notes that DFOP provided the best visual fit of the data (with SFO likely underestimating the DT90) and that failure of the t-test alone is not a reason to reject to the fit. However, the applicant's SFO fit is accepted on this occasion because the DT90 from all fits were less than the 365d accumulation trigger, the DT50 values are sufficiently small to not impact on the final PECsoil calculations and the DT50s were sufficiently similar so as not to impact on the persistence assessment.

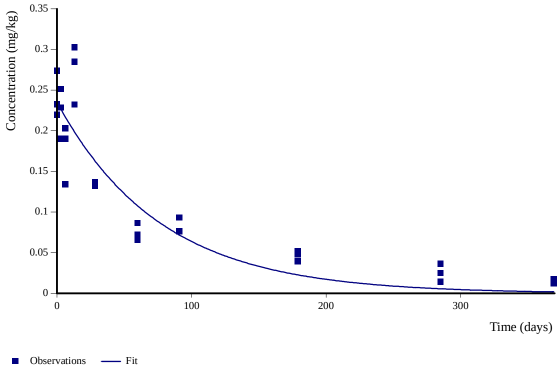
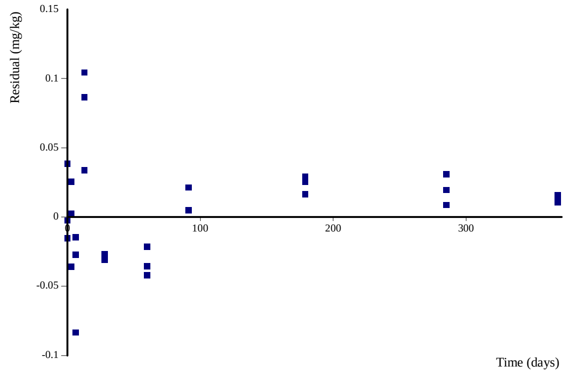
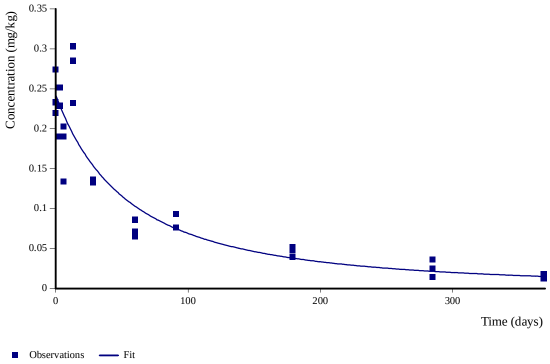
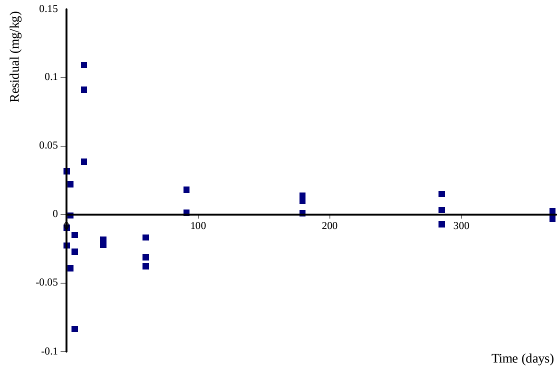
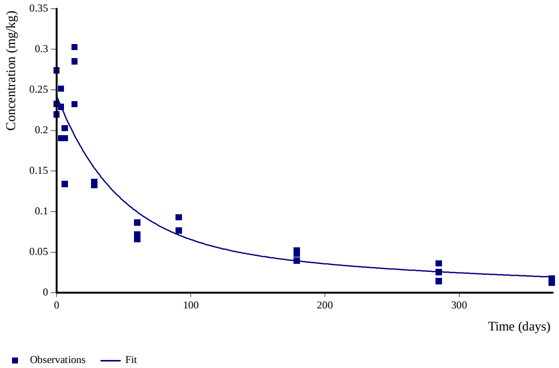
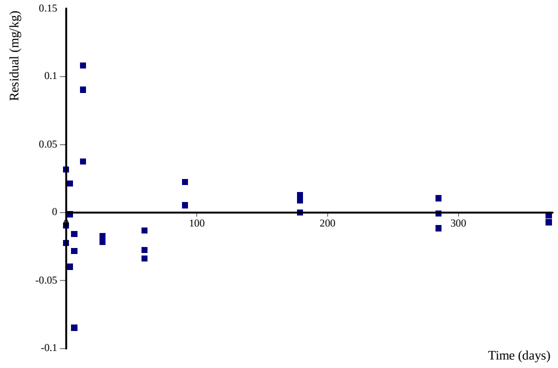
A summary of the applicant's kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.2-1** and the graphical outputs in Figure CA.B.**8.1.2.3.2-1**; the applicant's values are accepted by the CA, although, the CA considers the visual fit to be acceptable rather than "good". The final endpoints selected are summarised in Table CA.B.**8.1.2.3.2-3**.

Table CA.B.8.1.2.3.2-1: Summary of FR01 T2 best-fit kinetic modelling

Modelling		**Applicant's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Good**	Good	Good
DT_{50} (days)		**53.2**	46.3	44.7
DT_{90} (days)		**177**	261	304
DT_{90}/3.32 (days)		**n/a**	78.5	91.6
DT_{50} (days) - Slow phase		**n/a**	n/a	215
χ^2 error (%)		**20.1**	20.0	20.9
k ($days^{-1}$)	k or k_1	**0.01304**	n/a	0.02234
	k_2	**n/a**	n/a	0.003226
P value	k or k_1	**1.61E-6**	n/a	0.1039
	k_2	**n/a**	n/a	0.318
g		**n/a**	n/a	0.7352
alpha		**n/a**	1.74	n/a
beta		**n/a**	94.6	n/a
95% CI (lower/upper)	alpha	**n/a**	-1.073 / 4.552	n/a
	beta	**n/a**	-121.4 / 310.6	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.2-1: Applicant's FR01 T2 parent and metabolite kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided a good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.2-2** and the graphical outputs in Figure CA.B.**8.1.2.3.2-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.2-3**.

Table CA.B.8.1.2.3.2-2: Summary of FR01 T2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good
DT_{50} (days)		**53.9**	47.8
DT_{90} (days)		**179**	209
DT_{50} (days) - Slow phase		**n/a**	98.7
χ^2 error (%)		**19.2**	21.0
k ($days^{-1}$)	k or k_1	**0.01286**	0.02181
	k_2	**n/a**	0.00703
P value	k or k_1	**1.33E-7**	0.3286
	k_2	**n/a**	0.3789
g		**n/a**	0.5924

Selected model shown in bold

Figure CA.B.8.1.2.3.2-2: Applicant's FR01 T2 parent-only modelling kinetic fits

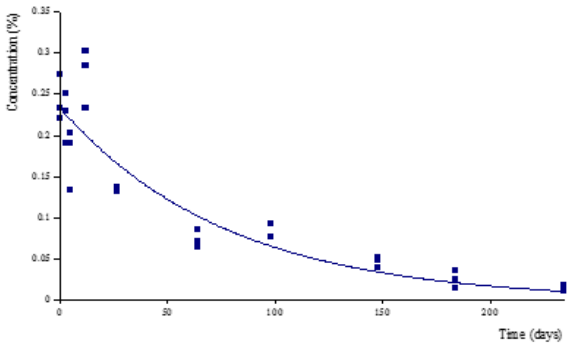
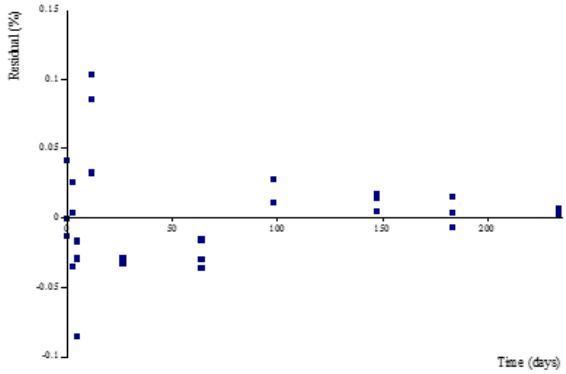
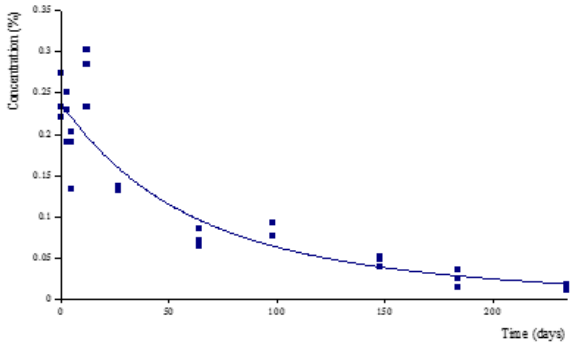
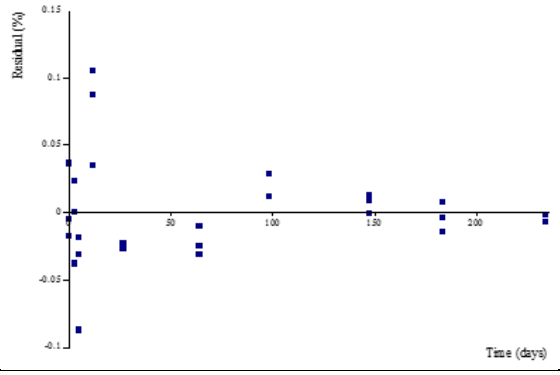
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.2-3: Summary of FR01 T2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	53.2	177
Modelling endpoints			
Bixlozone	SFO	53.9	179

CA.B.8.1.2.3.3. FR01 T3

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted DFOP provided the best visual fit of the models and had the lowest χ^2 value, however, k1 failed the t-test. Nevertheless, due to the DFOP fit providing the best fit of the initial rapid decline phase, the applicant concludes the DFOP model best-fits the data. The applicant then re-ran the DFOP parent fit with the metabolite data modelled in sequence (both SFO and DFOP models were ran for the metabolite). The applicant concludes the DFOP – SFO fit was visually and statistically acceptable, therefore, these endpoints were selected as the best-fit for this test system.

To validate the applicant's modelling, the CA ran parent-only SFO and parent-plus-metabolite DFOP – SFO fits of the non-normalised data. The CA concurs with the applicant that the parent DFOP fit best describes the data. As this assessment is determining the best-fit kinetics, the CA accepts the applicant's argument to disregard the k1 t-test result; furthermore, the CA notes the k1 t-test value is acceptable in the parent-plus-metabolite simulation.

The CA notes χ^2 values >20% are obtained for the metabolite simulations. However, the CA considers this to be due to the dataset and not the kinetic model selected. As such, the CA agrees with the applicant in selecting SFO as the best-fit model for the metabolite data. As the CA's values were similar to the applicant's (DT50 differences of <1 day), the applicant's values are accepted on this occasion.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.3-1** and the graphical outputs in Figure CA.B.**8.1.2.3.3-1** and Figure CA.B.**8.1.2.3.3-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.3-4**.

Table CA.B.8.1.2.3.3-1: Summary of FR01 T3 best-fit kinetic modelling

Modelling		**Applicant's modelling**							**CA's modelling**		
Pathway		**Bixlozone only**			**Bixlozone → 2,4-DBA**				**Bixlozone only**	**Bixlozone → 2,4-DBA**	
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**Bixlozone**	**2,4-DBA**
Model		SFO	FOMC	DFOP	**DFOP**	**SFO**	DFOP	DFOP	SFO	DFOP	SFO
Visual fit		Unacceptable	Very good	Very good	**Very good**	**Acceptable**	Very good	Acceptable	Poor	Good	Acceptable
DT_{50} (days)		7.86	1.95	0.472	**0.2**	**6.4**	2.88	3.29	8.12	0.007	6.63
DT_{90} (days)		26.1	250	118	**108**	**21.3**	147	15.7	27.0	119	22.03
DT_{90}/3.32 (days)		n/a	75.1	35.5	**32.5**	**n/a**	44.3	4.73	n/a	35.8	n/a
DT_{50} (days) - Slow phase		n/a	n/a	60.7	**53.1**	**n/a**	87.9	37.5	n/a	61.2	n/a
χ^2 error (%)		41.2	13.2	7.55	**8.04**	**27.1**	14.7	25.7	41.1	7.63	33.7
k ($days^{-1}$)	k or k_1	0.08819	n/a	3.529	**9.253**	**0.1083**	0.4452	0.2456	0.0854	244.6	0.1045
	k_2	n/a	n/a	0.01141	**0.01305**	**n/a**	0.00789	0.01849	n/a	0.01133	n/a
P value	k or k_1	0.003997	n/a	0.4939	**5.03E-44**	**5.45E-4**	0.03344	0.08394	0.0042	2.0E-16	0.2843
	k_2	n/a	n/a	0.04297	**0.02818**	**n/a**	0.0807	0.2507	n/a	0.0023	n/a
g		n/a	n/a	0.6139	**0.5925**	**n/a**	0.6816	19.8	n/a	0.6130	n/a
alpha		n/a	0.3414	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
beta		n/a	0.2941	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
95% CI (lower/upper)	alpha	n/a	0.09286 / 0.59	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
	beta	n/a	-0.5653 / 1.154	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.3-1: Applicant's FR01 T3 parent-only kinetic fits

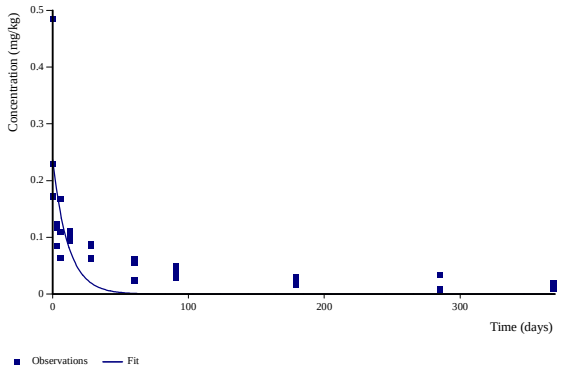
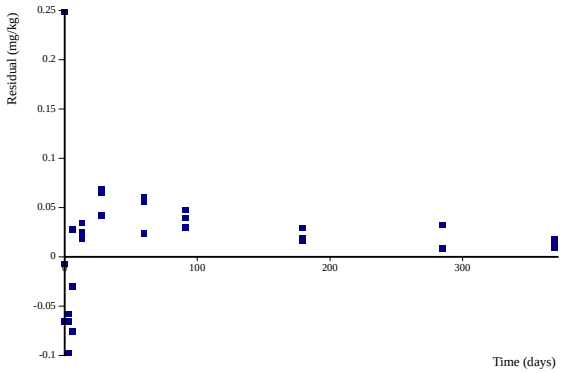
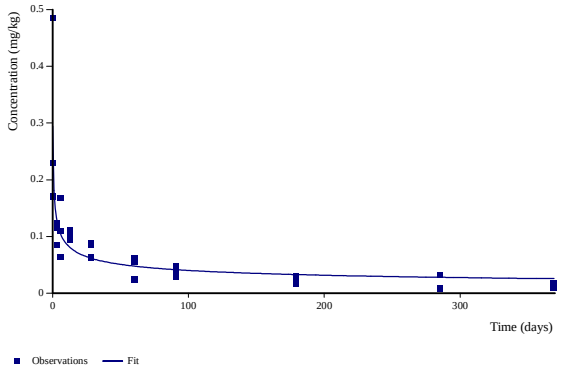
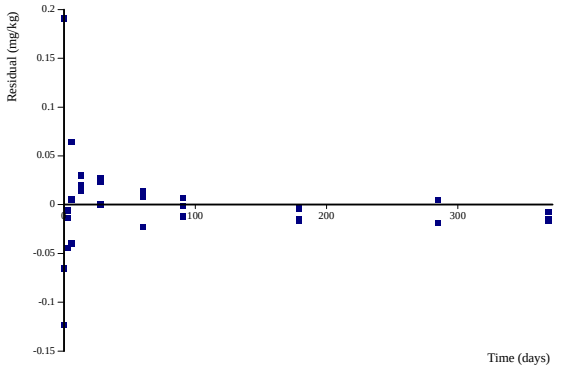
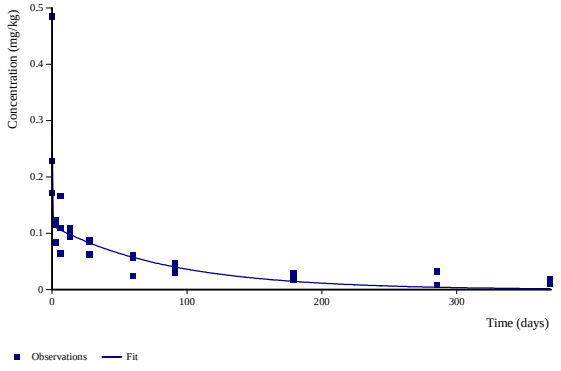
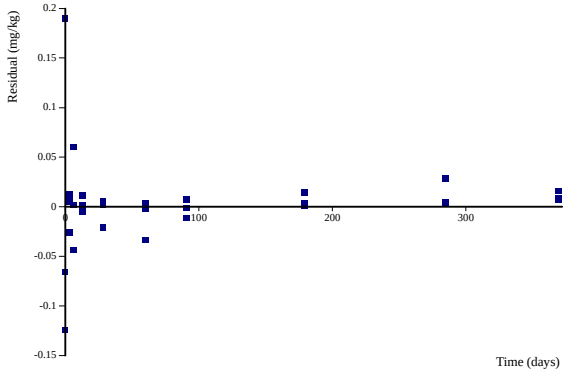
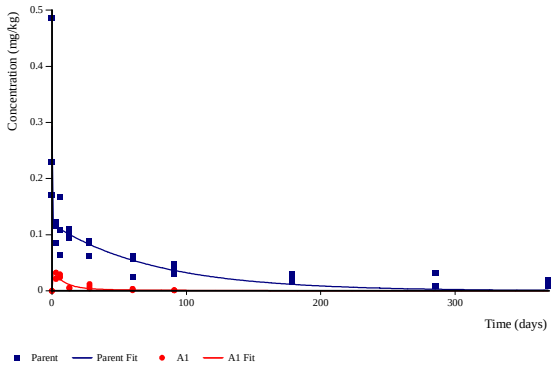
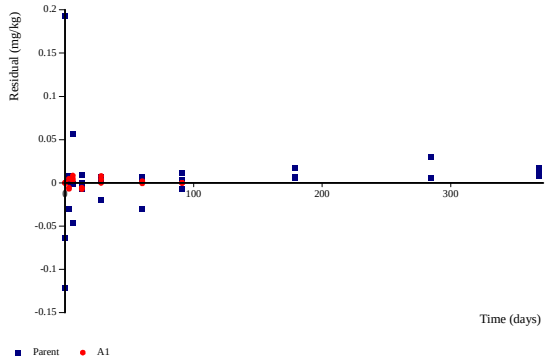
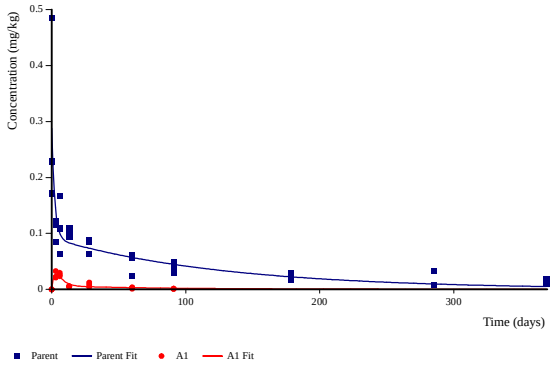
Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

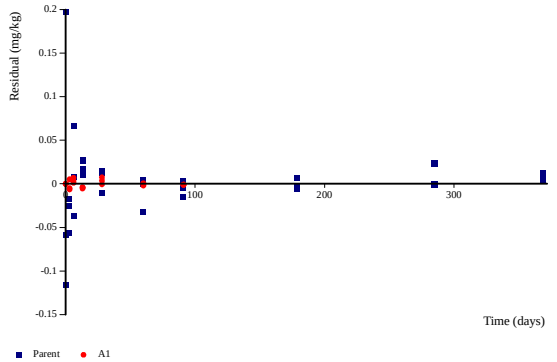
Figure CA.B.8.1.2.3.3-2: Applicant's FR01 T3 parent and metabolite kinetic fits

Fitted Model	**Residuals**
DFOP-SFO	DFOP-SFO
DFOP-DFOP	DFOP-DFOP









Persistence endpoints

As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided an acceptable visual and statistical fit of the data. Although the biphasic fits were visually better than the SFO fit, because the DT50 values were all significantly less than the 120d persistence trigger, the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.3-2** and Figure CA.B.**8.1.2.3.3-3**.

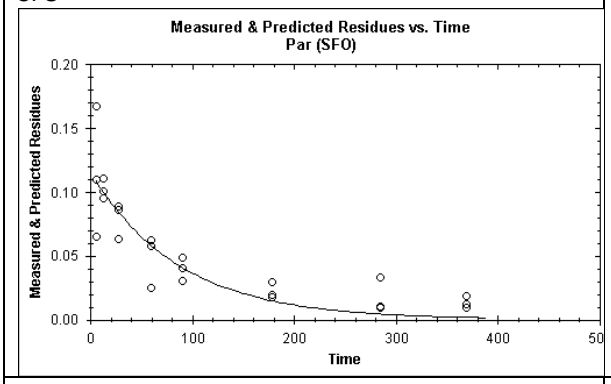
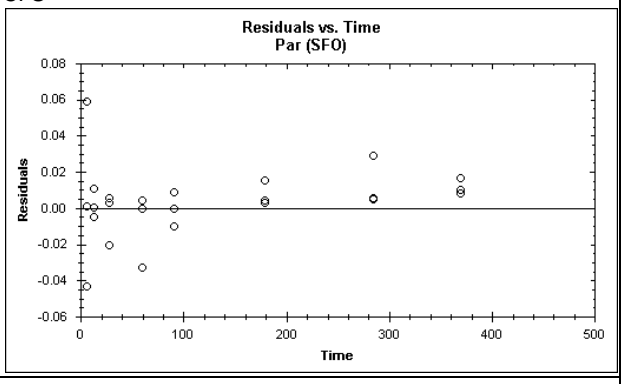
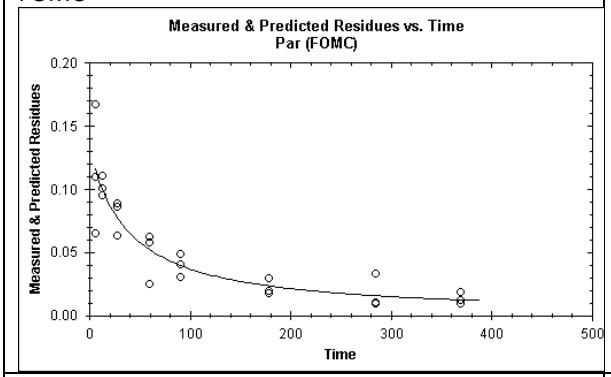
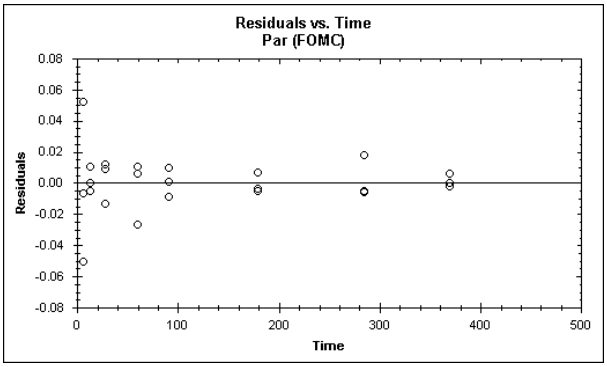
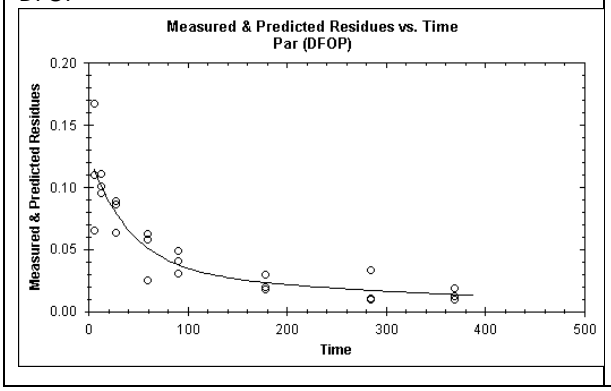
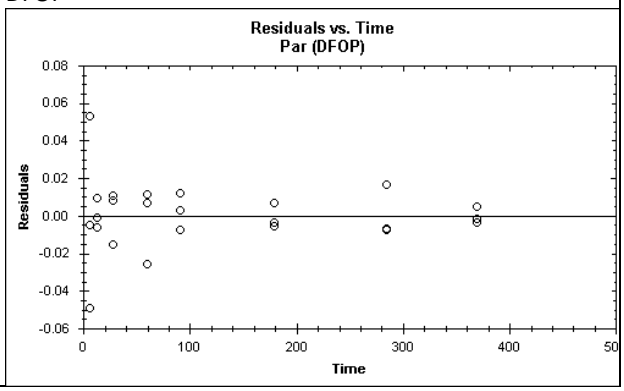
The CA has not considered 2,4-DBA in the persistence kinetic evaluation as formation occurred prior to 10 mm rainfall.

Table CA.B.8.1.2.3.3-2: Summary of FR01 T3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Acceptable**	Very good	Very good
DT_{50} (days)		**59.4**	38.1	41.7
DT_{90} (days)		**197**	329	381
DT_{90}/3.32 (days)		**n/a**	99.1	115
DT_{50} (days) - Slow phase		**n/a**	n/a	263
χ^2 error (%)		**11.7**	3.15	2.36
k ($days^{-1}$)	k or k_1	**0.01166**	n/a	0.025081
	k_2	**n/a**	n/a	0.002638
P value	k or k_1	**3.67E-5**	n/a	0.1104
	k_2	**n/a**	n/a	0.3001
g		**n/a**	n/a	0.7271
alpha		**n/a**	1.0369	n/a
beta		**n/a**	40.047	n/a
95% CI (lower/upper)	alpha	**n/a**	-0.15257 / 2.226	n/a
	beta	**n/a**	-51.3579 / 131.45	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.3-3: CA's FR01 T3 parent-only persistence kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). As >5% 2,4-DBA was formed prior to 10 mm rainfall, it has not been considered in the kinetic assessment. The applicant concluded the SFO fit provided a very good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.3-3** and the graphical outputs in Figure CA.B.**8.1.2.3.3-4**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.3-4**.

Table CA.B.8.1.2.3.3-3: Summary of FR01 T3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Very good**	Very good
DT_{50} (days)		**59.1**	44.4
DT_{90} (days)		**196**	227
DT_{50} (days) - Slow phase		**n/a**	88.1
χ^2 error (%)		**6.24**	2.89
k ($days^{-1}$)	k or k_1	**0.01174**	0.03671
	k_2	**n/a**	0.00787
P value	k or k_1	**2.41E-6**	0.3095
	k_2	**n/a**	0.1537
g		**n/a**	0.4027

Selected model shown in bold

Figure CA.B.8.1.2.3.3-4: Applicant's FR01 T3 parent-only modelling kinetic fits

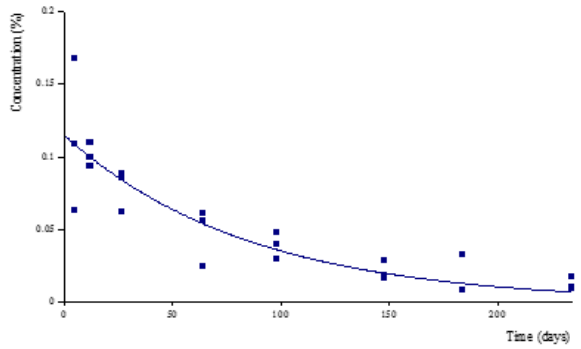
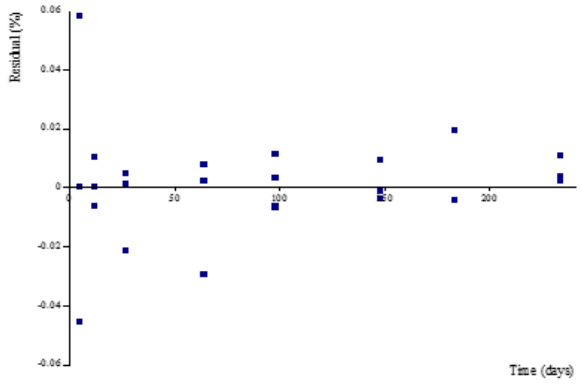
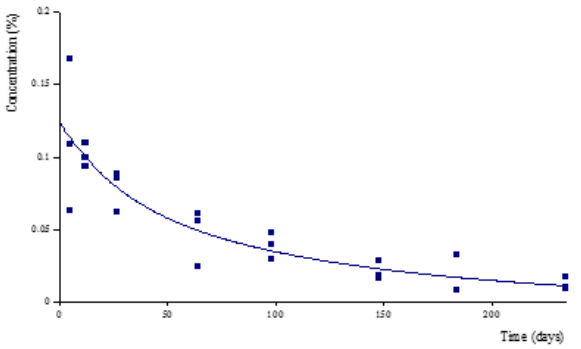
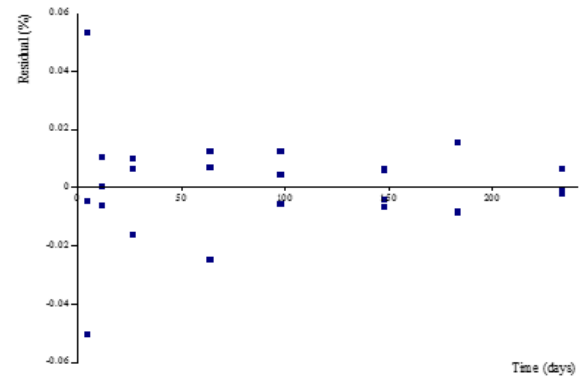
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.3-4: Summary of FR01 T3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	0.2	108
2,4-DBA	SFO	6.4	21.3
Persistence endpoints			
Bixlozone	SFO	59.4	197
Modelling endpoints			
Bixlozone	SFO	59.1	196

CA.B.8.1.2.3.4. FR01 T4

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted SFO provided a good visual and statistical fit of the data, whereas, the biphasic models were statistically unacceptable. Therefore, the applicant concludes SFO as the best-fit model.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs and obtained similar results to the applicant. The CA concurs the SFO model provided the best visual and statistical fit and so accepts the applicant's conclusion this was the best-fit model.

The CA notes residues of 2,4-DBA were detected in the trial at 6 DALA and 28 DALA. However, as this only covers two time points and the residues were <LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA considers a parent-only evaluation appropriate.

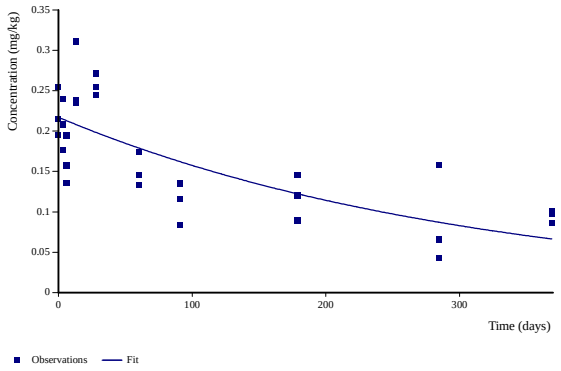
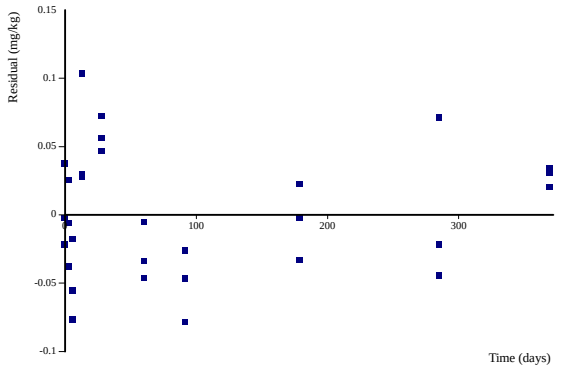
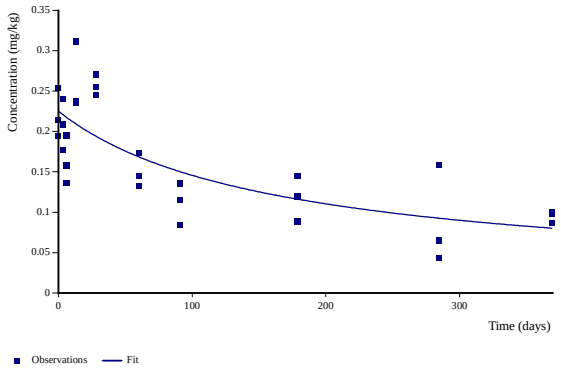
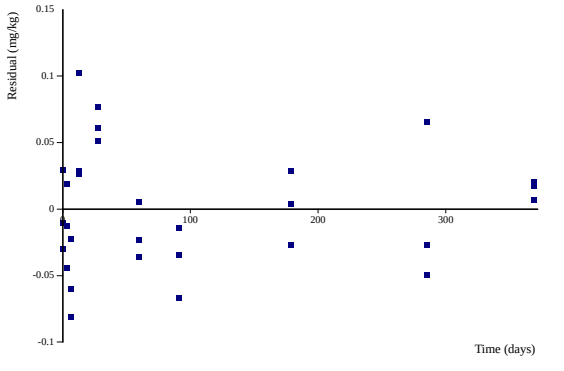
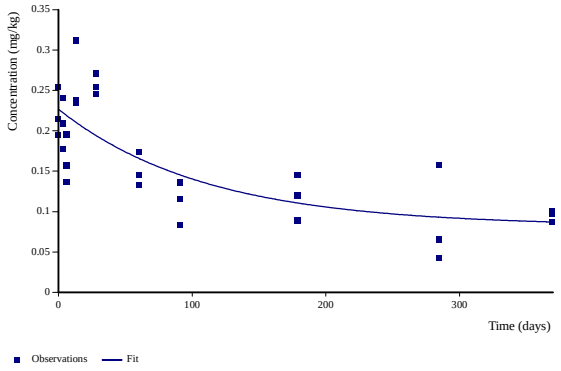
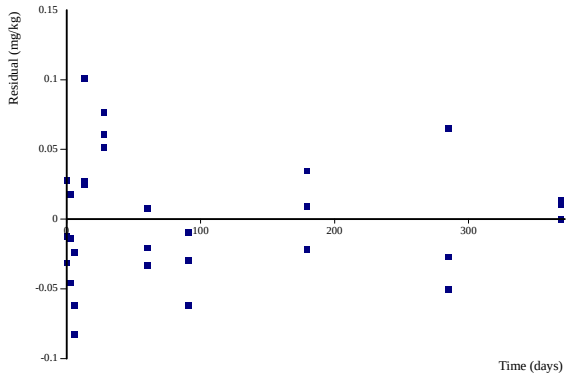
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.4-1** and the graphical outputs in Figure CA.B.**8.1.2.3.4-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.4-3**.

Table CA.B.8.1.2.3.4-1: Summary of FR01 T4 best-fit kinetic modelling

Modelling		**Applicant's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Acceptable**	Acceptable	Acceptable
DT_{50} (days)		**216**	192	169
DT_{90} (days)		**719**	2.46E+03	>10,000
DT_{90}/3.32 (days)		**n/a**	740	3012
DT_{50} (days) - Slow phase		**n/a**	n/a	>10,000
χ^2 error (%)		**17.3**	17.4	18
k ($days^{-1}$)	k or k_1	**0.003204**	n/a	0.009146
	k_2	**n/a**	n/a	2.35E-005
P value	k or k_1	**3.29E-5**	n/a	0.312
	k_2	**n/a**	n/a	0.4989
g		**n/a**	n/a	0.6335
alpha		**n/a**	0.7752	n/a
beta		**n/a**	132.9	n/a
95% CI (lower/upper)	alpha	**n/a**	-1.031 / 2.581	n/a
	beta	**n/a**	-358.2 / 623.9	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.4-1: Applicant's FR01 T4 parent kinetic fits

Fitted Model	**Residuals**
Applicant's kinetic fits	
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.4-2** and the graphical outputs in Figure CA.B.**8.1.2.3.4-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.4-3**.

Table CA.B.8.1.2.3.4-2: Summary of FR01 T4 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**152**	152
DT_{90} (days)		**504**	509
DT_{50} (days) - Slow phase		**n/a**	157
χ^2 error (%)		**15.8**	17.6
k ($days^{-1}$)	k or k_1	**0.004567**	0.006312
	k_2	**n/a**	0.004419
P value	k or k_1	**4.56E-6**	0.4855
	k_2	**n/a**	0.3804
g		**n/a**	0.0846

Selected model shown in bold

Figure CA.B.8.1.2.3.4-2: Applicant's FR01 T4 parent-only modelling kinetic fits

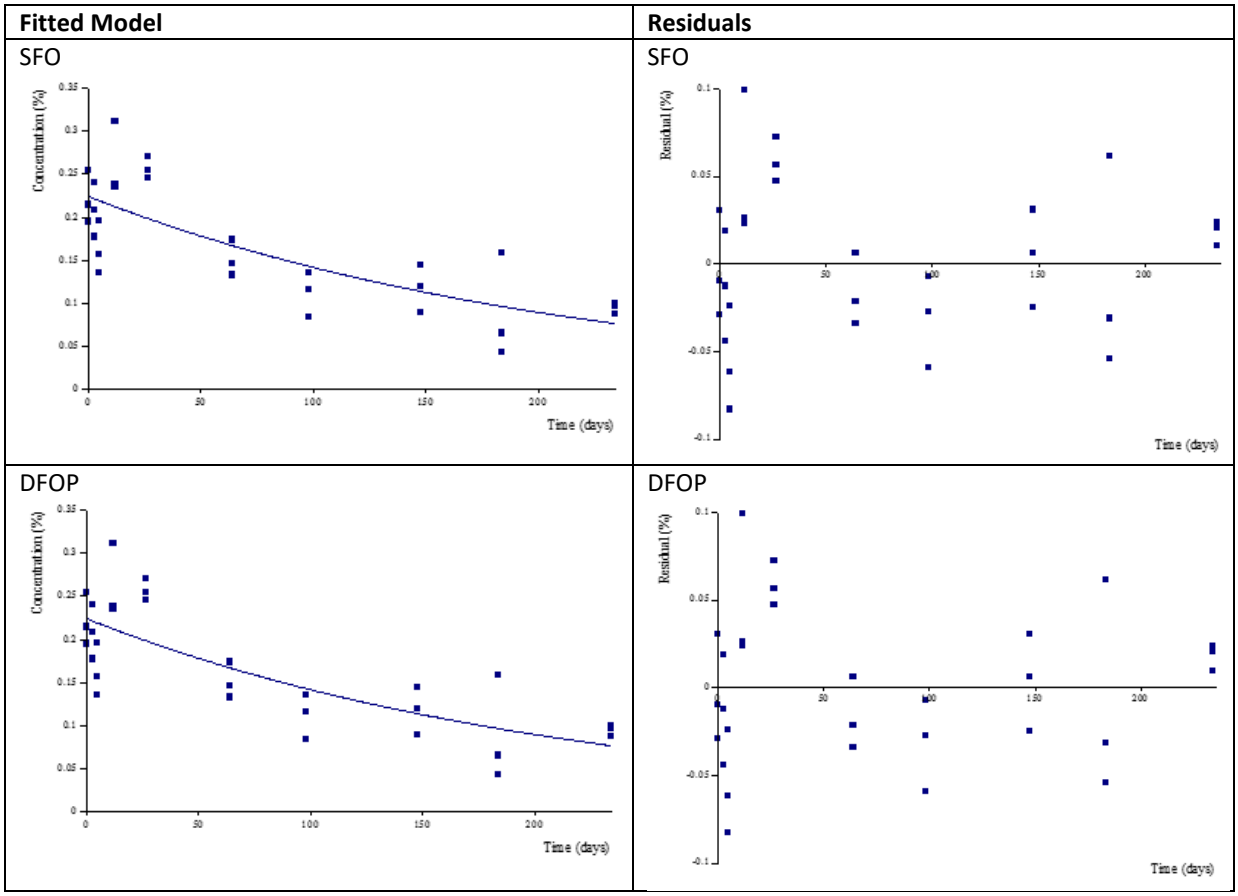


Table CA.B.8.1.2.3.4-3: Summary of FR01 T4 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	216	719
Modelling endpoints			
Bixlozone	SFO	152	504

CA.B.8.1.2.3.5. IT01 T1

Best-fit endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted there was very little difference between the three visual fits, however, the FOMC and DFOP fits were statistically unacceptable. Therefore, the applicant concluded upon SFO as the best-fit model for parent-only.

The applicant then re-ran the SFO parent fit with the metabolite data modelled in sequence (both SFO and DFOP models were ran for the metabolite). The applicant concludes the SFO – SFO fit was visually and statistically acceptable, therefore, these endpoints were selected as the best-fit for this test system.

To validate the applicant's modelling, the CA ran parent-only SFO and DFOP models and parent-plus-metabolite SFO – SFO models. The CA concurs with the applicant that the parent SFO fit best describes the data. The CA notes that in both the applicant's parent-plus-metabolite modelling and the CA's, the metabolite SFO fit resulted in a t-test value >0.1 and χ^2 values >20%. However, the CA considers the passing of the t-test to be less important in best-fit kinetics. Also, the CA considers the high χ^2 values to be as a result of the variable nature of the field study dataset, and not due to the kinetic model selected. Although peak metabolite formation is not reached by the curve, because the metabolite residues are plotted on either side of the residual curve (demonstrating reasonable fit relative to the high variance of the residues), the fit is nevertheless accepted. Therefore, the CA agrees with the applicant that the SFO – SFO fit is acceptable for deriving best-fit kinetics.

The CA obtained a slightly longer parent DT50 value to the applicant, and a slightly lower metabolite DT50; this is believed to be due to the slightly different data handling measures undertaken by the CA, meaning slightly different summed-residues were obtained. The CA considers its evaluation to be more appropriate and so the results of the CA's SFO – SFO fit is considered further.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.5-1** and the graphical outputs in Figure CA.B.**8.1.2.3.5-1** and Figure CA.B.**8.1.2.3.5-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.5-4**.

Table CA.B.8.1.2.3.5-1: Summary of IT01 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**							**CA's modelling**			
Pathway		**Bixlozone only**			**Bixlozone → 2,4-DBA**				**Bixlozone only**		**Bixlozone → 2,4-DBA**	
Compound		**bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**
Model		SFO	FOMC	DFOP	SFO	SFO	SFO	DFOP	SFO	DFOP	**SFO**	**SFO**
Visual fit		Very good	Very good	Very good	Good	Acceptable	Good	Acceptable	Good	Good	**Good**	**Acceptable**
DT_{50} (days)		28.3	18.1	17.5	19.6	6.91	20.1	6.72	28.6	17.6	**28.5**	**4.98**
DT_{90} (days)		94.1	186	173	65.2	23	66.8	22.3	95.0	177	**94.6**	**16.5**
DT_{90}/3.32 (days)		n/a	55.9	52.1	n/a	n/a	n/a	6.72	n/a	53.4	**n/a**	**n/a**
DT_{50} (days) - Slow phase		n/a	n/a	80.4	n/a	n/a	n/a	6.76	n/a	83.1	**n/a**	**n/a**
χ^2 error (%)		13.9	6.31	6.74	16.4	24.4	16.1	34.9	13.9	6.75	**13.9**	**37.0**
k ($days^{-1}$)	k or k_1	0.02447	n/a	0.0888	0.03533	0.1003	0.03447	0.1031	0.0242	0.0872	**0.0244**	**0.1392**
	k_2	n/a	n/a	0.008618	n/a	n/a	n/a	0.1026	n/a	0.0083	**n/a**	**n/a**
P value	k or k_1	0.006208	n/a	0.2249	0.003867	0.01848	0.00785	0.4972	0.0062	0.2209	**0.0002**	**0.3886**
	k_2	n/a	n/a	0.221	n/a	n/a	n/a	0.5	n/a	0.2212	**n/a**	**n/a**
g		n/a	n/a	0.556	n/a	n/a	n/a	0.9985	n/a	0.5616	**n/a**	**n/a**
alpha		n/a	0.9011	n/a	n/a	n/a	n/a	n/a	n/a	n/a	**n/a**	**n/a**
beta		n/a	15.64	n/a	n/a	n/a	n/a	n/a	n/a	n/a	**n/a**	**n/a**
95% CI (lower/upper)	alpha	n/a	-0.6388 / 2.441	n/a	n/a	n/a	n/a	n/a	n/a	n/a	**n/a**	**n/a**
	beta	n/a	-34.96 / 66.25	n/a	n/a	n/a	n/a	n/a	n/a	n/a	**n/a**	**n/a**

Best-fit model shown in bold

Figure CA.B.8.1.2.3.5-1: Applicant's IT01 T1 parent-only kinetic fits

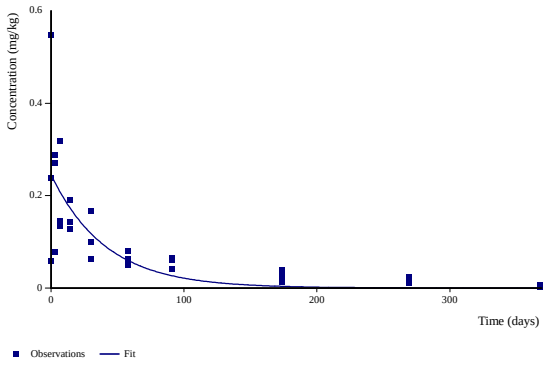
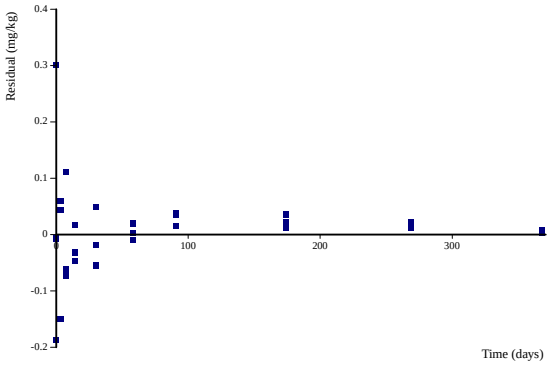
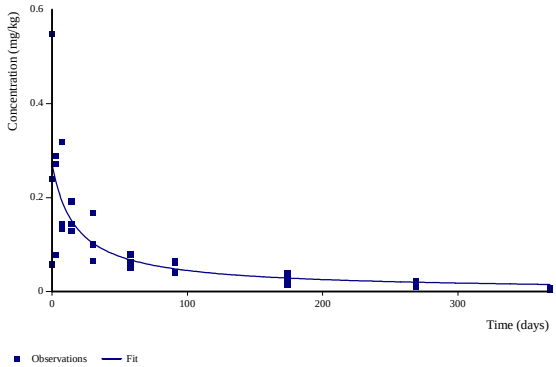
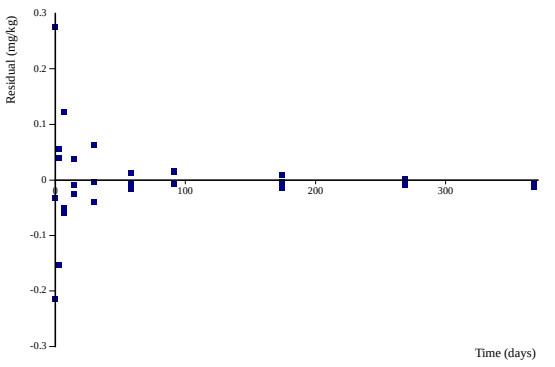
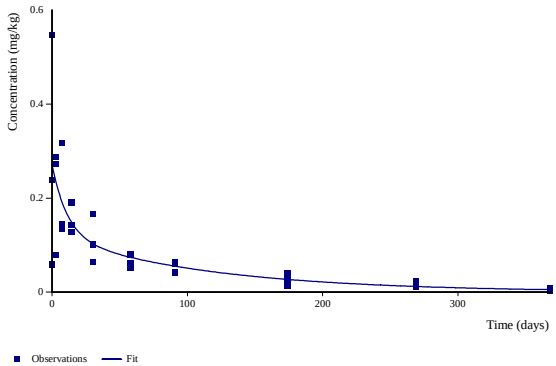
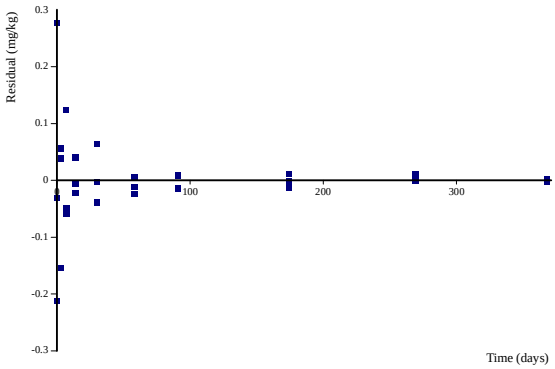
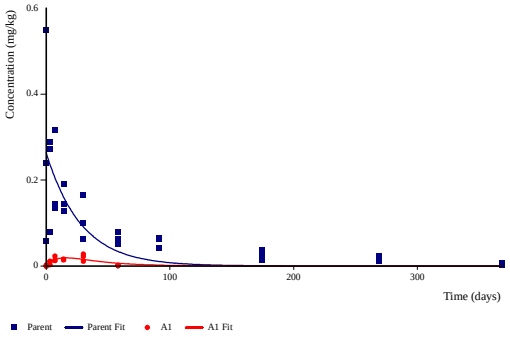
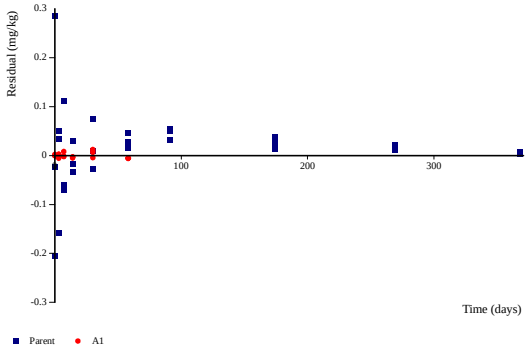
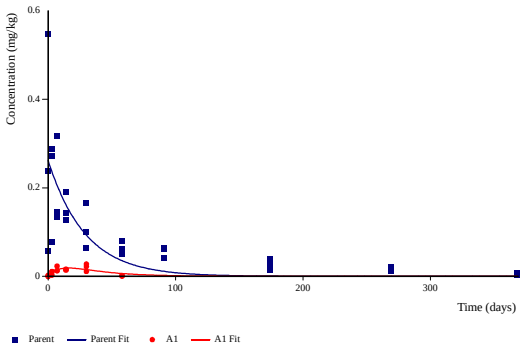
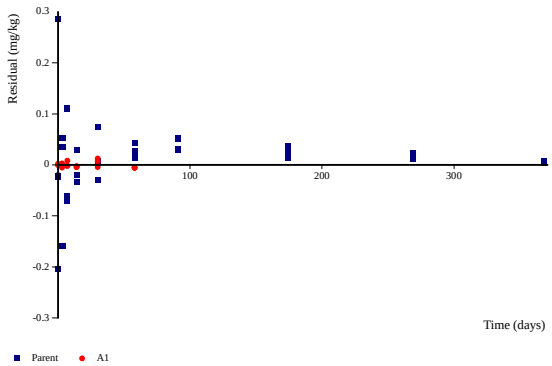
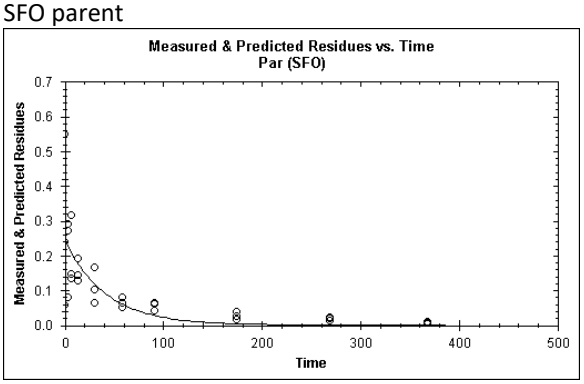
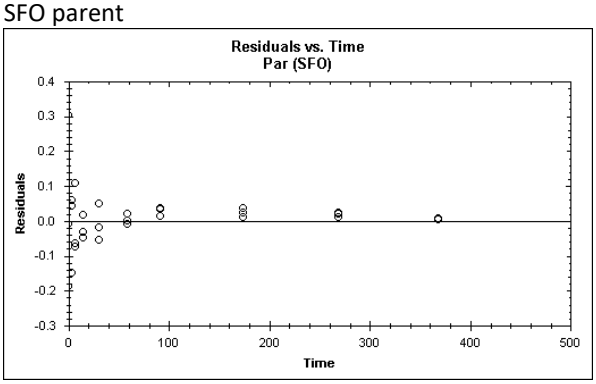
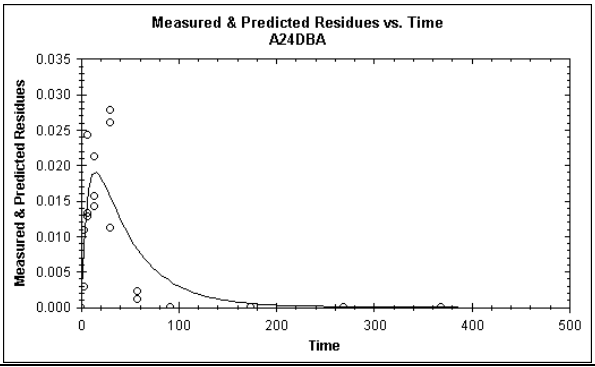
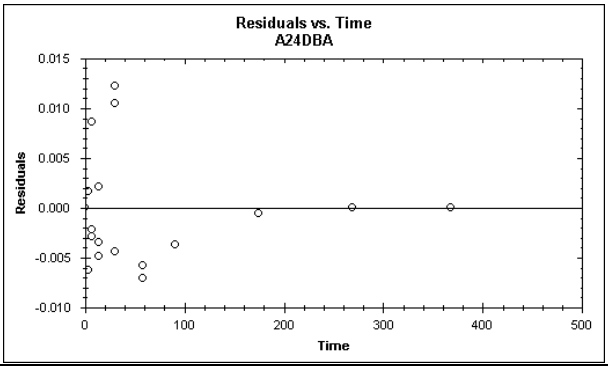
Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Figure CA.B.8.1.2.3.5-2: Applicant's and CA's IT01 T1 parent and metabolite kinetic fits

Fitted Model	Residuals
Applicant's kinetic fits	
SFO-SFO 	SFO-SFO 
SFO-DFOP 	SFO-DFOP 
CA's SFO – SFO kinetic fits	
SFO parent 	SFO parent 
SFO metabolite 	SFO metabolite 

Persistence endpoints

As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a very good visual and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.5-2** and Figure CA.B.**8.1.2.3.5-3**.

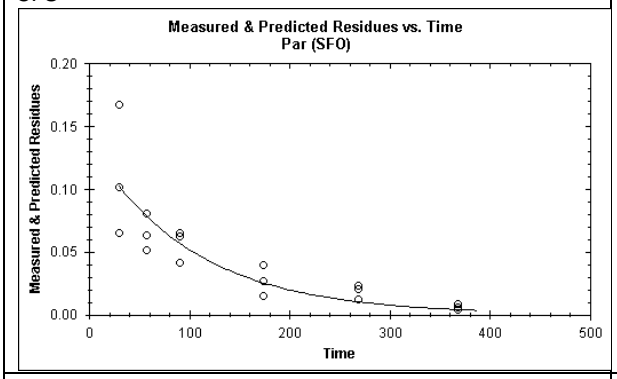
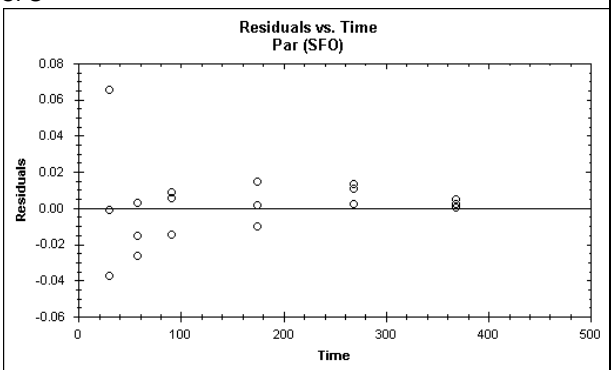
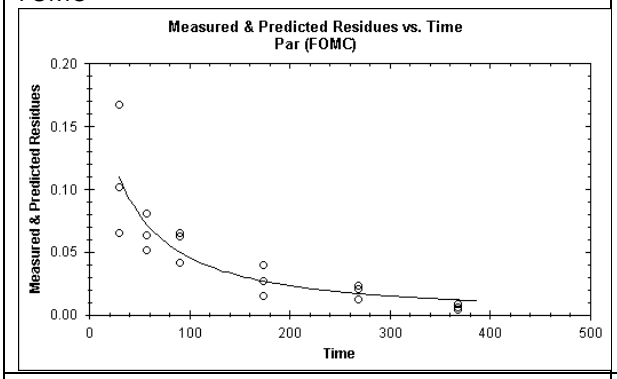
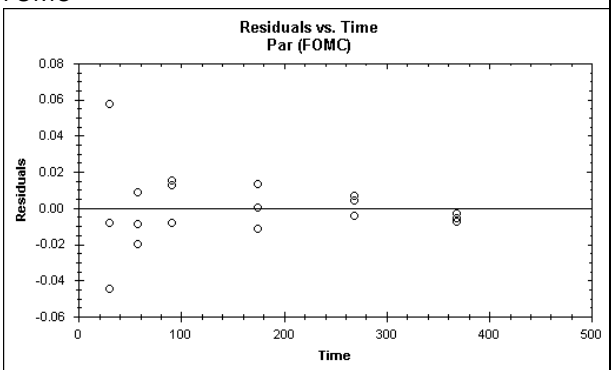
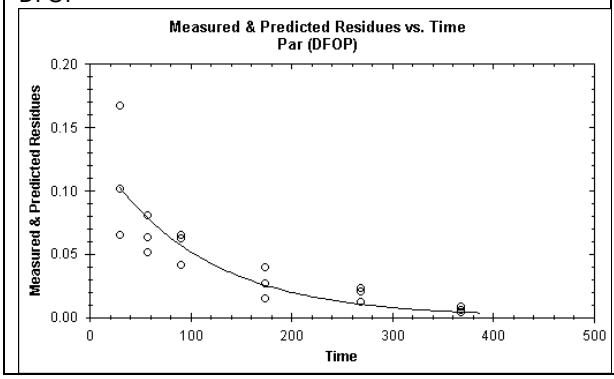
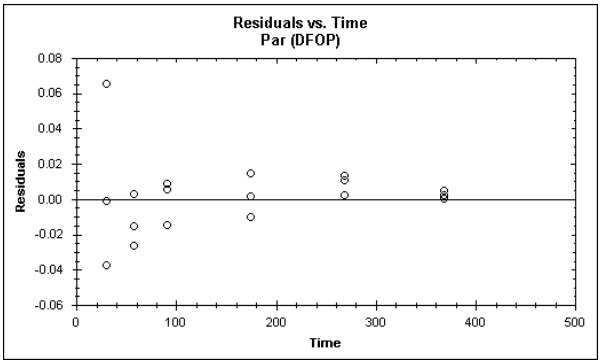
The CA has not considered 2,4-DBA in the persistence kinetic evaluation as formation occurred prior to 10 mm rainfall.

Table CA.B.8.1.2.3.5-2: Summary of IT01 T1 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Very good**	Very good	Very good
DT_{50} (days)		**70.9**	29.1	70.9
DT_{90} (days)		**236**	204	235
DT_{90}/3.32 (days)		**n/a**	61.3	70.9
DT_{50} (days) - Slow phase		**n/a**	n/a	71.4
χ^2 error (%)		**12.5**	8.51	15.8
k ($days^{-1}$)	k or k_1	**0.00978**	n/a	9.780E-3
	k_2	**n/a**	n/a	9.707E-3
P value	k or k_1	**5.99E-4**	n/a	1.25E-3
	k_2	**n/a**	n/a	<2E-16
g		**n/a**	n/a	1.00
alpha		**n/a**	1.2826	n/a
beta		**n/a**	40.544	n/a
95% CI (lower/upper)	alpha	**n/a**	-0.9605 / 3.526	n/a
	beta	**n/a**	-152.76 / 233.85	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.5-3: CA's IT01 T1 parent-only persistence kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). As >5% 2,4-DBA was formed prior to 10 mm rainfall, it has not been considered in the kinetic assessment. The applicant concluded the SFO fit provided a good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar SFO results to the applicant. The CA obtained substantially different DFOP results to the applicant, believed to be due to differing ways the models have handled the lack of data prior to 43.1 normalised days. As the applicant notes, in their CAKE assessment the fast phase occurs outside the dataset (see Figure CA.B.**8.1.2.3.5-4**). Whereas in the CA's Kingui assessment, k_1 fits the dataset and the g value is 1. However, as the SFO fit of the data is good enough, this is accepted as the appropriate modelling endpoint fit and so the differing DFOP fits are not considered further. As the CA obtained similar SFO results to the applicant, the applicant's results are accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.5-3** and the graphical outputs in Figure CA.B.**8.1.2.3.5-4**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.5-4**.

Table CA.B.8.1.2.3.5-3: Summary of IT01 T1 modelling endpoint kinetics

Modelling		**Applicant's modelling**		**CA's modelling**	
Pathway		**Bixlozone only**		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP	SFO	DFOP
Visual fit		**Good**	Good	Good	Good
DT_{50} (days)		**57.8**	4.11	58.4	58.4
DT_{90} (days)		**192**	52	194	194
DT_{50} (days) - Slow phase		**n/a**	57.8	n/a	69.2
χ^2 error (%)		**9.72**	12.3	9.19	11.6
k ($days^{-1}$)	k or k_1	**0.01199**	0.2252	0.0118	0.0118
	k_2	**n/a**	0.01199	n/a	0.0100
P value	k or k_1	**3.48E-5**	0.4535	3.21E-5	9.51E-5
	k_2	**n/a**	0.6.33E-5	n/a	<2E-16
g		**n/a**	0.7744	n/a	1.0000

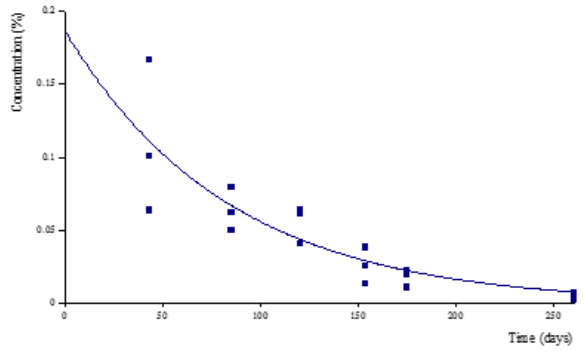
Selected model shown in bold

Figure CA.B.8.1.2.3.5-4: Applicant's and CA's IT01 T1parent-only modelling kinetic fits

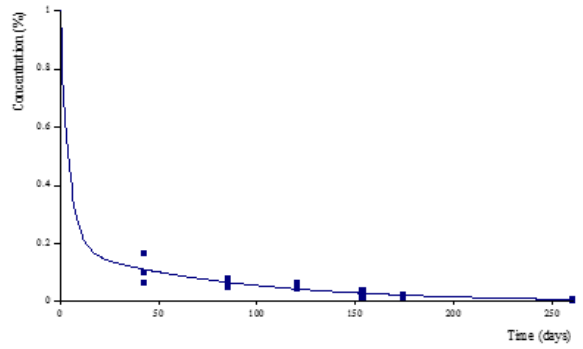
Applicant's modelling

Fitted Model

SFO

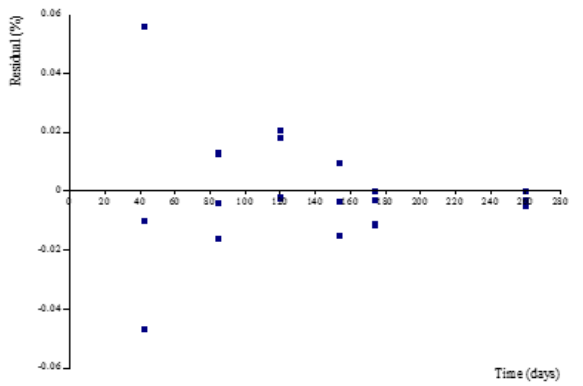


DFOP

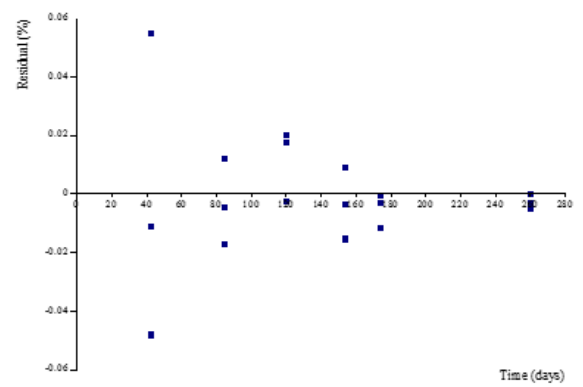


Residuals

SFO

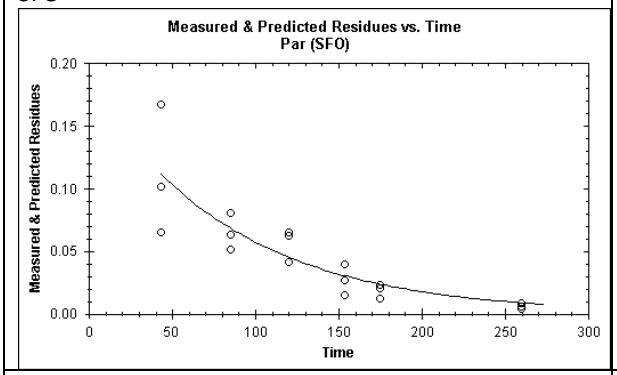


DFOP

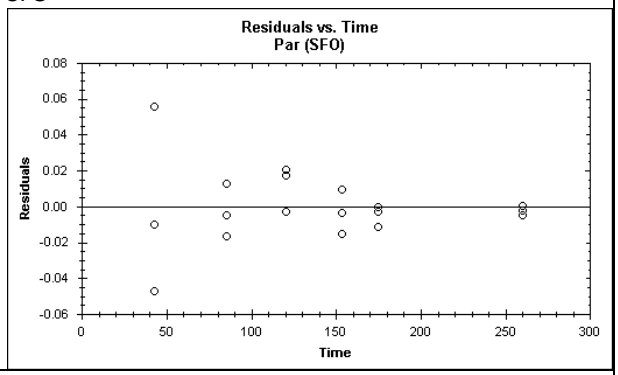


CA's modelling

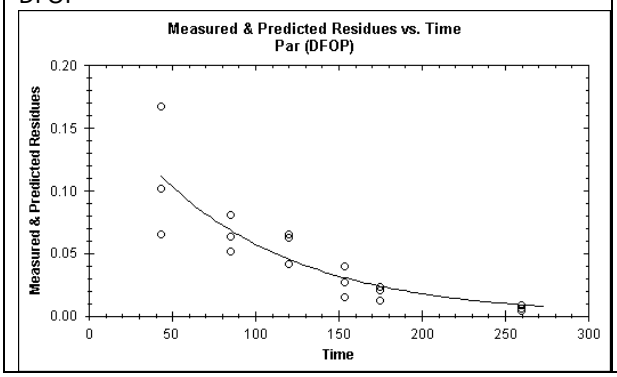
SFO



SFO



DFOP



DFOP

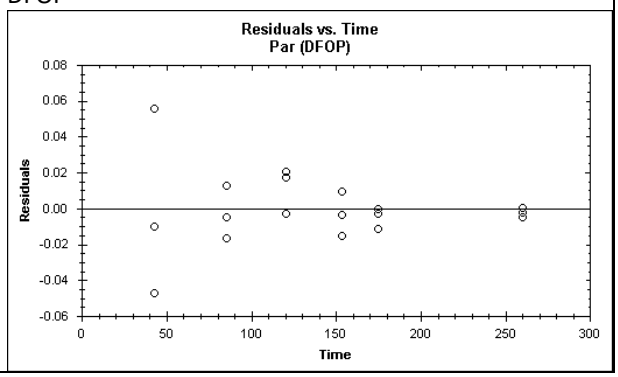


Table CA.B.8.1.2.3.5-4: Summary of IT01 T1 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	28.5	94.6
2,4-DBA	SFO	4.98	16.5
Persistence endpoints			
Bixlozone	SFO	70.9	236
Modelling endpoints			
Bixlozone	SFO	57.8	192

CA.B.8.1.2.3.6. IT01 T2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted SFO provided a good visual and statistical fit of the data which was not improved by the biphasic fits. Therefore, the applicant concludes SFO as the best-fit model.

The CA has repeated the applicant's SFO and DFOP model runs. The CA obtained similar results to the applicant and concurs the SFO model provided the best visual and statistical fit. The applicant's SFO fit is therefore accepted.

The CA notes 2,4-DBA was also detected in this field trial at three sampling events which the applicant has not assessed. The CA therefore proceeded to run a SFO – SFO fit of the data; however, the 2,4-DBA fit was unacceptable. The CA considers this as a result of there being insufficient data in the decline phase. As such, the CA does not consider it achievable to obtain a robust kinetic fit of the 2,4-DBA data.

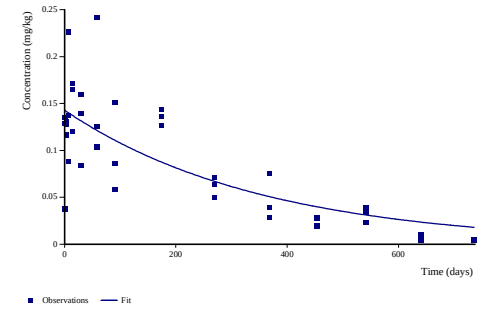
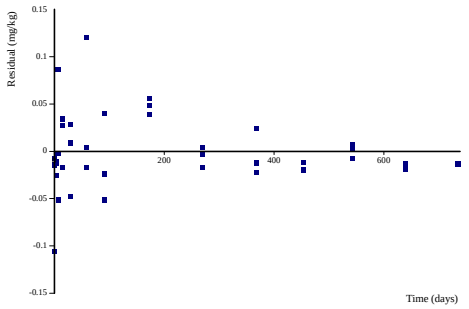
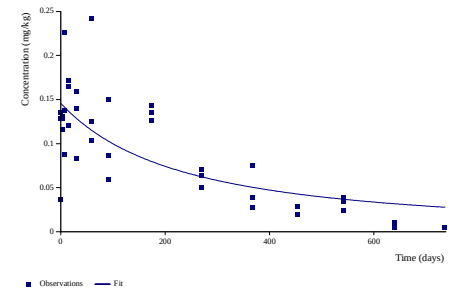
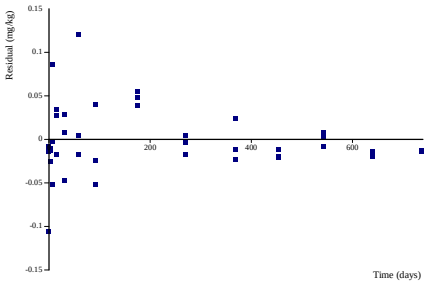
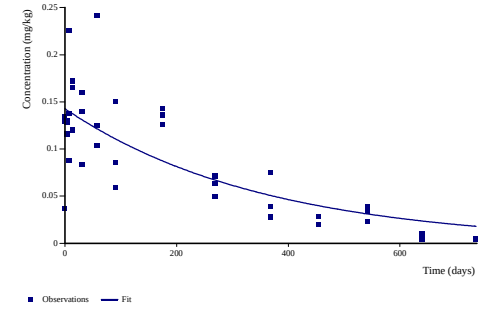
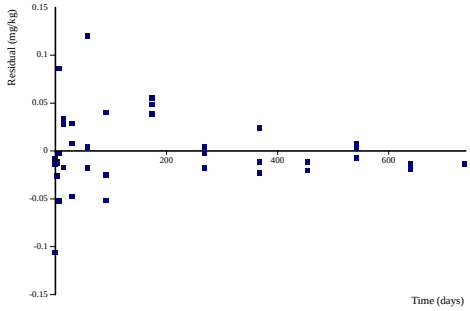
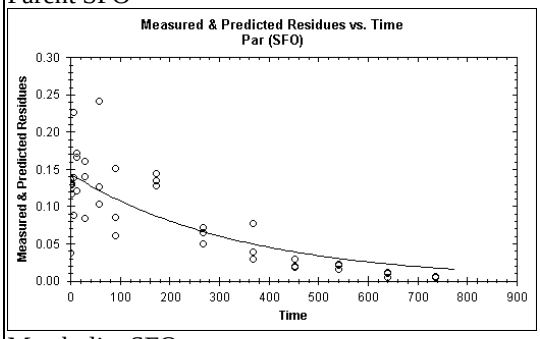
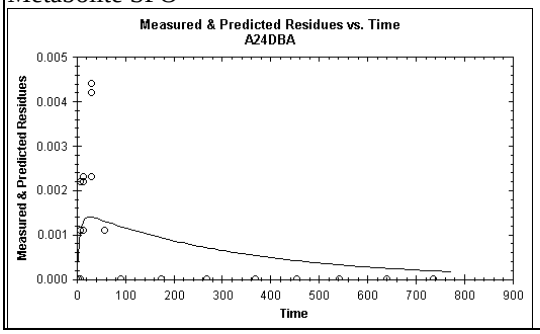
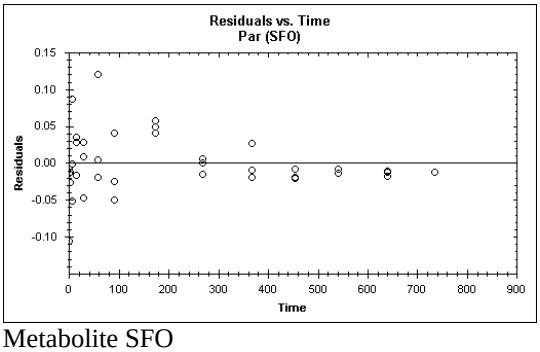
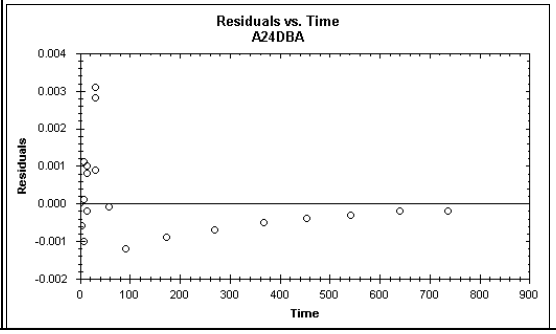
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.6-1** and the graphical outputs in Figure CA.B.**8.1.2.3.6-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.6-3**.

Table CA.B.8.1.2.3.6-1: Summary of IT01 T2 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**		
Pathway		**Bixlozone only**			**Bixlozone only**	**Bixlozone → 2,4-DBA**	
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**
Model		**SFO**	FOMC	DFOP	SFO	SFO	SFO
Visual fit		**Acceptable**	Acceptable	Acceptable	Acceptable	Acceptable	Poor
DT_{50} (days)		**247**	207	247	238	238	3.57
DT_{90} (days)		**819**	1.37E+3	819	791	791	11.9
DT_{90}/3.32 (days)		**n/a**	417	247	n/a	n/a	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	247	n/a	n/a	n/a
χ^2 error (%)		**20.7**	21.4	22.2	21.1	21.1	106
k ($days^{-1}$)	k or k_1	**0.00281**	n/a	0.002873	0.002910	0.002910	0.1940
	k_2	**n/a**	n/a	0.002806	n/a	n/a	n/a
P value	k or k_1	**4.25E-6**	n/a	0.4985	4.43E-6	1.48E-10	0.483
	k_2	**n/a**	n/a	0.4781	n/a	n/a	n/a
g		**n/a**	n/a	0.06641	n/a	n/a	n/a
alpha		**n/a**	1.378	n/a	n/a	n/a	n/a
beta		**n/a**	317	n/a	n/a	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	-83.28 / 86.04	n/a	n/a	n/a	n/a
	beta	**n/a**	239.9 / 394	n/a	n/a	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.6-1: IT01 T2 parent (and CA's metabolite) kinetic fits

Fitted Model	**Residuals**
Applicant's kinetic fits	
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 
CA's SFO – SFO kinetic fits	
Parent SFO  Metabolite SFO 	Parent SFO  Metabolite SFO 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.6-2** and the graphical outputs in Figure CA.B.**8.1.2.3.6-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.6-3**.

Table CA.B.8.1.2.3.6-2: Summary of IT01 T2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**187**	187
DT_{90} (days)		**619**	620
DT_{50} (days) - Slow phase		**n/a**	188
χ^2 error (%)		**24.8**	26.5
k ($days^{-1}$)	k or k_1	**0.003717**	0.003717
	k_2	**n/a**	0.003694
P value	k or k_1	**4.89E-6**	0.4945
	k_2	**n/a**	0.4999
g		**n/a**	0.9821

Selected model shown in bold

Figure CA.B.8.1.2.3.6-2: Applicant's IT01 T2 parent-only modelling kinetic fits

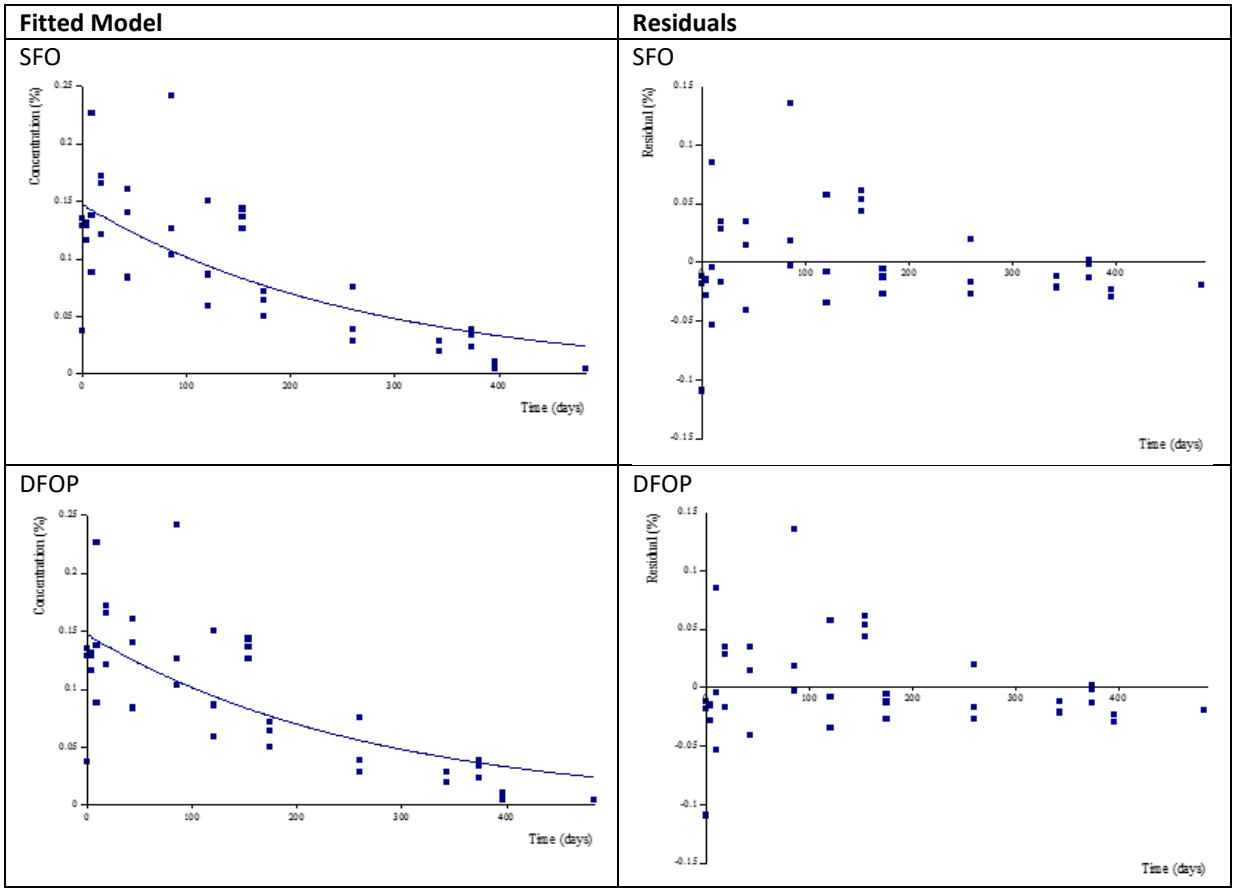


Table CA.B.8.1.2.3.6-3: Summary of IT01 T2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	247	819
Modelling endpoints			
Bixlozone	SFO	187	619

CA.B.8.1.2.3.7. IT01 T3

Best-fit endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted DFOP provided the best visual and statistical fit of the models and so re-ran the DFOP parent fit with the metabolite data modelled in sequence (both SFO and DFOP models were ran for the metabolite). The applicant concludes the DFOP – SFO fit was visually and statistically acceptable, therefore, these endpoints were selected as the best-fit for this test system.

To validate the applicant's modelling, the CA ran parent-only SFO and DFOP models and parent-plus-metabolite DFOP – SFO fits of the data. The CA concurs with the applicant that the parent DFOP fit best describes the data.

The CA notes its DFOP – SFO fit of the metabolite data was poor; the visual fit was poor, exhibited by a χ^2 value of 40.5% and t-test value of 0.18. However, the applicant's DFOP – SFO fit was much better. The difference in results is believed to be due to the different data handling techniques used and/or the applicant selecting IRLS as the optimisation method. Because the applicant's modelling resulted in an acceptable DFOP – SFO fit, the CA considers their results acceptable.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.7-1** and the graphical outputs in Figure CA.B.**8.1.2.3.7-1** and Figure CA.B.**8.1.2.3.7-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.7-4**.

Table CA.B.8.1.2.3.7-1: Summary of IT01 T3 best-fit kinetic modelling

Modelling		**Applicant's modelling**							**CA's modelling**		
Pathway		**Bixlozone only**			**Bixlozone → 2,4-DBA**				**Bixlozone only**	**Bixlozone → 2,4-DBA**	
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**2,4-DBA**	**Bixlozone**	**Bixlozone**	**2,4-DBA**
Model		SFO	FOMC	DFOP	**DFOP**	**SFO**	DFOP	DFOP	SFO	DFOP	SFO
Visual fit		Unacceptable	Very good	Very good	**Very good**	**Acceptable**	**Very good**	Acceptable	Unacceptable	Very good	Unacceptable
DT_{50} (days)		10.6	4.18	3.57	**7.36**	**15.7**	7.36	15.7	10.8	3.72	22.3
DT_{90} (days)		35.2	206	140	**219**	**52.1**	219	52.1	36.0	144	74.2
DT_{90}/3.32 (days)		n/a	62	42.2	**66.0**	**n/a**	66.0	15.7	n/a	43.4	n/a
DT_{50} (days) - Slow phase		n/a	n/a	73	**178**	**n/a**	179	15.7	n/a	76.4	n/a
χ^2 error (%)		32.3	11.0	12.7	**19.7**	**13.7**	19.7	19.6	32.3	12.6	40.5
k ($days^{-1}$)	k or k_1	0.06535	n/a	0.4262	**0.1404**	**0.04416**	0.1404	0.04417	0.0640	0.3989	0.0310
	k_2	n/a	n/a	0.009498	**0.003885**	**n/a**	0.003883	0.04416	n/a	0.0091	n/a
P value	k or k_1	4.74E-4	n/a	0.02134	**0.001113**	**0.001659**	0.002238	0.04539	0.0005	0.0012	0.1806
	k_2	n/a	n/a	0.0204	**0.1274**	**n/a**	0.08387	0.01965	n/a	0.0019	n/a
g		n/a	n/a	0.6238	**0.7661**	**n/a**	0.7661	0.4246	n/a	0.6305	n/a
alpha		n/a	0.4382	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
beta		n/a	1.081	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
95% CI (lower/upper)	alpha	n/a	0.2073 / 0.669	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a
	beta	n/a	-0.6507 / 2.812	n/a	**n/a**	**n/a**	n/a	n/a	n/a	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.7-1: Applicant's IT01 T3 parent-only kinetic fits

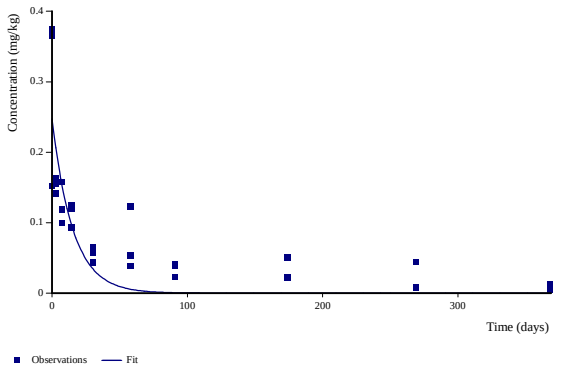
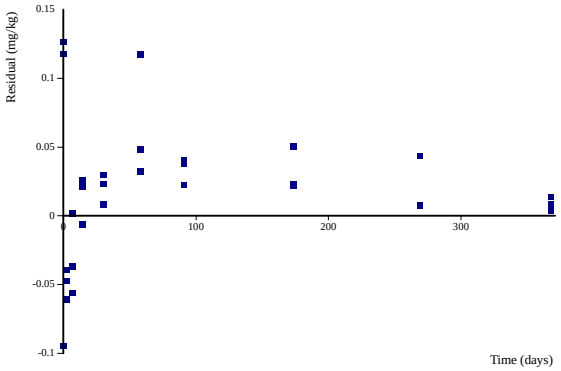
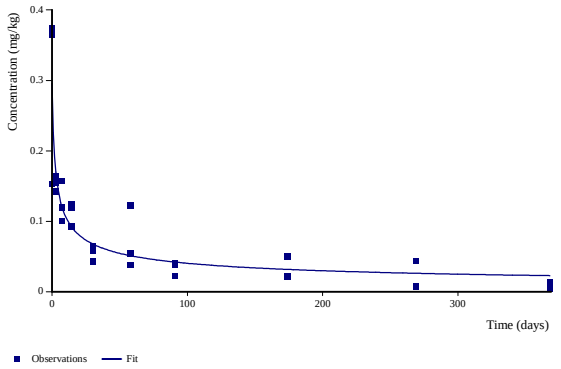
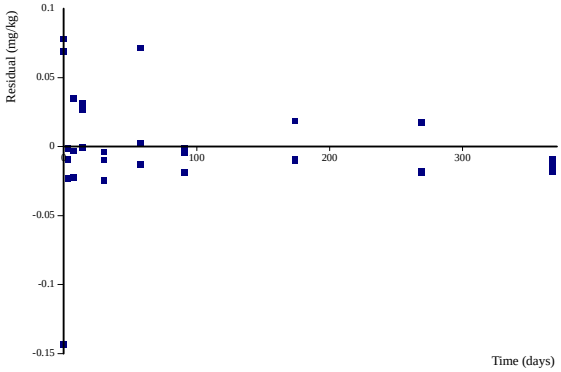
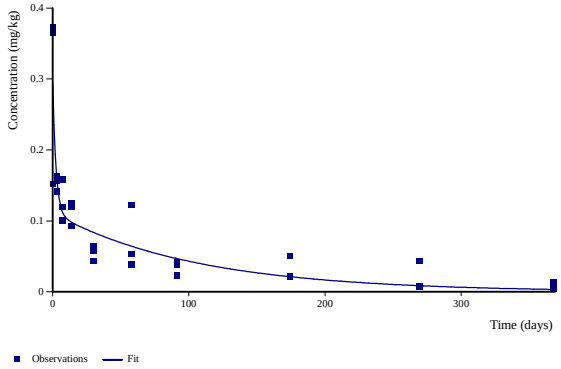
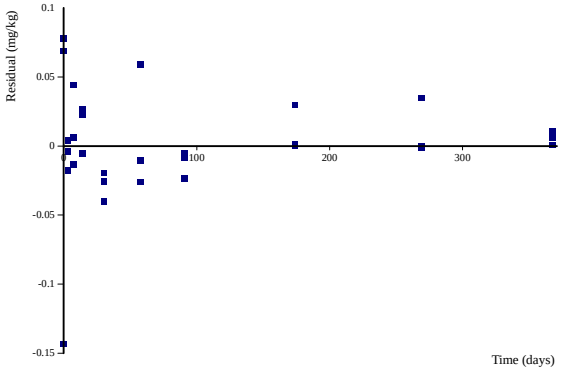
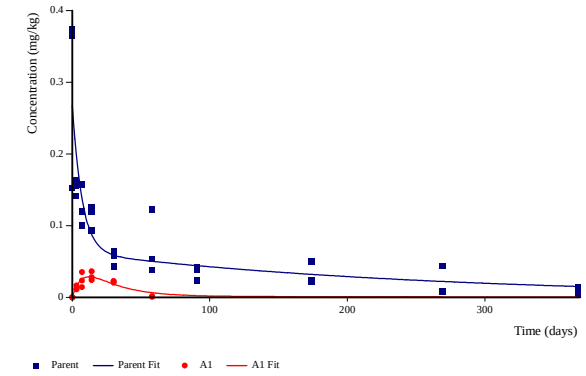
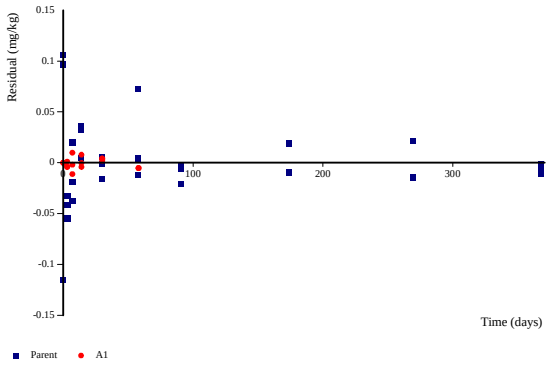
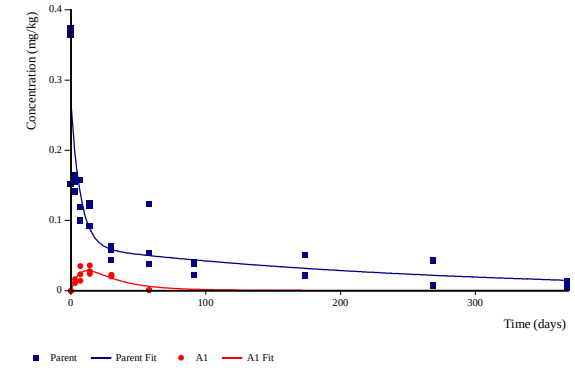
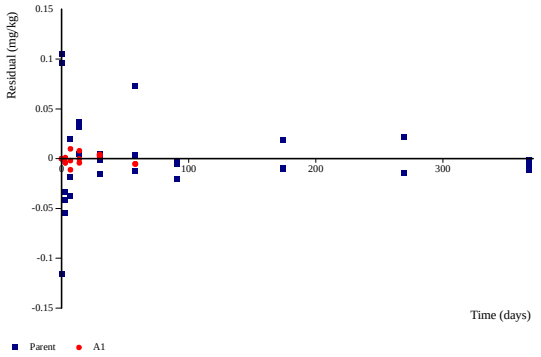
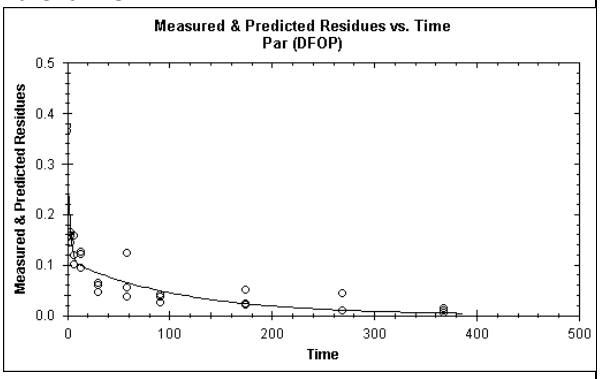
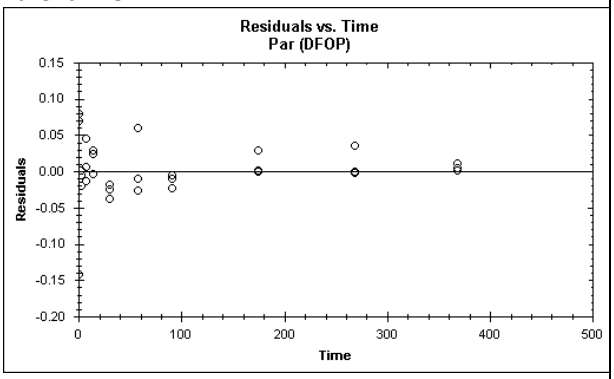
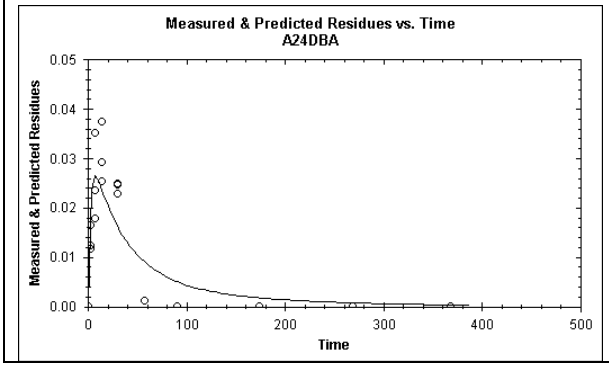
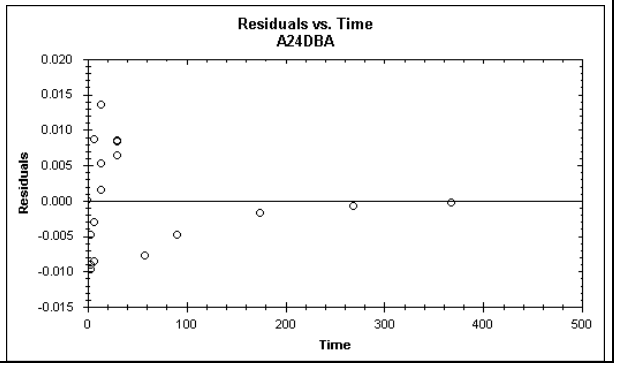
Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Figure CA.B.8.1.2.3.7-2: Applicant's and CA's IT01 T3 parent and metabolite kinetic fits

Fitted Model	**Residuals**
Applicant's kinetic fits	
DFOP-SFO	DFOP-SFO
	
DFOP-DFOP	DFOP-DFOP
	
CA's DFOP – SFO kinetic fits	
Parent DFOP	Parent DFOP
	
Metabolite SFO	Metabolite SFO
	

Persistence endpoints

As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a very good visual fit and an acceptable statistical fit of the data; the χ^2 value was >15%, however, the CA considers this unavoidable due to the immediate sampling occasions being omitted from the evaluation. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.7-2** and Figure CA.B.**8.1.2.3.7-3**.

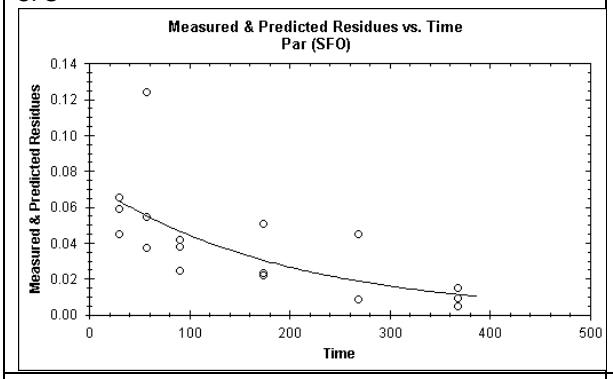
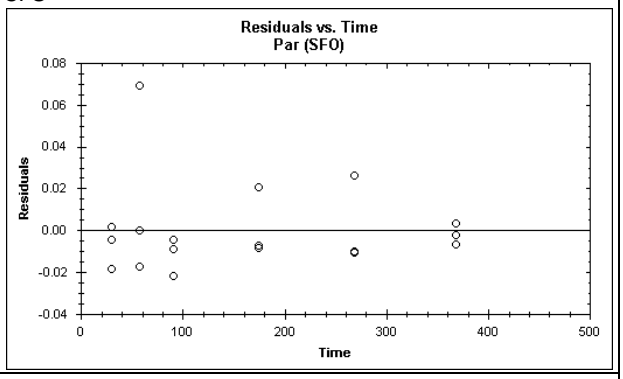
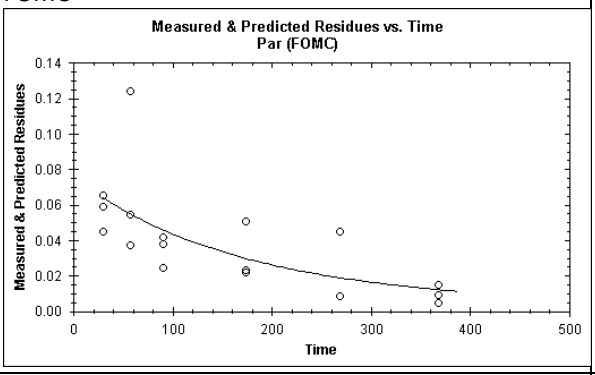
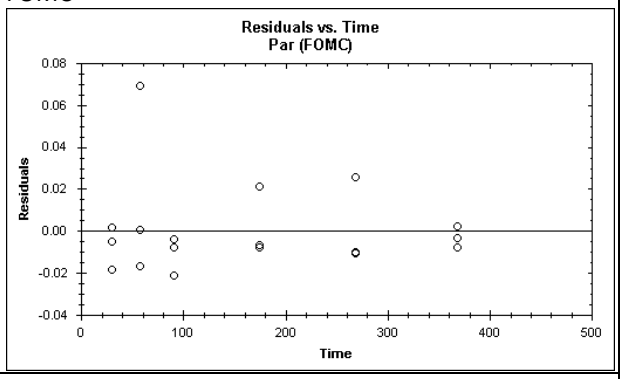
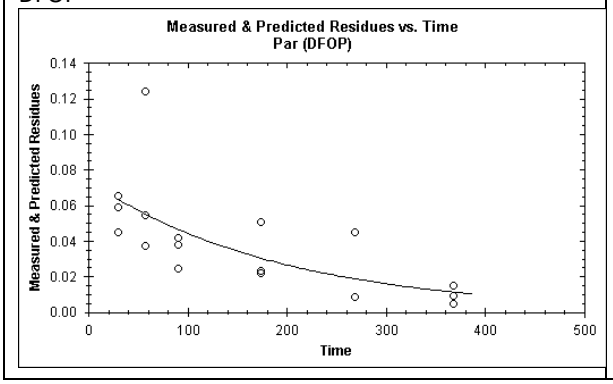
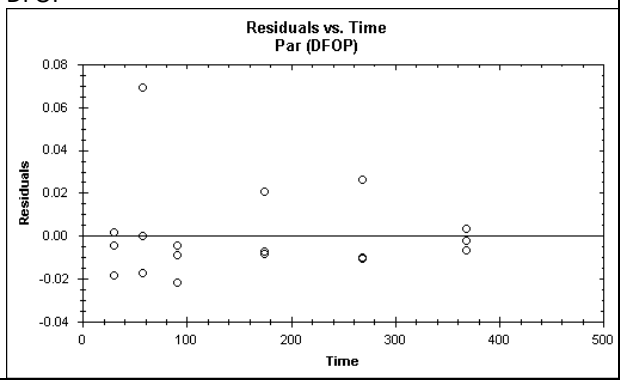
The CA has not considered 2,4-DBA in the persistence kinetic evaluation as formation occurred prior to 10 mm rainfall.

Table CA.B.8.1.2.3.7-2: Summary of IT01 T3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Very good**	Very good	Very good
DT_{50} (days)		**135**	126	43.2
DT_{90} (days)		**447**	477	355
DT_{90}/3.32 (days)		**n/a**	144	107
DT_{50} (days) - Slow phase		**n/a**	n/a	134
χ^2 error (%)		**19.3**	21.4	24.33
k ($days^{-1}$)	k or k_1	**0.00516**	n/a	7.153E-1
	k_2	**n/a**	n/a	5.155E-3
P value	k or k_1	**0.00847**	n/a	<2E-16
	k_2	**n/a**	n/a	0.0129
g		**n/a**	n/a	0.3753
alpha		**n/a**	6.272	n/a
beta		**n/a**	1.076E+3	n/a
95% CI (lower/upper)	alpha	**n/a**	-1.127E+2 / 125.272	n/a
	beta	**n/a**	-2.195E+4 / 24103	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.7-3: CA's IT01 T3 parent-only persistence kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). As >5% 2,4-DBA was formed prior to 10 mm rainfall, it has not been considered in the kinetic assessment. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.7-3** and the graphical outputs in Figure CA.B.**8.1.2.3.7-4**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.7-4**.

Table CA.B.8.1.2.3.7-3: Summary of IT01 T3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**98**	98
DT_{90} (days)		**326**	>10000
DT_{50} (days) - Slow phase		**n/a**	326
χ^2 error (%)		**23.5**	29.6
k ($days^{-1}$)	k or k_1	**0.007074**	0.007075
	k_2	**n/a**	3.00E-95
P value	k or k_1	**6.93E-3**	0.4688
	k_2	**n/a**	0.5000
g		**n/a**	0.9999

Selected model shown in bold

Figure CA.B.8.1.2.3.7-4: Applicant's IT01 T3 parent-only modelling kinetic fits

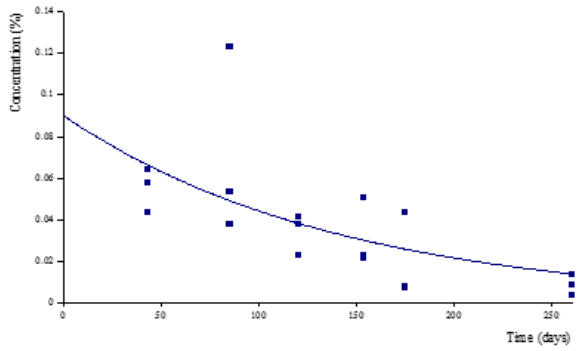
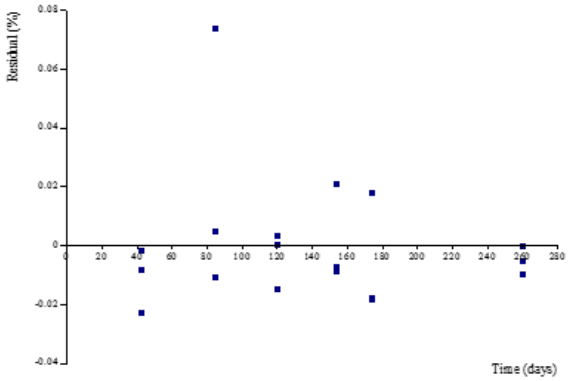
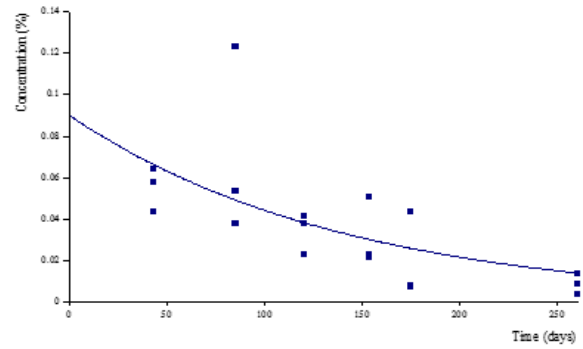
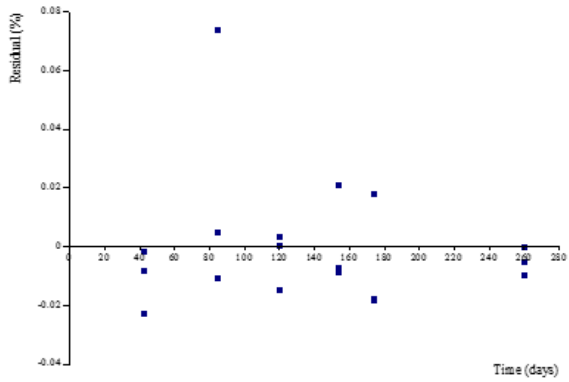
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.7-4: Summary of IT01 T3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	7.36	219
2,4-DBA	SFO	15.7	52.1
Persistence endpoints			
Bixlozone	SFO	135	447
Modelling endpoints			
Bixlozone	SFO	98	326

CA.B.8.1.2.3.8. IT01 T4

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted SFO provided a good visual and statistical fit of the data, whereas, the biphasic models were statistically unacceptable. Therefore, the applicant concludes SFO as the best-fit model.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA concurs the SFO model provided the best visual and statistical fit and is able to replicate the applicant's DT50 value. Therefore, the applicant's SFO modelling is accepted on this occasion.

The CA notes residues of 2,4-DBA were detected in sub-plot A of the trial. However, as the first residue (3 DALA) was 0.0015 mg/kg and the last residue (30 DALA) was 0.0021 mg/kg, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

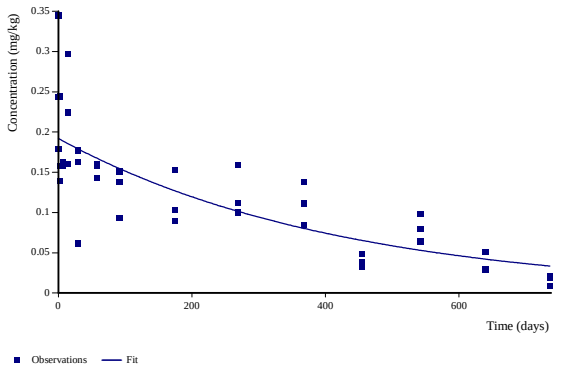
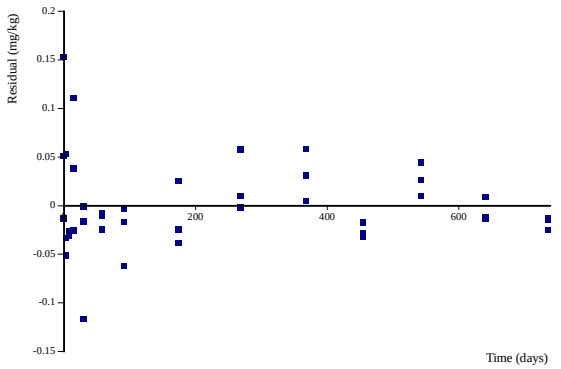
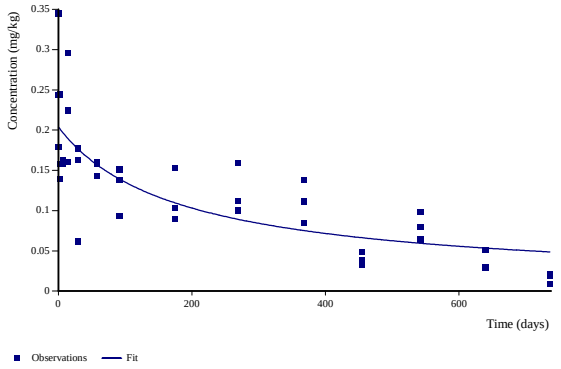
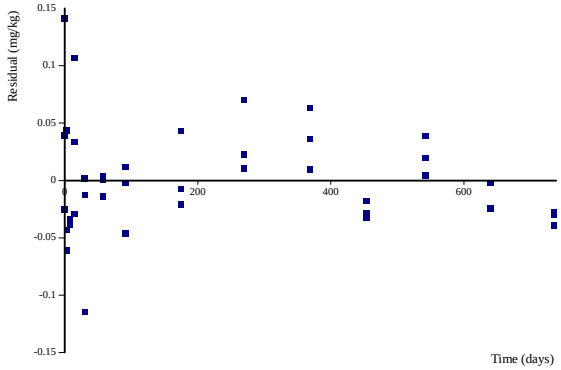
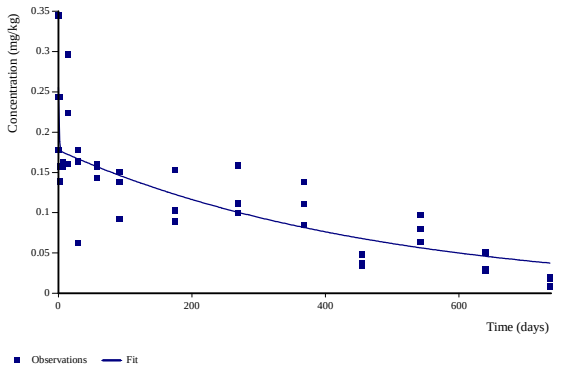
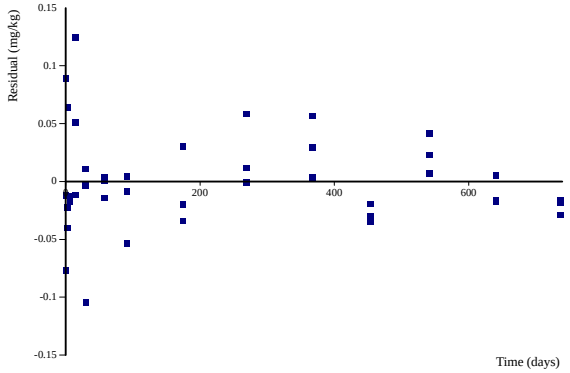
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.8-1** and the graphical outputs in Figure CA.B.**8.1.2.3.8-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.8-3**.

Table CA.B.8.1.2.3.8-1: Summary of IT01 T4 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Acceptable**	Acceptable	Acceptable	Acceptable
DT_{50} (days)		**292**	205	156	292
DT_{90} (days)		**971**	2.44E+3	918	971
DT_{90}/3.32 (days)		**n/a**	734	277	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	328	n/a
χ^2 error (%)		**19.7**	20.2	16.6	19.9
k ($days^{-1}$)	k or k_1	**0.002372**	n/a	1.047	0.002371
	k_2	**n/a**	n/a	0.002113	n/a
P value	k or k_1	**5.58E-7**	n/a	0.3532	5.95E-7
	k_2	**n/a**	n/a	2.62E-6	n/a
g		**n/a**	n/a	0.3054	n/a
alpha		**n/a**	0.8128	n/a	n/a
beta		**n/a**	152.5	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	-0.4394 / 2.065	n/a	n/a
	beta	**n/a**	-264 / 568.9	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.8-1: IT01 T4 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.8-2** and the graphical outputs in Figure CA.B.**8.1.2.3.8-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.8-3**.

Table CA.B.8.1.2.3.8-2: Summary of IT01 T4 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**195**	124
DT_{90} (days)		**646**	625
DT_{50} (days) - Slow phase		**n/a**	216
χ^2 error (%)		**17.6**	15.5
k ($days^{-1}$)	k or k_1	**0.003564**	2.862
	k_2	**n/a**	3.21E-3
P value	k or k_1	**3.95E-8**	0.4995
	k_2	**n/a**	3.74E-7
g		**n/a**	0.2552

Selected model shown in bold

Figure CA.B.8.1.2.3.8-2: Applicant's IT01 T4 parent-only modelling kinetic fits

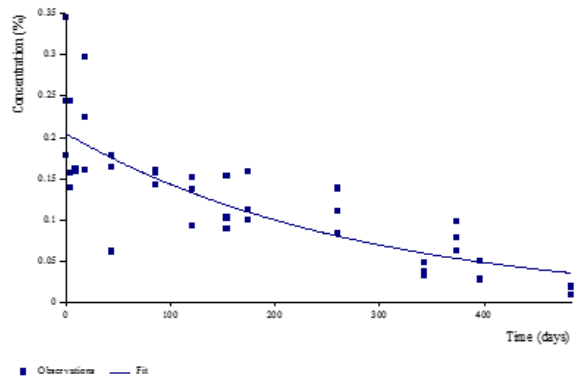
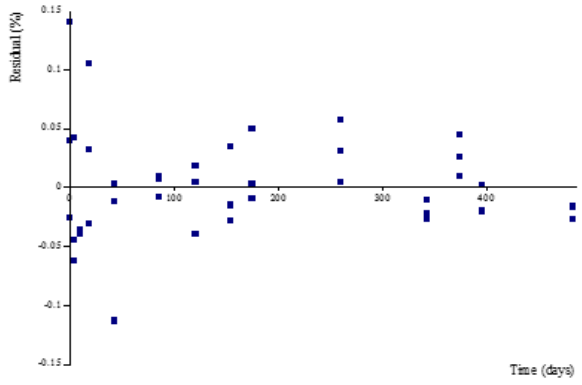
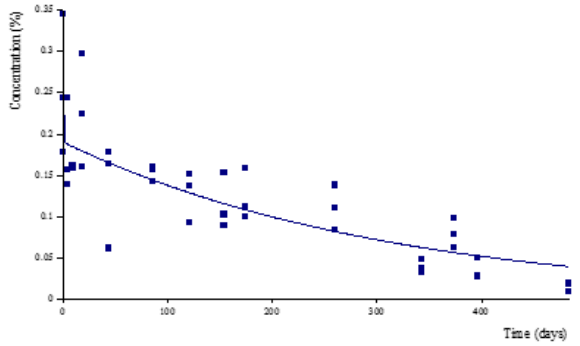
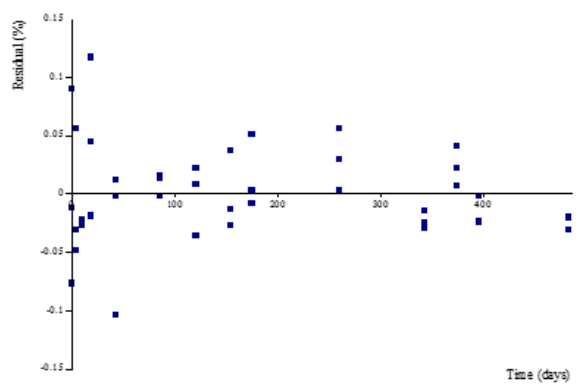
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.8-3: Summary of IT01 T4 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	292	971
Modelling endpoints			
Bixlozone	SFO	195	646

CA.B.8.1.2.3.9. IT02 T2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted the SFO fit was visually and statistically unacceptable. As the DFOP resulted in a good visual fit, the lowest χ^2 value and acceptable t-test values, the applicant concluded upon this model as the best-fit model.

The CA has repeated the applicant's SFO, FOMC and DFOP kinetic fits and agrees with their assessment.

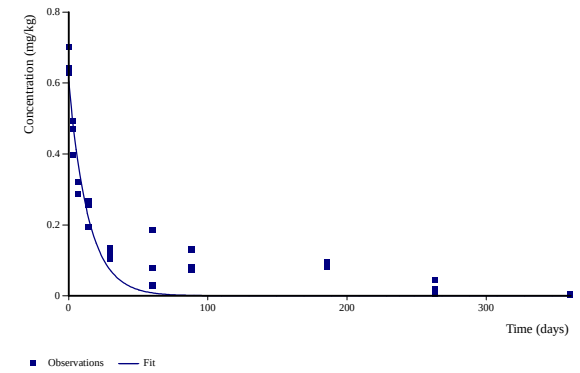
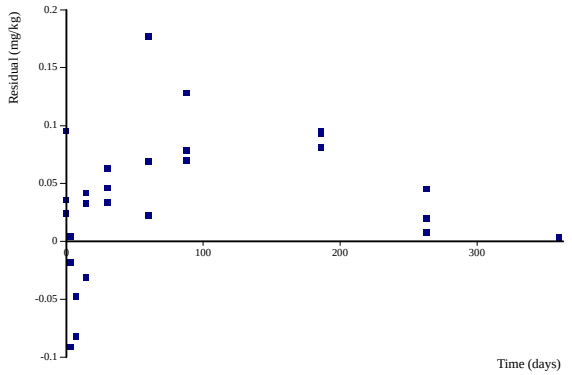
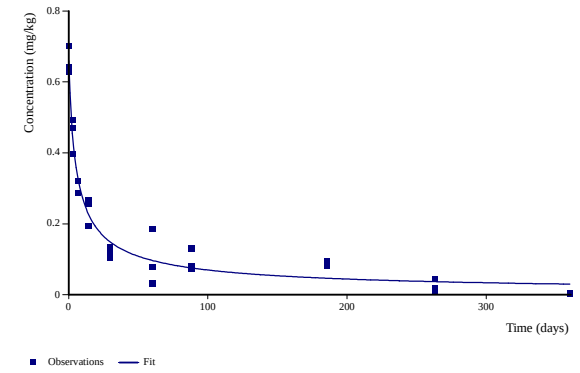
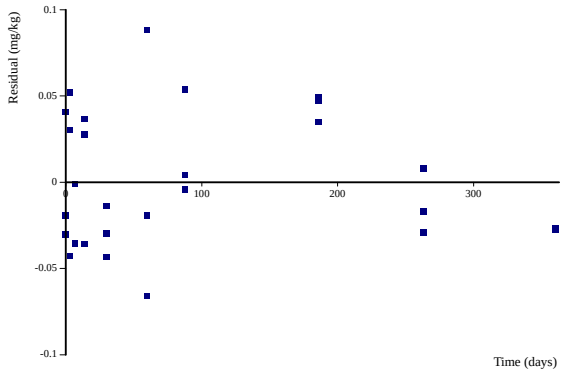
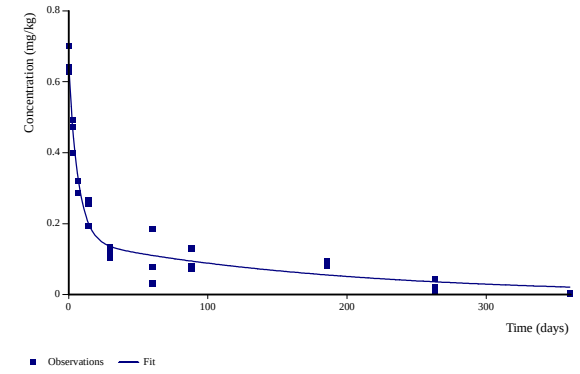
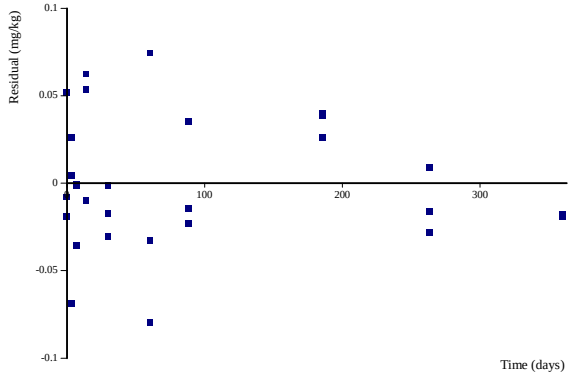
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.9-1** and the graphical outputs in Figure CA.B.**8.1.2.3.9-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.9-3**.

Table CA.B.8.1.2.3.9-1: Summary of IT02 T2 best-fit kinetic modelling

Modelling		**Applicant's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	**DFOP**
Visual fit		Unacceptable	Good	**Good**
DT_{50} (days)		9.75	6.6	**6.9**
DT_{90} (days)		32.4	109	**157**
DT_{90}/3.32 (days)		n/a	32.7	**47.3**
DT_{50} (days) - Slow phase		n/a	n/a	**126**
χ^2 error (%)		22.8	8.48	**8.30**
k ($days^{-1}$)	k or k_1	0.07106	n/a	**0.1496**
	k_2	n/a	n/a	**0.005509**
P value	k or k_1	5.85E-8	n/a	**8.50E-8**
	k_2	n/a	n/a	**0.002424**
g		n/a	n/a	**0.7631**
alpha		n/a	0.6723	**n/a**
beta		n/a	3.658	**n/a**
95% CI (lower/upper)	alpha	n/a	0.4925 / 0.852	**n/a**
	beta	n/a	1.478 / 5.838	**n/a**

Best-fit model shown in bold

Figure CA.B.8.1.2.3.9-1: Applicant's IT02 T2 parent-only kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an unacceptable visual fit of the data, whereas, the DFOP visual and statistical fits were acceptable. Therefore, the applicant considered the DFOP endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant. The CA notes that both fits resulted in similar DT90 values and a DFOP back-calculated DT50 (DT90/3/32; since the DT90 was reached during the trial) would be very similar to the SFO DT50. Therefore, for simplicity, the SFO fit is considered acceptable by the CA. As the CA obtained very similar SFO endpoints to the applicant, the applicant's SFO fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.9-2** and the graphical outputs in Figure CA.B.**8.1.2.3.9-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.9-3**.

Table CA.B.8.1.2.3.9-2: Summary of IT02 T2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable***	Acceptable
DT_{50} (days)		**9.38**	5.95
DT_{90} (days)		**31.2**	39.5
DT_{50} (days) - Slow phase		**n/a**	64.7
χ^2 error (%)		**19.3**	6.4
k ($days^{-1}$)	k or k_1	**0.07392**	0.192
	k_2	**n/a**	0.01756
P value	k or k_1	**7.76E-10**	1.35E-5
	k_2	**n/a**	3.99E-3
g		**n/a**	0.6886

Selected model shown in bold

* The applicant considered the visual fit unacceptable

Figure CA.B.8.1.2.3.9-2: IT02 T2 parent-only modelling kinetic fits

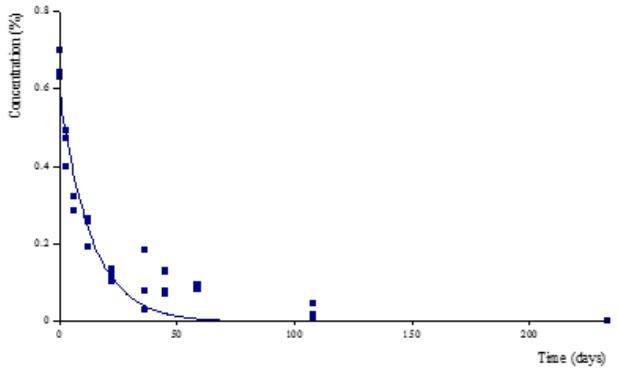
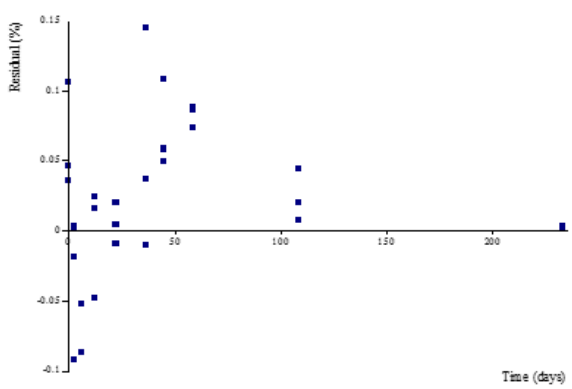
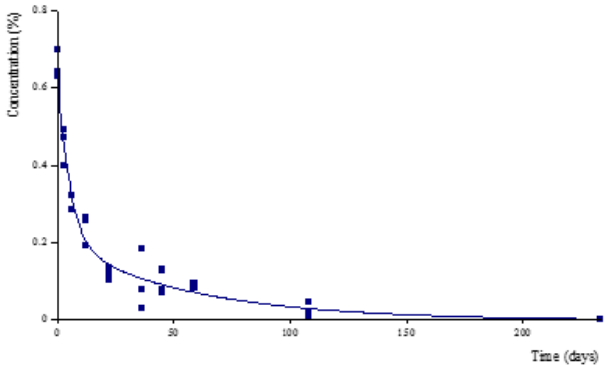
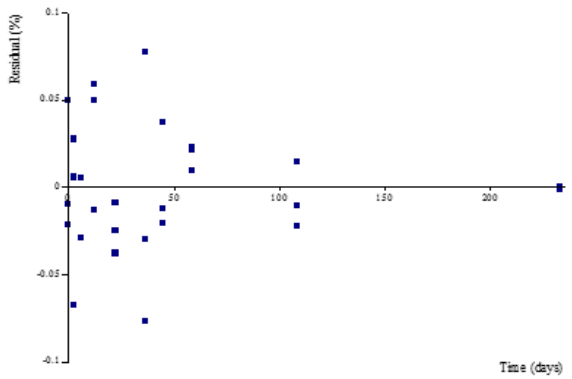
Applicant's modelling	
Fitted Model	**Residuals**
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.9-3: Summary of IT02 T2 modelling and best-fit endpoints

Compound	**Model**	**DT50 (d)**	**DT90 (d)**
Best-fit endpoints			
Bixlozone	DFOP	6.9	157
Modelling endpoints			
Bixlozone	SFO	9.38	31.2

CA.B.8.1.2.3.10. GE01 T1

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA obtained a very similar SFO DT50 value to the applicant (difference <1.5 days); therefore, the applicant's SFO results are accepted on this occasion.

The CA notes residues of 2,4-DBA were detected in the trial at 29 DALA and 58 DALA. However, as this only covers two time points and all the residues were <LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

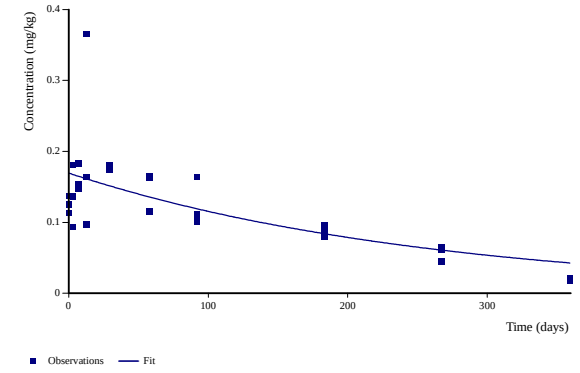
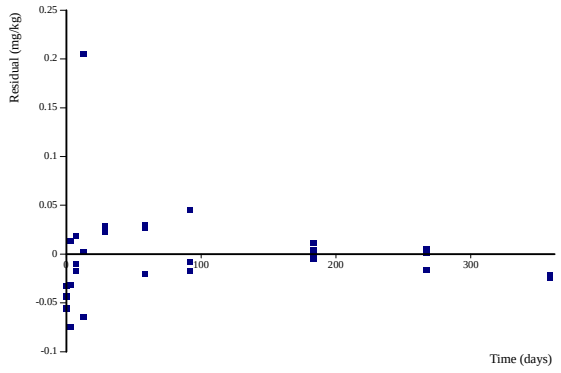
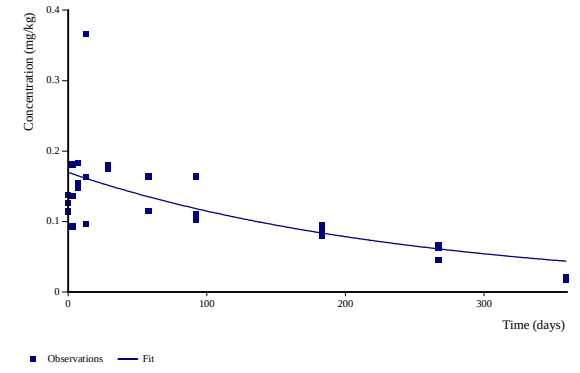
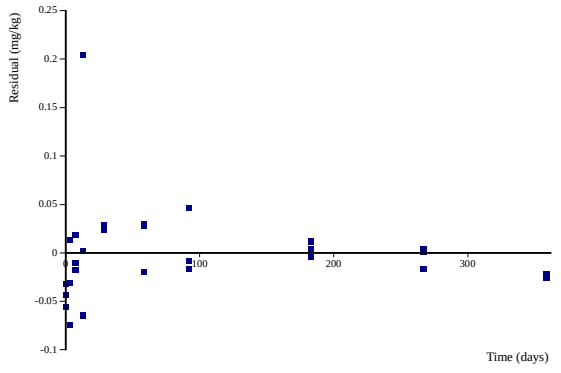
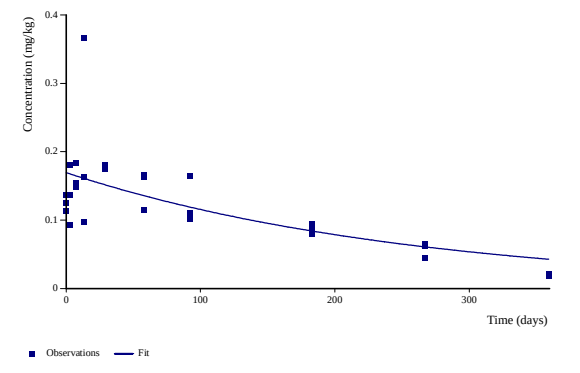
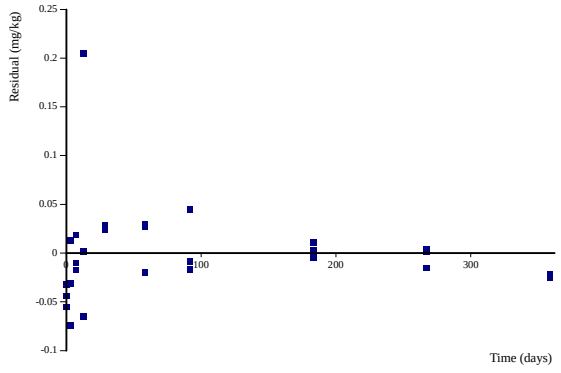
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.10-1** and the graphical outputs in Figure CA.B.**8.1.2.3.10-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.10-4**.

Table CA.B.8.1.2.3.10-1: Summary of GE01 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**		
Pathway		**bixlozone only**		
Compound		**bixlozone**	**bixlozone**	**bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Good**	Good	Good
DT_{50} (days)		**181**	180	181
DT_{90} (days)		**601**	631	601
DT_{90}/3.32 (days)		**n/a**	190	181
DT_{50} (days) - Slow phase		**n/a**	n/a	181
χ^2 error (%)		**16.5**	17.5	18.3
k ($days^{-1}$)	k or k_1	**0.003833**	n/a	0.003835
	k_2	**n/a**	n/a	0.003826
P value	k or k_1	**6.74E-4**	n/a	0.4991
	k_2	**n/a**	n/a	0.4999
g		**n/a**	n/a	0.8932
alpha		**n/a**	14.4	n/a
beta		**n/a**	3.64E+3	n/a
95% CI (lower/upper)	alpha	**n/a**	6.463 / 22.35	n/a
	beta	**n/a**	3.48E+3 / 3.81E+3	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.10-1: GE01 T1 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Persistence endpoints

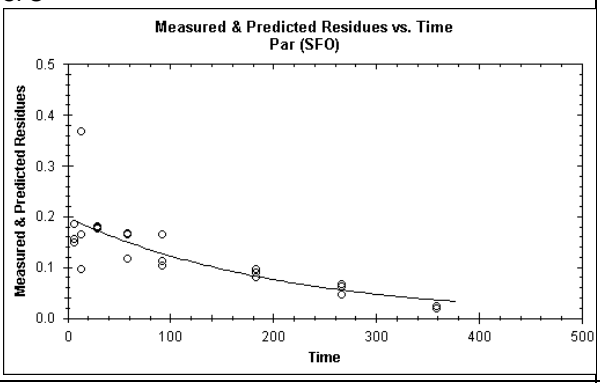
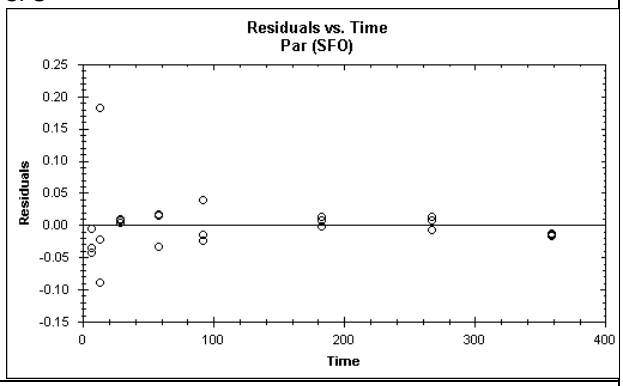
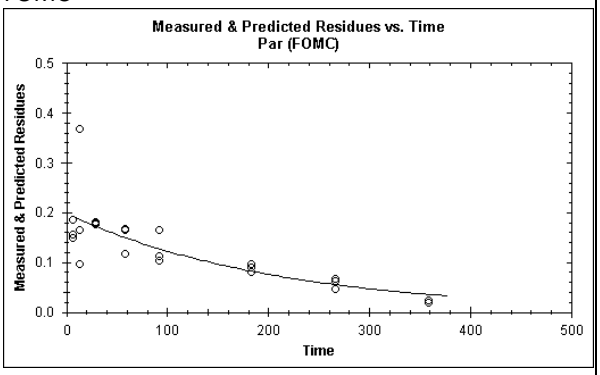
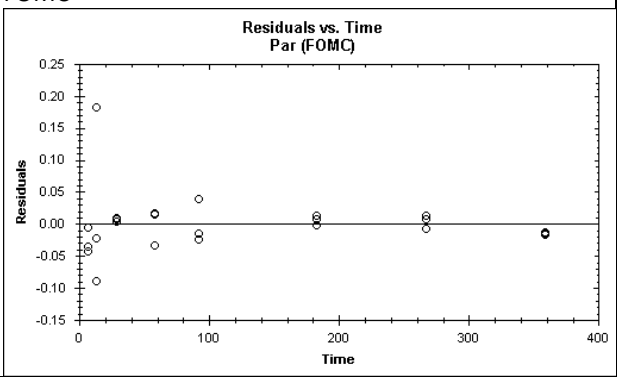
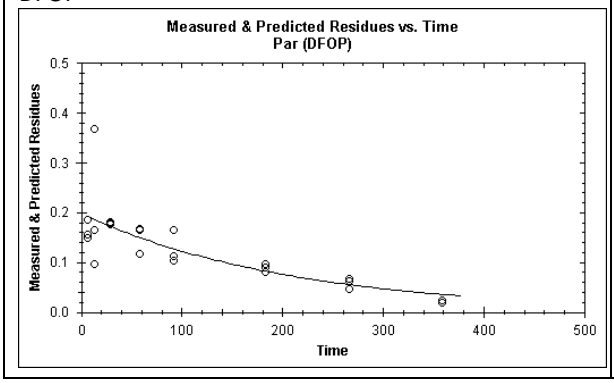
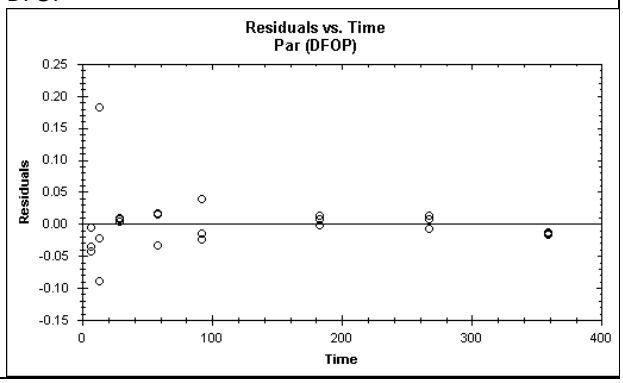
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a good visual fit and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.10-2** and Figure CA.B.**8.1.2.3.10-2**.

Table CA.B.8.1.2.3.10-2: Summary of GE01 T1 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Good**	Good	Good
DT_{50} (days)		**144**	144	144
DT_{90} (days)		**477**	477	477
DT_{90}/3.32 (days)		**n/a**	144	144
DT_{50} (days) - Slow phase		**n/a**	n/a	144
χ^2 error (%)		**9.39**	10.0	10.8
k ($days^{-1}$)	k or k_1	**0.00483**	n/a	1.641E-1
	k_2	**n/a**	n/a	4.829E-3
P value	k or k_1	**2.74E-4**	n/a	<2E-16
	k_2	**n/a**	n/a	1.75E-3
g		**n/a**	n/a	2.22E-14
alpha		**n/a**	4.657E+3	n/a
beta		**n/a**	9.641E+5	n/a
95% CI (lower/upper)	alpha	**n/a**	4.657E+3 / 4.657E+3	n/a
	beta	**n/a**	9.641E+5 / 9.641E+5	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.10-2: CA's GE01 T1 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.10-3** and the graphical outputs in Figure CA.B.**8.1.2.3.10-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.10-4**.

Table CA.B.8.1.2.3.10-3: Summary of GE01 T1 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**49.3**	47.7
DT_{90} (days)		**164**	178
DT_{50} (days) - Slow phase		**n/a**	89.6
χ^2 error (%)		**9.003**	10.3
k ($days^{-1}$)	k or k_1	**0.01406**	0.01781
	k_2	**n/a**	7.73E-3
P value	k or k_1	**6.93E-4**	0.4572
	k_2	**n/a**	0.4863
g		**n/a**	0.7269

Selected model shown in bold

Figure CA.B.8.1.2.3.10-3: Applicant's GE01 T1 parent-only modelling kinetic fits

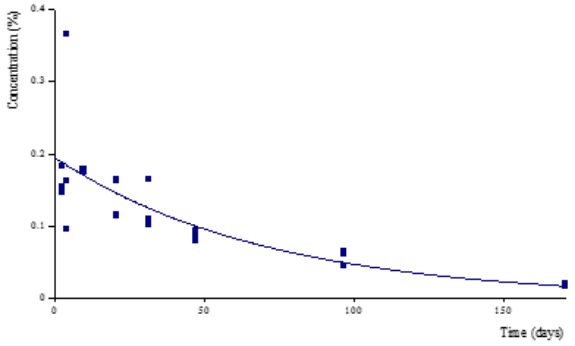
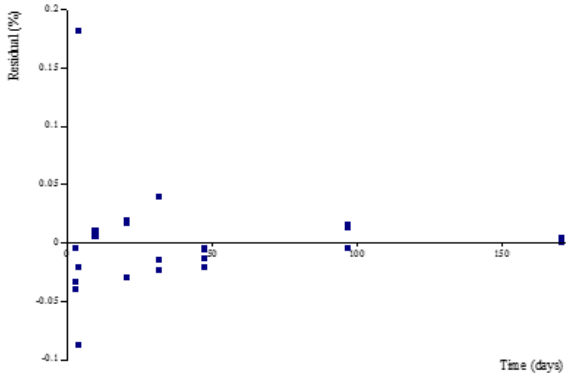
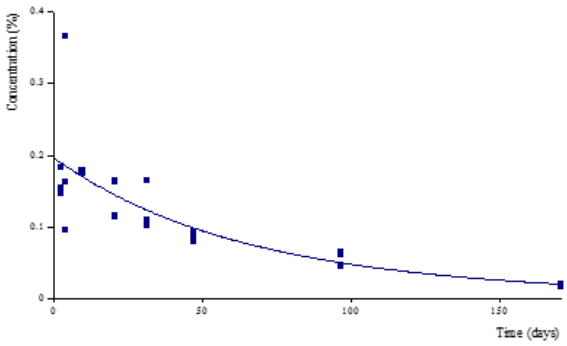
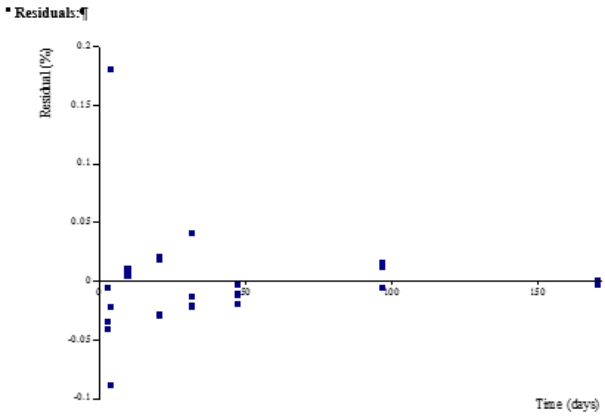
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.10-4: Summary of GE01 T1 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	181	601
Persistence endpoints			
Bixlozone	SFO	144	477
Modelling endpoints			
Bixlozone	SFO	49.3	164

CA.B.8.1.2.3.11. GE01 T2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA obtained a very similar SFO DT50 value to the applicant (difference <1 day); therefore, the applicant's SFO results are accepted on this occasion.

The CA notes residues of 2,4-DBA were detected in the trial at 29 DALA and 58 DALA. However, as this only covers two time points and all the residues were <LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

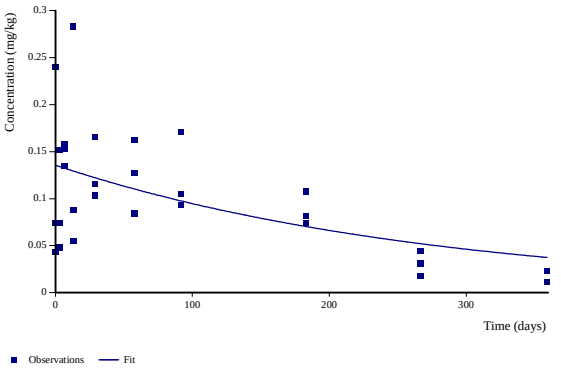
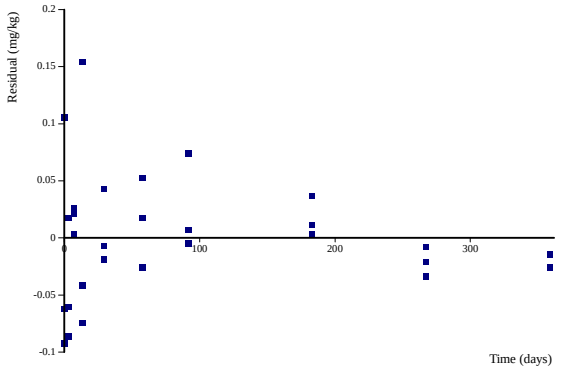
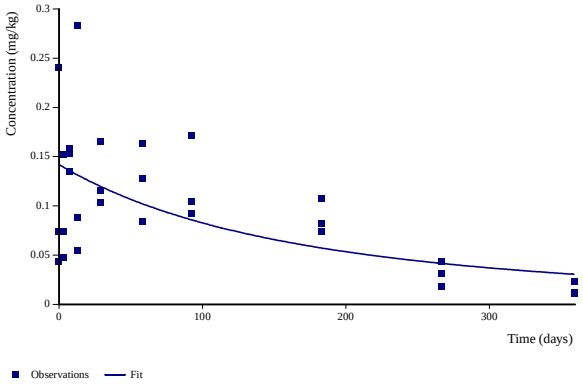
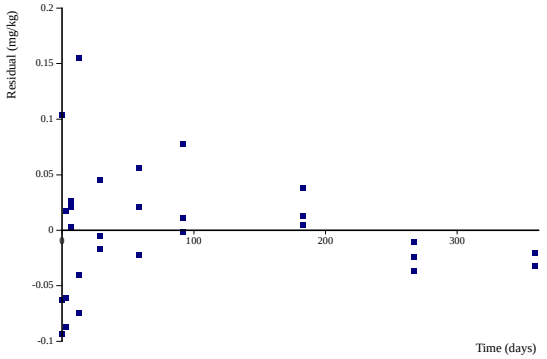
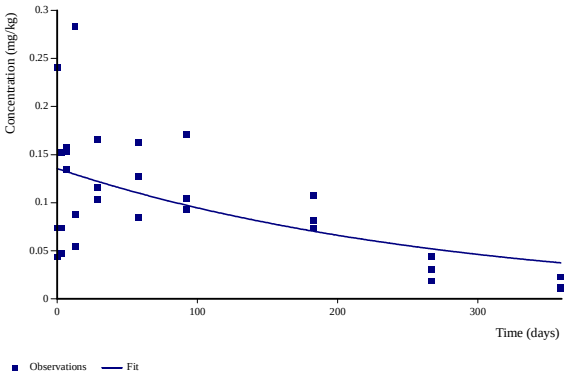
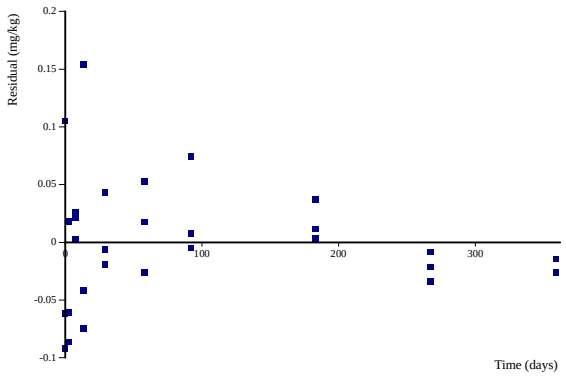
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.11-1** and the graphical outputs in Figure CA.B.**8.1.2.3.11-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.11-3**.

Table CA.B.8.1.2.3.11-1: Summary of GE01 T2 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Acceptable**	Acceptable	Acceptable	Acceptable
DT_{50} (days)		**193**	133	193	194
DT_{90} (days)		**642**	652	650	644
DT_{90}/3.32 (days)		**n/a**	196	196	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	>10,000	n/a
χ^2 error (%)		**17.2**	19.7	19.1	17.2
k ($days^{-1}$)	k or k_1	**0.003588**	n/a	0.003611	0.003578
	k_2	**n/a**	n/a	3.59E-9	n/a
P value	k or k_1	**0.00834**	n/a	0.452	0.00835
	k_2	**n/a**	n/a	0.5	n/a
g		**n/a**	n/a	0.9953	n/a
alpha		**n/a**	2.29	n/a	n/a
beta		**n/a**	376	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	1.184 / 3.395	n/a	n/a
	beta	**n/a**	nd	n/a	n/a

Best-fit model shown in bold
nd – Not determined: parameter could not be calculated by the CAKE model.

Figure CA.B.8.1.2.3.11-1: GE01 T2 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.11-2** and the graphical outputs in Figure CA.B.**8.1.2.3.11-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.11-3**.

Table CA.B.8.1.2.3.11-2: Summary of GE01 T2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Unacceptable
DT_{50} (days)		**68.9**	68.9
DT_{90} (days)		**229**	>10000
DT_{50} (days) - Slow phase		**n/a**	234
χ^2 error (%)		**15.5**	17.3
k ($days^{-1}$)	k or k_1	**0.01006**	0.01019
	k_2	**n/a**	1.46E-7
P value	k or k_1	**0.01204**	0.4664
	k_2	**n/a**	0.5000
g		**n/a**	0.9919

Selected model shown in bold

Figure CA.B.8.1.2.3.11-2: Applicant's GE01 T2 parent-only modelling kinetic fits

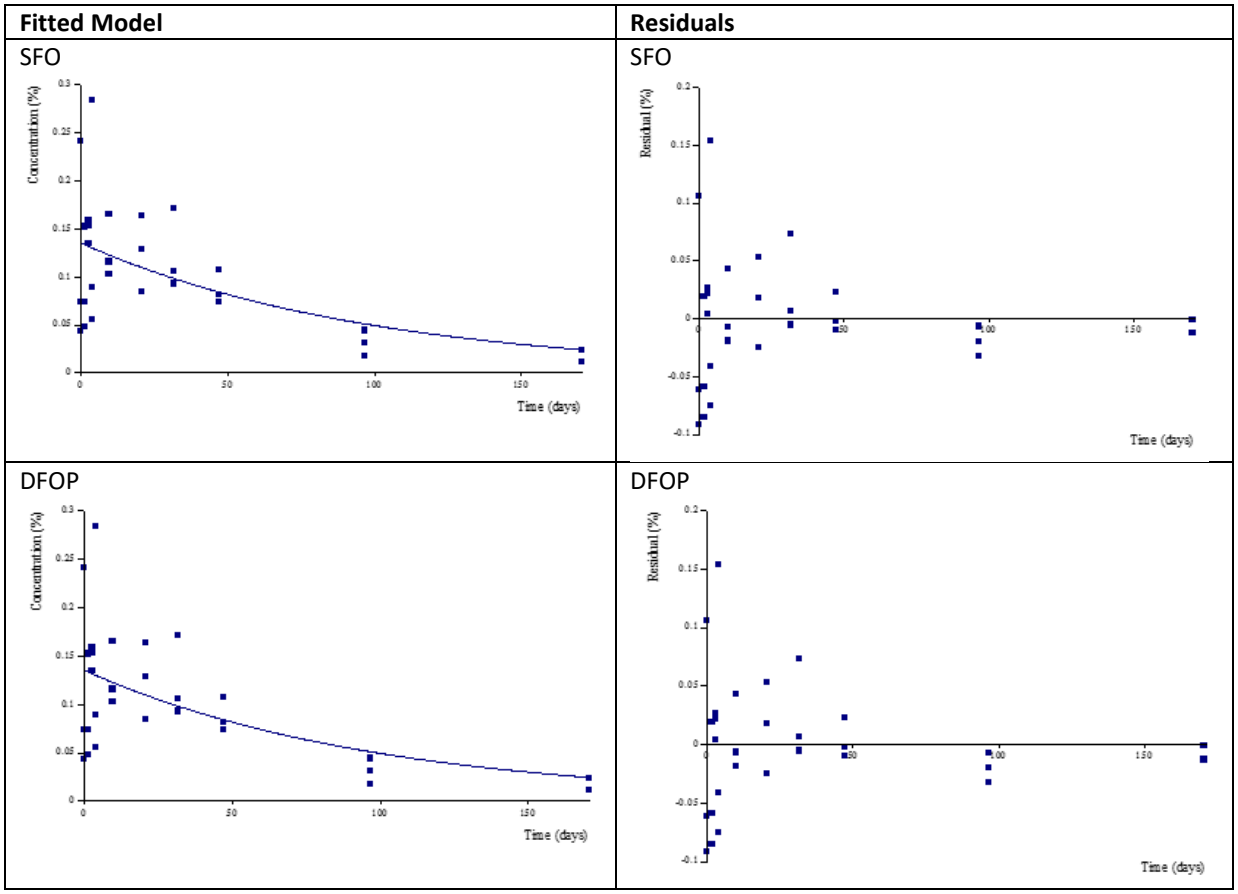


Table CA.B.8.1.2.3.11-3: Summary of GE01 T2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	193	642
Modelling endpoints			
Bixlozone	SFO	68.9	229

CA.B.8.1.2.3.12. GE01 T3

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA obtained a very similar SFO DT50 value to the applicant (difference <2 days); therefore, the applicant's SFO results are accepted on this occasion.

The CA notes residues of 2,4-DBA were detected in the trial at 29 DALA. However, as this is only one time points and the residues were <LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

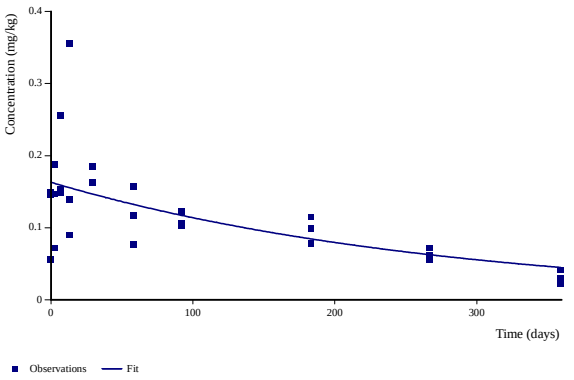
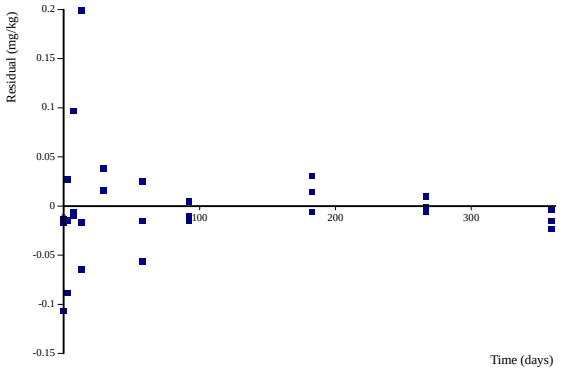
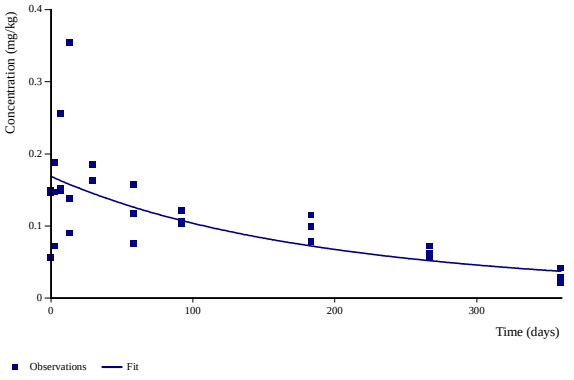
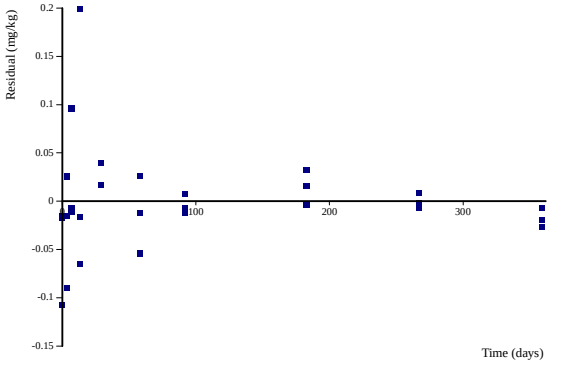
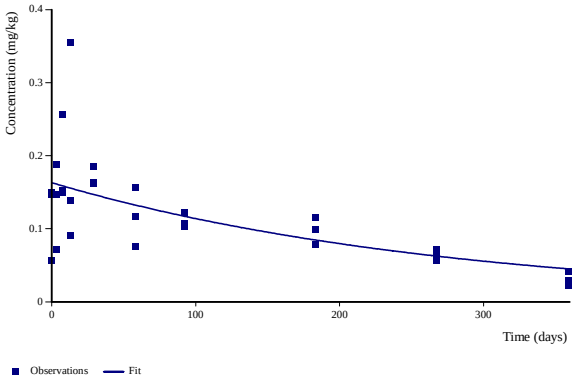
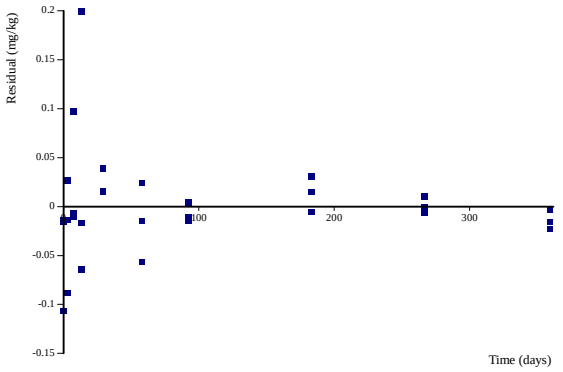
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.12-1** and the graphical outputs in Figure CA.B.**8.1.2.3.12-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.12-4**.

Table CA.B.8.1.2.3.12-1: Summary of GE01 T3 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Good**	Good	Good	Good
DT_{50} (days)		**194**	147	194	196
DT_{90} (days)		**643**	616	644	651
DT_{90}/3.32 (days)		**n/a**	185	194	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	195	n/a
χ^2 error (%)		**16.3**	17.5	18.1	16.3
k ($days^{-1}$)	k or k_1	**0.00358**	n/a	0.003586	0.00354
	k_2	**n/a**	n/a	0.003564	n/a
P value	k or k_1	**0.002356**	n/a	0.4989	0.0025
	k_2	**n/a**	n/a	0.499	n/a
g		**n/a**	n/a	0.5107	n/a
alpha		**n/a**	3.672	n/a	n/a
beta		**n/a**	706.2	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	nd	n/a	n/a
	beta	**n/a**	nd	n/a	n/a

Best-fit model shown in bold
nd – Not determined: parameter could not be calculated by the CAKE model.

Figure CA.B.8.1.2.3.12-1: GE01 T3 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Persistence endpoints

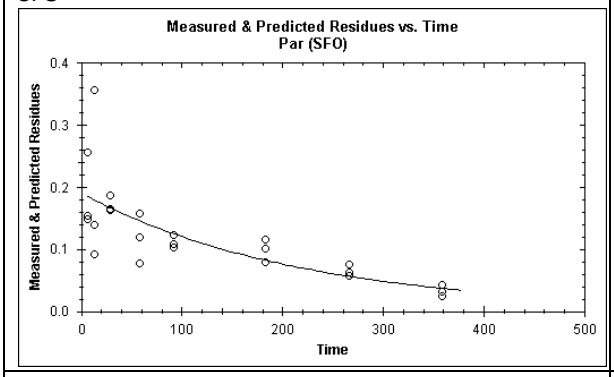
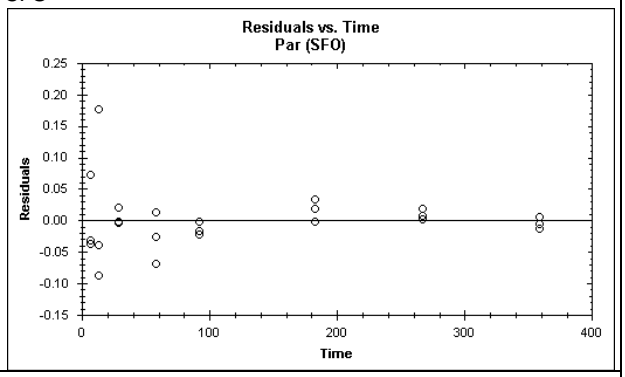
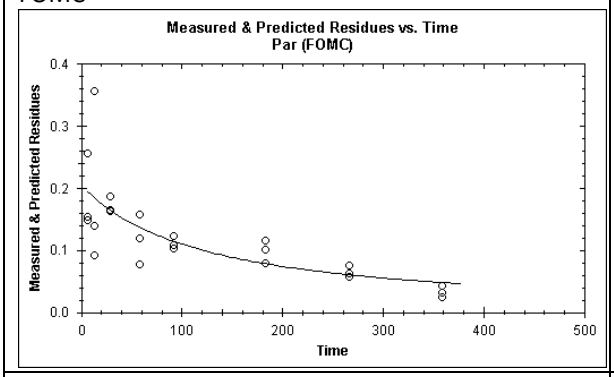
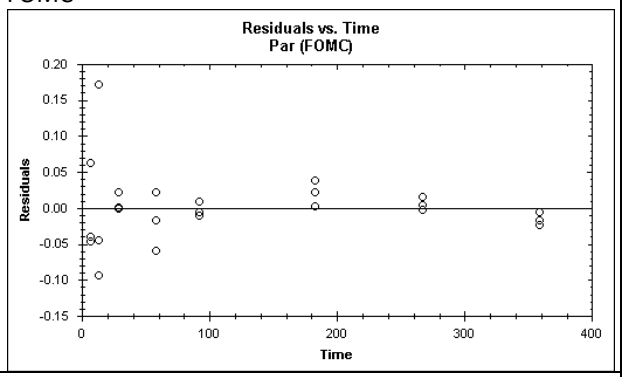
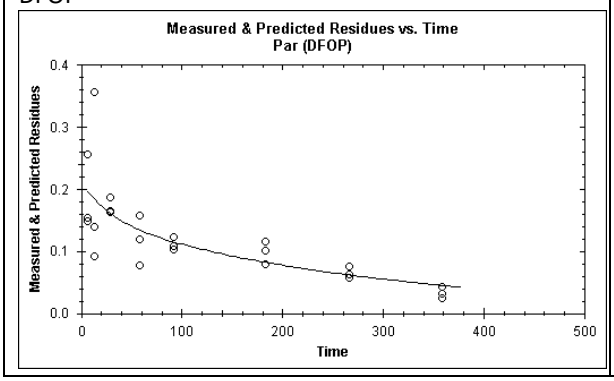
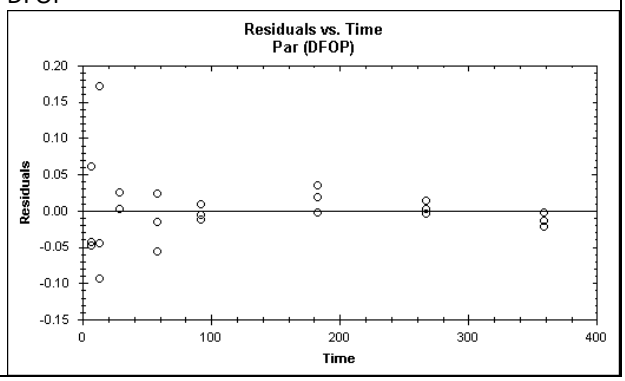
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a good visual fit and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.12-2** and Figure CA.B.**8.1.2.3.12-2**.

Table CA.B.8.1.2.3.12-2: Summary of GE01 T3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**bixlozone only**		
Compound		**bixlozone**	**bixlozone**	**bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Good**	Good	Good
DT_{50} (days)		**151**	114	112
DT_{90} (days)		**500**	911	575
DT_{90}/3.32 (days)		**n/a**	275	173
DT_{50} (days) - Slow phase		**n/a**	n/a	201
χ^2 error (%)		**9.28**	8.84	8.66
k ($days^{-1}$)	k or k_1	**4.60E-3**	n/a	0.0333
	k_2	**n/a**	n/a	0.0034
P value	k or k_1	**7.5E-4**	n/a	0.3882
	k_2	**n/a**	n/a	0.1401
g		**n/a**	n/a	0.2755
alpha		**n/a**	1.118	n/a
beta		**n/a**	133.3	n/a
95% CI (lower/upper)	alpha	**n/a**	-2.177 / 4.413	n/a
	beta	**n/a**	-493.3 / 759.8	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.12-2: CA's GE01 T3 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.12-3** and the graphical outputs in Figure CA.B.**8.1.2.3.12-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.12-4**.

Table CA.B.8.1.2.3.12-3: Summary of GE01 T3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Good
DT_{50} (days)		**50.5**	34.6
DT_{90} (days)		**168**	229
DT_{50} (days) - Slow phase		**n/a**	86.4
χ^2 error (%)		**8.78**	5.39
k ($days^{-1}$)	k or k_1	**0.01372**	0.07774
	k_2	**n/a**	8.02E-3
P value	k or k_1	**1.60E-3**	0.3351
	k_2	**n/a**	0.1595
g		**n/a**	0.3734

Selected model shown in bold

Figure CA.B.8.1.2.3.12-3: Applicant's GE01 T3 parent-only modelling kinetic fits

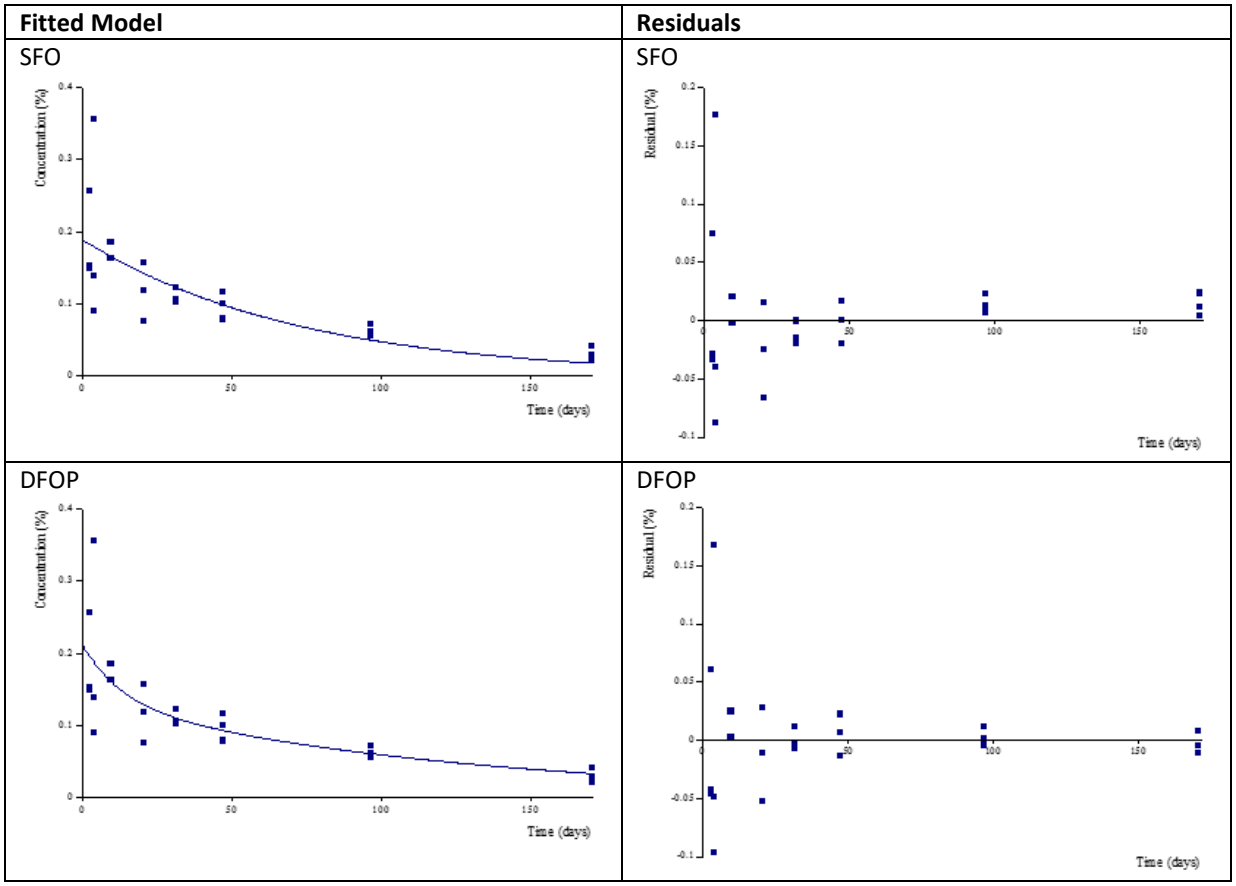


Table CA.B.8.1.2.3.12-4: Summary of GE01 T3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	194	643
Persistence endpoints			
Bixlozone	SFO	151	500
Modelling endpoints			
Bixlozone	SFO	50.5	168

CA.B.8.1.2.3.13. GE01 T4

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. However, the CA obtains slightly longer DT50 and DT90 values to the applicant, likely due to the slightly different data handling techniques used. As the CA's considers its approach more appropriate, the CA's SFO fit is considered further.

The CA notes residues of 2,4-DBA were detected in the trial at 29 DALA and 267 DALA. However, as this only represents two time points and only one of the residues was >LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts a parent-only evaluation for this trial.

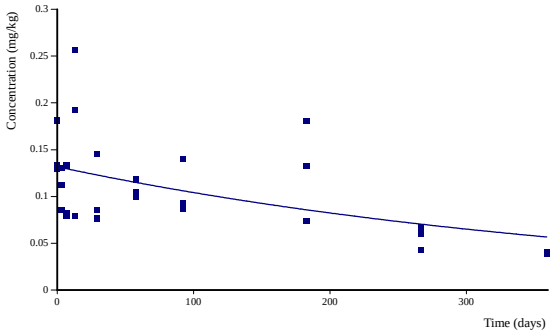
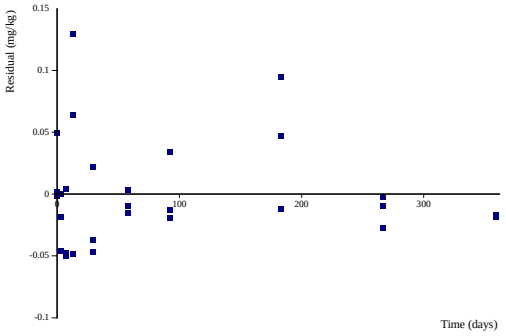
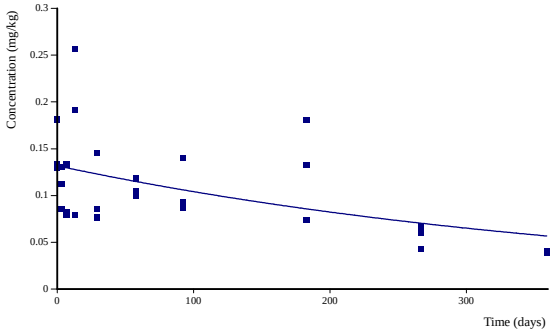
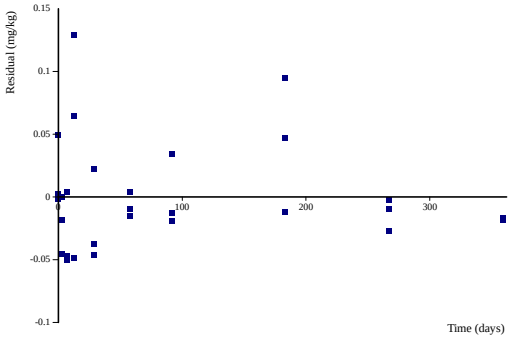
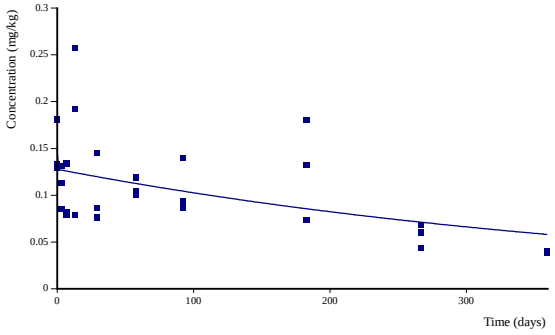
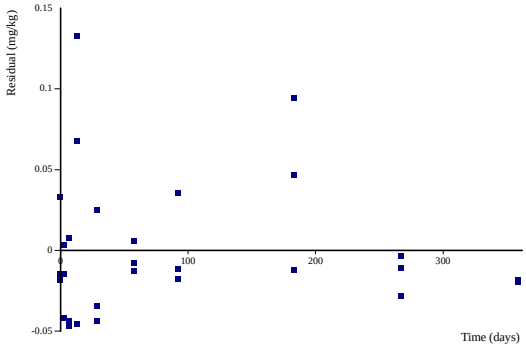
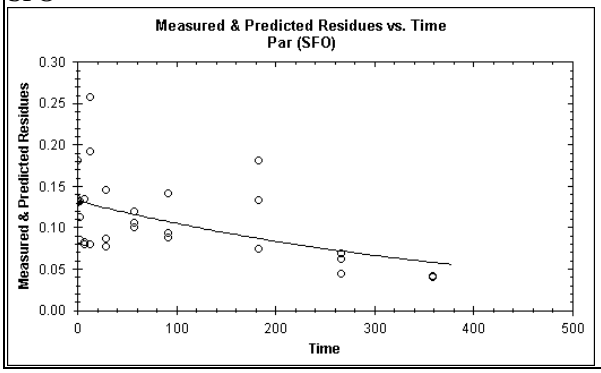
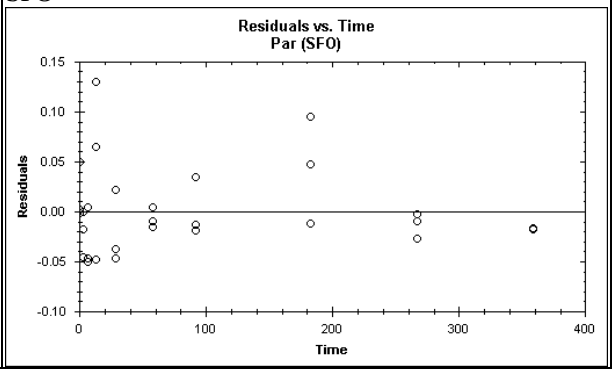
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.13-1** and the graphical outputs in Figure CA.B.**8.1.2.3.13-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.13-3**.

Table CA.B.8.1.2.3.13-1: Summary of GE01 T4 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	DFOP	**SFO**
Visual fit		Acceptable	Acceptable	Acceptable	**Acceptable**
DT_{50} (days)		296	296	248	**300**
DT_{90} (days)		982	986	983	**997**
DT_{90}/3.32 (days)		n/a	297	296	**n/a**
DT_{50} (days) - Slow phase		n/a	n/a	316	**n/a**
χ^2 error (%)		19.5	20.5	21.1	**19.5**
k ($days^{-1}$)	k or k_1	0.002346	n/a	5.506	**0.00231**
	k_2	n/a	n/a	0.002191	**n/a**
P value	k or k_1	0.005498	n/a	0.4853	**0.006**
	k_2	n/a	n/a	0.004877	**n/a**
g		n/a	n/a	0.1384	**n/a**
alpha		n/a	200	n/a	**n/a**
beta		n/a	8.51E+4	n/a	**n/a**
95% CI (lower/upper)	alpha	n/a	-2653 / 3.05E+003	n/a	**n/a**
	beta	n/a	-1.078E+6 / 1.25E+6	n/a	**n/a**

Best-fit model shown in bold

Figure CA.B.8.1.2.3.13-1: GE01 T4 parent kinetic fits

Fitted Model	**Residuals**
Applicant's kinetic fits	
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	
CA's SFO kinetic fit	
SFO	SFO
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.13-2** and the graphical outputs in Figure CA.B.**8.1.2.3.13-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.13-3**.

Table CA.B.8.1.2.3.13-2: Summary of GE01 T4 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Unacceptable
DT_{50} (days)		**105**	105
DT_{90} (days)		**350**	106
DT_{50} (days) - Slow phase		**n/a**	350
χ^2 error (%)		**17.8**	19.7
k ($days^{-1}$)	k or k_1	**6.59E-3**	0.1588
	k_2	**n/a**	6.57E-3
P value	k or k_1	**4.95E-3**	0.0851
	k_2	**n/a**	0.0066
g		**n/a**	0.0031

Selected model shown in bold

Figure CA.B.8.1.2.3.13-2: Applicant's GE01 T4 parent-only modelling kinetic fits

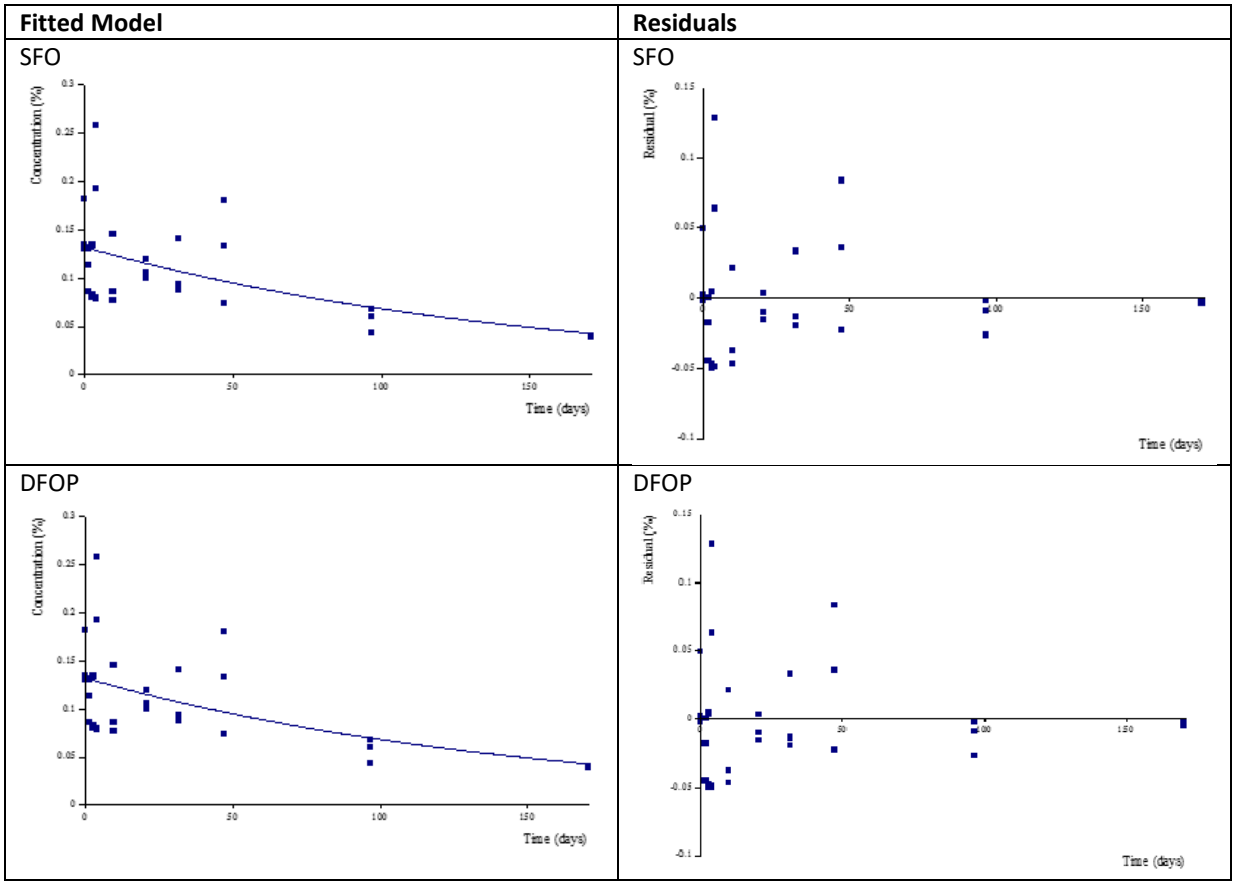


Table CA.B.8.1.2.3.13-3: Summary of GE01 T4 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	300	997
Modelling endpoints			
Bixlozone	SFO	105	350

CA.B.8.1.2.3.14. FR02 T1

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA obtained a very similar SFO DT50 value to the applicant (difference <0.5 days); therefore, the applicant's SFO results are accepted on this occasion.

The CA notes residues of 2,4-DBA were detected in the trial at 6 DALA, 13 DALA and 30 DALA. However, as the residues were all <LOQ and the 30 DALA residues were slightly larger than the 6 DALA residues (and so no decline phase can be observed), the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

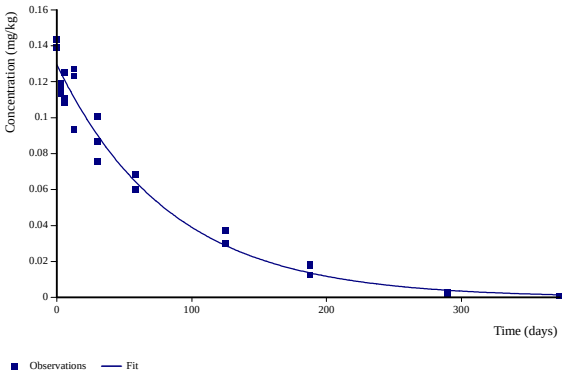
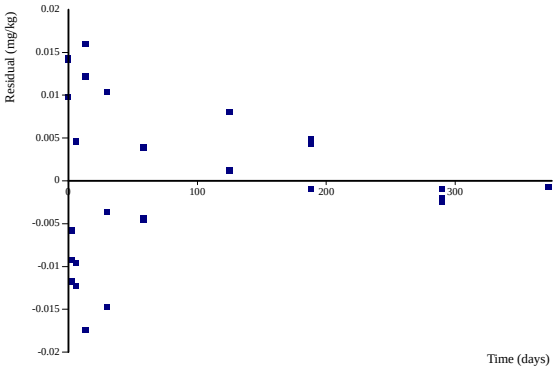
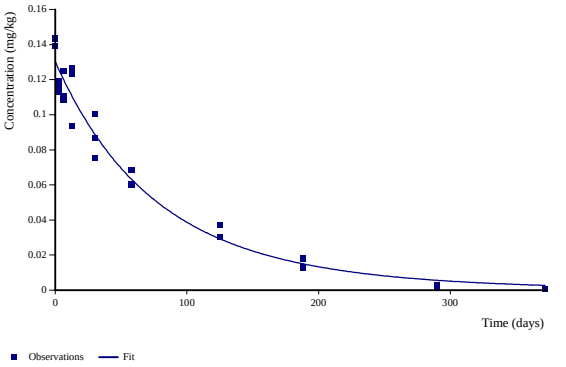
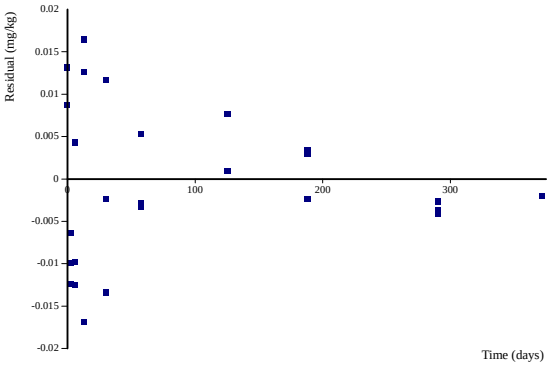
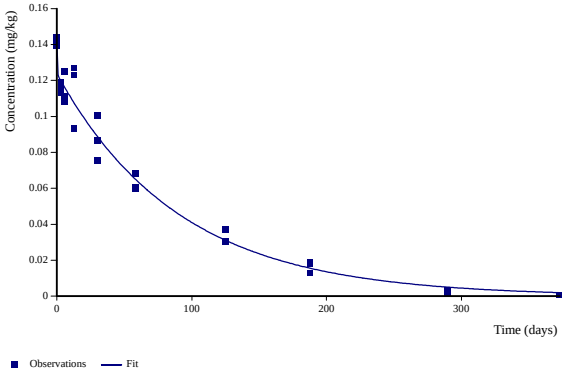
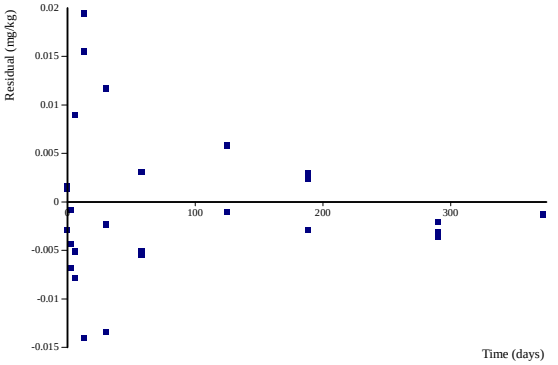
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.14-1** and the graphical outputs in Figure CA.B.**8.1.2.3.14-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.14-4**.

Table CA.B.8.1.2.3.14-1: Summary of FR02 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Very good**	Very good	Very good	Very good
DT_{50} (days)		**57.8**	55.4	50.2	58.0
DT_{90} (days)		**192**	202	196	193
DT_{90}/3.32 (days)		**n/a**	60.9	59	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	62.8	n/a
χ^2 error (%)		**6.69**	6.88	3.88	6.52
k ($days^{-1}$)	k or k_1	**0.01198**	n/a	4.033	0.0119
	k_2	**n/a**	n/a	0.01104	n/a
P value	k or k_1	**4.85E-13**	n/a	0.4668	3.88E-13
	k_2	**n/a**	n/a	1.38E-13	n/a
g		**n/a**	n/a	0.1297	n/a
alpha		**n/a**	8.675	n/a	n/a
beta		**n/a**	665.6	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	-18.7 / 36.05	n/a	n/a
	beta	**n/a**	-1602 / 2.93E+003	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.14-1: FR02 T1 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Persistence endpoints

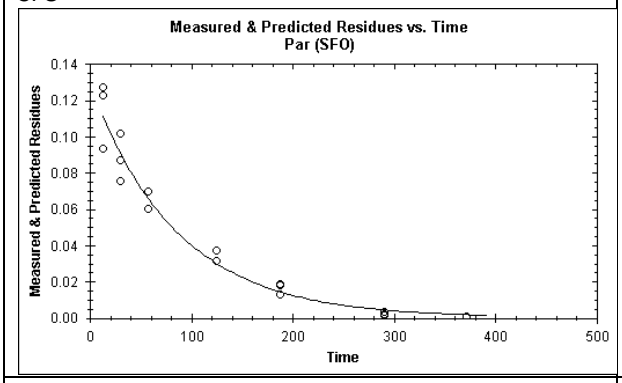
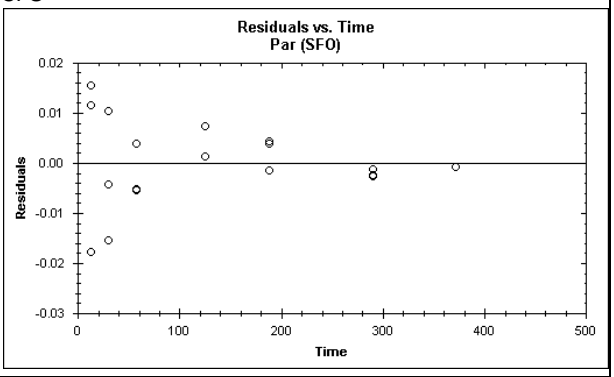
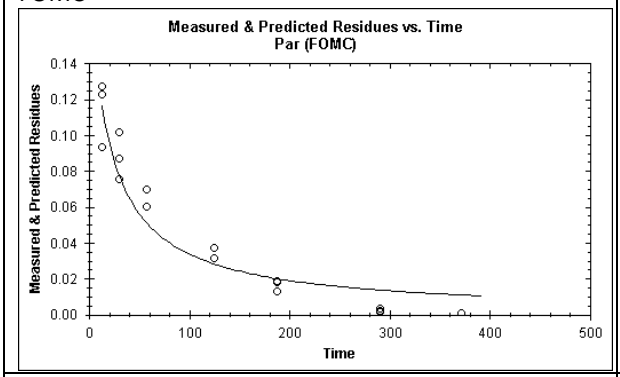
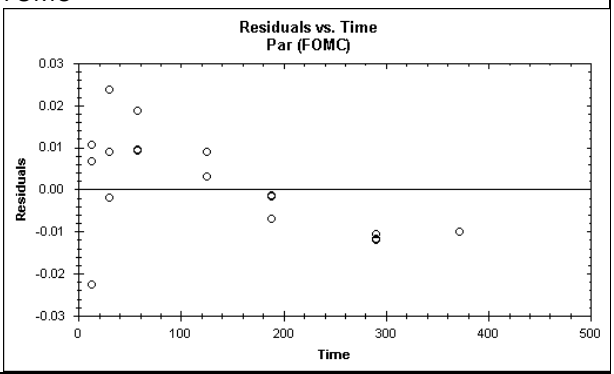
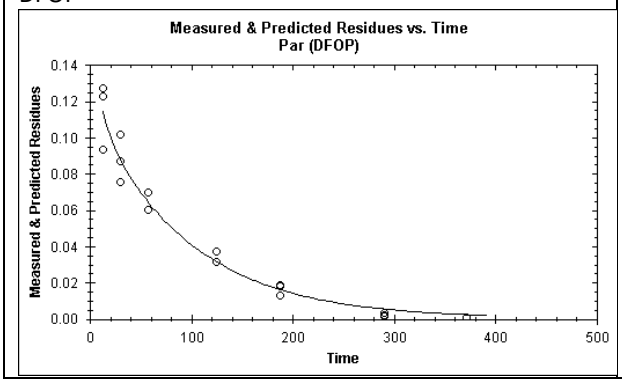
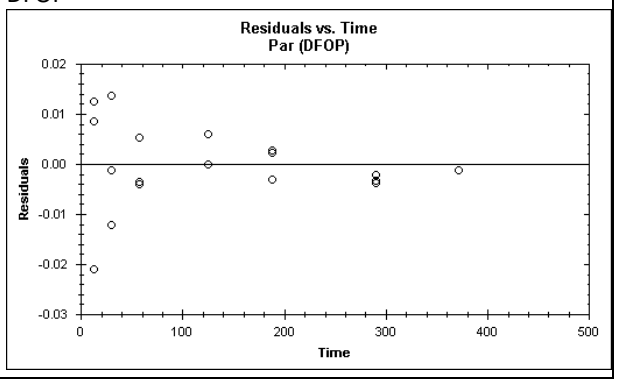
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a very good visual fit and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.14-2** and Figure CA.B.**8.1.2.3.14-2**.

Table CA.B.8.1.2.3.14-2: Summary of FR02 T1 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Very good**	Acceptable	Very good
DT_{50} (days)		**58.9**	20.9	33.4
DT_{90} (days)		**196**	199	182
DT_{90}/3.32 (days)		**n/a**	59.8	54.9
DT_{50} (days) - Slow phase		**n/a**	n/a	64.6
χ^2 error (%)		**4.80**	16.6	3.60
k ($days^{-1}$)	k or k_1	**0.01177**	n/a	0.11770
	k_2	**n/a**	n/a	0.01073
P value	k or k_1	**1.65E-9**	n/a	0.391
	k_2	**n/a**	n/a	1.43E-5
g		**n/a**	n/a	0.2925
alpha		**n/a**	0.9562	n/a
beta		**n/a**	19.648	n/a
95% CI (lower/upper)	alpha	**n/a**	0.2835 / 1.629	n/a
	beta	**n/a**	-22.313 / 61.609	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.14-2: CA's FR02 T1 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the SFO fit provided a very good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.14-3** and the graphical outputs in Figure CA.B.**8.1.2.3.14-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.14-4**.

Table CA.B.8.1.2.3.14-3: Summary of FR02 T1 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Very good**	Very good
DT_{50} (days)		**23.1**	21.3
DT_{90} (days)		**76.7**	79.6
DT_{50} (days) - Slow phase		**n/a**	31.5
χ^2 error (%)		**2.73**	2.32
k ($days^{-1}$)	k or k_1	**0.03001**	0.0465
	k_2	**n/a**	0.02204
P value	k or k_1	**7.58E-10**	0.325
	k_2	**n/a**	0.2757
g		**n/a**	0.4917

Selected model shown in bold

Figure CA.B.8.1.2.3.14-3: Applicant's FR02 T1 parent-only modelling kinetic fits

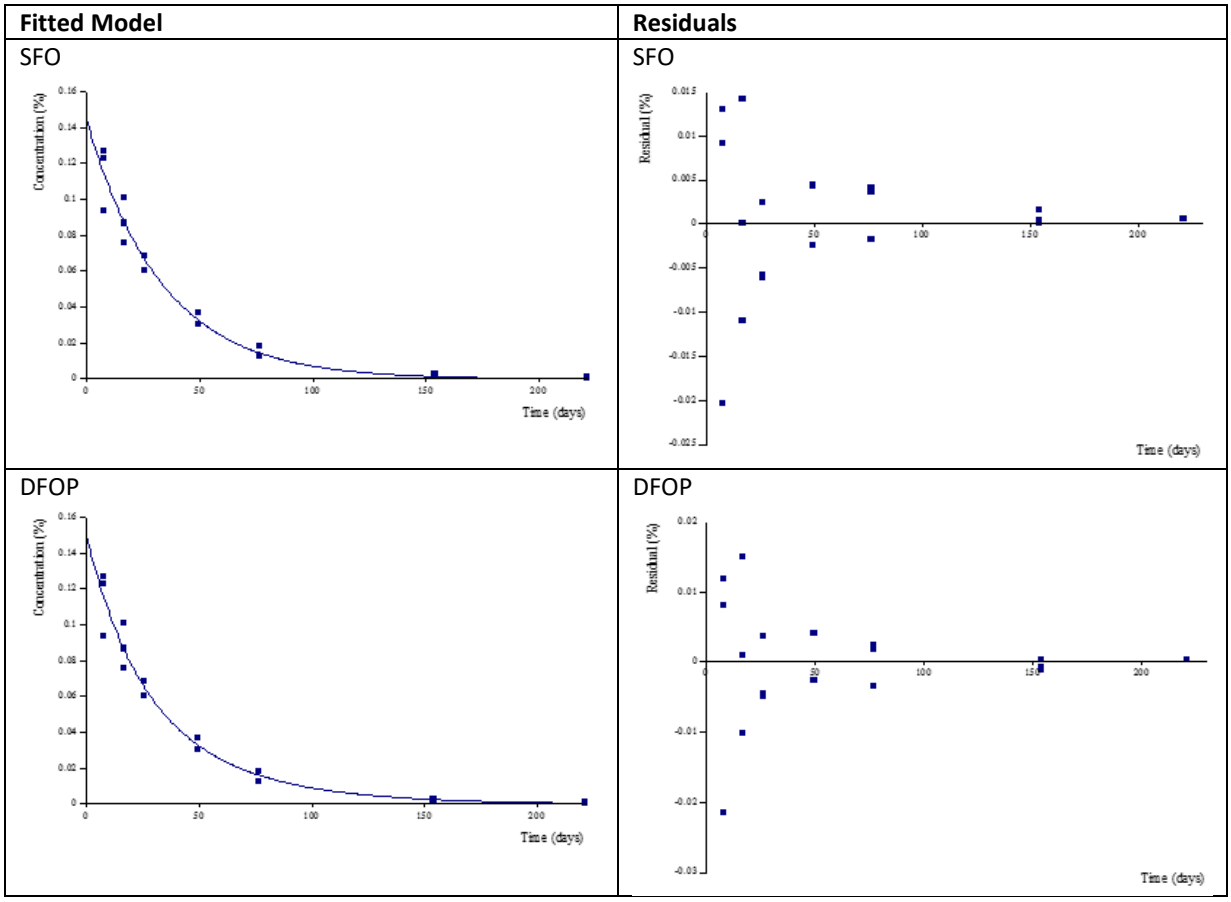


Table CA.B.8.1.2.3.14-4: Summary of FR02 T1 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	57.8	192
Persistence endpoints			
Bixlozone	SFO	58.9	196
Modelling endpoints			
Bixlozone	SFO	23.1	76.7

CA.B.8.1.2.3.15. FR02 T2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA is able to replicate the applicant's SFO results and so considers them appropriate for further consideration.

The CA notes only one residue of 2,4-DBA was detected in the trial (at 30 DALA). Therefore, no kinetic evaluation can be undertaken and so the CA accepts the applicant's parent-only evaluation.

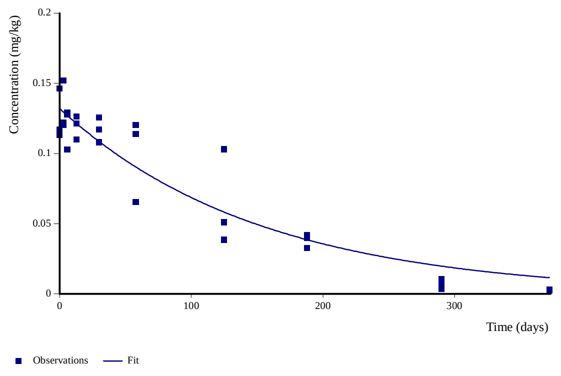
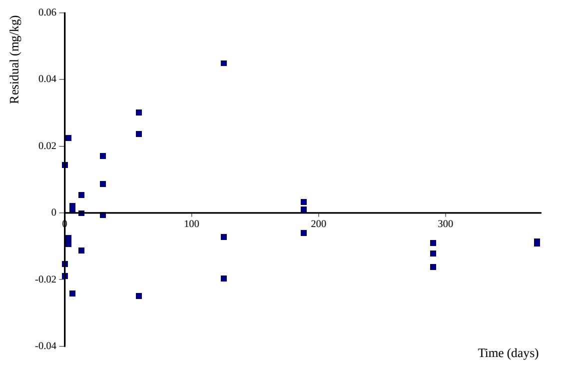
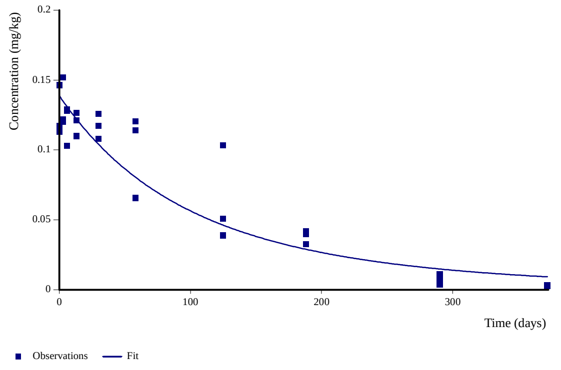
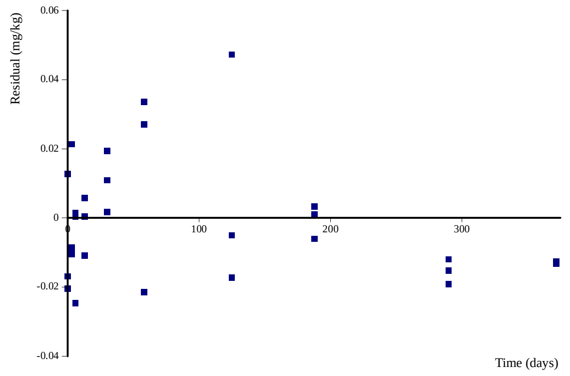
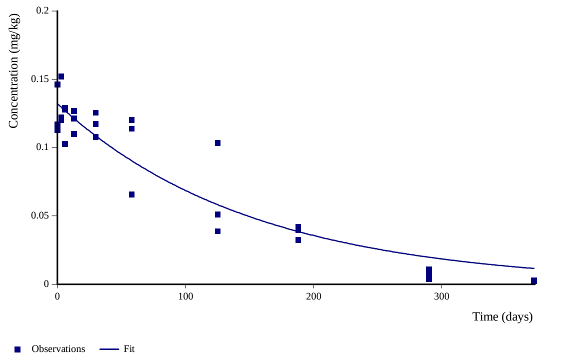
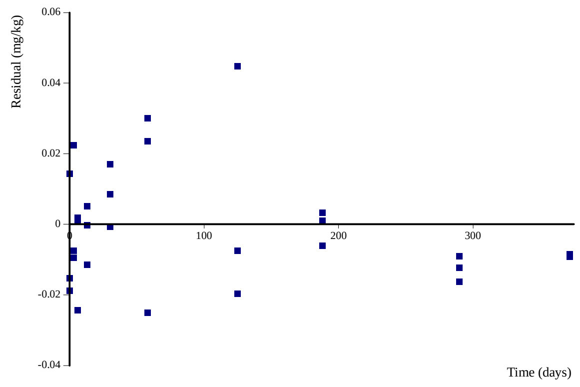
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.15-1** and the graphical outputs in Figure CA.B.**8.1.2.3.15-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.15-3**.

Table CA.B.8.1.2.3.15-1: Summary of FR02 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Good**	Good	Good	Good
DT_{50} (days)		**106**	75.2	106	106
DT_{90} (days)		**352**	301	352	352
DT_{90}/3.32 (days)		**n/a**	90.7	106	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	106	n/a
χ^2 error (%)		**7.12**	9.65	7.87	7.12
k ($days^{-1}$)	k or k_1	**0.006544**	n/a	0.173	0.006544
	k_2	**n/a**	n/a	0.006544	n/a
P value	k or k_1	**2.15E-9**	n/a	n.d.	2.15E-9
	k_2	**n/a**	n/a	6.06E-10	n/a
g		**n/a**	n/a	1.14E-7	n/a
alpha		**n/a**	4.509	n/a	n/a
beta		**n/a**	452.4	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	n.d.	n/a	n/a
	beta	**n/a**	n.d.	n/a	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.15-1: <u>FR02 T2 applicant's parent kinetic fits</u>

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided a good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.15-2** and the graphical outputs in Figure CA.B.**8.1.2.3.15-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.15-3**.

Table CA.B.8.1.2.3.15-2: Summary of FR02 T2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good
DT_{50} (days)		**47.9**	47.9
DT_{90} (days)		**159**	159
DT_{50} (days) - Slow phase		**n/a**	48.0
χ^2 error (%)		**6.45**	7.16
k ($days^{-1}$)	k or k_1	**0.01446**	0.01758
	k_2	**n/a**	0.01445
P value	k or k_1	**5.98E-9**	0.4979
	k_2	**n/a**	0.1821
g		**n/a**	0.0074

Selected model shown in bold

Figure CA.B.8.1.2.3.15-2: Applicant's FR02 T2 parent-only modelling kinetic fits

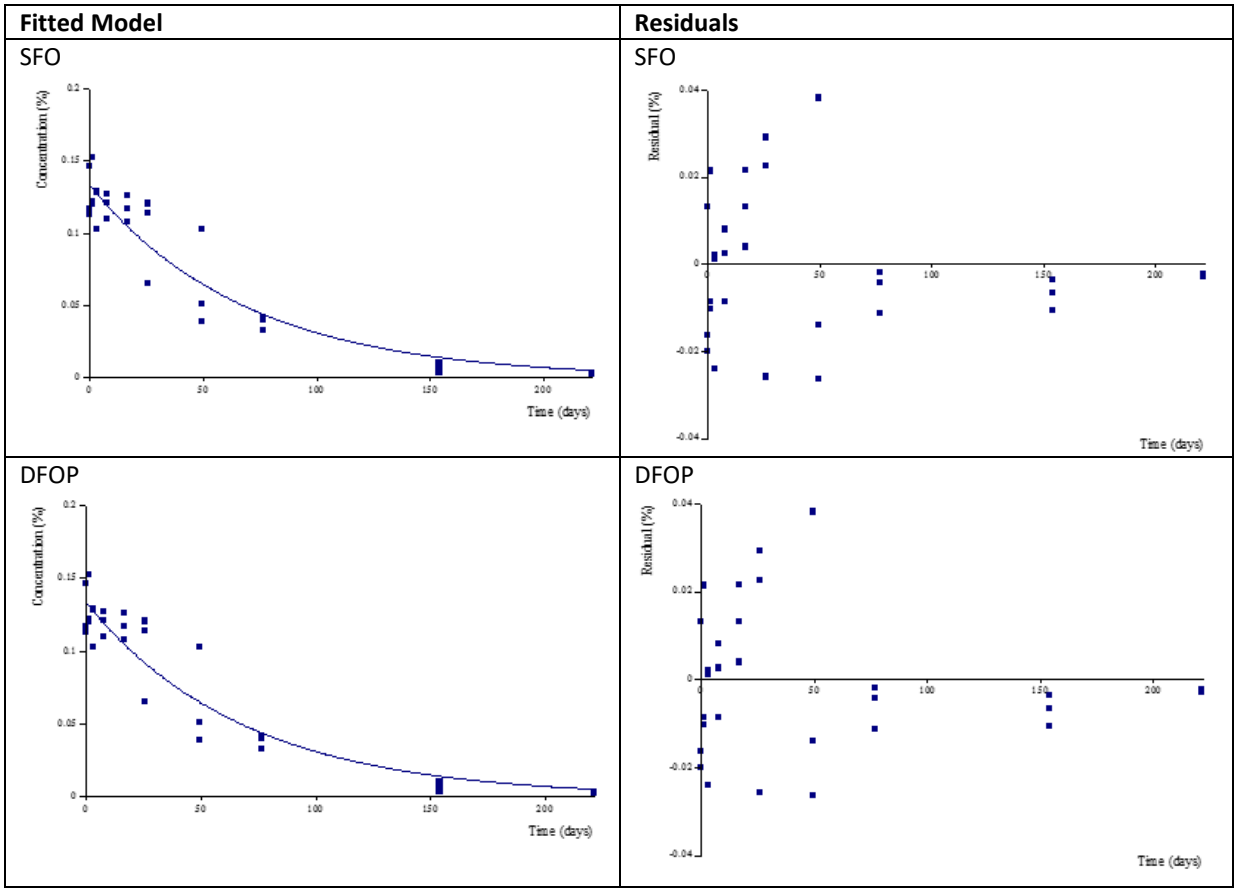


Table CA.B.8.1.2.3.15-3: Summary of FR02 T2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	106	352
Modelling endpoints			
Bixlozone	SFO	47.9	159

CA.B.8.1.2.3.16. FR02 T3

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA is able to replicate the applicant's SFO results and so considers them appropriate for further consideration.

The CA notes residues of 2,4-DBA were detected in the trial at 13 DALA and 30 DALA. However, as this only covers two time points and all the residues were <LOQ, the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

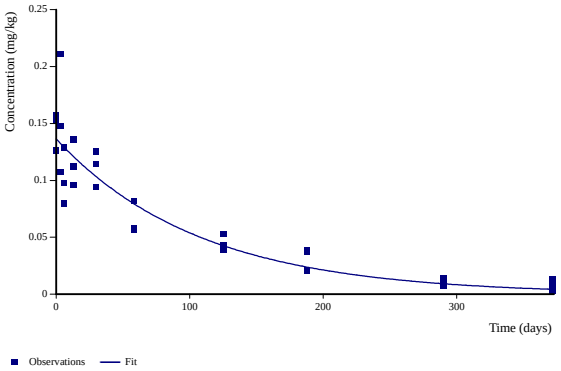
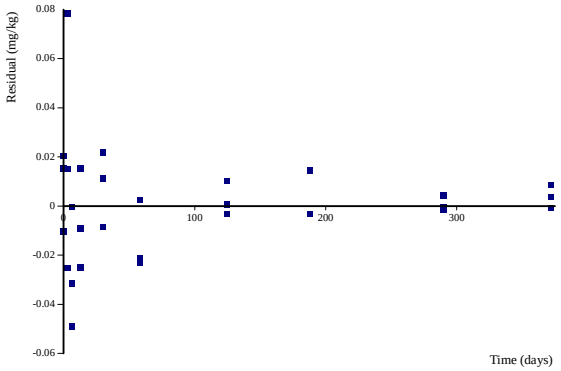
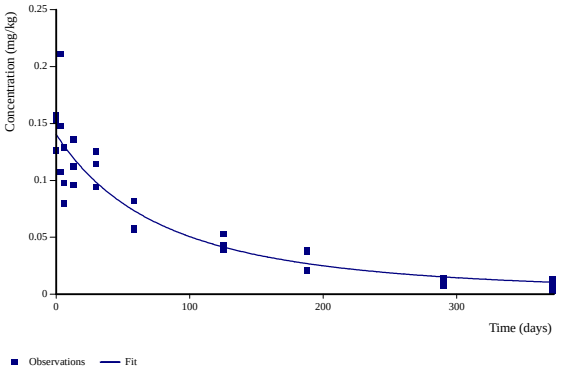
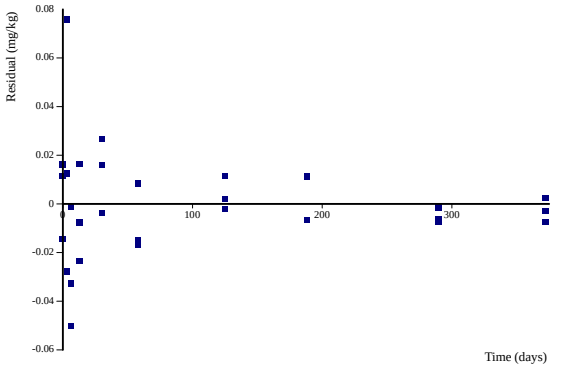
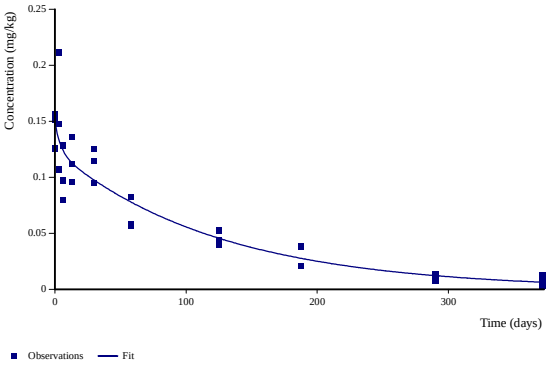
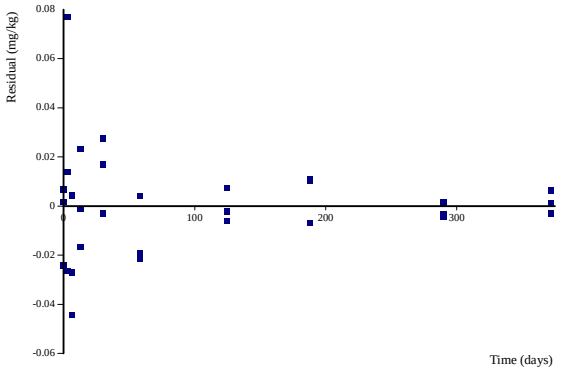
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.16-1** and the graphical outputs in Figure CA.B.**8.1.2.3.16-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.16-4**.

Table CA.B.8.1.2.3.16-1: Summary of FR02 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Very good**	Very good	Very good	Very good
DT_{50} (days)		**74.7**	63.1	62.8	74.7
DT_{90} (days)		**248**	308	264	248
DT_{90}/3.32 (days)		**n/a**	92.7	79.5	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	86.8	n/a
χ^2 error (%)		**13.3**	13.3	13.2	13.3
k ($days^{-1}$)	k or k_1	**0.009279**	n/a	0.2325	0.009278
	k_2	**n/a**	n/a	0.007985	n/a
P value	k or k_1	**1.10E-6**	n/a	0.2686	1.10E-6
	k_2	**n/a**	n/a	1.16E-4	n/a
g		**n/a**	n/a	0.1747	n/a
alpha		**n/a**	2.316	n/a	n/a
beta		**n/a**	180.8	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	-2.761 / 7.392	n/a	n/a
	beta	**n/a**	-339.4 / 701	n/a	n/a

Best-fit model shown in bold
nd – Not determined: parameter could not be calculated by the CAKE model.

Figure CA.B.8.1.2.3.16-1: FR02 T3 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Persistence endpoints

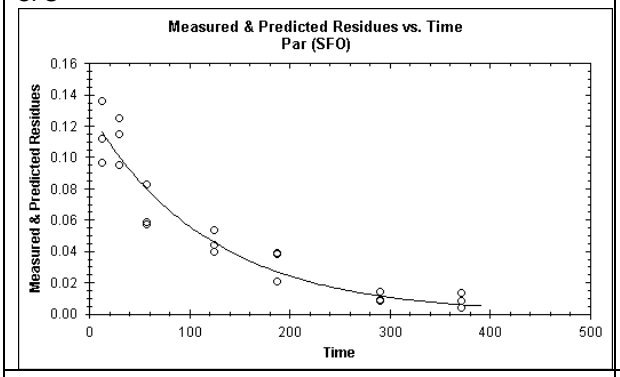
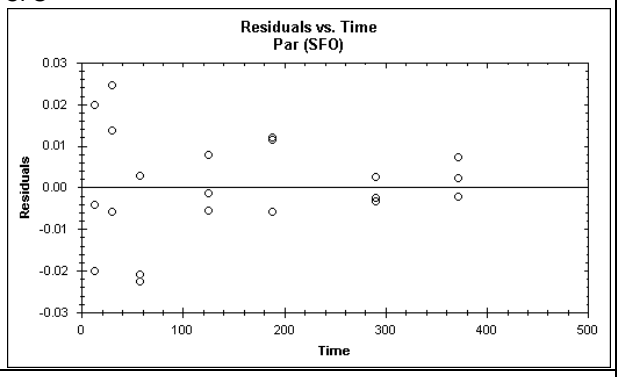
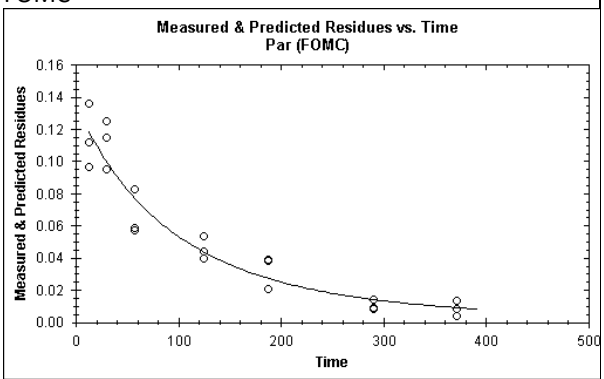
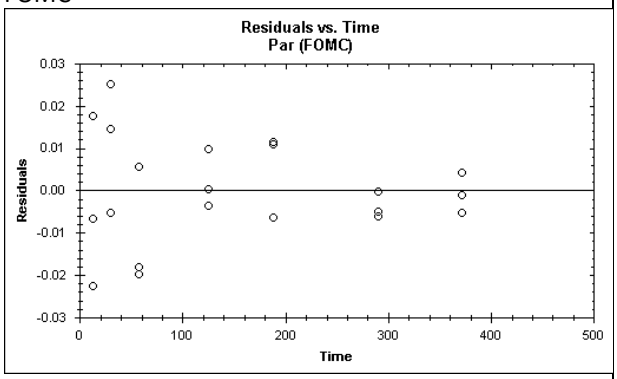
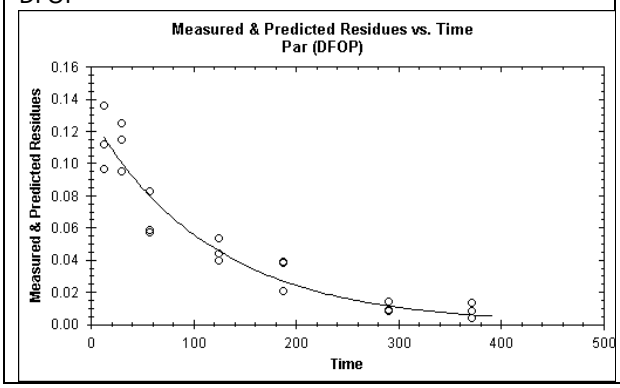
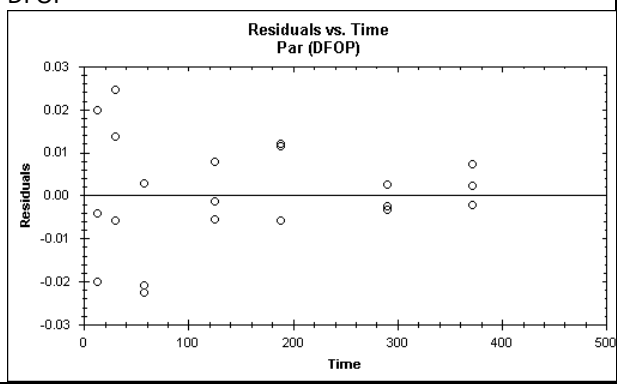
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a very good visual fit and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.16-2** and Figure CA.B.**8.1.2.3.16-2**.

Table CA.B.8.1.2.3.16-2: Summary of FR02 T3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Very good**	Very good	Very good
DT_{50} (days)		**82.1**	71.4	82.1
DT_{90} (days)		**273**	294	273
DT_{90}/3.32 (days)		**n/a**	88.7	82.1
DT_{50} (days) - Slow phase		**n/a**	n/a	82.1
χ^2 error (%)		**10.1**	10.3	12.0
k ($days^{-1}$)	k or k_1	**0.008444**	n/a	0.3137
	k_2	**n/a**	n/a	8.444E-3
P value	k or k_1	**3.76E-8**	n/a	<2E-16
	k_2	**n/a**	n/a	1.74E-6
g		**n/a**	n/a	0.0000
alpha		**n/a**	3.9661	n/a
beta		**n/a**	374.00	n/a
95% CI (lower/upper)	alpha	**n/a**	-6.1542 / 14.086	n/a
	beta	**n/a**	-800.68 / 1548.7	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.16-2: CA's FR02 T3 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the SFO fit provided a good visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.16-3** and the graphical outputs in Figure CA.B.**8.1.2.3.16-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.16-4**.

Table CA.B.8.1.2.3.16-3: Summary of FR02 T3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good
DT_{50} (days)		**33.0**	28.3
DT_{90} (days)		**110**	131
DT_{50} (days) - Slow phase		**n/a**	113
χ^2 error (%)		**12.4**	13.3
k ($days^{-1}$)	k or k_1	**0.02102**	0.0307
	k_2	**n/a**	0.00616
P value	k or k_1	**4.19E-7**	0.1149
	k_2	**n/a**	0.3599
g		**n/a**	0.8089

Selected model shown in bold

Figure CA.B.8.1.2.3.16-3: Applicant's FR02 T3 parent-only modelling kinetic fits

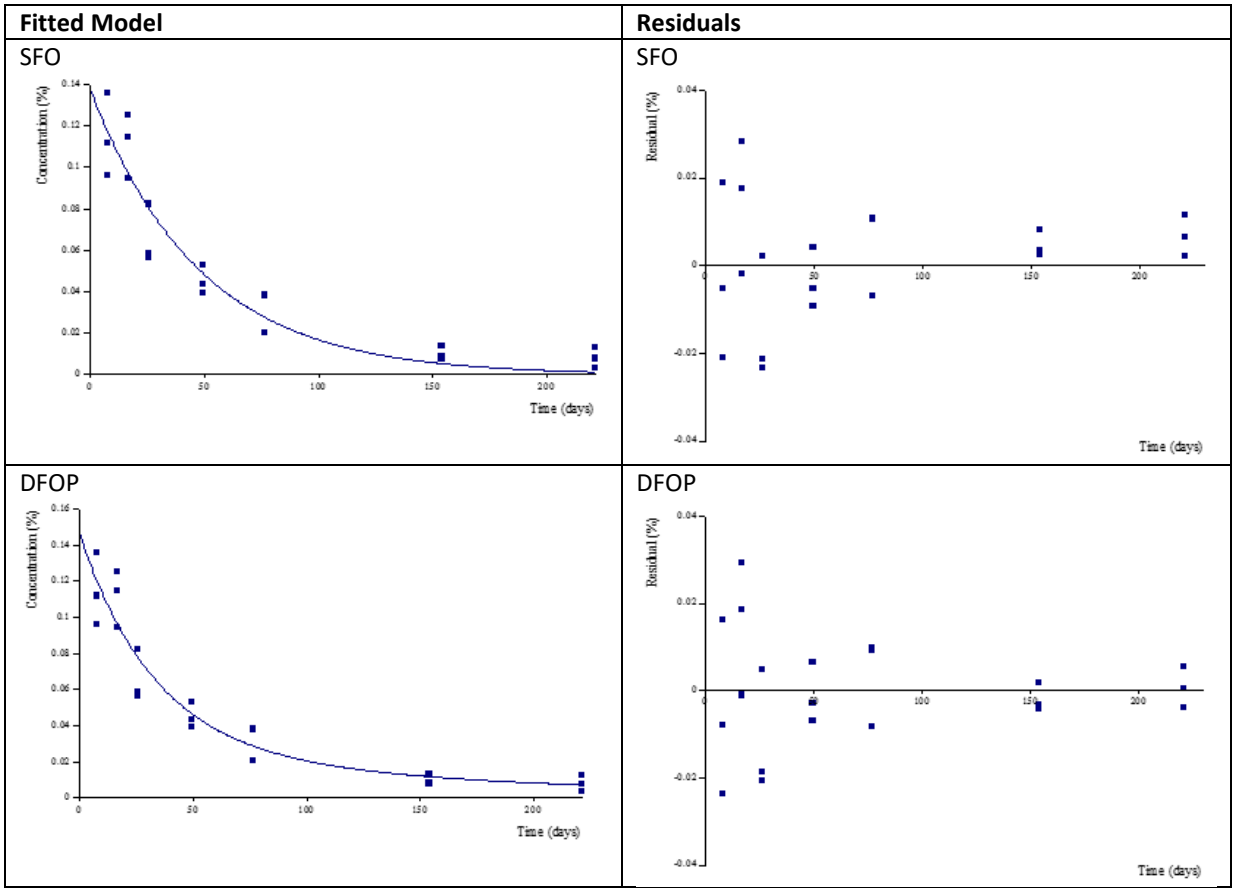


Table CA.B.8.1.2.3.16-4: Summary of FR02 T3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	74.7	248
Persistence endpoints			
Bixlozone	SFO	82.1	273
Modelling endpoints			
Bixlozone	SFO	33.0	110

CA.B.8.1.2.3.17. FR02 T4

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared (no metabolites were detected in this trial). The applicant concluded SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA notes the biphasic fits were not visually or statistically better than SFO and so the CA concurs with the applicant that SFO provides the best-fit model. The CA obtained a very similar SFO DT50 value to the applicant (difference <1 day); therefore, the applicant's SFO results are accepted on this occasion.

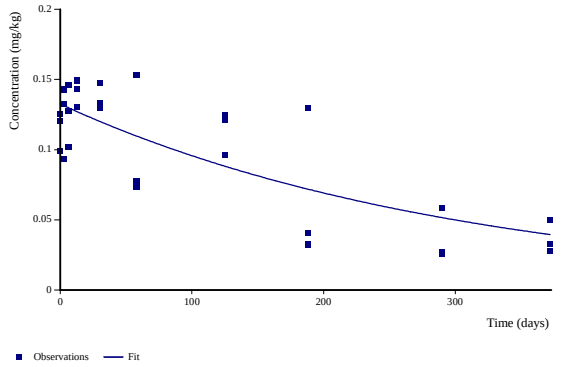
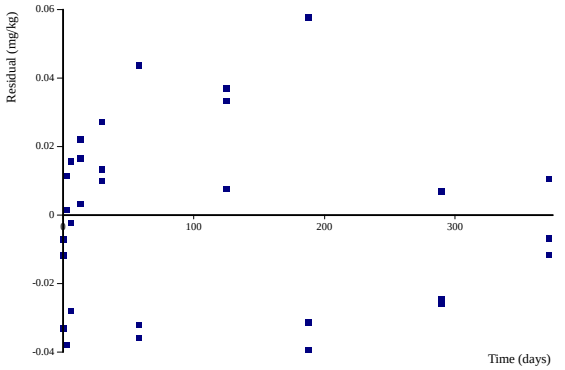
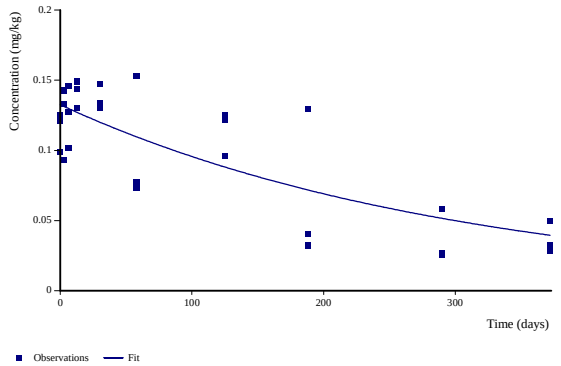
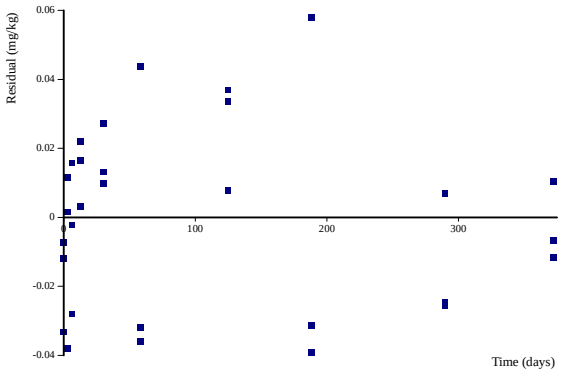
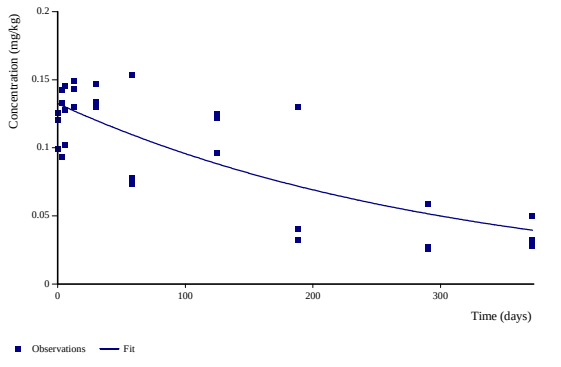
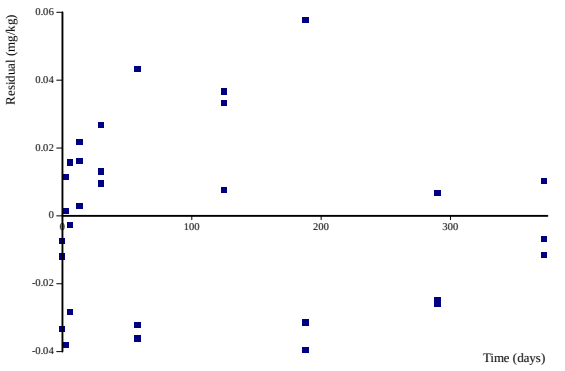
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.17-1** and the graphical outputs in Figure CA.B.**8.1.2.3.17-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.17-3**.

Table CA.B.8.1.2.3.17-1: Summary of FR02 T1 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP	SFO
Visual fit		**Acceptable**	Acceptable	Acceptable	Very good
DT_{50} (days)		**213**	213	213	214
DT_{90} (days)		**708**	712	708	709
DT_{90}/3.32 (days)		**n/a**	214	213	n/a
DT_{50} (days) - Slow phase		**n/a**	n/a	213	n/a
χ^2 error (%)		**10.9**	11.5	12.1	10.9
k ($days^{-1}$)	k or k_1	**0.003251**	n/a	0.1216	0.003247
	k_2	**n/a**	n/a	0.003254	n/a
P value	k or k_1	**9.10E-6**	n/a	n.d.	9.08E-6
	k_2	**n/a**	n/a	2.98E-6	n/a
g		**n/a**	n/a	2.39E-6	n/a
alpha		**n/a**	137.2	n/a	n/a
beta		**n/a**	4.2E-4	n/a	n/a
95% CI (lower/upper)	alpha	**n/a**	-1927 / 2.20E+003	n/a	n/a
	beta	**n/a**	-5.78E+5 / 6.62E+5	n/a	n/a

Best-fit model shown in bold
nd – Not determined: parameter could not be calculated by the CAKE model.

Figure CA.B.8.1.2.3.17-1: FR02 T4 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.17-2** and the graphical outputs in Figure CA.B.**8.1.2.3.17-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.17-3**.

Table CA.B.8.1.2.3.17-2: Summary of FR02 T4 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable**	Acceptable
DT_{50} (days)		**106**	105
DT_{90} (days)		**351**	350
DT_{50} (days) - Slow phase		**n/a**	106
χ^2 error (%)		**10.6**	11.7
k ($days^{-1}$)	k or k_1	**6.56E-3**	7.21E-3
	k_2	**n/a**	6.56E-3
P value	k or k_1	**1.93E-5**	0.5000
	k_2	**n/a**	0.4993
g		**n/a**	0.0322

Selected model shown in bold

Figure CA.B.8.1.2.3.17-2: Applicant's FR02 T4 parent-only modelling kinetic fits

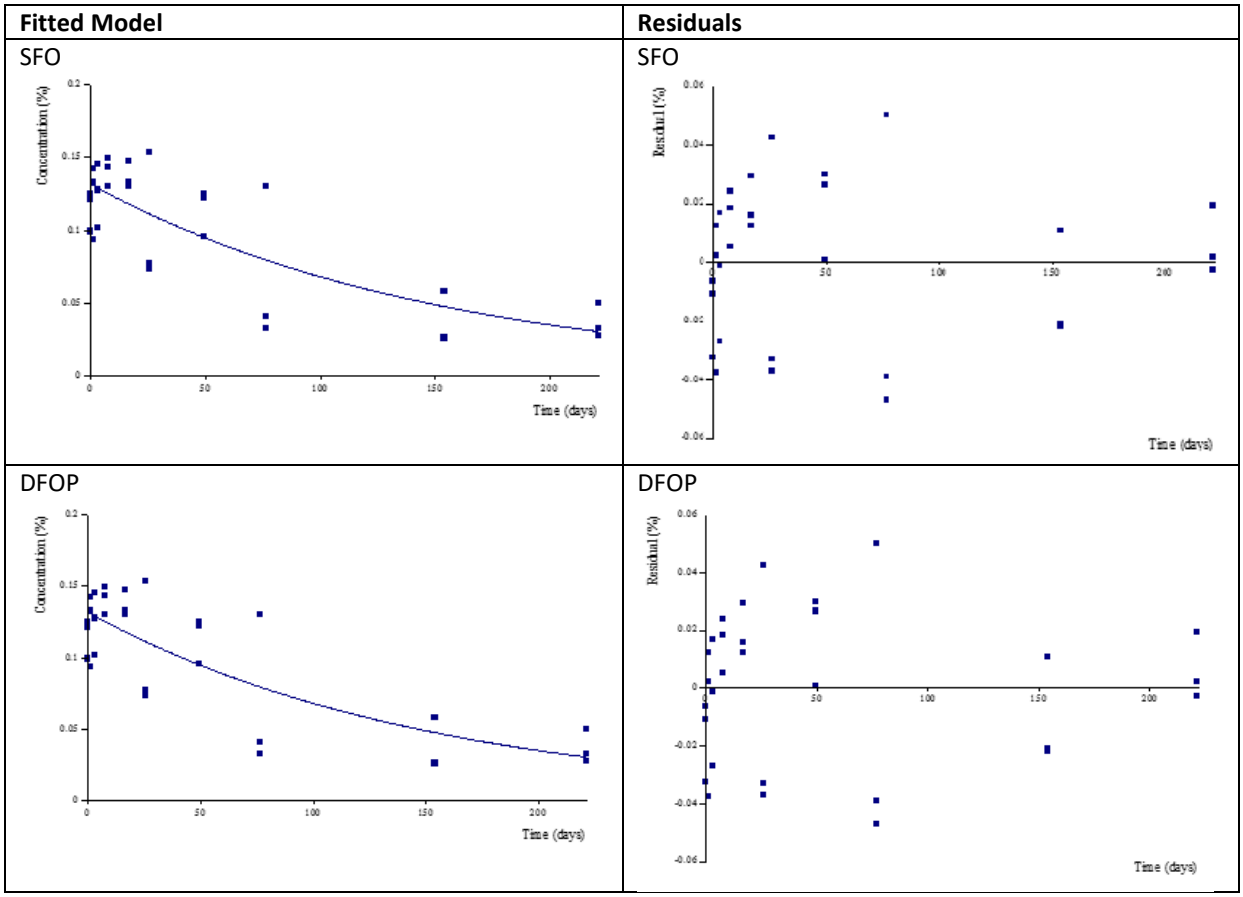


Table CA.B.8.1.2.3.17-3: Summary of FR02 T4 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	SFO	213	708
Modelling endpoints			
Bixlozone	SFO	106	351

CA.B.8.1.2.3.18. GE02 Plot 2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded DFOP provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA agrees the biphasic visual fits were better than SFO. The CA also agrees DFOP provided the slightly better visual and statistical fit of the data. Therefore, the CA concurs with the applicant's conclusion of DFOP as the best-fit model; the CA is able to replicate the applicant's DFOP results.

The CA notes residues of 2,4-DBA were detected in the trial at 3 DALA, 28 DALA, 177 DALA and 272 DALA. However, as the residues were sporadic in nature with the largest residues in the 272 DALA samples (and so no clear decline phase can be observed), the CA does not consider this data robust enough to perform kinetic analysis. Therefore, the CA accepts the applicant's parent-only evaluation.

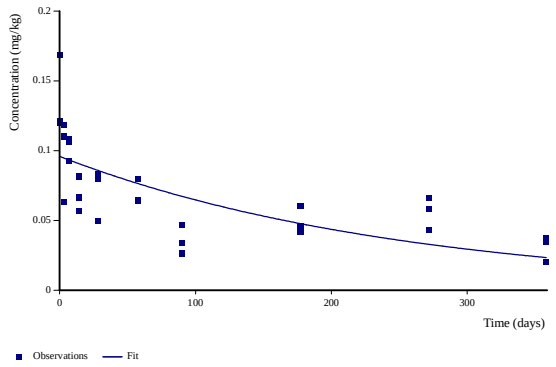
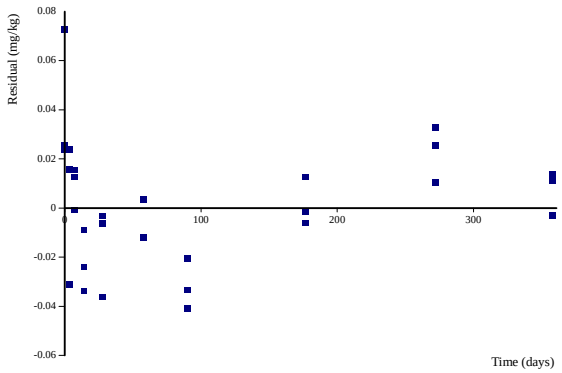
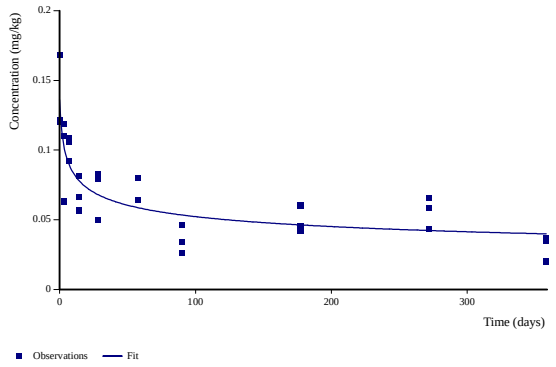
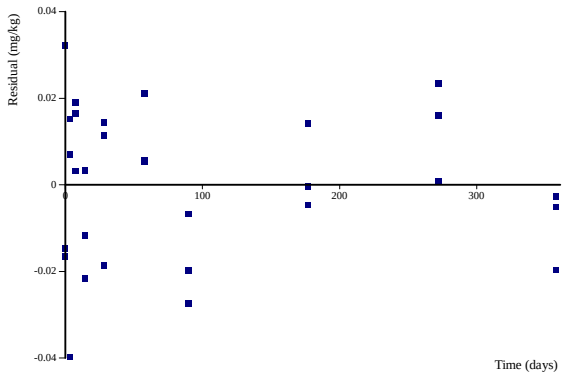
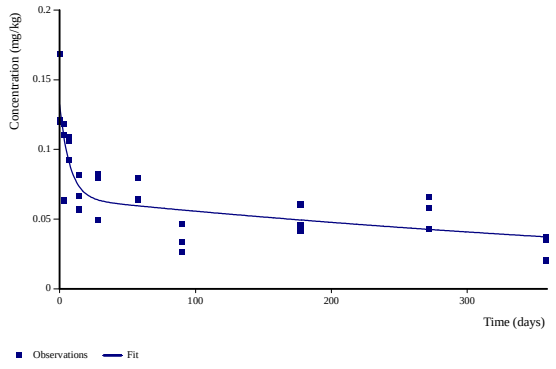
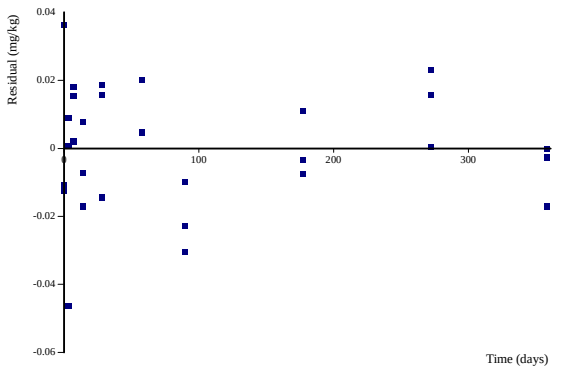
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.18-1** and the graphical outputs in Figure CA.B.**8.1.2.3.18-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.18-3**.

Table CA.B.8.1.2.3.18-1: Summary of GE02 P2 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	**DFOP**	DFOP
Visual fit		Acceptable	Good	**Good**	Good
DT_{50} (days)		176	28.1	**22.6**	22.6
DT_{90} (days)		584	>10000	**1.03E+3**	>1000
DT_{90}/3.32 (days)		n/a	>10000	**310**	301
DT_{50} (days) - Slow phase		n/a	n/a	**446**	446
χ^2 error (%)		22.8	11.9	**13.3**	13.3
k ($days^{-1}$)	k or k_1	0.003941	n/a	**0.1339**	0.1339
	k_2	n/a	n/a	**0.001554**	0.001554
P value	k or k_1	2.46E-4	n/a	**0.01733**	0.01733
	k_2	n/a	n/a	**0.03576**	0.03576
g		n/a	n/a	**0.5077**	0.5077
alpha		n/a	0.2131	**n/a**	n/a
beta		n/a	1.13	**n/a**	n/a
95% CI (lower/upper)	alpha	n/a	0.1192 / 0.307	**n/a**	n/a
	beta	n/a	-0.9552 / 3.216	**n/a**	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.18-1: GE02 P2 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the SFO fit provided an unacceptable visual fit of the data, whereas, the DFOP visual and statistical fits were acceptable. Therefore, the applicant considered the DFOP endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant. However, the CA disagrees with the applicant's visual fit assessment of the SFO fit. Whilst the DFOP visual fit is superior to the SFO fit, the CA considers the SFO fit acceptable for determining modelling endpoints because SFO adequately fits the latter datapoints. Furthermore, the CA considers the large χ^2 (20.6%) to be due to the systematic variation of the data; for example, the residues reported for the last three time points are greater than, or equal to, the 76.4 normalised time point residues. Therefore, the CA considers the SFO model good enough for determining the modelling endpoints. As the CA obtained very similar SFO endpoints to the applicant, the applicant's SFO fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.18-2** and the graphical outputs in Figure CA.B.**8.1.2.3.18-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.18-3**.

Table CA.B.8.1.2.3.18-2: Summary of GE02 P2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Acceptable***	Acceptable
DT_{50} (days)		**103**	23.4
DT_{90} (days)		**343**	487
DT_{50} (days) - Slow phase		**n/a**	200
χ^2 error (%)		**20.6**	12.9
k ($days^{-1}$)	k or k_1	**6.71E-3**	0.3057
	k_2	**n/a**	3.47E-3
P value	k or k_1	**3.19E-3**	0.0307
	k_2	**n/a**	0.0106
g		**n/a**	0.4581

Selected model shown in bold

* The applicant considered the visual fit unacceptable

Figure CA.B.8.1.2.3.18-2: Applicant's GE02 P2 parent-only modelling kinetic fits

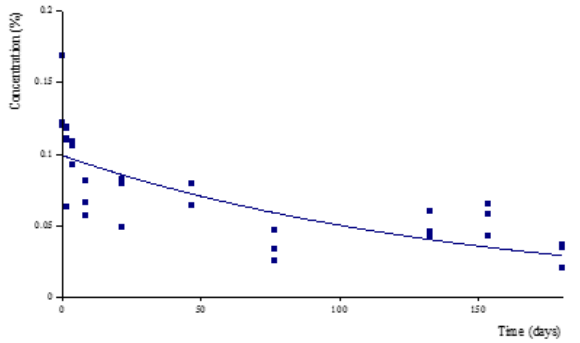
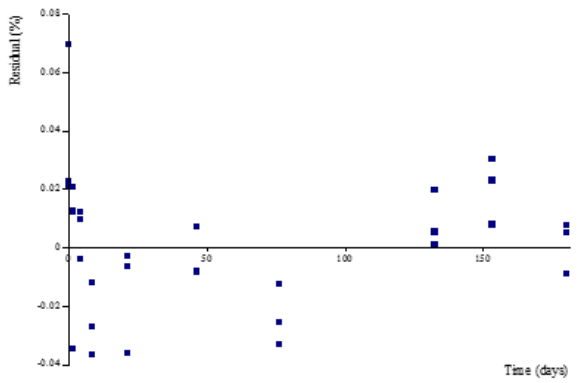
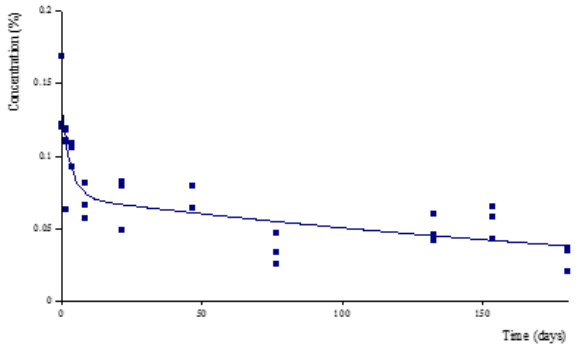
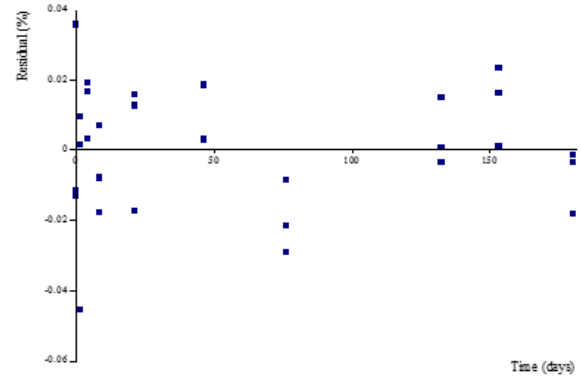
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.18-3: Summary of GE02 P2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	22.6	>1000
Modelling endpoints			
Bixlozone	SFO	103	343

CA.B.8.1.2.3.19. GE02 Plot 3

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant concluded DFOP provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA agrees the biphasic visual fits were better than SFO. The CA also agrees DFOP provided the slightly better visual and statistical fit of the data. Therefore, the CA concurs with the applicant's conclusion of DFOP as the best-fit model; the CA is able to replicate the applicant's DFOP DT50 value and the applicant's results are accepted.

The CA notes residues of 2,4-DBA were detected in the trial at 3 DALA, 7 DALA, 28 DALA, 177 DALA and 272 DALA. However, as the residues were sporadic in nature with the largest residues in the 272 DALA samples (and so no clear decline phase can be observed), the CA does not consider this data robust enough to perform kinetic analysis. The CA also notes that the peak occurrence of 2,4-DBA (272 DALA sample) was 99.53% on a molar basis. Therefore, a formation fraction of 1 is appropriate and so kinetic fittings to derive a formation fraction is not required.

The CA also notes that a single residue of 3-OH propenamide was also detected in the study, however, as it is a single residue, no kinetic analysis can be performed. Therefore, the CA accepts the applicant's parent-only evaluation.

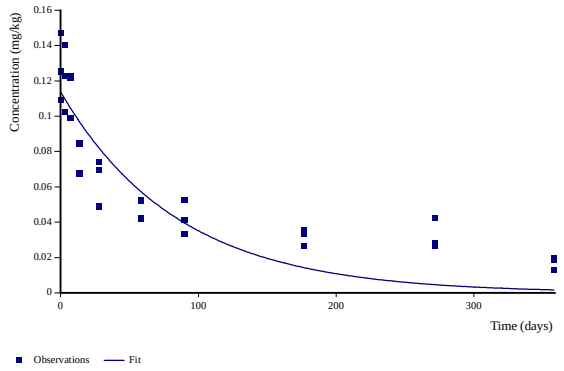
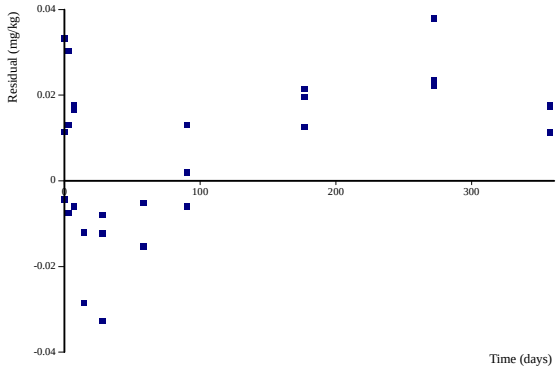
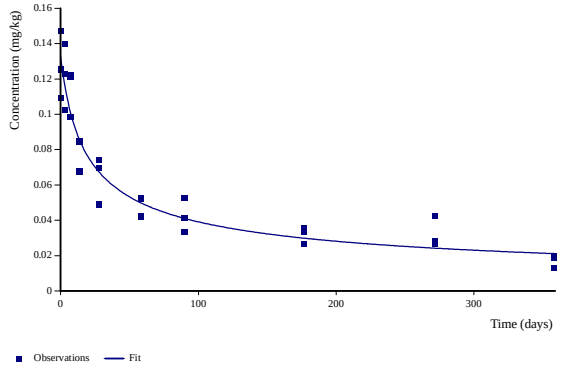
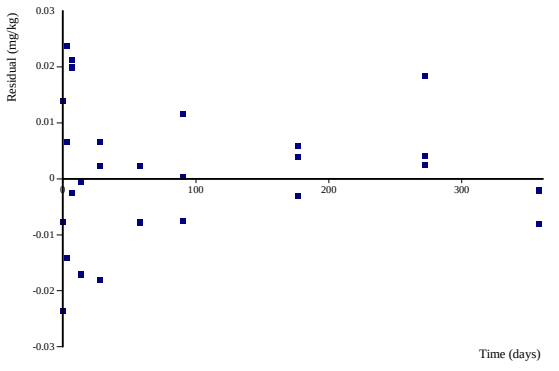
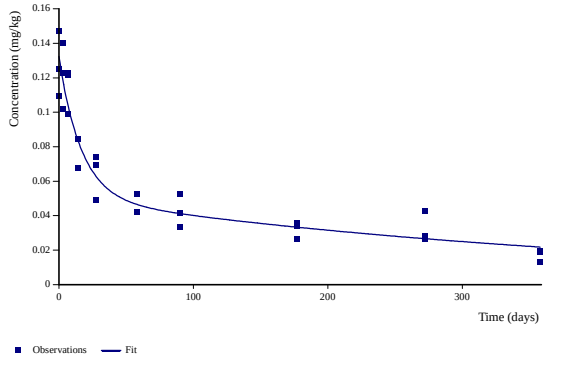
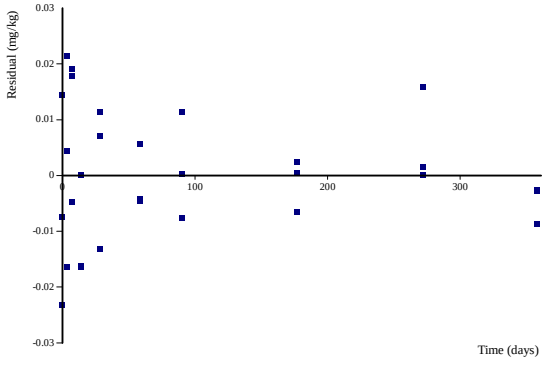
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.19-1** and the graphical outputs in Figure CA.B.**8.1.2.3.19-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.19-4**.

Table CA.B.8.1.2.3.19-1: Summary of GE02 P3 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	**DFOP**	DFOP
Visual fit		Unacceptable	Very good	**Very good**	Very good
DT_{50} (days)		59.1	28.9	**24.7**	24.7
DT_{90} (days)		196	908	**571**	573
DT_{90}/3.32 (days)		n/a	273	**172**	173
DT_{50} (days) - Slow phase		n/a	n/a	**296**	294
χ^2 error (%)		19.9	8.76	**7.69**	7.77
k ($days^{-1}$)	k or k_1	0.01174	n/a	**0.06019**	0.06104
	k_2	n/a	n/a	**0.002343**	0.002354
P value	k or k_1	3.91E-6	n/a	**6.85E-4**	7.54E-4
	k_2	n/a	n/a	**0.01648**	0.01556
g		n/a	n/a	**0.6188**	0.6144
alpha		n/a	0.5085	**n/a**	n/a
beta		n/a	9.919	**n/a**	n/a
95% CI (lower/upper)	alpha	n/a	0.3026 / 0.714	**n/a**	n/a
	beta	n/a	-0.1724 / 20.01	**n/a**	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.19-1: GE02 P3 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO 	SFO 
FOMC 	FOMC 
DFOP 	DFOP 

Persistence endpoints

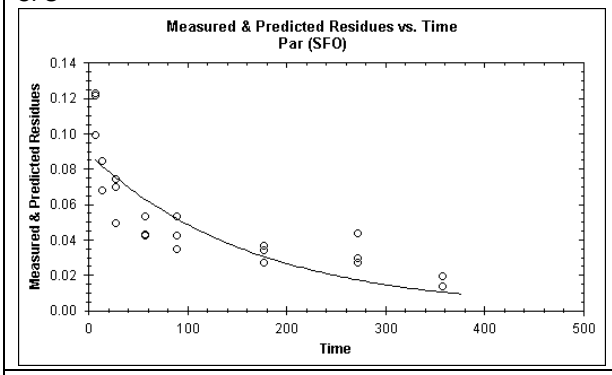
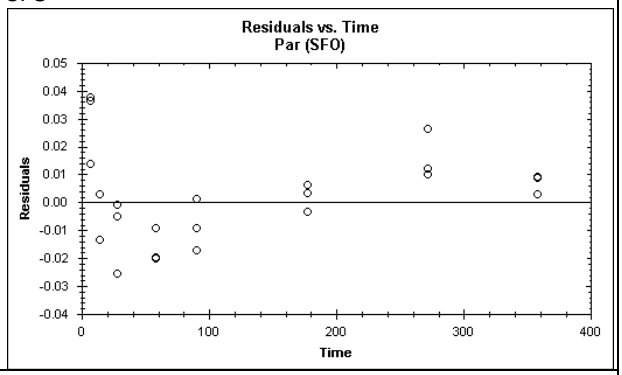
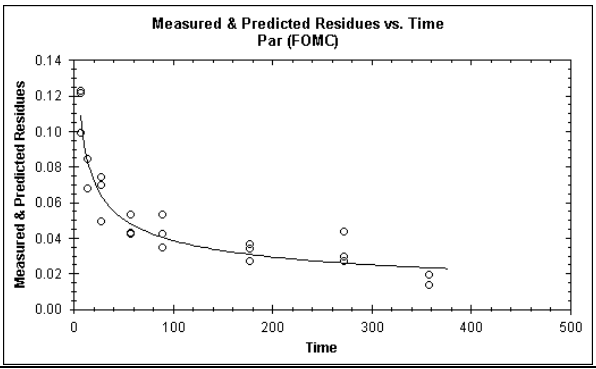
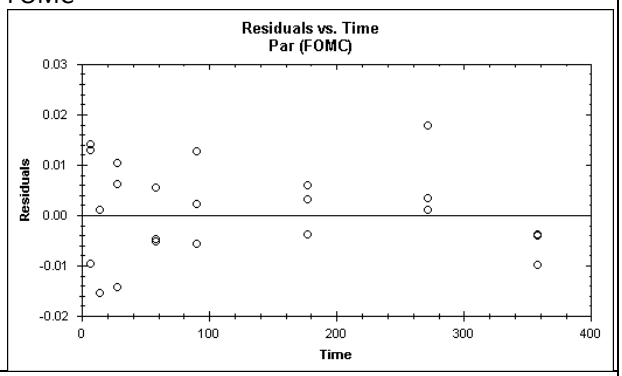
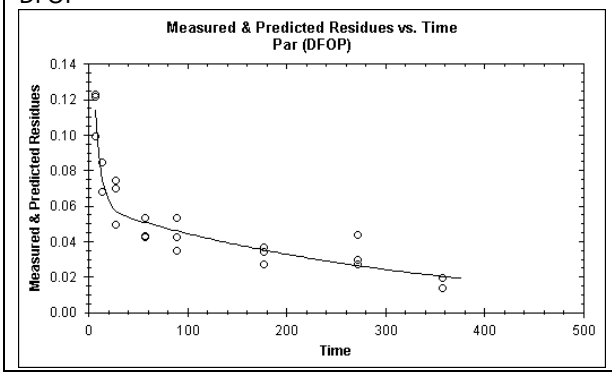
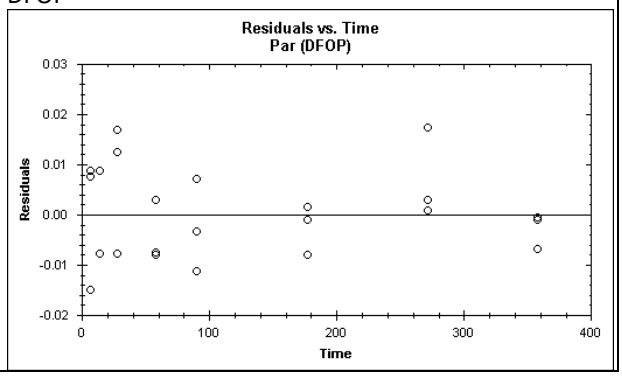
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided an acceptable visual fit and statistical fit of the data, however, the biphasic fits were visually and/or statistically better. The CA considers the DFOP model to provide the best visual and statistical fit of the data and so the CA considers the DFOP fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.19-2** and Figure CA.B.**8.1.2.3.19-2**.

Table CA.B.8.1.2.3.19-2: Summary of GE02 P3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	**DFOP**
Visual fit		Acceptable	Very good	**Very good**
DT_{50} (days)		114	2.33	**7.03**
DT_{90} (days)		380	157	**318**
DT_{90}/3.32 (days)		n/a	47.3	**95.5**
DT_{50} (days) - Slow phase		n/a	n/a	**226**
χ^2 error (%)		21.8	8.74	**7.37**
k ($days^{-1}$)	k or k_1	0.00607	n/a	**0.1588**
	k_2	n/a	n/a	**3.068E-3**
P value	k or k_1	3.09E-5	n/a	**0.0048**
	k_2	n/a	n/a	**0.0001**
g		n/a	n/a	**0.735**
alpha		n/a	0.4003	**n/a**
beta		n/a	0.5000	**n/a**
95% CI (lower/upper)	alpha	n/a	0.245 / 0.556	**n/a**
	beta	n/a	-7.730 / 8.730	**n/a**

Best-fit model shown in bold

Figure CA.B.8.1.2.3.19-2: CA's GE02 P3 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the DFOP fit provided a better visual and statistical fit than the SFO fit and, therefore, considered the DFOP endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant. However, the CA considers the SFO visual and statistical fit good enough to obtain modelling endpoints. As the CA obtained very similar SFO endpoints to the applicant, the applicant's SFO fit is accepted and considered further in the evaluation.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.19-3** and the graphical outputs in Figure CA.B.**8.1.2.3.19-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.19-4**.

Table CA.B.8.1.2.3.19-3: Summary of GE02 P3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good*
DT_{50} (days)		**72.6**	26.5
DT_{90} (days)		**241**	285
DT_{50} (days) - Slow phase		**n/a**	111
χ^2 error (%)		**18.9**	6.75
k ($days^{-1}$)	k or k_1	**9.55E-3**	0.3931
	k_2	**n/a**	6.22E-3
P value	k or k_1	**1.13E-6**	0.0292
	k_2	**n/a**	7.13E-6
g		**n/a**	0.4106

Selected model shown in bold

* The applicant notes, because the Mo value was outside the dataset in the initial fitting, the kinetics were calculated resetting the first time point to zero.

Figure CA.B.8.1.2.3.19-3: Applicant's GE02 P3 parent-only modelling kinetic fits

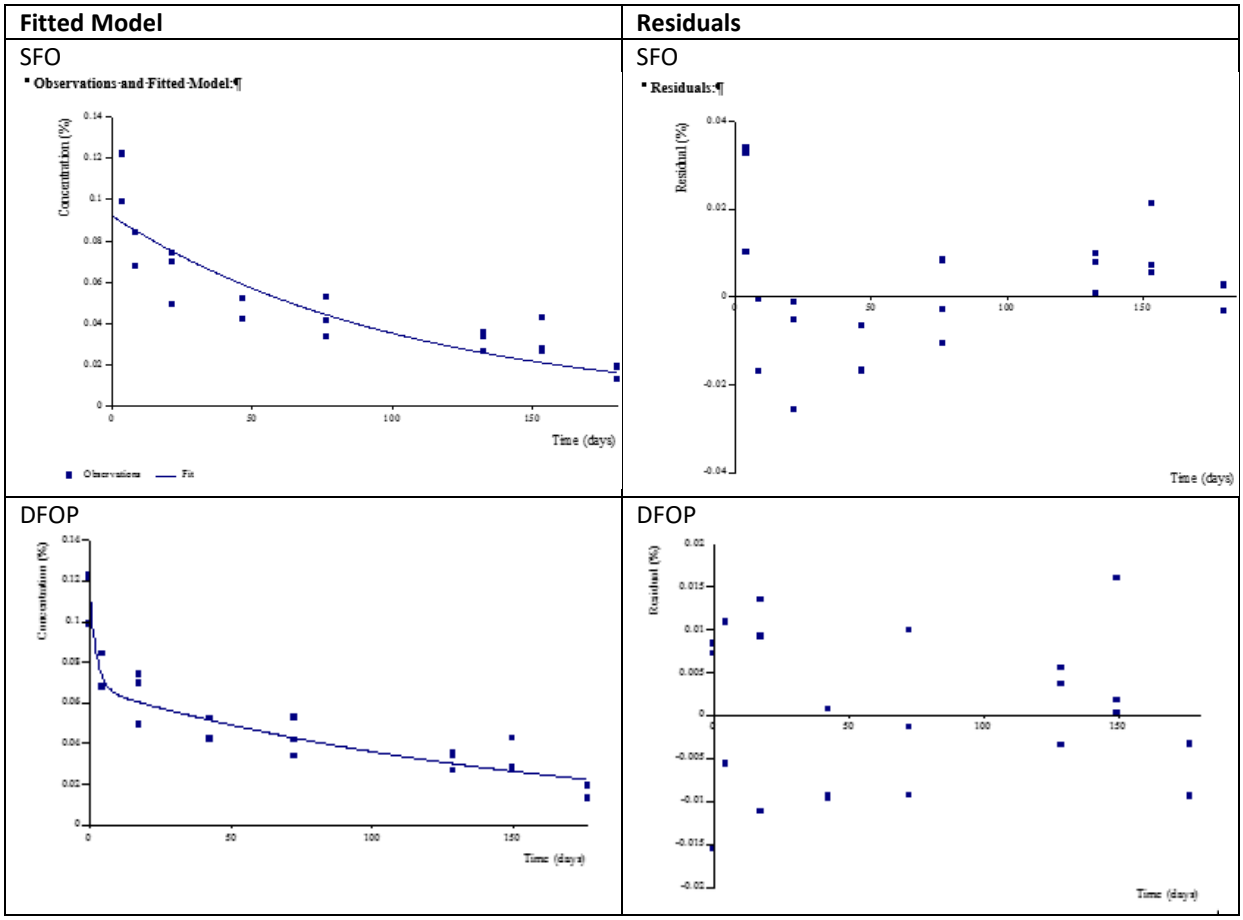


Table CA.B.8.1.2.3.19-4: Summary of GE02 P3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	24.7	571
Persistence endpoints			
Bixlozone	DFOP	7.03	318
Modelling endpoints			
Bixlozone	SFO	72.6	241

CA.B.8.1.2.3.20. UK01 Plot 2

Best-fit endpoints

In order to determine triggering, persistence and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared; no metabolites were detected in this trial. The applicant concluded DFOP provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA agrees the biphasic visual fits were better than SFO. The CA also agrees DFOP provided the slightly better visual and statistical fit of the data. Therefore, the CA concurs with the applicant's conclusion of DFOP as the best-fit model; the CA is able to replicate the applicant's DFOP results.

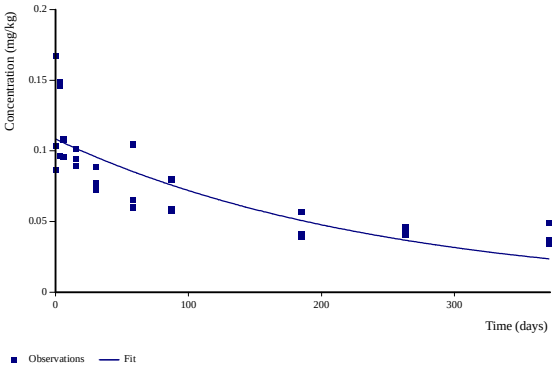
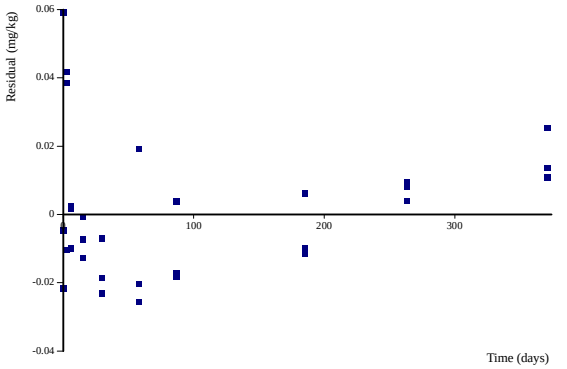
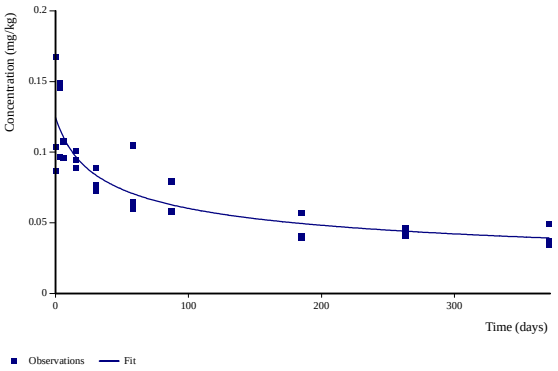
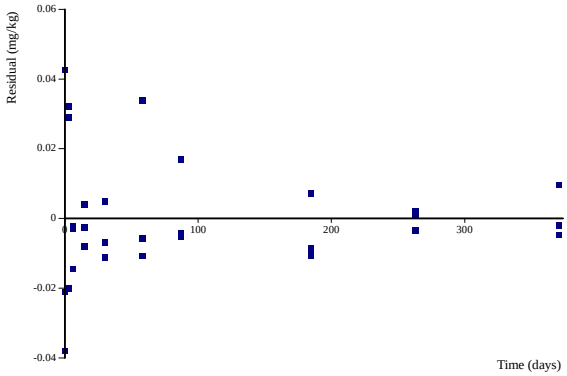
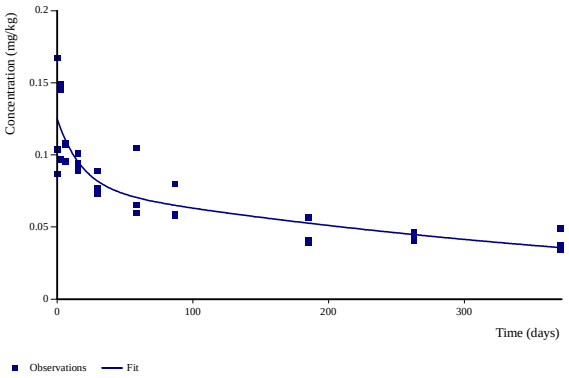
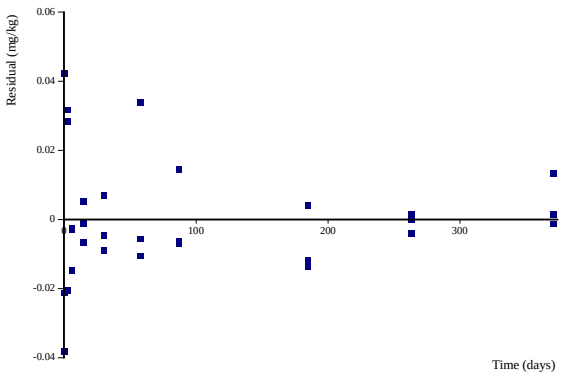
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.20-1** and the graphical outputs in Figure CA.B.**8.1.2.3.20-1**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.20-3**.

Table CA.B.8.1.2.3.20-1: Summary of UK01 P2 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	**DFOP**	DFOP
Visual fit		Good	Very good	**Very good**	Very good
DT_{50} (days)		169	89.5	**105**	105
DT_{90} (days)		562	>10,000	**873**	873
DT_{90}/3.32 (days)		n/a	3.05E+3	**263**	263
DT_{50} (days) - Slow phase		n/a	n/a	**331**	331
χ^2 error (%)		12.4	6.17	**6.74**	6.74
k ($days^{-1}$)	k or k_1	0.004099	n/a	**0.05586**	0.05593
	k_2	n/a	n/a	**0.002092**	0.002093
P value	k or k_1	4.27E-6	n/a	**0.0989**	0.0988
	k_2	n/a	n/a	**0.02083**	0.02065
g		n/a	n/a	**0.379**	0.379
alpha		n/a	0.3512	**n/a**	n/a
beta		n/a	14.44	**n/a**	n/a
95% CI (lower/upper)	alpha	n/a	0.09971 / 0.603	**n/a**	n/a
	beta	n/a	-13.43 / 42.3	**n/a**	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.20-1: UK01 P2 applicant's parent kinetic fits

Fitted Model	**Residuals**
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data. The applicant concluded the DFOP fit provided a better visual and statistical fit than the SFO fit and, therefore, considered the DFOP endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained very similar results to the applicant. However, the CA considers the SFO visual and statistical fit good enough to obtain modelling endpoints. As the CA obtained very similar SFO endpoints to the applicant, the applicant's SFO fit is accepted and considered further in the evaluation.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.20-2** and the graphical outputs in Figure CA.B.**8.1.2.3.20-2**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.20-3**.

Table CA.B.8.1.2.3.20-2: Summary of UK01 P2 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good
DT_{50} (days)		**78.8**	63.4
DT_{90} (days)		**262**	325
DT_{50} (days) - Slow phase		**n/a**	113
χ^2 error (%)		**9.4**	6.78
k ($days^{-1}$)	k or k_1	**8.80E-3**	0.1465
	k_2	**n/a**	6.16E-3
P value	k or k_1	**1.84E-7**	0.2071
	k_2	**n/a**	4.31E-6
g		**n/a**	0.261

Selected model shown in bold

Figure CA.B.8.1.2.3.20-2: Applicant's UK01 P2 parent-only modelling kinetic fits

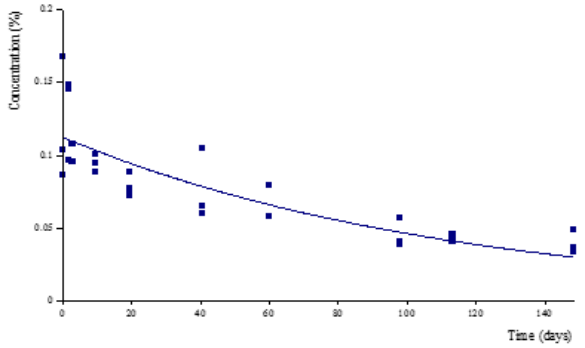
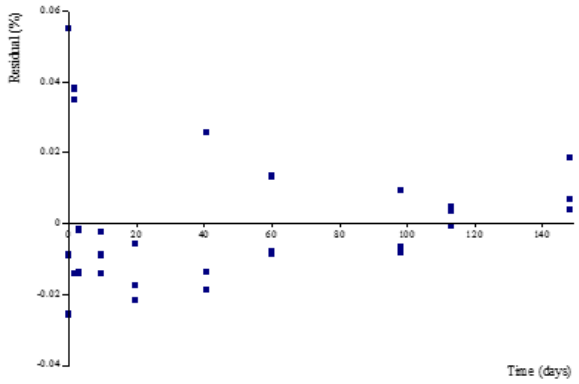
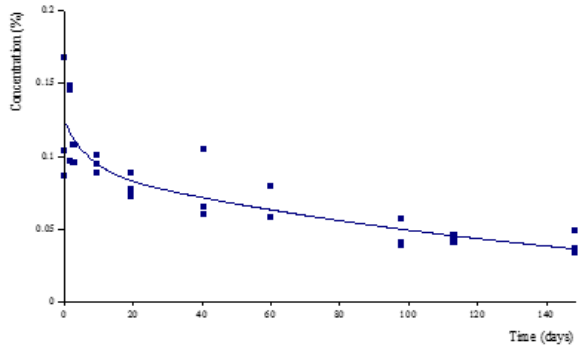
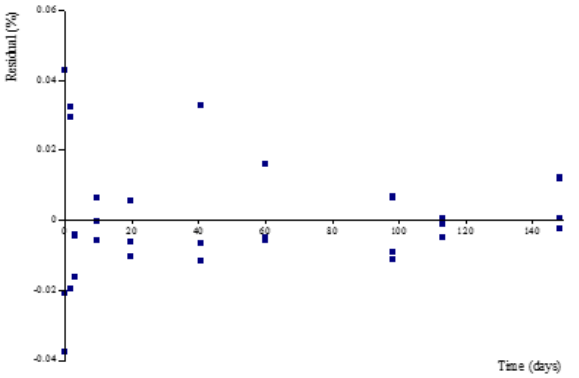
Fitted Model	Residuals
SFO 	SFO 
DFOP 	DFOP 

Table CA.B.8.1.2.3.20-3: Summary of UK01 P2 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	105	873
Modelling endpoints			
Bixlozone	SFO	78.8	262

CA.B.8.1.2.3.21. UK01 Plot 3

Triggering and PECsoil endpoints

In order to determine triggering and PECsoil endpoints, the applicant used the non-normalised data to conclude upon the best-fit kinetics. As an initial step, parent-only SFO, FOMC and DFOP models were run and the results compared. The applicant noted all three fits were visually acceptable, however, the SFO χ^2 was >15%, the FOMC beta confidence interval contains 0 and the DFOP k1 t-test value was >0.1. Therefore, the applicant considered that, despite the larger χ^2 value, SFO provided the best visual and statistical fit of the data.

The CA has repeated the applicant's SFO, FOMC and DFOP model runs. The CA agrees all three visual fits were acceptable. In the CA's modelling, the DFOP t-test values were also acceptable for both k1 and k2. As the DFOP fit had the slightly lower χ^2 value and a slightly better visual fit, the CA considers DFOP to provide the best-fit of the data. The CA considers the difference in DFOP statistical results from the applicant's modelling is due to the different data handling techniques used and/or the CA selecting NLLS as the optimisation method.

The CA notes residues of 2,4-DBA were detected in the trial at 3 DALA, 6 DALA and 15 DALA. However, the CA does not consider the data to show a sufficient decline phase, therefore, the CA does not consider the data robust enough to perform kinetic analysis.

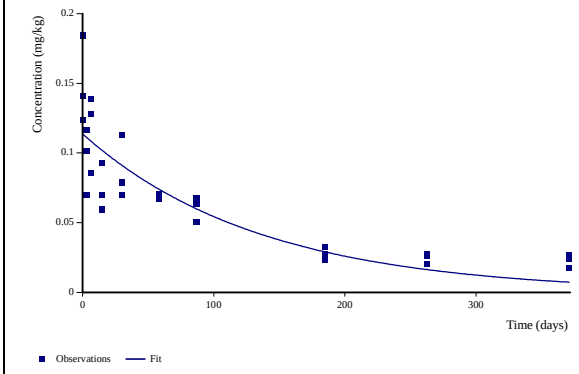
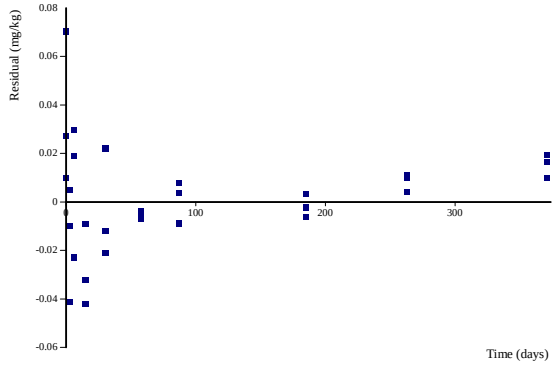
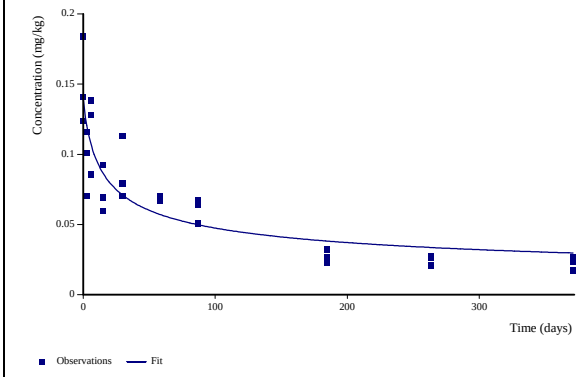
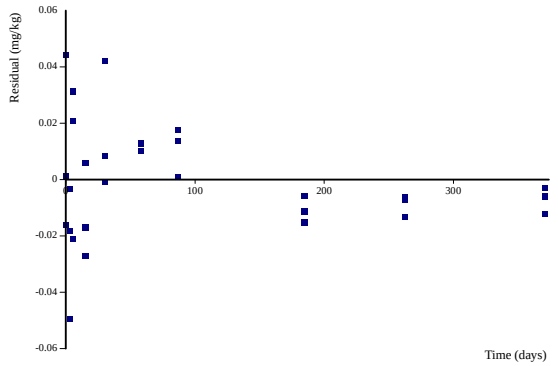
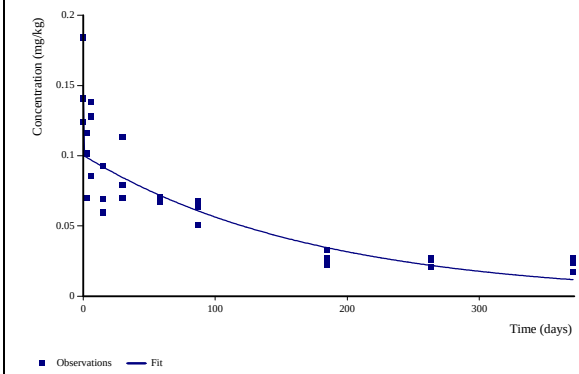
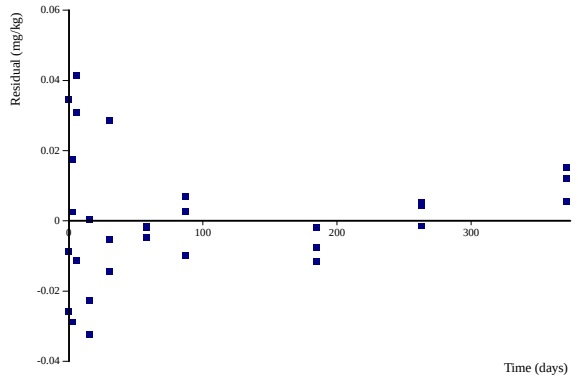
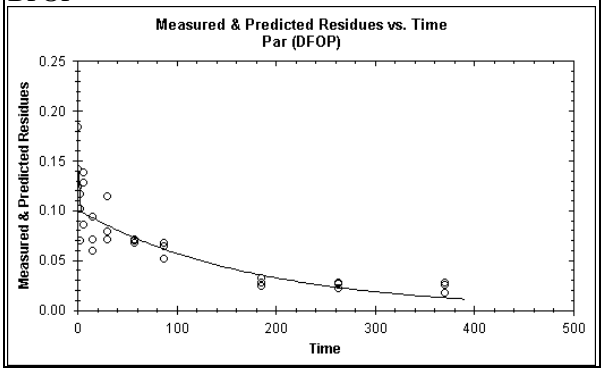
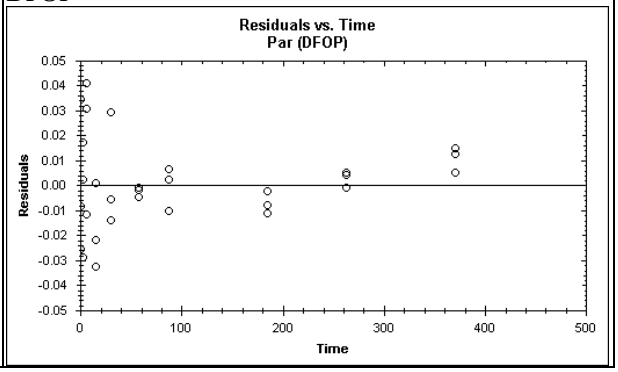
A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.21-1** and the graphical outputs in Figure CA.B.8.1.2.3.21-1. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.21-4**.

Table CA.B.8.1.2.3.21-1: Summary of UK01 P3 best-fit kinetic modelling

Modelling		**Applicant's modelling**			**CA's modelling**
Pathway		**Bixlozone only**			**Bixlozone only**
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		SFO	FOMC	DFOP	**DFOP**
Visual fit		Acceptable	Acceptable	Acceptable	**Good**
DT_{50} (days)		94	31.3	51.1	**51.8**
DT_{90} (days)		312	2.91E+3	330	**333**
DT_{90}/3.32 (days)		n/a	n/a	99	**100**
DT_{50} (days) - Slow phase		n/a	876	120	**121**
χ^2 error (%)		18.2	15.1	11.9	**11.8**
k ($days^{-1}$)	k or k_1	0.007372	n/a	4.168	**6.806**
	k_2	n/a	n/a	0.005772	**5.733E-3**
P value	k or k_1	1.23E-5	n/a	0.4986	**2.0E-16**
	k_2	n/a	n/a	8.43E-5	**6.90E-6**
g		n/a	n/a	0.328	**0.327**
alpha		n/a	0.3683	n/a	**n/a**
beta		n/a	5.615	n/a	**n/a**
95% CI (lower/upper)	alpha	n/a	0.1587 / 0.578	n/a	**n/a**
	beta	n/a	-4.081 / 15.31	n/a	**n/a**

Best-fit model shown in bold

Figure CA.B.8.1.2.3.21-1: UK01 P3 parent kinetic fits

Fitted Model	**Residuals**
Applicant's kinetic fits	
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	
CA's DFOP kinetic fit	
DFOP	DFOP
	

Persistence endpoints

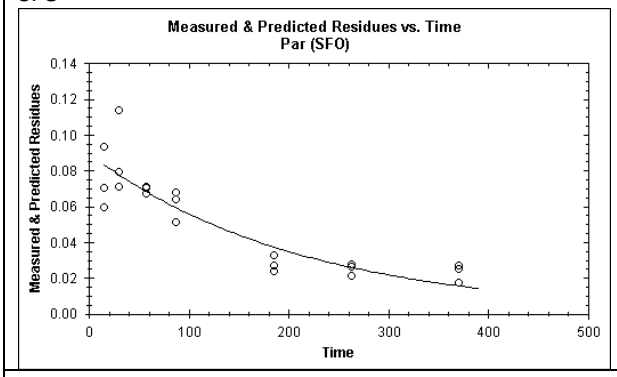
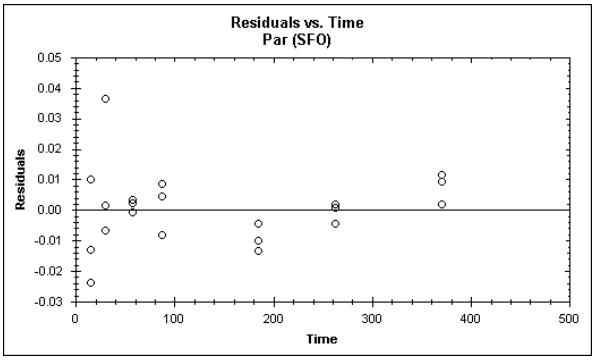
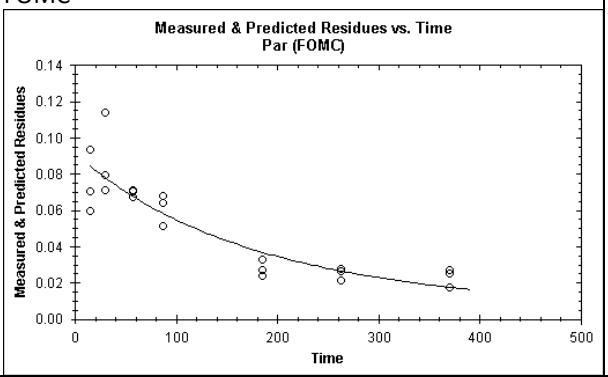
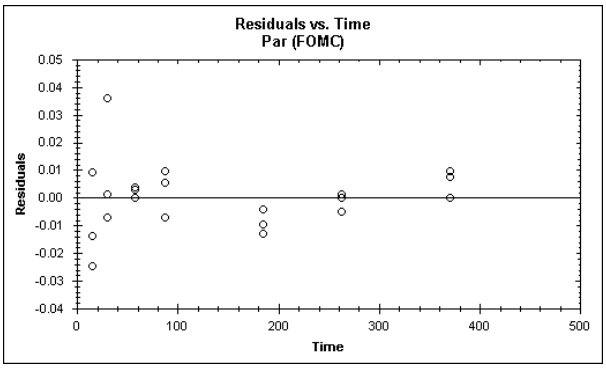
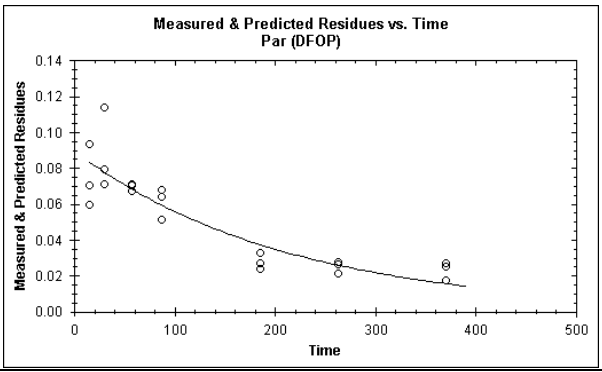
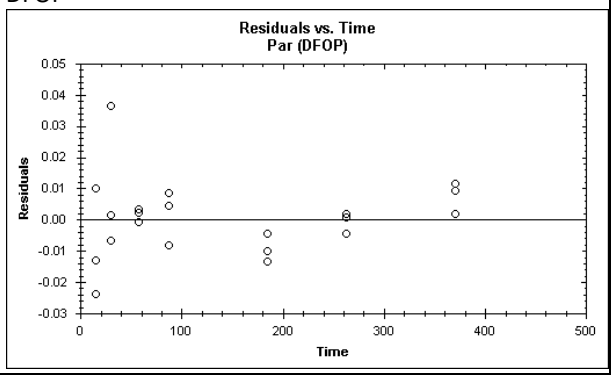
As this trial did not include incorporation, separate non-normalised persistence calculations are required. The CA removed the sampling occasions prior to 10 mm of rainfall from the dataset and proceeded to run SFO, FOMC and DFOP models. The SFO model provided a good visual fit and statistical fit of the data. The biphasic fits were not visually or statistically better than the SFO fit and so the CA considers the SFO fit to provide suitable persistence endpoints. The kinetic fit results are presented in Table CA.B.**8.1.2.3.21-2** and Figure CA.B.**8.1.2.3.21-2**.

Table CA.B.8.1.2.3.21-2: Summary of UK01 P3 persistence kinetic modelling

Modelling		**CA's modelling**		
Pathway		**Bixlozone only**		
Compound		**Bixlozone**	**Bixlozone**	**Bixlozone**
Model		**SFO**	FOMC	DFOP
Visual fit		**Good**	Good	Good
DT_{50} (days)		**146**	137	146
DT_{90} (days)		**486**	554	486
DT_{90}/3.32 (days)		**n/a**	167	146
DT_{50} (days) - Slow phase		**n/a**	n/a	146
χ^2 error (%)		**10.5**	11.2	12.5
k ($days^{-1}$)	k or k_1	**4742E-3**	n/a	0.6106
	k_2	**n/a**	n/a	4.742E-3
P value	k or k_1	**1.91E-6**	n/a	<2E-16
	k_2	**n/a**	n/a	6.35E-6
g		**n/a**	n/a	4.788E-5
alpha		**n/a**	4.250	n/a
beta		**n/a**	770.9	n/a
95% CI (lower/upper)	alpha	**n/a**	-19.97 / 28.47	n/a
	beta	**n/a**	-4329 / 5870	n/a

Best-fit model shown in bold

Figure CA.B.8.1.2.3.21-2: CA's UK01 P3 parent-only persistence kinetic fits

Fitted Model	Residuals
SFO	SFO
	
FOMC	FOMC
	
DFOP	DFOP
	

Modelling endpoints

In order to determine suitable modelling endpoints, the applicant initially produced SFO and DFOP fits of the normalised parent-only data (after 10 mm rain had fallen). The applicant concluded the SFO fit provided an acceptable visual and statistical fit of the data and so considered the SFO endpoints appropriate for modelling. The CA has repeated the applicant's modelling and obtained similar results to the applicant, therefore, the applicant's fit is accepted.

A summary of the kinetic statistical outputs is presented in Table CA.B.**8.1.2.3.21-3** and the graphical outputs in Figure CA.B.**8.1.2.3.21-3**. The final endpoints selected are summarised in Table CA.B.**8.1.2.3.21-4**.

Table CA.B.8.1.2.3.21-3: Summary of UK01 P3 modelling endpoint kinetics

Modelling		**Applicant's modelling**	
Pathway		**Bixlozone only**	
Compound		**Bixlozone**	**Bixlozone**
Model		**SFO**	DFOP
Visual fit		**Good**	Good
DT_{50} (days)		**69.2**	69.2
DT_{90} (days)		**230**	230
DT_{50} (days) - Slow phase		**n/a**	69.2
χ^2 error (%)		**12.2**	14.5
k ($days^{-1}$)	k or k_1	**0.01002**	0.01002
	k_2	**n/a**	0.01002
P value	k or k_1	**1.23E-6**	0.5000
	k_2	**n/a**	0.5000
g		**n/a**	0.0833

Selected model shown in bold

Figure CA.B.8.1.2.3.21-3: Applicant's UK01 P3 parent-only modelling kinetic fits

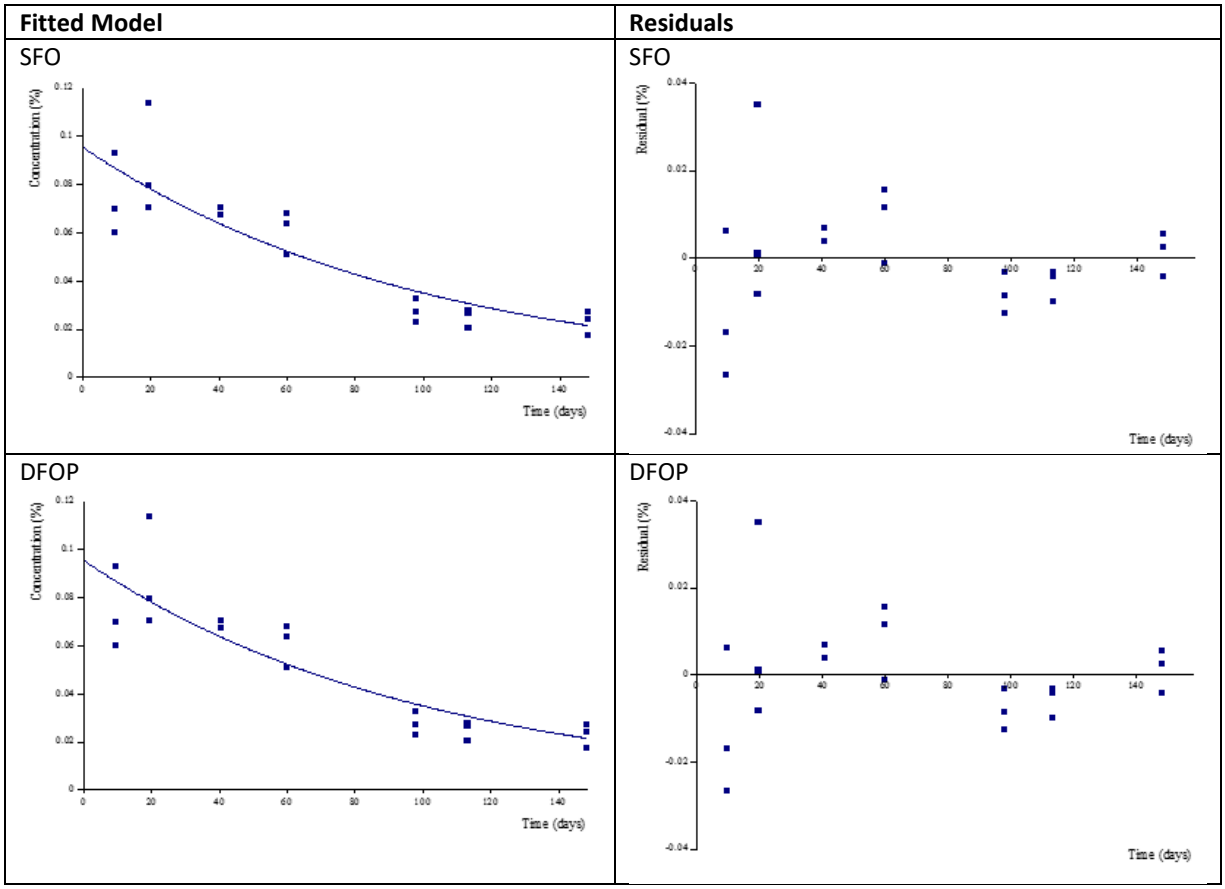


Table CA.B.8.1.2.3.21-4: Summary of UK01 P3 modelling and best-fit endpoints

Compound	Model	DT50 (d)	DT90 (d)
Best-fit endpoints			
Bixlozone	DFOP	51.8	333
Bixlozone	SFO	146	186
Modelling endpoints			
Bixlozone	SFO	69.2	230

CA.B.8.1.2.3.22. Conclusion

The field dissipation triggering and PECsoil DT_{50} and DT_{90} values for bixlozone and 2,4-dichlorobenzoic acid are summarised in Table CA.B.8.1.2.3.22-1Table CA.B. and Table CA.B.8.1.2.3.22-2 below. Further consideration of pH dependence, combining lab and field DT50s and persistence are presented in sections CA.B.8.1.4, CA.B.8.1.3 and CA.B.8.1.5 respectively.

As DT_{90} values >365 days were determined at 11 trial sites, the CA considers it appropriate to consider bixlozone accumulation as part of the assessment. As no specific bixlozone accumulation studies have been submitted, accumulation is to be considered as part of the PECsoil assessment. The longest non-normalised bixlozone DT_{50} is 300 d (site GE01) from the CS formulation. The longest non-normalised bixlozone DT_{50} from the SC formulation trials was 247 d (site IT01). In line with the discussion on modelling endpoints below, the longest SC formulation DT_{50} is considered appropriate for use in the PECsoil assessment to support the representative SC formulation product.

Table CA.B.8.1.2.3.22-1: Summary of field dissipation triggering and PECsoil DT_{50} and DT_{90} values for bixlozone

Trial	Site	F9600-4 SC			F9600-21 CS		
		DT_{50} (days)	**DT_{90} (days)**	**Model**	**DT_{50} (days)**	**DT_{90} (days)**	**Model**
Incorporated							
15SGS088	FR01	53.2	177	SFO	216	719	SFO
	IT01	247	819	SFO	292	971	SFO
	IT02	6.90	157	DFOP	-	-	-
15SGS111	GE01	193	642	SFO	300	997	SFO
	FR02	106	352	SFO	213	708	SFO
S16-02441	GE02	22.6	>1000	DFOP	-	-	-
	UK01	105	873	DFOP	-	-	-
Bare soil							
15SGS088	FR01	19.6	125	DFOP	0.2	108	DFOP
	IT01	28.5	94.6	SFO	7.36	219	DFOP
	IT02	-	-	-	-	-	-
15SGS111	GE01	181	601	SFO	194	643	SFO
	FR02	57.8	192	SFO	74.7	248	SFO
S16-02441	GE02	24.7	571	DFOP	-	-	-
	UK01	51.8	333	DFOP	-	-	-

Metabolite, 2,4-dichlorobenzoic acid, was generally only observed at low concentrations for short time periods only in the field dissipation trials. Field dissipation DT_{50} and DT_{90} values could only be determined for the bare soil plots in trials 15SGS088 FR01 and IT02, in which 2,4-dichlorobenzoic acid is shown to dissipate quickly under field conditions.

As DT_{90} values were less than the 365 day trigger value, accumulation does not need to be considered for this metabolite. The longest non-normalised 2,4-DBA DT_{50} is 15.7 d (from site IT01).

Table CA.B.8.1.2.3.22-2: Summary of field dissipation triggering and PECsoil DT_{50} and DT_{90} values for 2,4-dichlorobenzoic acid

Trial	Site	F9600-4 SC			F9600-21 CS		
		DT_{50} (days)	**DT_{90} (days)**	**Model**	**DT_{50} (days)**	**DT_{90} (days)**	**Model**
Bare soil							
15SGS088	FR01	2.77	9.22	SFO	6.40	21.3	SFO
	IT01	4.98	16.5	SFO	15.7	52.1	SFO

The bixlozone DT_{50} and DT_{90} values for the Persistence assessment are summarised in Table CA.B.8.1.2.3.22-3; the bare soil DT_{50} values differ to the triggering and PECsoil endpoints due to the differing treatment of results prior to 10 mm rainfall. A full assessment of bixlozone persistence in soil is presented in section CA.B.8.1.5.

Table CA.B.8.1.2.3.22-3: Summary of Persistence DT_{50} and DT_{90} values for bixlozone

Trial	Site	F9600-4 SC			F9600-21 CS		
		DT_{50} (days)	**DT_{90} (days)**	**Model**	**DT_{50} (days)**	**DT_{90} (days)**	**Model**
Incorporated							
15SGS088	FR01	53.2	177	SFO	216	719	SFO
	IT01	247	819	SFO	292	971	SFO
	IT02	6.90	157	SFO	-	-	-
15SGS111	GE01	193	642	SFO	300	997	SFO
	FR02	106	352	SFO	213	708	SFO
S16-02441	GE02	22.6	>1000	DFOP	-	-	-
	UK01	105	873	DFOP	-	-	-
Bare soil							
15SGS088	FR01	41.9	139	SFO	59.4	197	SFO
	IT01	70.9	236	SFO	135	447	SFO
	IT02	-	-	-	-	-	-
15SGS111	GE01	144	477	SFO	151	500	SFO
	FR02	58.9	196	SFO	82.1	273	SFO
S16-02441	GE02	7.03	318	DFOP	-	-	-
	UK01	146	486	SFO	-	-	-

The bixlozone modelling endpoints are summarised in Table CA.B.. As >5% 2,4-DBA was formed prior to 10 mm rainfall, or residues were detected sporadically, robust modelling kinetic fits cannot be obtained for the metabolite.

Table CA.B.8.1.2.3.22-4: Summary of modelling DT_{50} and DT_{90} values for bixlozone

Trial	Site	F9600-4 SC			F9600-21 CS		
		DT_{50} (days)	**DT_{90} (days)**	**Model**	**DT_{50} (days)**	**DT_{90} (days)**	**Model**
Incorporated							
15SGS088	FR01[a)]	53.9	179	SFO	152	504	SFO
	IT01[a)]	187	619	SFO	195	646	SFO
	IT02[b)]	9.38	31.2	SFO	-	-	-
15SGS111	GE01[b)]	68.9	229	SFO	105	350	SFO
	FR02[b)]	47.9	159	SFO	106	351	SFO
S16-02441	GE02[c)]	103	343	SFO	-	-	-
	UK01[c)]	78.8	262	SFO	-	-	-
Geomean DT_{50} (d)		59.1		-	135		-
Bare soil							
15SGS088	FR01[a)]	43.5	145	SFO	59.1	196	SFO
	IT01[a)]	57.8	192	SFO	98.0	326	SFO
	IT02[b)]	-	-	-	-	-	-
15SGS111	GE01[b)]	49.3	164	SFO	50.5	168	SFO
	FR02[b)]	23.1	76.7	SFO	33.0	110	SFO
S16-02441	GE02[c)]	72.6	241	SFO	-	-	-
	UK01[c)]	69.2	230	SFO	-	-	-
Geomean DT_{50} (d)		49.3		-	55.7		-

[a)] Test item applied in summer [b)] Test item applied in autumn [c)] Test item applied in spring

Looking at the modelling endpoints in Table CA.B.8.1.2.3.22-4 above, based on individual site comparisons and geometric means for the 4 combinations of trial design and formulation, there are potentially differences between the behaviour seen with the two different formulations, as well as between the different application methods. This might influence selection of the overall modelling endpoint (for example whether it is appropriate to treat all data as a single population for the purposes of deriving a geometric mean value, or to separate them into subpopulations based on formulation and/or application method).

Due to the possibility that distinct populations exist, the CA considered it appropriate to use the EFSA DegT50 endpoint selector tool for the statistical analysis. This tool can be used to determine whether laboratory and field

dissipation values come from the same population, or whether the field values are statistically significantly shorter than the laboratory values. In this example the tool was used to first determine whether the bare soil study design gave shorter DT50 values than the incorporated study design. The DT50 values from the bare soil plots are derived from sampling points after 10mm rainfall to minimise short term surface processes and to produce a bulk soil matrix degT50 which should be equivalent to the values from the incorporated study design. However it is possible that taking sampling points after 10mm rainfall is insufficient to account for all surface processes, and that the bare soil study design gives shorter DT50s. The hypothesis that was tested in the EFSA endpoint selector was therefore whether or not the bare soil plots gave shorter DT50s than the incorporated plots. Due to the possible additional influence of formulation type, the comparison between incorporated and bare soil application methods was assessed for each formulation separately.

Considering the SC formulation, the EFSA DegT50 tool indicated that the incorporated and bare soil application method gave DT50 values that were from the same population (that is that the results from the bare soil plots were not shorter than from the incorporated plots). The results for the SC formulation could therefore be treated as a single population, irrespective of application method.

Considering the CS formulation, the EFSA DegT50 tool indicated that the bare soil application method did give shorter DT50 values compared with the incorporated plots. The results for the CS formulation should not therefore be treated as a single population.

A further comparison was undertaken to compare the formulation types. Since the analysis above indicated that for the CS formulation there was a difference between application methods, the formulation comparison was performed for each application method separately. Here the hypothesis that was tested in the EFSA endpoint selector was whether or not the SC formulation gave shorter DT50s than the CS formulation. This would be the case if the CS formulation had longer DT50s, for example because the capsule suspension led to slow release of the active substance over time where it becomes available to degradation, or due to reduced loss via volatilisation.

For the incorporated study design, the EFSA DegT50 tool indicated that the SC formulation did give shorter DT50 values compared to the CS formulation. However, for the bare soil plots, the EFSA tool indicated there was no significant difference between formulation types. This indicates for the incorporated plots at least, it may not be appropriate to combine data from both formulations.

The analysis above was used by the CA to determine the appropriate values to use for modelling endpoint selection. Note that the incorporated study design is in line with the recommendations for modern field dissipation study conduct from the EFSA DegT50 guidance in order to derive a long term bulk soil matrix value. Note also that the representative formulation for the purposes of active substance approval is an SC formulation.

The CA proposes to use all of the data from plots treated with the SC formulation (incorporated and bare soil) for the purposes of selecting a modelling endpoint for the representative SC formulation. The CA considered using just the data from the incorporated plots for the SC formulation. This would have used data that was generated in line with the EFSA DegT50 guidance. However the analysis above indicated that for the SC formulation, there was no statistical difference between the results from the incorporated and bare soil plots. The DegT50 values for the bare soil plots were also derived following the EFSA DegT50 guidance (i.e. using sampling points after 10mm rain had fallen). The CA considered there was therefore no reason to exclude the information from the bare soil plots in this case and that it would be preferable to use all valid data in determining a modelling endpoint. The geometric mean of all plots treated with the SC formulation was 54.4 d (n=13; treating each separate result as an individual replicate in determining the geometric mean).

The appropriateness of using a t-test (utilised in the EFSA DegT50 spreadsheet and typically only used for comparing 2 datasets) for comparing multiple datasets was considered further. The CA approached a statistician (within the CA) to perform independent statistical analysis on the field study results, to see if their analysis confirmed the CA's original conclusion. The statistician noted that trial results are available for 4 combinations of trial design and formulation with between 4 and 7 DT50 values for each. The box-plot of the untransformed data (left-hand plot, Figure CA.B.8.1.2.3.22-1) shows some evidence of skew and carrying out formal comparisons and statistical testing on the natural log transformed data would be appropriate (as is the case in the EFSA DegT50 spreadsheet). Informally, the box-plots suggest that the mean half-life is longer for the incorporated trials with the CS formulation than the other three combinations, and perhaps some suggestion that the mean is higher for the incorporated design with the SC formulation than both of the bare soil combinations.

The results of pairwise independent sample T-tests, and alternative non-parametric (Mann-Whitney) statistical tests, of the means for the natural log-transformed data are shown in Table CA.B.8.1.2.3.22-5. Note that in the opinion of the independent statistician, the relative simplicity of the data, resulting in effectively 4 combinations of trial design and formulation type meant that applying the t-test to the different pairs of data was a reasonable approach in this case. However the independent statistician did also note that the EFSA degT50 tool used a relatively high alpha significance value of 0.25, which would mean that statistically significant differences were more likely to be determined using the EFSA tool than more conventional significance levels. These tests indicate that for the CS formulation, DT50 values are statistically significantly longer for the incorporated than for the bare soil trial design at the conventional 5% significance level (P=0.017). The mean DT50 value was longer for the CS vs the SC formulation for trial type 1, though formal testing showed that the statistical significance was weaker with only the Mann-Whitney test reaching the conventional level of 5% (P=0.042). The statistician recommended that a 2-way Analysis of Variance (ANOVA) be used to test the effect of both the trial design and formulation factors, and this confirms the findings from the pair-wise comparisons (Table CA.B.8.1.2.3.22-5). The analysis suggests that both the trial design and formulation may be having an effect on DT50 values, though neither effect reached conventional levels of statistical significance (P=0.081 for trial type and P=0.118 for formulation). Although informal examination of the data suggests that it is specifically the CS formulation in the context of the incorporated design that produces longer DT50 values, the interaction between the trial design and the formulation factors in the ANOVA was not statistically significant. Thus, in formal terms, given the degree of variability in the results due to the relatively small number of trials, it is not clear whether it is the incorporated trial type alone or the CS formulation in combination with the incorporated trial type design that is most important in producing longer DT50 values.

Table CA.B.8.1.2.3.22-5: Outputs of t-test and Mann-Whitney statistical tests

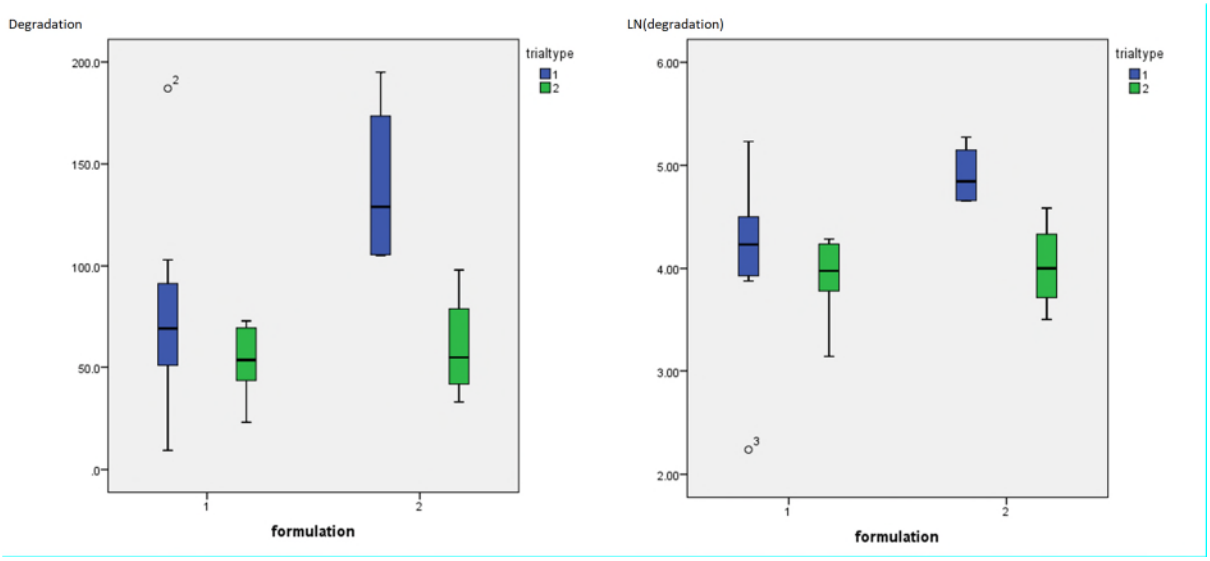
Trial design	Formulation	
	1. SC	2. CS
1. Incorporated	Mean days=78.4, SD=56.0 N=7 Mean $\log_e$ days=4.08 SD=0.93	Mean days=139.5 SD=43.0 N=4 Mean $\log_e$ days=4.90 SD=0.30 Tests of CS vs SC (trial type1) T-test: P=0.126 Mann-Whitney test: P=0.042
2. Bare soil	Mean days=52.6 SD=18.3 N=6 Mean $\log_e$ days=3.90 SD=0.42 Test of Bare vs Incorporated (SC formulation): T-test: P=0.668 Mann-Whitney test: P=0.446	Mean days=60.2 SD=27.5 N=4 Mean $\log_e$ days=4.02 SD=0.45 Test of Bare vs Incorporated (CS formulation): T-test: P=0.017 Mann-Whitney test: P=0.029 Tests of CS vs SC (trial type 2): T-test: P=0.672 Mann-Whitney test: P=0.762

Table CA.B.8.1.2.3.22-6: Outputs of ANOVA statistical test

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	2.741[a]	3	0.914	2.238	0.121	0.283
Intercept	352.918	1	352.918	864.491	0.000	0.981
formulation	1.105	1	1.105	2.708	0.118	0.137

Trial type	1.400	1	1.400	3.430	0.081	0.168
formulation * trial type	0.607	1	0.607	1.487	0.239	0.080
Error	6.940	17	0.408	-	-	-
Total	375.488	21	-	-	-	-
Corrected Total	9.681	20	-	-	-	-

Figure CA.B.8.1.2.3.22-1: Box plots of field DT50 values (formulation 1 = SC, formulation 2 = CS, trial type 1 = incorporated, trial type 2 = bare soil



The CA has concluded that the additional independent statistical advice was broadly supportive of the original assessment, indicating statistically significant differences for the CS formulation tested with the incorporated study design. The CA also notes that the additional 2-way Analysis of Variance (ANOVA) tests indicated that the interaction between trial design and formulation factors was not statistically significant, and given the variability and relatively small number of trials, there remains a degree of uncertainty over whether it is the incorporated trial type alone or the CS formulation in combination with the incorporated trial type design that is most important in producing longer DT50 values. Overall the CA concluded that in this case it is most appropriate to consider solely the SC field data (both bare soil and incorporated) further in the representative product groundwater and higher tier drainflow simulations, noting that this is directly applicable for the representative SC formulation product. This conclusion is in line with the original assessment. Should authorisation for a CS formulated product be sought in the future, detailed consideration and justification should be provided at that point to determine the appropriate DT50 value for use in the exposure calculations.

In order to reduce any bias in the data set caused by some sites having replicate DT50 values in the population, the CA also calculated a geomean value of each site before calculating a geomean of the different trial sites. The individual site and overall geomean values are summarised in Table CA.B.8.1.2.3.22-7.

Table CA.B.8.1.2.3.22-7: Summary of SC formulation field DT50 and geomean values

Soil	Plot	Formulation	DT50 (days)	Geomean DT50 (days)
Field study – Gemrot, F, 2018a				
FR01	Incorporated	F9600-4 SC	53.9	48.4
	Bare soil	F9600-4 SC	43.5	
IT01	Incorporated	F9600-4 SC	187	104
	Bare soil	F9600-4 SC	57.8	
IT02	Incorporated	F9600-4 SC	9.38	9.38
Field study – Gemrot, F, 2018b				
GE01	Incorporated	F9600-4 SC	68.9	58.3
	Bare soil	F9600-4 SC	49.3	
FR02	Incorporated	F9600-4 SC	47.9	33.3
	Bare soil	F9600-4 SC	23.1	
Field study – Gezahegne, W, 2018				
GE02	Incorporated	F9600-4 SC	103	86.5
	Bare soil	F9600-4 SC	72.6	
UK01	Incorporated	F9600-4 SC	78.8	73.8
	Bare soil	F9600-4 SC	69.2	
Geomean DT50 (d)			54.4	48.0

In the CA's original representative product exposure calculations, a DT50 of 54.4 days was used in the groundwater and higher tier drainflow calculations. For future product submissions based on SC formulations (or other formulation types not expected to influence the environmental fate and behaviour of the active substance), it is considered a DT50 of 48.0 days is most appropriate for use in the groundwater and higher tier drainflow exposure calculations. This updated DT50 (48.0 d) is considered to be sufficiently similar to the original DT50 (54.4 d) used in the exposure calculations to not warrant re-performing the exposure calculations as any change is expected to be insignificant. As noted above, should authorisation for a CS formulated product be sought in the future, detailed consideration and justification should be provided at that point to determine the appropriate DT50 value for use in the exposure calculations.

CA.B.8.1.3. **Comparison of laboratory and field modelling endpoints**

The CA has undertaken a comparison of the laboratory and field kinetics using the EFSA DegT50 calculator in order to determine the appropriate modelling endpoint for use in the exposure models. As explained in section CA.B.8.1.2.3 above, the CA considers it appropriate to populate the EFSA DegT50 calculator with the laboratory data and the geomean of each field study site for the SC formulation endpoints only (i.e. not the CS formulation – see section above for reasoning). The modelling endpoints input into the EFSA calculator are summarised in Table CA.B.**8.1.3-1**; see sections CA.B.8.1.1.4.1 and CA.B.8.1.2.3 for further information on how the endpoints were derived.

Table CA.B.8.1.3-1: Summary of bixlozone modelling endpoints

Soil	**Plot**	**Formulation**	**DT_{50} (days)**	**Geomean DT_{50} of field sites (days)**
Laboratory study – Simmonds, R, 2015a				
Lufa 6S	n/a	n/a	117.4	-
Lufa 5M	n/a	n/a	103.8	-
Lufa 2.2	n/a	n/a	330	-
RefeSol 02-A	n/a	n/a	184.3	-
CA-SL	n/a	n/a	138.3	-
Iowa	n/a	n/a	52.5	-
LAD-SCL-PF	n/a	n/a	140.7	-
Field study – Gemrot, F, 2018a				
FR01	Incorporated	F9600-4 SC	53.9	48.4
	Bare soil	F9600-4 SC	43.5	
IT01	Incorporated	F9600-4 SC	187	104
	Bare soil	F9600-4 SC	57.8	
IT02	Incorporated	F9600-4 SC	9.38	9.38
Field study – Gemrot, F, 2018b				
GE01	Incorporated	F9600-4 SC	68.9	58.3
	Bare soil	F9600-4 SC	49.3	
FR02	Incorporated	F9600-4 SC	47.9	33.3
	Bare soil	F9600-4 SC	23.1	
Field study – Gezahegne, W, 2018				
GE02	Incorporated	F9600-4 SC	103	86.5
	Bare soil	F9600-4 SC	72.6	
UK01	Incorporated	F9600-4 SC	78.8	73.8
	Bare soil	F9600-4 SC	69.2	
Geomean DT_{50} (d)				48.0

These endpoints confirm the hypothesis that field studies show shorter DegT50 than laboratory studies. Therefore, it is appropriate to only consider the field endpoints in the modelling. The resulting geomean value, of the SC field DT_{50}'s, is 48.0 days. This value is therefore appropriate for use in the exposure models. Please note, as explained above, in the CA's original representative product exposure calculations, a DT50 of 54.4 days was used in the groundwater and higher tier drainflow calculations. This updated DT50 (48.0 d) is considered to be sufficiently similar to the original DT50 (54.4 d) used in the exposure calculations for the SC formulation to not warrant re-performing the exposure calculations as any change is expected to be insignificant and to have no impact on the overall regulatory conclusion.

CA.B.8.1.4. **pH dependent degradation**

The CA has investigated the possibility of pH dependent degradation of bixlozone and metabolite 2,4-dichlorobenzoic acid (2,4-DBA); the CA has only investigated the pH dependency of this metabolite as this is the only 'major' soil aerobic metabolite. The CA notes that bixlozone's chemical properties do not indicate a mechanistic reason for a pH effect: the log Pow is stable over pH 4, 7 and 9 (3.3, 20°C), the solubility in water was not significantly impacted by pH (42.3, 39.6 and 41.9 mg/L at pH 4, 7 and 9 respectively (20 °C)) and it does not contain any ionisable groups within environmentally relevant ranges.

The CA has used the modelling endpoints from both the laboratory and SC field studies for the assessment of bixlozone and just the laboratory study for 2,4-DBA. For the pH dependency calculations, it was considered acceptable to consider the individual DT50 values from the field trial sites (as opposed to using the geomean DT50 of each site). The modelling DT50 values and soil pH of bixlozone are summarised in Table CA.B.**8.1.4-1** and 2,4-DBA in Table CA.B.**8.1.4-2**; see sections CA.B.8.1.1.4.1, CA.B.8.1.1.4.3 and CA.B.8.1.2.3 for further information on how the endpoints were derived.

Table CA.B.8.1.4-1: Summary of bixlozone soil pH and modelling DT50 values

Soil	pH (water, 0-30 cm horizon)	Plot	Formulation	DT_{50} (days)
Laboratory study – Simmonds, R, 2015a				
Lufa 6S	7.1	n/a	n/a	117.4
Lufa 5M	7.5	n/a	n/a	103.8
Lufa 2.2	5.7	n/a	n/a	330
RefeSol 02-A	6.3	n/a	n/a	184.3
CA-SL	7.4	n/a	n/a	138.3
Iowa	7.2	n/a	n/a	52.5
LAD-SCL-PF	8.1	n/a	n/a	140.7
Field study – Gemrot, F, 2018a				
FR01	5.9	Incorporated	F9600-4 SC	53.9
		Bare soil	F9600-4 SC	43.5
IT01	6.7	Incorporated	F9600-4 SC	187
		Bare soil	F9600-4 SC	57.8
IT02	6.7	Incorporated	F9600-4 SC	9.38
Field study – Gemrot, F, 2018b				
GE01	5.9	Incorporated	F9600-4 SC	68.9
		Bare soil	F9600-4 SC	49.3
FR02	5.1	Incorporated	F9600-4 SC	47.9
		Bare soil	F9600-4 SC	23.1
Field study – Gezahegne, W, 2018				
GE02	5.2	Incorporated	F9600-4 SC	103
		Bare soil	F9600-4 SC	72.6
UK01	7.1	Incorporated	F9600-4 SC	78.8
		Bare soil	F9600-4 SC	69.2

Table CA.B.8.1.4-2: Summary of 2,4-DBA soil pH and modelling DT50 values

Soil	pH ($CaCl_2$)	DT_{50} (days)
Laboratory study – Göcer, M., 2016b		
Lufa 2.1	4.84	8.9
Lufa 2.4	7.41	3.5
St. Bauzille 12-060	7.53	5.2

The CA has performed the Kendall test with the data, as well as producing linear regression plots. The results of the Kendall test are summarised in Table CA.B.**8.1.4-3** and the regression plots in Figure CA.B.**8.1.4-1** and Figure CA.B.**8.1.4-2**.

Table CA.B.8.1.4-3: Results of Kendall test

	Bixlozone lab DT_{50}'s	Bixlozone field DT_{50}'s	2,4-DBA lab DT_{50}'s
Tau	-0.333	0.223	-0.333
p-value	0.368	0.346	1.000

Figure CA.B.8.1.4-1: Bixlozone linear regression plot

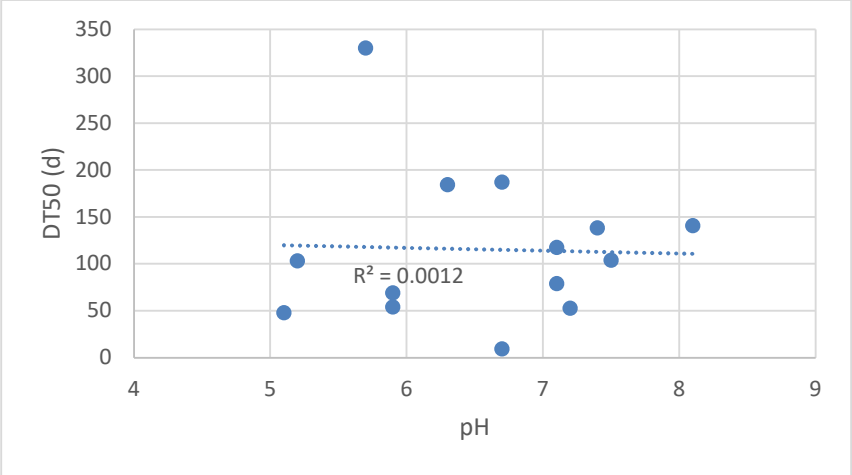
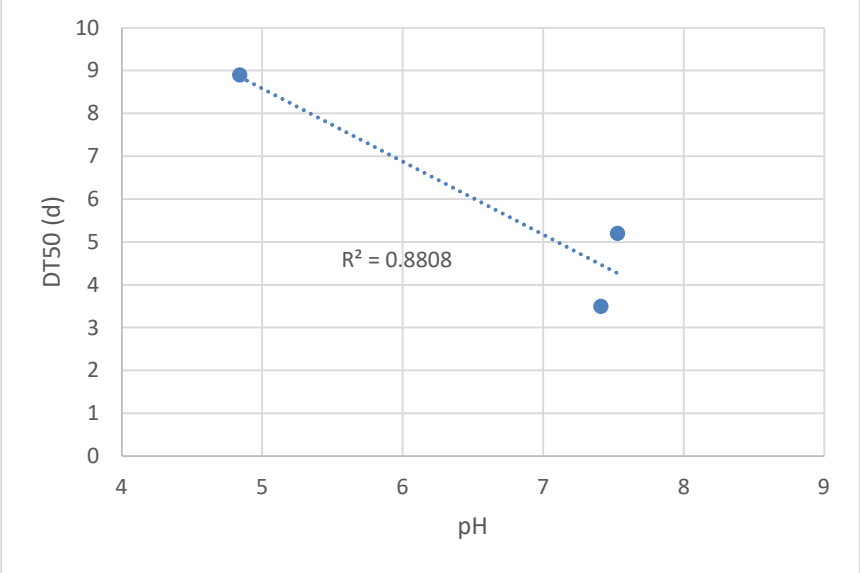


Figure CA.B.8.1.4-2: 2,4-DBA linear regression plot



The outcome of the Kendall test indicated a correlation between soil pH and DT_{50} for both bixlozone (in lab and field data) and 2,4-DBA did not exist. This is further evidenced by the bixlozone regression plot, which does not clearly identify a trend in the data. The 2,4-DBA regression plot indicates the possibility of a trend, however, the CA does not consider there to be sufficient data points to draw a definitive conclusion. Due to this uncertainty and because of the lack of correlation for 2,4-DBA shown with the Kendall test, the CA does not consider it necessary to consider pH dependant degradation in the bixlozone or 2,4-DBA exposure calculations.

CA.B.8.1.5. **Summary of Persistence assessment in soil**

The criteria for a pesticide to be classed as 'Persistent' or 'very Persistent' is outlined within Regulation 1107/2009 as it applies in Great Britain. For the soil compartment, these are as follows:

An active substance, safener or synergist fulfils the persistence criterion where:

- *The half-life in soil is higher than 120 days.*

An active substance, safener or synergist fulfils the 'very persistent' criterion where:

- *the half-life in soil is higher than 180 days.*

The relevant endpoints for the persistence assessment were identified based on the DG SANCO working document on "Evidence Needed to Identify POP, PBT and vPvB Properties for Pesticides" [*SANCO 2012. DG SANCO Working Document on "Evidence Needed to Identify POP, PBT and vPvB Properties for Pesticides". Brussels: European Commission Health and Consumers Directorate-General. Report 25.09.2012 - rev. 3.*]. According to this document, when available, field degradation half-lives are relevant for the P and vP assessment.

The degradation of bixlozone was investigated in a laboratory soil degradation study in 7 aerobic soils (see section CA.B.8.1.1.1). Additionally, the degradation of bixlozone was investigated under field conditions at 7 test sites (see section CA.B.8.1.2.1), which consisted of 21 trials where persistence endpoints could be determined. The persistence endpoints from both the laboratory and the field studies are summarised in Table CA.B.**8.1.5-1**.

Table CA.B.8.1.5-1: Summary of Persistence endpoints in soil

Soil	Plot	Formulation	DT_{50} (days)	Model
Laboratory study – Simmonds, R, 2015a				
Lufa 6S	n/a	n/a	136	FOMC
Lufa 5M	n/a	n/a	115	SFO
Lufa 2.2	n/a	n/a	1000	FOMC
RefeSol 02-A	n/a	n/a	358	DFOP
CA-SL	n/a	n/a	154	SFO
Iowa	n/a	n/a	64.1	SFO
LAD-SCL-PF	n/a	n/a	176	SFO
Field study – Gemrot, F, 2018a				
FR01	Incorporated	F9600-4 SC	53.2	SFO
		F9600-21 CS	216	SFO
	Bare soil	F9600-4 SC	41.9	SFO
		F9600-21 CS	59.4	SFO
IT01	Incorporated	F9600-4 SC	247	SFO
		F9600-21 CS	292	SFO
	Bare soil	F9600-4 SC	70.9	SFO
		F9600-21 CS	135	SFO
IT02	Incorporated	F9600-4 SC	6.90	SFO
Field study – Gemrot, F, 2018b				
GE01	Incorporated	F9600-4 SC	193	SFO
		F9600-21 CS	300	SFO
	Bare soil	F9600-4 SC	144	SFO
		F9600-21 CS	151	SFO
FR02	Incorporated	F9600-4 SC	106	SFO
		F9600-21 CS	213	SFO
	Bare soil	F9600-4 SC	58.9	SFO
		F9600-21 CS	82.1	SFO
GE02	Incorporated	F9600-4 SC	22.6	DFOP
	Bare soil	F9600-4 SC	7.03	DFOP
UK01	Incorporated	F9600-4 SC	105	DFOP

	Bare soil	F9600-4 SC	146	SFO

As can be seen, there is a great deal of variability in the DT_{50} values calculated, with values ranging from 6.90 days to 1000 days. Of the 7 laboratory DT_{50} values, 5 were above the 120 day trigger value. Of the 21 field DT_{50} values, 10 were above the 120 day trigger. Furthermore, DT_{50} values greater than the 120 day trigger were recorded at each test site (i.e. in France, Italy, Germany and the UK). Therefore, the CA considers there is sufficient weight of evidence to categorise bixlozone as persistent in soil.

Of the 21 field DT_{50} values, 6 were above the 180 day 'very Persistent' trigger. The trials that recorded DT_{50} values >180 days were located in Italy, France and Germany. Therefore, although the weight of evidence to consider the substance as very persistent is weaker than the persistent assessment above, overall the CA considers that bixlozone potentially fulfils the 'very Persistent' criteria.

CA.B.8.1.6. **Adsorption and desorption in soil**

CA.B.8.1.6.1. ***Adsorption and desorption of the active substance***

Report:	KCA 7.1.3.1.1 Simmonds, M.; Hawkins, T., (2016)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Adsorption to and Desorption from Eight Soils (Amended Final Report)
Testing facility:	Battelle UK Ltd., Springfield, UK.
Document No:	Study no. KW/14/005, FMC Tracking no. 2013EFT-ISX1025
Guidelines:	OECD Guideline 106 (2000); US EPA Guideline, OPPTS 835-1230 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

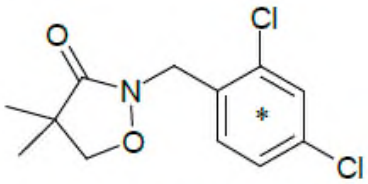
CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

The adsorption and desorption behaviour of [^{14}C]-bixlozone was examined in five European and three US soils (pH range 5.4 to 8.0 in 0.01M $CaCl_2$, % OC 0.3 – 2.1). Adsorption K_{FOC} values for [^{14}C]-bixlozone were 334 – 465 mL/g (geometric mean 381.5 mL/g, arithmetic mean 1/n = 0.874) and desorption $K_{FOC\text{-}des}$ values were 481 – 754 mL/g (geometric mean 564 mL/g, arithmetic mean 1/n= 0.876) indicating that there is a degree of irreversibility to [^{14}C]-bixlozone adsorption. There was no evidence of any pH dependence.

MATERIALS

Test substance [phenyl-U-^{14}C]-bixlozone



* Position of [^{14}C]-radiolabel

Lot/Batch no. CFQ42017
Purity Radiochemical Purity 97.48% (from Radio-HPLC)
CAS No 81777-95-9

The study was conducted using the batch equilibrium method, with five different European soils and three different US soils where there had been no pesticide use in the case of the EU soils (for previous 5 years for Lufa 6S, Lufa 5M, Lufa 2.2 and Refesol 02-A and previous 1 year for Ickingham) and no use of analogous compounds for the US soil CA-SL prior to collection. No pesticide history was provided in the study report for the US soils Iowa and LAD-SCL-PF, however the applicant confirmed no pesticide had been used for the previous 5 years. CA-SL soil was treated with metalaxyl-M 2 years and 4 years prior to sampling, with pendimethalin 3 years prior to sampling, and trifluralin 4 years prior to sampling.

The OECD 106 guideline states that detail on the pesticide history is necessary, however, due to the fact that the test item in this study is radiolabelled, and that the substances applied are not within the same chemical class as bixlozone (isoxazilidone herbicides), it is not expected that this will affect the outcome of this study. Further information on the pesticide use for the US soils Iowa and LAD-SCL-PF was not requested from the applicant. The European soils, except Icklingham, were not fertilised for 4 years prior to sampling. Icklingham, CA-SL, Iowa and LAD-SCL-PF soils' fertiliser history is not reported, and RefeSol 02-A had 30 kg P/ha applied once 2 years prior to sampling.

The soils were freshly collected, oven-dried, and sieved to 2 mm prior to use. A summary of the physical and chemical properties of the soils is provided in Table CA.B.**8.1.6.1-1**.

Table CA.B.8.1.6.1-1: Soil physiochemical properties

Soil Characterisation	**Lufa 6S**	**Lufa 5M**	**Lufa 2.2**	**Refesol 02-A**	**CA-SL**	**Iowa**	**LAD-SCL-PF**	**Icklingham**
Sampling location	Siebeldingen, Germany	Mechtersheim, Germany	Hanhofen Germany	Schmallenberg, Germany	Hughston, USA	Jackson, USA	Fermont USA	Icklingham, UK
Particle size distribution								
Sand (%)	29	56	84	22	77	15	27	94
Silt (%)	26	27	9	61	18	62	26	1
Clay (%)	45	17	7	17	5	23	47	5
Textural classification (USDA)	Clay	Sandy loam	Loamy sand	Silt loam	Loamy sand	Silt loam	Clay	Sand
pH ($CaCl_2$)	6.9	7.2	5.4	6.1	6.9	6.8	8.0	7.4
% Organic matter	3.6	2.2	2.6	2.1	0.59	3.6	1.8	2.1
% Organic carbon†	2.1	1.3	1.5	1.2	0.3	2.1	1.0	1.2
CEC (meq/100g)	21.0	9.7	7.3	11.2	5.5	13.6	31.1	9.4
Bulk density, disturbed (g/cm^3)	1.2	1.1	1.2	1.1	1.3	1.0	1.1	1.4
% Moisture at pF2.0	31.0	20.8	11.3	37.0	13.4	42.3	40.9	11.3
% Moisture at pF2.5	26.1	13.1	8.4	18.3	7.2	30.0	29.7	10.8

† % organic carbon = organic matter / 1.724

It is noted that soils tested did not include any with >2.1 % organic carbon, with a narrow range of 0.3 – 2.1 %. However, OECD 106 only states a minimum % organic carbon content and recommends a wide range of soils are used; no high value of % organic carbon is given. An adequate pH range of 5.4-8 is covered by the soils tested and the active is not expected to be ionisable at environmentally relevant pH.

METHOD

Experimental conditions

Test solutions were prepared by evaporating the test item to dryness under compressed air, then diluted to 10 mL with acetonitrile. 200 µL of this solution was diluted to 10 mL with acetonitrile, with a concentration of 0.51 mg/mL measured via LSC for this stock solution. Treatment solutions were prepared by diluting a suitable volume of the stock solution with 0.01M $CaCl_2$, so as to ensure that the organic solvent would not exceed 0.1 %.

During both preliminary and definitive tests, all vessels were shaken in the dark at a temperature of 20 ± 2°C. Centrifugation was performed at 4000 rpm and 20°C unless otherwise mentioned.

Preliminary tests

Preliminary investigations were carried out to check for adsorption to the PTFE tubes (and glass tubes for the adsorption to vessel determination), to determine any background radioactivity in the soils, to determine the soil to solution ratio to be used (1:10, 1:5 and 1:2.5) at an initial aqueous concentration of 0.30 mg/L, to check the stability of the test item in 0.01M calcium chloride, and to determine the time required for the compound to equilibrate between soil and water under both adsorption (ca 2, 4, 24, 48, and 72 hours) and desorption conditions (1, 2, 24 and 48 hours) at an initial aqueous concentration of 0.30 mg/L.

To determine the time required for the compound to equilibrate between soil and water, a treatment solution was made with an aliquot of stock solution diluted to a final volume of 100 mL with 0.01M $CaCl_2$. Appropriate amounts of soil (2, 4 or 8 g) were added to vessels with 0.01M $CaCl_2$ and shaken overnight to pre-equilibrate.

Following this, each vessel was treated with 1 mL treatment solution and shaken over a period of 72 hours. Individual vessels were removed after 2, 4, 24, 48, and 72 hours, centrifuged for 10 minutes, and aliquots of the supernatants analysed via LSC. The 48 and 72 hour samples were extracted 3 times using acetonitrile:water:formic acid (50:50:1, v/v/v) and shaken for 20 minutes. They were then centrifuged for 10 minutes, the supernatants combined and aliquots analysed by LSC. Aliquots of supernatant and extract were analysed by reverse-phase HPLC.

Following the adsorption phase, the method was repeated with fresh vessels, allowing adsorption for 48 hours, centrifugation of vessels for 10 minutes and removal of the supernatant. Fresh 0.01M $CaCl_2$, equal to the volume of supernatant removed, was added, and vessels were shaken for a further 48 hours. Individual vessels were removed at 1, 2, 24 and 48 hours. At each time point, vessels were centrifuged for 10 minutes and supernatant removed, with aliquots analysed by LSC.

Definitive test

For the definitive study, all soils were pre-equilibrated overnight (*ca* 16 hours) in a 0.01M $CaCl_2$ solution (*ca* 19 mL). Soils were present at 2, 4, or 8 g oven-dried equivalents in each vessel, appropriate to the soil:solution ratios determined during preliminary testing. Treatment solutions were prepared in order to give 1.0, 0.3, 0.1, 0.03 and 0.01 mg/L nominal concentration when 1 mL treatment solution is added to the 19 mL 0.01M $CaCl_2$ and soil to give an overall volume of 20 mL. After pre-equilibration, [^{14}C]-bixlozone treatment solutions were then added to each soil in duplicate (initial aqueous concentrations 0.94, 0.30, 0.10, 0.03 and 0.01 mg/L) and shaken for 48 hours (as determined during preliminary testing for equilibrium time) in the dark at 20ºC in PTFE vessels. Vessels were then centrifuged for 10 minutes, and supernatant removed. Aliquots of the supernatant were analysed by LSC.

A single desorption phase was also undertaken. Following centrifugation of the adsorption solution, the decanted solution was replaced by an approximately equal volume of fresh 0.01M $CaCl_2$ solution. This was then shaken for a further 2 hours, centrifuged for 10 minutes, and aliquots of the supernatant analysed by LSC.

Following the desorption phase, all vessels were solvent extracted twice using acetonitrile:water:formic acid (50:50:1, v/v/v) and shaken for 20 minutes. They were then centrifuged for 10 minutes and aliquots of the supernatants analysed by LSC. Vessels were then allowed to air dry, and the soil was homogenised and combusted, then analysed by LSC.

Analytical procedures

During the preliminary study to determine the equilibrium time (initial aqueous concentrations 0.30 mg/L), parental mass balances were determined in the supernatants and combined solvent extracts by LSC, following solubilisation in scintillation cocktail. Confirmatory and stability analysis was also performed by reverse-phase HPLC with in-line UV detection using a gradient elution with 1 % formic acid in water, and 1 % formic acid in acetonitrile as solvents. The LOQ <0.01% AR in the supernatant for both methods (0.004 % for HPLC, with the highest LOQ of 0.22 % for LSC with 0.01 µg/g nominal concentration). All supernatant and soil samples were stored refrigerated for a maximum of two days before analysis.

In the definitive isotherm study the aqueous supernatant was separated by centrifugation after adsorption and desorption. The concentration of radioactivity in the supernatant and combined duplicate extracts of each of the soil pellets was analysed by LSC after solubilisation in scintillation cocktail. Following extraction, the soil was combusted and the trapped CO_2 was trapped in Carbo-sorb®E absorbent and mixed with Permafluor®E+ scintillation cocktail, then quantified by LSC to determine the overall recovery. All supernatant and soil samples were stored refrigerated for a maximum of six days before analysis.

The concentration of [^{14}C]-bixlozone in the residual water is assumed to be equal to that in the supernatant. The soil concentration is calculated using the equation below:

$$C_S = \frac{CM_T - (C_W M + C_W R)}{W_{ode}}$$

C_s = Soil concentration after adsorption (µg g^{-1})
C = Concentration of treatment solution (µg g^{-1})
C_w = Concentration of adsorption supernatant (µg g^{-1})
M_T = Weight of treatment (g)
M = Weight of adsorption supernatant decanted (g)
R = Residual water (adsorption phase) (g)
W_{ode} = Oven dried soil equivalent (g)

For the desorption phase, the equation below is used to calculate the adsorbed substance:

$$C_{sx} = \frac{(C_{s(x-1)} W_{ode} + C_{w(x-1)} R_{(x-1)}) - (C_{wx} M_x + C_{wx} R_x)}{W_{ode}}$$

and $R_x = W_x - W_{ode}$

C_{sx} = Soil concentration (µg g^{-1})
$C_{s(x-1)}$ = Soil concentration (µg g^{-1}) previous cycle
C_{wx} = Concentration in aqueous phase at equilibrium (µg g^{-1})
$C_{w(x-1)}$ = Concentration in aqueous phase, previous cycle (µg g^{-1})
R_x = Residual water after this desorption cycle (g)
$R_{(x-1)}$ = Residual water (after previous cycle) (g)
M_x = Weight of supernatant decanted (g)
W_{ode} = Oven dried equivalent weight of soil (g)
W_x = Weight of soil pellet after decanting supernatant (g)

The concentration of radioactivity in the soil was then calculated by difference based on the LSC results from the aqueous phase (indirect method). The parameters of the Freundlich adsorption isotherm (K_F and 1/n) were estimated from a linear regression analysis using the $\log_{10}$-transformed measured aqueous concentrations and calculated soil concentrations.

The identity of [^{14}C]-bixlozone was confirmed by reverse-phase HPLC with a certified reference standard, using a Luna C18 column, and a gradient elution with water/trifluoroacetic acid (1000:0.5) and acetonitrile/trifluoroacetic acid (1000:0.5) at 22°C.

RESULTS

Mass Balance

The mass balances for preliminary tests and the definitive test are within the 90 – 110 % recovery range considered acceptable for radiolabelled studies. [^{14}C]-bixlozone was shown to be stable in the preliminary test, accounting for 92−96.4% of the total radioactivity in the supernatant and soil extracts for all soils after 72 hour of equilibration. The recovery of radioactivity in the overall system from the definitive isotherm study was also shown to be acceptable, with a range of 93.9 – 96.8%.

Preliminary tests

There was no evidence of significant adsorption to test vessels (98.9%AR and 101.5%AR recovered in PTFE and glass vessels, respectively).

Soil:solution ratios should be selected where the percentage adsorbed is above 20 %, and preferably above 50 %. The CA confirms these criteria are reached at the soil:solution ratios selected by the applicant for use in the definitive test. These are 1:2.5 for the CA-SL soil, 1:5 for the Lufa 5M soil, Refesol 02-A, LAD-SCL-PF and Icklingham soils and 1:10 for the Lufa 6S, Lufa 2.2 and Iowa soils.

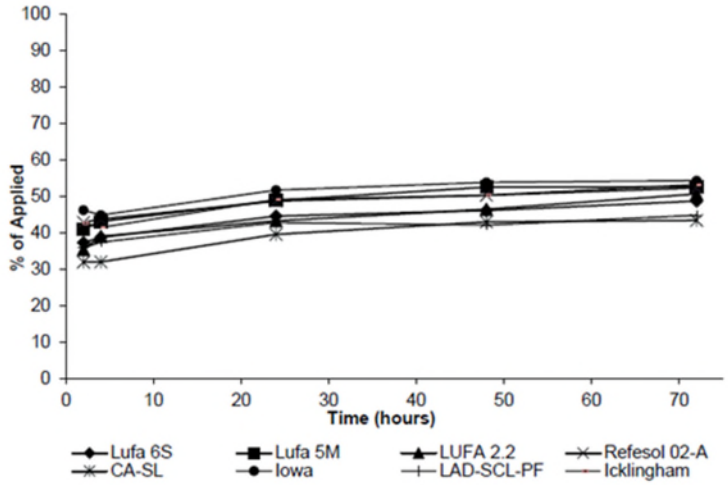
Table CA.B.8.1.6.1-2: Recoveries at different soil:solution ratios

Soil:Solution ratio	Soil	µg/g		Adsorbed to soil (% AR)
		Aqueous	Soil	
1:10	**Lufa 6S***	0.151	1.407	48.7
	Lufa 5M	0.195	0.950	32.6
	Lufa 2.2*	0.157	1.337	46.2
	RefeSol 02-A	0.188	1.018	34.9
	CA-SL	0.243	0.474	16.5
	Iowa*	0.139	1.506	51.8
	LAD-SCL-PF	0.206	0.846	29.0
	Icklingham	0.190	1.005	34.6
1:5	Lufa 6S	0.104	0.928	64.1
	Lufa 5M*	0.144	0.722	49.9
	Lufa 2.2	0.109	0.898	61.8
	RefeSol 02-A*	0.145	0.726	50.0
	CA-SL	0.214	0.377	25.9
	Iowa	0.090	1.001	68.8
	LAD-SCL-PF*	0.167	0.603	41.4
	Icklingham*	0.142	0.740	51.1
1:2.5	Lufa 6S	0.062	0.570	78.5
	Lufa 5M	0.093	0.492	67.7
	Lufa 2.2	0.063	0.567	77.9
	RefeSol 02-A	0.095	0.482	66.4
	CA-SL*	0.175	0.283	38.8
	Iowa	0.052	0.594	81.7
	LAD-SCL-PF	0.115	0.435	59.8
	Icklingham	0.090	0.499	68.8

* Ratio's used for each soil in the definitive test

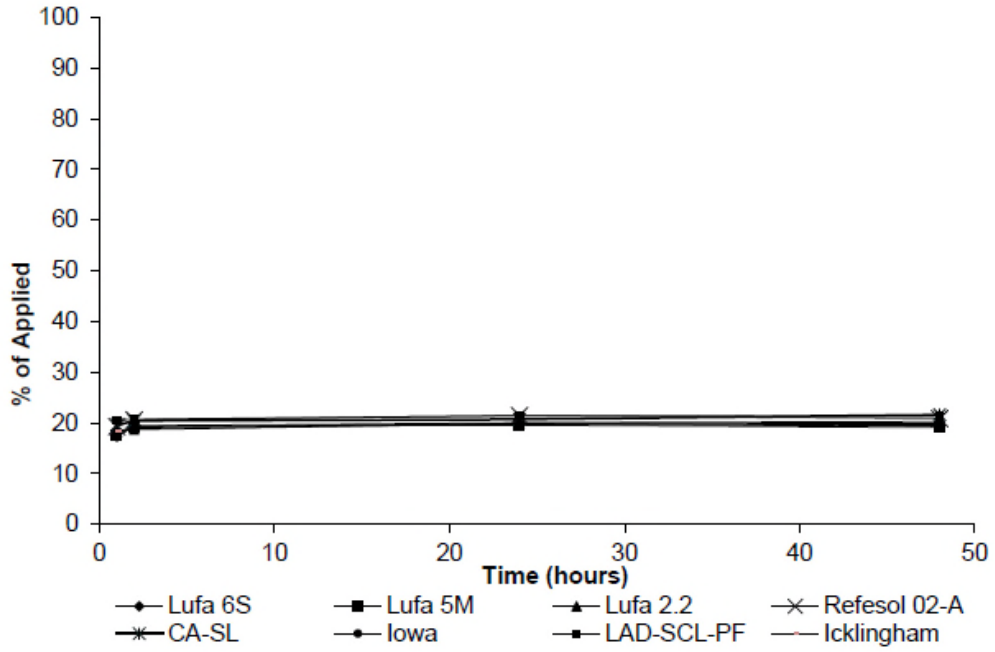
An equilibrium time of 48 hours was selected, as the applicant judged there to be little significant increase in adsorption between 48 and 72 hour samples. The plots of adsorption vs time show that a plateau has been reached (see Figure CA.B.**8.1.6.1-1**) and therefore the CA agrees with the choice of 48 hour equilibrium time.

Figure CA.B.8.1.6.1-1: Adsorption equilibrium time (preliminary experiment) of [^{14}C]-bixlozone



Stability of [^{14}C]-bixlozone was confirmed for the time period of 72 hours. For the determination of the desorption equilibrium time, there was no significant change in the levels of radioactivity in solution between 2 and 48 hours in any of the soils tested. Therefore 2 hours was selected as the equilibrium time for desorption.

Figure CA.B.8.1.6.1-2: Desorption equilibrium time (preliminary experiment) of [^{14}C]-bixlozone



Control samples containing soil and 0.01M $CaCl_2$ only showed negligible background radioactivity and therefore correction was not needed for recovery data.

Definitive test

In the definitive adsorption test, the amount of applied test material adsorbed ranged from 38.0-68.9% depending on soil and treatment concentration (Table CA.B.**8.1.6.1-3** to Table CA.B.**8.1.6.1-10**). There was no evidence of pH dependence and no evidence of any different behaviour between the European and US soils. Therefore all eight soils were considered suitable to determine mean values.

Table CA.B.8.1.6.1-3: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Lufa 6S soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Lufa 6S	1.0	4.102	0.512	44.15	2.543	0.194	38.0
		4.127	0.511	44.14	2.564	0.195	37.87
	0.3	1.437	0.155	47.23	0.922	0.062	35.83
		1.401	0.156	47.03	0.915	0.061	34.73
	0.1	0.52	0.047	52.16	0.348	0.021	33.05
		0.507	0.049	50.63	0.342	0.02	32.53
	0.03	0.163	0.014	53.72	0.112	0.006	31.24
		0.161	0.014	53.76	0.111	0.006	31.35
	0.01	0.059	0.004	57.89	0.042	0.002	27.87
		0.058	0.004	58.45	0.043	0.002	26.99

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-4: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Lufa 5M soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Lufa 5M	1.0	2.208	0.482	47.07	1.457	0.194	34.02
		2.269	0.468	48.66	1.541	0.19	32.09
	0.3	0.784	0.139	52.49	0.542	0.061	30.94
		0.784	0.14	51.95	0.546	0.06	30.37
	0.1	0.286	0.042	56.93	0.206	0.02	28.12
		0.285	0.042	56.87	0.205	0.02	28.03
	0.03	0.094	0.011	62.34	0.072	0.005	23.52
		0.094	0.011	62.27	0.071	0.006	24.86
	0.01	0.033	0.003	66.16	0.026	0.002	22.24
		0.033	0.003	65.73	0.026	0.002	20.79

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-5: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Lufa 2.2 soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Lufa 2.2	1.0	4.156	0.533	43.52	2.668	0.184	35.80
		4.189	0.525	44.21	2.707	0.185	35.39
	0.3	1.428	0.157	47.53	0.941	0.06	34.12
		1.435	0.157	47.54	0.951	0.06	33.77
	0.1	0.524	0.049	51.38	0.362	0.019	30.97
		0.532	0.049	51.86	0.37	0.019	30.47
	0.03	0.167	0.013	55.17	0.12	0.006	27.99
		0.1633	0.014	54.75	0.116	0.006	28.88
	0.01	0.0623	0.004	61.29	0.046	0.002	25.77
		0.0622	0.004	60.87	0.047	0.002	24.85

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-6: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Refesol 02-A soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Refesol 02-A	1.0	2.299	0.503	48.45	1.502	0.206	34.64
		2.329	0.496	48.85	1.538	0.023	33.96
	0.3	0.821	0.142	54.09	0.565	0.064	31.18
		0.798	0.148	52.41	0.538	0.064	32.59
	0.1	0.287	0.044	57.19	0.203	0.02	29.23
		0.287	0.044	57.12	0.205	0.02	28.85
	0.03	0.092	0.012	60.67	0.068	0.006	26.39
		0.092	0.012	60.48	0.068	0.006	26.23
	0.01	0.032	0.004	63.26	0.025	0.002	23.48
		0.032	0.004	62.97	0.025	0.002	22.92

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-7: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in CA-SL soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
CA-SL	1.0	0.864	0.583	36.70	0.531	0.203	38.5
		0.922	0.556	39.32	0.565	0.209	38.77
	0.3	0.329	0.169	43.37	0.215	0.065	34.67
		0.338	0.167	44.49	0.224	0.065	33.7
	0.1	0.126	0.05	49.53	0.088	0.021	29.83
		0.124	0.051	48.74	0.084	0.021	31.86
	0.03	0.037	0.015	49.07	0.026	0.006	31.08
		0.037	0.015	48.90	0.026	0.006	31.12
	0.01	0.013	0.005	52.76	0.01	0.002	28.16
		0.013	0.005	52.15	0.01	0.002	28.08

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-8: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Iowa soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Iowa	1.0	3.831	0.382	43.32	2.623	0.152	31.51
		4.316	0.475	49.26	2.9	0.185	32.81
	0.3	1.529	0.136	54.63	1.079	0.057	29.43
		1.522	0.136	54.50	1.081	0.057	28.96
	0.1	0.558	0.04	59.98	0.41	0.019	26.53
		0.557	0.041	59.38	0.409	0.019	26.59
	0.03	0.175	0.011	63.48	0.135	0.005	22.85
		0.169	0.011	61.08	0.128	0.005	24.02
	0.01	0.064	0.003	68.50	0.052	0.002	18.79
		0.066	0.003	69.29	0.054	0.001	18.47

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-9: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in LAD-SCL-PF soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
LAD-SCL-PF	1.0	1.886	0.537	43.41	1.207	0.206	36.02
		1.951	0.515	45.14	1.278	0.202	34.51
	0.3	0.645	0.159	46.68	0.427	0.064	33.78
		0.641	0.16	46.28	0.43	0.062	32.91
	0.1	0.223	0.051	48.61	0.151	0.021	32.14
		0.218	0.052	47.69	0.145	0.021	33.73
	0.03	0.069	0.015	50.10	0.048	0.006	30.67
		0.068	0.015	49.84	0.047	0.006	31.08
	0.01	0.023	0.005	50.23	0.016	0.002	30.43
		0.023	0.005	49.56	0.016	0.002	29.46

† % Desorbed compared to the adsorbed amount.

Table CA.B.8.1.6.1-10: Concentration of [^{14}C]-bixlozone in the solid and liquid phases at the end of the adsorption and desorption periods in Icklingham soil

	Concentration (mg/L)	Adsorption			Desorption		
		Soil (mg/kg)	Solution (mg/L)	Adsorbed (%)	Soil (mg/kg)	Solution (mg/L)	Desorbed (%)†
Icklingham	1.0	2.278	0.491	47.20	1.476	0.197	35.2
		2.332	0.472	48.32	1.55	0.199	33.54
	0.3	0.82	0.137	53.44	0.564	0.061	31.13
		0.822	0.137	53.54	0.567	0.061	30.99
	0.1	0.301	0.041	58.50	0.218	0.02	27.5
		0.3	0.041	58.39	0.217	0.02	27.82
	0.03	0.097	0.011	63.38	0.073	0.006	25.12
		0.098	0.011	63.86	0.075	0.005	23.5
	0.01	0.035	0.003	67.83	0.028	0.002	21.4
		0.036	0.003	68.97	0.028	0.002	21.09

† % Desorbed compared to the adsorbed amount.

The CA performed all relevant quality checks (OECD 106 evaluators checklist, November 2017) as part of confirming the acceptability of the study and of the reported endpoints. These checks confirmed that the % adsorption of 36.7-69.29 % was acceptable. The mass balance of the test substance was between 92.0 and 96.4 %. The acceptability of the analytical method was confirmed over the entire range of concentrations measured; the LOQ and LOD were acceptable. The use of the indirect method was appropriate based on a Kd * soil/solution ratio > 0.3 in all soils. The graphical fits of the Freundlich equation are presented below based on the standard linear regression form using log-log transformed data alongside the associated residual plots. The R^2 of the standard linear regression ranged from 0.997 to 1 and the visual fit of both the standard regression and the residual plots were acceptable. The K_{fE}/K_f ratio was less than 1.2 for all soils and is therefore considered acceptable. The values calculated by the CA are provided in Table CA.B.**8.1.6.1-11** and Table CA.B.**8.1.6.1-12**.

Table CA.B.8.1.6.1-11: Results table for the OECD 106 evaluators checklist calculated by the CA

Soil	Units	Lufa 6S	Lufa 5M	Lufa 2.2	Refesol 02-A
Adsorption method	-	Indirect	Indirect	Indirect	Indirect
Soil: solution ratio	g dw/mL	10:1	5:1	10:1	5:1
Mass balance of radioactivity (at highest tested conc.)	%	96.1	94.7	95.9	96.4
Adsorbed percentage	%	44.1-58.5	47.1-66.2	43.5-60.9	48.5-63.3
Kd x (soil: solution ratio)	-	0.80-1.48	0.92-2.20	0.78-1.56	0.91-1.60
^{ads}KF (95% confidence interval)	L/kg dw	7.356 (6.957-7.778)	4.079 (3.938-4.225)	7.138 (6.766-7.530)	4.408 (4.141-4.693)

$^{ads}1/n$ (95% confidence interval)	-	0.882 (0.866-0.897)	0.833 (0.823-0.842)	0.865 (0.849-0.880)	0.884 (0.866-0.901)
$^{ads}R^2$	-	1.0	1.0	1.0	0.999
$^{ads}K_{F,OC}$	L/kg OC	350.3	313.8	475.9	367.3
K_{fE}/K_f	-	1.093-1.094	1.118-1.122	1.102-1.105	1.083-1.084

Table CA.B.8.1.6.1-12: Results table for the OECD 106 evaluators checklist calculated by the CA

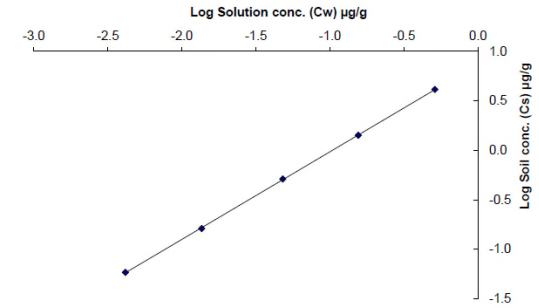
Soil	Units	CA-SL	Iowa	LAD-SCL-PF	Icklingham
Adsorption method	-	Indirect	Indirect	Indirect	Indirect
Soil: solution ratio	g dw/mL	1:2.5	1:10	1:5	1:5
Mass balance of radioactivity (at highest tested conc.)	%	93.6	92.0	92.8	95.3
Adsorbed percentage	%	36.7-52.76	43.32-69.29	43.41-50.23	47.20-68.97
Kd x (soil: solution ratio)	-	0.59-1.04	0.91-2.20	0.70-0.92	0.93-2.40
^{ads}KF (95% confidence interval)	L/kg dw	1.602 (1.407-1.824)	8.192 (7.600-8.831)	3.619 (3.446-3.801)	4.205 (4.030-4.388)
^{ads}1/n (95% confidence interval)	-	0.896 (0.858-0.934)	0.841 (0.821-0.861)	0.949 (0.935-0.964)	0.826 (0.815-0.838)
$^{ads}R^2$	-	0.997	0.999	1.000	1.000
$^{ads}K_{F,OC}$	L/kg OC	534.1	390.1	361.9	350.5
K_{fE}/K_f	-	1.186-1.203	1.156-1.193	1.189-1.202	1.104-1.109

The ^{ads}1/n and $^{ads}K_{FOC}$ values calculated by the CA are slightly different to those reported by the applicant however they are considered to be marginal and likely due to rounding errors. The study is considered acceptable by the CA and the applicants values will be used in the exposure modelling.

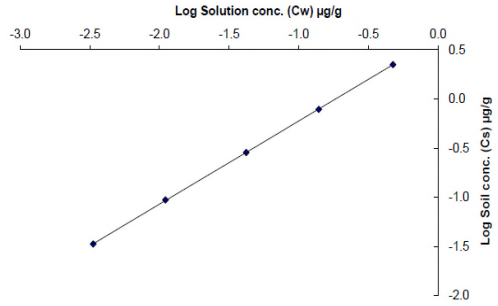
The applicant's adsorption and desorption parameters are provided in Table CA.B.**8.1.6.1-13**. Isotherms are shown in Figure CA.B.**8.1.6.1-3** below.

Figure CA.B.8.1.6.1-3: Freundlich isotherms for [^{14}C]-bixlozone in all soils

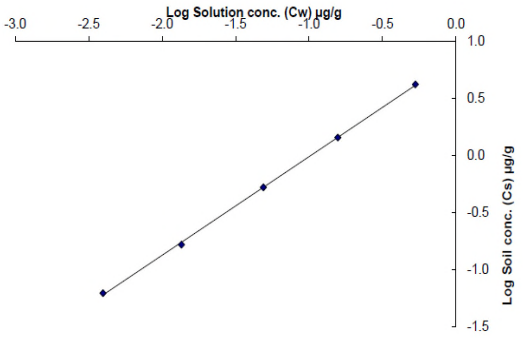
Lufa 6S



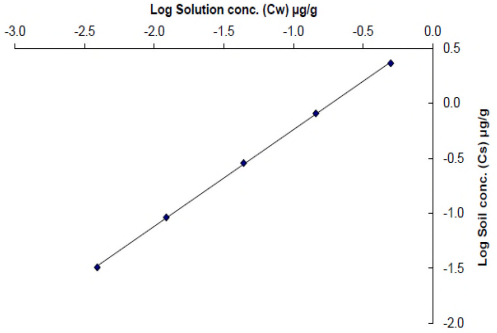
Lufa 5M



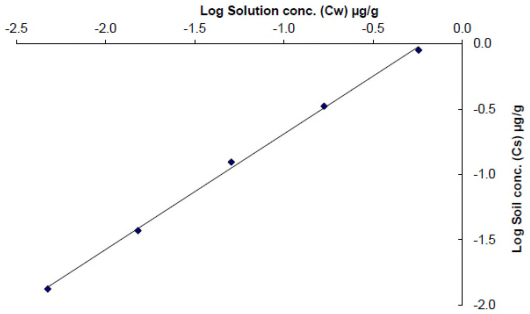
Lufa 2.2



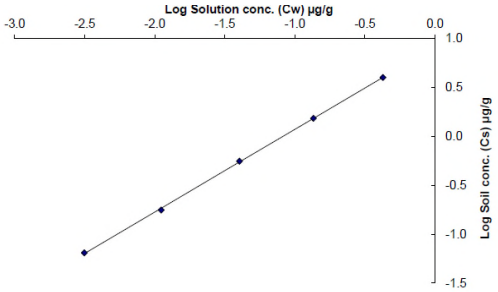
RefeSol 02-A



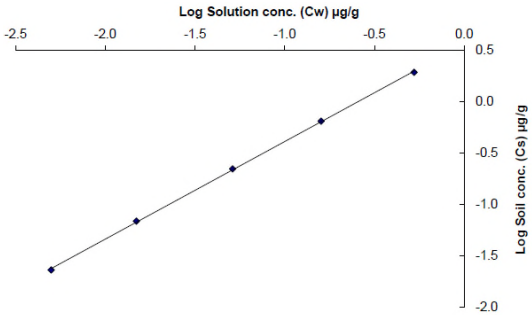
CA-SL



Iowa



LAD-SCL-PF



Icklingham

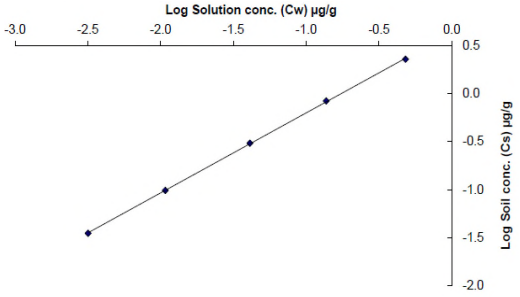


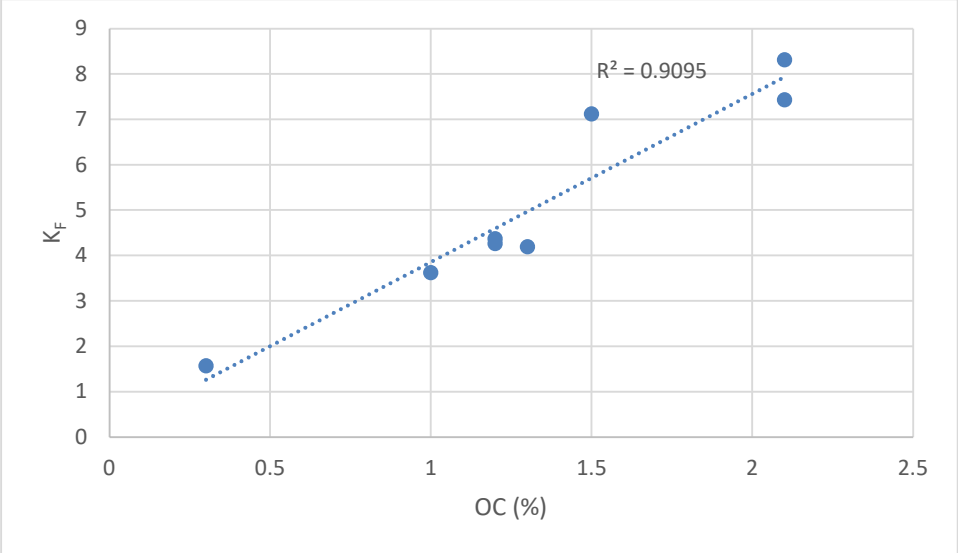
Table CA.B.8.1.6.1-13: Adsorption characteristics of [^{14}C]-bixlozone on eight soils

Soil	Organic carbon (%)	pH (0.01M $CaCl_2$)	Adsorption			Desorption		
			K_F^{ads}	K_{Foc}	1/n	K_F^{des}	$K_{Foc-des}$	1/n
Lufa 6S	2.1	6.9	7.43	352.9	0.885	10.9	518.8	0.890
Lufa 5M	1.3	7.2	4.19	334.2	0.846	6.03	481.0	0.854
Lufa 2.2	1.5	5.4	7.12	464.9	0.864	11.6	754.0	0.878
Refesol 02-A	1.2	6.1	4.37	364.1	0.879	6.07	505.7	0.874
CA-SL	0.3	6.9	1.57	458.4	0.885	2.37	692.8	0.884
Iowa	2.1	6.8	8.31	397.0	0.848	12.1	577.4	0.843
LAD-SCL-PF	1.0	8	3.62	354.8	0.949	5.66	554.2	0.943
Icklingham	1.2	7.4	4.26	348.1	0.832	5.94	485.5	0.843
Arithmetic mean					**0.874**		**571.2**	**0.876**
Geometric mean				**381.5**			**564.0**	

$K_F^{ads/des}$ = Freundlich adsorption/desorption distribution coefficient
K_{Foc} = Coefficient of adsorption per unit organic carbon, $K_{Foc-des}$ = Coefficient of desorption per unit organic carbon
1/n = Slope of the Freundlich adsorption isotherm

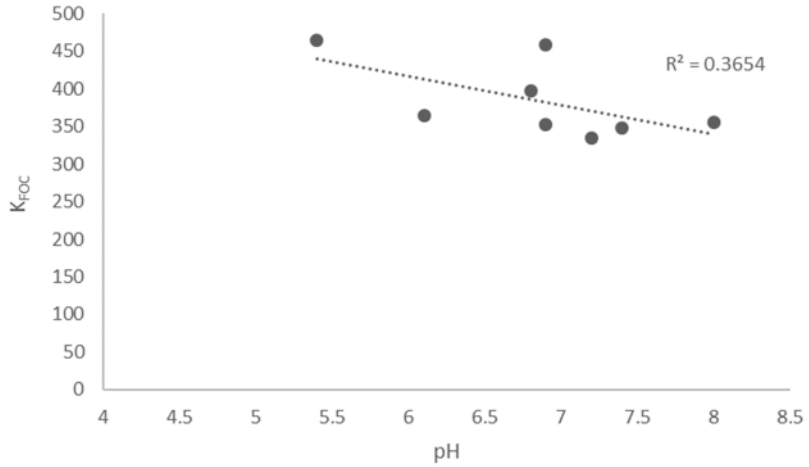
As can be seen in Figure CA.B.**8.1.6.1-4**, the K_F value increases with the organic carbon content, and therefore it is appropriate to calculate Kfoc values. The K_{OC} values for desorption are higher than those measured for adsorption, indicating a degree of irreversibility to the adsorption of bixlozone.

Figure CA.B.8.1.6.1-4: K_F compared with organic carbon



In order to explore the potential relationship between K_{OC} and pH, the CA has used the Kendall rank correlation coefficient to measure the association between K_{FOC} and pH. The results indicated that there was no statistically significant association (-0.327, p = 0.319). The CA also carried out a regression analysis which also showed no significant relationship (0.3654, p = 0.112), see Figure CA.B.**8.1.6.1-5**. This is as expected due to bixlozone's log Pow (3.3 - pH 4, 7 and 9, 20°C), water solubility (42.3, 39.6 and 41.9 mg/L at pH 4, 7 and 9 respectively (20 °C)) and it not containing any ionisable groups within environmentally relevant ranges.

Figure CA.B.8.1.6.1-5: K_{FOC} and pH correlation for bixlozone in all tested soils



CONCLUSION

The adsorption and desorption behaviour of [^{14}C]-bixlozone was studied in eight soils. Adsorption correlated well with organic carbon and K_{Foc} values for [^{14}C]-bixlozone were 334 – 465 mL/g (geometric mean 381.5 mL/g, arithmetic mean 1/n = 0.874) and desorption $K_{Foc\text{-}des}$ values were 481 – 754 mL/g (geometric mean 564 mL/g, arithmetic mean 1/n = 0.876), indicating that there is a degree of irreversibility to [^{14}C]-bixlozone adsorption. There was no evidence of any pH dependence.

CA.B.8.1.6.2. ***Bixlozone-3-OH-Propanamide Determination of Adsorption/Desorption Behaviour in Four Soils***

Report:	KCA 7.1.3.1.2/01, Gahm, F.; Kirchherr, M. (2017, amended 2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	F9600-3-OH Propanamide Adsorption/Desorption Behaviour in Four Soils
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01056, FMC Tracking no. 2016EFT-ISX2464
Guidelines:	OECD Guideline 106 (2000); US EPA Guideline, OPPTS 835-1230 (October 2008) SANCO/3029/99 rev.4
GLP:	Yes (laboratory certified by German National Authority)

CA Comments	Tier 1 mass balance failed in non-sterilised soil. Applicant then sterilised soil samples and repeated tier 1 study. The CA does not believe this to have affected the outcome of the study and considers it a reasonable adjustment. However, 3-OH is only a major metabolite under anaerobic conditions. Furthermore, as justification was provided and accepted excluding 3-OH anaerobic degradation from the exposure calculations, the results of this study are not considered further in the DAR. **This study is <u>not</u> relied upon.**

INTRODUCTION

The adsorption/desorption properties of bixlozone-3-OH-Propanamide, a metabolite of bixlozone, were determined in four different soils of European origin (LUFA 2.1, LUFA 2.4, St. Bauzille 12-060 and Fraunhofer Refesol 06-A) applying the batch equilibrium method according to OECD guideline 106 (2000).

Preliminary investigations were conducted to check for adsorption to the glass test vessels, to confirm the stability of the test item in 0.01M calcium chloride solution, to determine the soil to solution ratio to be used (1:1, 1:5 and 1:25) at an initial aqueous concentration of 1.0 mg/L, and to determine the time required for the compound to equilibrate between soil and water under both adsorption (2, 4, 6, 24 and 48 hours) and desorption conditions (2, 4, 6, 24 and 48 hours) at an initial aqueous concentration of 1.0 mg/L and a soil to solution ratio of 1:1. Initial testing indicated bixlozone-3-OH-propanamide was not stable in the presence of soil, therefore preliminary and definitive tests were performed under sterile conditions using γ-sterilised soil and autoclaved $CaCl_2$ solutions.

For the definitive test, the sterilised soils were pre-equilibrated overnight (minimum 12 hours) in 0.01M $CaCl_2$ solution (*ca* 18 mL) in glass flasks with PTFE sealed screw caps. Bixlozone-3-OH-propanamide dissolved in 0.01M $CaCl_2$ solution was added to four different soils (20 g oven-dried equivalents) and the solutions made up to 20 mL in total, to give initial nominal concentrations of 1.0, 0.5, 0.1, 0.05 and 0.01 mg/L. Test vessels were prepared in duplicate for each soil and each test item concentration. Test vessels were shaken for 24 hours (adsorption equilibrium time) in the dark at constant temperature (20 – 25°C). Following centrifugation of the adsorption solution, the decanted solution was replaced by an approximately equal volume of 0.01M $CaCl_2$ solution. Test vessels were then mixed for a further 24 hours (desorption equilibrium time).

Soil Collection

The study was carried out in four different soil types (LUFA 2.1, LUFA 2.4, St. Bauzille 12-060 and Fraunhofer Refesol 06-A) varying in their properties e.g. texture, pH, total organic carbon, or cation exchange capacity.

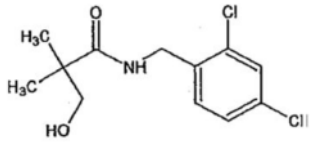
The soils 2.1, 2.4 and St. Bauzille 12-060 were delivered already sieved to a particle size ≤ 2 mm. The soil 06-A was sieved at the test facility to a particle size ≤ 2 mm. The soils were air-dried at the test facility at ambient temperature. The moisture content of the soils was determined by heating three aliquots at 105 °C until there was no significant change in weight (approx. 16 hours). For all calculations the mass of soil refers to oven dry mass, i.e. the weight of soil corrected for moisture content.

Table CA.B.8.1.6.2-1: Soil Properties

Soil Characterisation			**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**	**RefeSol 06-A**
Sampling location			Dudenhofen, Germany	Leimersheim, Germany	Herault, France	Schmallenberg, Germany
Date of collection			05 April 2016	05 April 2016	27 April 2016	14 March 2016
Sampling Depth (cm)			0 - 20 cm	0 - 20 cm	0 - 10 cm	0 - 25 cm
Storage conditions	Before drying		4 °C	4 °C	4 °C	4 °C
	After Drying		at ambient temperature	at ambient temperature	at ambient temperature	at ambient temperature
Particle size distribution		Sand (%)	86.0	34.5	13.7	12.5
		Silt (%)	10.5	40.6	46.0	38.8
		Clay (%)	3.5	24.9	40.4	48.7
Textural classification (USDA)			Loamy sand	Loam	Silty clay	Clay
pH ($CaCl_2$)			4.84	7.41	7.53	7.34
% Organic matter			1.17	3.26	3.62	4.52
% Organic carbon			0.68	1.89	2.10	2.62
CEC (meq/100g)			4.3	32.0	19.0	29.1
Bulk density, disturbed (g/L)			1447	1220	1210	1202
Maximum water holding capacity (%)			31.65	49.16	46.33	53.05
Pesticide use history at the collection site			None for previous 5 years	None for previous 5 years	None for previous 5 years	None for previous 4 years

Test compound details

Table CA.B.8.1.6.2-2: Test compound details

Name (IUPC):	Bixlozone-3-OH-Propanomide
Structure:	
Molecular weight:	276.2 g/mol
Batch Number:	ARD48P2
Radiochemical purity	98.5 % w/w
Storage:	Deep frozen (≤-18ºC) dark, dry

Preparation of Test Solutions

The stock solution containing 1152 mg/L bixlozone-3-OH-Propanamide in acetonitrile/water (1/1, v/v) was prepared by dissolving 11.7 mg of bixlozone-3-OH-Propanamide in 10 mL acetonitrile/water (1/1, v/v) and used as application solution in the non-sterile Tier 1 test.

An application solution containing 100 mg/L test item was prepared by diluting 1.736 mL stock solution with 0.01 M $CaCl_2$ to a final volume of 20 mL. The application solution was used for samples containing 1 mg/L (sterile Tier 1, Tier 2 and Tier 3 experiments) and 0.5 mg/L test item (Tier 3 experiment).

Furthermore two different application solutions were prepared for Tier 3 experiments in 0.01 M $CaCl_2$ containing 11 mg/L and 1 mg/L bixlozone-3-OH-Propanamide. Aliquots of these solutions were used for application of samples containing the following concentrations of the test item: 0.1, 0.05 and 0.01 mg/L bixlozone-3-OH-Propanamide in 0.01 M $CaCl_2$ solution.

The CA notes that the stock solution contains more than 1 % solvent. (4.34%). This is not considered to have affected the overall results of the experiment because the application solution was further diluted giving less than 0.1% (0.0037%) of solvent when applied to soil.

TIER 1 PRELIMINARY TEST METHOD

Selection of Optimal Soil/ Solution Ratios

Initial testing indicated bixlozone-3-OH-propanamide was not stable in the presence of soil, therefore the tests were also performed under sterile conditions using γ-sterilised soil and autoclaved $CaCl_2$ solutions in order to obtain an acceptable mass balance. Both methods are described below.

Non-sterile

Tier 1 was performed with four soils (2.1, 2.4, 12-060 and 06-A) at soil/solution ratios of 1/1, 1/5 and 1/25. The mass adsorbed to each soil at all samplings was determined by the indirect method (determination of test item concentration in the aqueous solution and calculation by difference of the test item concentration in soil). Each sample contained an actual initial concentration of 0.9396 mg/L test item.

For each of the four soils, three soil/solution ratios were used.
- 20 g soil and 20 cm^3 aqueous solution of the test substance (ratio 1/1)
- 10 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/5)
- 2 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/25)

The experiments of Tier 1, including controls, were performed with the serial method in duplicate.

43.4 µL of the test item solution were applied (corresponding to 50 µg test item) to the 1/5 and 1/25 samples and 17.36 µL (corresponding to 20 µg test item) of the test item solution were applied to the 1/1 samples for the non-sterile Tier 1 experiments (duplicates were used for each soil type).

The sampling times were 0 (only control samples) and 24 hours after application. Two control samples in 0.01 M $CaCl_2$ solution without soil were treated with the test item and were subjected to the same steps as the test systems, in order to check the stability of the test item in 0.01 M $CaCl_2$ solution and its possible adsorption on the surfaces of the test vessels.

Sterile

Based on the results of the Tier 1 test (mass balance < 90 %), the Tier 1 test was performed again with four soils (2.1, 2.4, 12-060 and 06-A). The mass adsorbed from the soil at all samplings was determined by the indirect method (determination of test item concentration in the aqueous solution and calculation by difference of the test item concentration in soil). Each sample contained an actual initial concentration of 1.0331 mg/L test item.

Four soils 2.1, 2.4, 12-060 and 06-A and three soil/solution ratios were used.
- 20 g soil and 20 cm^3 aqueous solution of the test substance (ratio 1/1);
- 10 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/5);
- 2 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/25);

The experimental procedure was identical to the non-sterile Tier 1 test except using sterile conditions (γ-sterilized soils and sterile 0.01 M $CaCl_2$ solution) and the following application procedure.

500 µL of the application solution (100 mg/L) were applied (corresponding to 50 µg test item) to the 1/5 and 1/25 samples and 200 µL (corresponding to 20 µg test item) of application solution were applied to the 1/1 samples for the sterile Tier 1 experiments (duplicates were used for each soil type).

During the adsorption test, the pH of the aqueous phase was measured before and after contact with the soil.

Mass balance

The mass balance was evaluated for all soil/solution ratios in Tier 1 (non-sterile and sterile) tests after 24 hours. For this purpose, the aqueous and the solid phase were separated by centrifugation. The aqueous phase was recovered as completely as possible before being analysed. The extraction of the soil was performed twice with each 80 mL of acetonitrile/water (80/20, v/v) at ambient temperature and shaking for 30 minutes. The extracts were separated from the soil by centrifugation at 2300 rpm for 4 minutes. The amount of the test item in the combined soil extracts was determined by HPLC-MS/MS and the mass balance was calculated.

TIER 1 RESULTS

Non-sterile mass balance

The mass balance was between 58.8 % and 97.7 % for soil 2.1, between 74.4 % and 101.2 % for soil 2.4, between 39.5 % and 97.2 % for soil 12-060 and between 17.2 % and 88.6 % for soil 06-A (see Table CA.B.**8.1.6.2-3**). Therefore, the test item was considered not stable for the time scale of the Tier 1 test. For this reason, Tier 1 was performed again under sterile conditions.

Table CA.B.8.1.6.2-3: Parental Mass balance after 24 hours of agitation (Tier 1 Non-sterile)

Sample	Ratio	Replicate	Recovery [%]	Mean Recovery [%]
Lufa 2.1	1/1	1	56.3	58.8
		2	61.4	
	1/5	1	78.9	78.7
		2	78.4	
	1/25	1	97.6	97.7
		2	97.7	
Lufa 2.4	1/1	1	71.5	74.4
		2	77.3	
	1/5	1	84.0	87.7
		2	91.3	
	1/25	1	101.4	101.2
		2	100.9	
St. Bauzille 12-060	1/1	1	38.9	39.5
		2	40.1	
	1/5	1	70.1	70.0
		2	69.9	
	1/25	1	96.8	97.2
		2	97.6	
Refesol 06-A	1/1	1	30.8	17.2
		2	3.7	
	1/5	1	62.1	47.5
		2	32.8	
	1/25	1	90.7	88.6
		2	86.5	

Sterile Mass balance

The amount of the test item in the soil extracts and the aqueous solution was determined and the mass balance was calculated. The mass balance was between 99.0 % and 100.9 % for soil 2.1, between 100.3 % and 103.8 % for soil 2.4, between 96.5 % and 102.2 % for soil 12-060 and between 98.2 % and 103.1 % for soil 06-A (see Table CA.B.**8.1.6.2-4**). Therefore, the test item was considered stable for the time scale of the test. For this reason, Tier 2 and Tier 3 were performed under sterile conditions.

Table CA.B.8.1.6.2-4: Parental Mass Balance after 24 hours of agitation (Tier 1 Sterile)

Sample	Ratio	Replicate	Recovery [%]	Mean recovery [%]
Lufa 2.1	1/1	1	99.2	100.9
		2	102.5	
	1/5	1	97.9	99.0
		2	100.2	
	1/25	1	99.6	99.3
		2	99.1	
Lufa 2.4	1/1	1	101.7	103.3
		2	104.9	
	1/5	1	101.7	103.8
		2	106.0	
	1/25	1	101.5	100.3
		2	99.2	
St. Bauzille 12-060	1/1	1	94.3	96.5
		2	98.7	
	1/5	1	102.7	102.2
		2	101.7	
	1/25	1	99.3	99.9
		2	100.5	
Refesol 06-A	1/1	1	99.9	98.2
		2	96.6	
	1/5	1	102.2	103.1
		2	104.1	
	1/25	1	101.8	102.7
		2	103.7	

The sterilised soil samples gave much improved mean recoveries. The CA agrees with the applicant's decision to use sterilised soil to gain adequate figures for mass balance recovery. The CA does not believe this to have affected the outcome of the study and considers it a reasonable adjustment.

Stability of the Test item in 0.01 M $CaCl_2$ solution and possible Adsorption on Test Vessel Surface

In the two control samples (only 0.01 M $CaCl_2$ solution containing the test item without soil) of the Tier 1 experiment, the mean recovery was 101.0 % after 24 hours of application. Therefore, the test item was stable in solution without soil during the entire experimental period. Furthermore, no adsorption of the test item to the surface of the test vessel was observed after 24 hours of agitation (see Table CA.B.**8.1.6.2**-**5**).

Table CA.B.8.1.6.2-5: Stability of bixlozone-OH-Propanamide in 0.01 M $CaCl_2$ Solution (tier 1 sterile)

	Sampling Interval [Hours]	Recovery Single Values [% of applied]	Mean Recovery [% of applied]
Control	0	100	100
		100	
	24	101	101
		102	

Determination of Appropriate Soil Solution Ratio

The adsorption at soil/solution ratio 1/25 was 2.7 % for soil 2.1, 3.2 % for soil 2.4, 5.3 % for soil 12-060 and 6.6 % for soil 06-A. At a soil/solution ratio of 1/5 the adsorption was 12.1 % for soil 2.1, 21.7 % for soil 2.4, 18.8 % for soil 12-060 and 32.2 % for soil 06-A. At a soil/solution ratio of 1/1 the adsorption was 38.7 % for soil 2.1, 59.2 % for soil 2.4, 59.2 % for soil 12-060 and 70.2 % for soil 06-A.

Table CA.B.8.1.6.2-6: (Tier 1 sterile) Concentration of test item in supernatant and adsorption coefficient (CA calculation)

Sample	Ratio	replicate	Concentration [µg/mL]	% adsorption
2.1	1/1	1	0.638	38.3
		2	0.628	39.2
	1/5	1	0.909	12.0
		2	0.906	12.3
	1/25	1	1.005	2.73
		2	1.005	2.73
2.4	1/1	1	0.450	56.4
		2	0.393	62.0
	1/5	1	0.830	19.7
		2	0.787	23.8
	1/25	1	1	3.20
		2	1	3.20
12-060	1/1	1	0.415	59.8
		2	0.428	58.6
	1/5	1	0.843	18.4
		2	0.836	19.1
	1/25	1	0.997	3.52
		2	0.959	7.15
06-A	1/1	1	0.314	69.6
		2	0.302	70.8
	1/5	1	0.701	32.2
		2	0.700	32.3
	1/25	1	0.990	4.15
		2	0.939	9.09

The % adsorption given in Table CA.B.**8.1.6.2-6** were calculated by the CA and concordant with the applicant's calculations. The highest rate of adsorption was at a ratio of 1/1 in each sample and is above the OECD guidance requirement that % adsorption is above 20 %, preferably 50 %. The CA agrees with the % adsorption ratio of 1/1 selected by the applicant during Tier 2.

TIER 1 CONCLUSION

After sterilisation of the soil samples the reported mass balance was concordant with OECD requirements (mass balance >90%). Stability of the test item in 0.01M $CaCl_2$ was proven for longer that the 24 hour test period, and no adsorption of the test item to the surface of the test vessel was observed. For the following higher tier experiments as soil: solution ratio of 1:1 was selected.

TIER 2 METHOD

Determination of Equilibration Time

Based on the results of Tier 1 experiments, the Tier 2 test was performed at a soil/solution ratio of 1/1 (20 g soil and 20 cm^3 aqueous solution) for all soils (2.1, 2.4, 12-060, 06-A) using the serial method in duplicate under sterile conditions by using γ-sterilized soils and autoclaving the 0.01 M aqueous $CaCl_2$ solution.

An aliquot of 0.200 mL test item solution at 100 mg/L was applied (corresponding to 20 µg test item) to each pre-equilibrated sample (duplicates were used for each soil type) and shaken at 150 rpm at a constant temperature between 20-25°C. The actual initial concentration was 1.0575 mg/L test item.

The sampling times for both adsorption and desorption were 0 (only control samples), 2, 4, 6, 24 and 48 hours after application. The desorption experiments were performed following a 24 hour adsorption phase.

Two control samples containing 0.01 M $CaCl_2$ solution were treated with the test item and were subjected to the same steps as the test systems, in order to check the stability of the test item in 0.01 M $CaCl_2$ solution. Furthermore, two blank systems (no test item) for each soil at a soil /solution ratio of 1/1 were prepared and treated in the same manner as the spiked soil samples. An aliquot of the aqueous phase of each blank was measured by LC-MS/MS.

TIER 2 RESULTS

The measured concentrations in supernatant are shown in Table CA.B.**8.1.6.2-7** and the results of % adsorption in shown in Table CA.B.**8.1.6.2-8**.

Table CA.B.8.1.6.2-7: Concentration of Test Item in Supernatant for soil to solution ratio 1/1 during Tier 2 Adsorption Experiment.

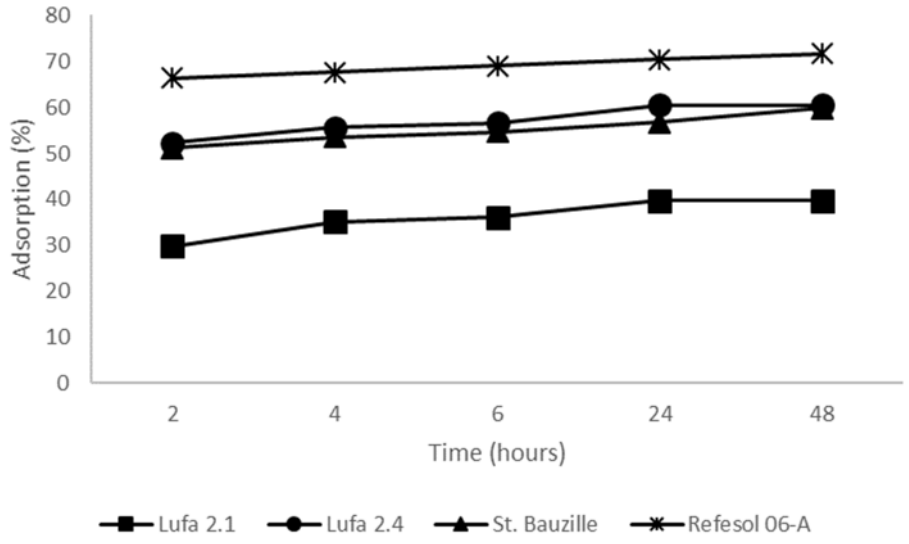
Lufa 2.1		**Concentration [µg/mL]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	0.7313	0.6742	0.6641	0.6427	0.6346
	Sample 2	0.7527	0.6999	0.6901	0.6385	0.6505
Lufa 2.4		**Concentration [µg/mL]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	0.5117	0.4931	0.4402	0.4234	0.4372
	Sample 2	0.4968	0.4488	0.4824	0.4169	0.4023
St. Bauzille 12-060		**Concentration [µg/mL]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	0.5116	0.4847	0.4789	0.4505	0.4206
	Sample 2	0.5202	0.4984	0.4831	0.4678	0.4341
Refesol 06-A		**Concentration [µg/mL]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	0.3511	0.3422	0.3281	0.3255	0.3126
	Sample 2	0.3586	0.3454	0.3281	0.3050	0.2908
Control		**Concentration [µg/mL]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Control	Sample 1	1.0679	1.0825	1.0874	1.0507	1.0849
	Sample 2	1.0140	1.0697	1.0480	1.0381	1.0741

Table CA.B.8.1.6.2-8: Time Dependent course of Adsorption for soil to solution Ratio 1/1 (soils 2.1, 2.4, 12-060 and 06-A) Applicant calculations

Lufa 2.1		**Adsorption [% of applied]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	30.8	36.4	37.5	39.6	40.5
	Sample 2	28.8	33.9	35.0	40.0	39.0
	Mean	29.8	35.2	36.2	39.8	39.7
Lufa 2.4		**Adsorption [% of applied]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	51.6	53.5	58.5	60.2	59.0
	Sample 2	53.0	57.6	54.6	60.8	62.3
	Mean	52.3	55.6	56.6	60.5	60.6
St. Bauzille 12-060		**Adsorption [% of applied]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	51.6	54.3	54.9	57.7	60.5
	Sample 2	50.8	53.0	54.5	56.0	59.3
	Mean	51.2	53.6	54.7	56.8	59.9
Refesol 06-A		**Adsorption [% of applied]**				
Time [h]		**2**	**4**	**6**	**24**	**48**
Ratio 1/1	Sample 1	66.8	67.7	69.1	69.4	70.7
	Sample 2	66.1	67.4	69.1	71.3	72.7
	Mean	66.4	67.6	69.1	70.4	71.7

The CA has independently calculated the applicant figures for adsorption and agrees with those stated figures in Table CA.B.8.1.2.2-8 above. There is marginal difference between the % adsorption at 24 and 48 hours and therefore the CA agrees that 24 hours is the most acceptable equilibration time (see Figure CA.B.**8.1.6.2-1**).

Figure CA.B.8.1.6.2-1: Adsorption equilibrium time (preliminary experiment) of bixlozone-3-OH-Propanamide



Mass Balance

Based on the results of Tier 1 the mass balance was evaluated for Tier 2 at soil/solution ratios of 1/1 for all soils after 48 hours adsorption. The extraction of the soil was performed twice with each 80 mL of acetonitrile/water (80/20, v/v) at ambient temperature and shaking for 30 minutes. The extracts were separated from the soil by centrifugation at 2300 rpm for 4 minutes. The amount of the test item in the combined soil extracts was determined by HPLC-MS/MS and the mass balance was calculated. The amount of the test item in the supernatant and the soil extracts was determined and the mass balance was calculated. The mass balance was between 90.1 % and 101.2 % (see Table CA.B.**8.1.6.2-9**). The test item was considered to be stable in the time scale of the test under sterile conditions.

Table CA.B.8.1.6.2-9: Parental Mass Balance after 48 hours of Agitation (tier 2 Adsorption)

Sample	Ratio	Replicate	Recovery [%]	Mean recovery [%]
Lufa 2.1	1/1	1	101.1	101.2
		2	101.4	
Lufa 2.4	1/1	1	97.7	99.2
		2	100.6	
St. Bauzille 12.060	1/1	1	90.1	90.1
		2	n/a	
Refesol 06-A	1/1	1	94.4	94.3
		2	94.1	

TIER 2 CONCLUSION

The mass balance of tier 2 adsorption was considered stable. The applicant chose an equilibration time of 24 hours. In each soil experiment, 48 hours showed the highest % adsorption however the difference between % adsorption at 24 hrs and 48 hrs was negligible. The CA accepts the applicants decision of 24 hr equilibration time.

TIER 3 METHOD

The samples contained actual initial amounts of 1.148, 0.5728, 0.1097, 0.0553 and 0.0097 µg/mL test item in 0.01 M sterilized $CaCl_2$, covering two orders of magnitude. The resulting aqueous equilibrium concentrations and the water solubility were taken into account when choosing these concentrations. The Tier 3 adsorption test was performed with all four soils at a soil/solution ratio of 1/1 (20 g soil and 20 cm^3 aqueous solution) for all soils (2.1, 2.4, 12-060, 06-A) using the serial method in duplicate under sterile conditions by using γ-sterilised soils and autoclaving the 0.01 M aqueous $CaCl_2$ solution. The desorption test followed using the soil samples from adsorption by adding 0.01 M $CaCl_2$ to re-adjust to 20 cm^3 aqueous solution.

The adsorption and desorption tests were performed as follows.

Each experiment (one soil and five treatment levels) was done with pre-equilibrated soils in duplicate to allow estimation of the variance of the results. Furthermore, two control samples were prepared containing 0.01 M $CaCl_2$ solution without soil.

The test was performed using the serial method in 100 mL glass bottles with PTFE sealed screw caps. Agitation was performed at a constant temperature between 20-25°C on a flatbed shaker with a frequency of around 150 rpm to keep the soil dispersed in the aqueous phase.

After 24 h the suspensions were centrifuged at 2300 rpm for 4 minutes. The aqueous phase was recovered as completely as possible. Then, the desorption test followed using the samples from adsorption by adding 0.01 M $CaCl_2$ to re-adjust to 20 cm^3.

The sampling times were 24 hours after application for the adsorption test and 24 h after adsorption for the desorption test.

The adsorbed mass per unit mass of soil was plotted as a function of the equilibrium concentration of the test item. The logarithm of the adsorbed mass was plotted as a function of the logarithm of the equilibrium concentration of the test item. The desorbed mass per unit mass of soil was plotted as a function of the equilibrium concentration of the test item. The logarithm of the desorbed mass was plotted as a function of the logarithm of the equilibrium concentration of the test item.

RESULTS

Table CA.B.8.1.6.2-10: Concentration and Logarithm of Concentration of bixlozone-3-OHPropanamide at Adsorption Equilibrium after 24 hours in the Water Phase and bound to Soil Lufa 2.1 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
1.15	Sample 1	0.660	0.488	-0.180	-0.312
	Sample 2	0.663	0.485	-0.178	-0.315
	Mean	0.662	0.486	-0.179	-0.313
0.573	Sample 1	0.329	0.244	-0.483	-0.613
	Sample 2	0.313	0.260	-0.504	-0.586
	Mean	0.321	0.252	-0.493	-0.599
0.110	Sample 1	0.0538	0.0559	-1.27	-1.25
	Sample 2	0.0584	0.0513	-1.23	-1.29
	Mean	0.0561	0.0536	-1.25	-1.27
0.0553	Sample 1	0.0275	0.0277	-1.56	-1.56
	Sample 2	0.0276	0.0276	-1.56	-1.56
	Mean	0.0276	0.0277	-1.56	-1.56
0.00969	Sample 1	0.00472	0.00497	-2.33	-2.30
	Sample 2	0.00476	0.00493	-2.32	-2.31
	Mean	0.00474	0.00495	-2.32	-2.31

Table CA.B.8.1.6.2-11: Concentration and Logarithm of Concentration of bixlozone-3-OHPropanamide at Adsorption Equilibrium after 24 hours in the Water Phase and bound to Soil Lufa 2.4 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
1.15	Sample 1	0.491	0.657	-0.309	-0.183
	Sample 2	0.469	0.679	-0.329	-0.168
	Mean	0.480	0.668	-0.319	-0.176
0.573	Sample 1	0.242	0.331	-0.616	-0.481
	Sample 2	0.236	0.337	-0.628	-0.472
	Mean	0.239	0.334	-0.622	-0.476
0.110	Sample 1	0.0410	0.0687	-1.39	-1.16
	Sample 2	0.0408	0.0689	-1.39	-1.16
	Mean	0.0409	0.0688	-1.39	-1.16
0.0553	Sample 1	0.0178	0.0374	-1.75	-1.43
	Sample 2	0.0172	0.0381	-1.76	-1.42
	Mean	0.0175	0.0378	-1.76	-1.42
0.00969	Sample 1	0.00353	0.00616	-2.45	-2.21
	Sample 2	0.00291	0.00678	-2.54	-2.17
	Mean	0.00322	0.00647	-2.49	-2.19

Table CA.B.8.1.6.2-12: Concentration and Logarithm of Concentration of bixlozone-3-OHPropanamide at Adsorption Equilibrium after 24 hours in the Water Phase and bound to Soil St. Bauzille 12-060 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
1.15	Sample 1	0.462	0.686	-0.335	-0.164
	Sample 2	0.468	0.680	-0.330	-0.167
	Mean	0.465	0.683	-0.333	-0.166
0.573	Sample 1	0.224	0.349	-0.650	-0.458
	Sample 2	0.229	0.343	-0.639	-0.464
	Mean	0.227	0.346	-0.644	-0.461
0.110	Sample 1	0.0421	0.0676	-1.38	-1.17
	Sample 2	0.0397	0.0700	-1.40	-1.15
	Mean	0.0409	0.0688	-1.39	-1.16
0.0553	Sample 1	0.0177	0.0376	-1.75	-1.42
	Sample 2	0.0189	0.0364	-1.72	-1.44
	Mean	0.0183	0.0370	-1.74	-1.43
0.00969	Sample 1	0.00299	0.00670	-2.52	-2.17
	Sample 2	0.00309	0.00660	-2.51	-2.18
	Mean	0.00304	0.00665	-2.52	-2.18

Table CA.B.8.1.6.2-13: Concentration and Logarithm of Concentration of bixlozone-3-OHPropanamide at Adsorption Equilibrium after 24 hours in the Water Phase and bound to Soil Refesol 06-A for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
1.15	Sample 1	0.331	0.817	-0.481	-0.0876
	Sample 2	0.316	0.832	-0.501	-0.0797
	Mean	0.323	0.825	-0.491	-0.0836
0.573	Sample 1	0.158	0.415	-0.801	-0.382
	Sample 2	0.153	0.420	-0.815	-0.377
	Mean	0.156	0.417	-0.808	-0.380
0.110	Sample 1	0.0263	0.0834	-1.58	-1.08
	Sample 2	0.0274	0.0823	-1.56	-1.08
	Mean	0.0269	0.0829	-1.57	-1.08
0.0553	Sample 1	0.0133	0.0420	-1.88	-1.38
	Sample 2	0.0142	0.0411	-1.85	-1.39
	Mean	0.0137	0.0415	-1.86	-1.38
0.00969	Sample 1	0.00228	0.00741	-2.64	-2.13
	Sample 2	0.00228	0.00741	-2.64	-2.13
	Mean	0.00228	0.00741	-2.64	-2.13

Table CA.B.8.1.6.2-14: Concentration and Logarithm of Concentration of bixlozone-3-OH-Propanamide at Desorption Equilibrium after 24 hours in the Water Phase and bound to Soil Lufa 2.1 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
0.682	Sample 1	0.355	0.328	-0.450	-0.484
	Sample 2	0.342	0.329	-0.466	-0.483
	Mean	0.348	0.329	-0.458	-0.483
0.343	Sample 1	0.180	0.157	-0.745	-0.805
	Sample 2	0.173	0.178	-0.763	-0.750
	Mean	0.176	0.167	-0.754	-0.777
0.0819	Sample 1	0.0337	0.0508	-1.47	-1.29
	Sample 2	0.0349	0.0463	-1.46	-1.33
	Mean	0.0343	0.0485	-1.46	-1.31
0.0365	Sample 1	0.0160	0.0195	-1.80	-1.71
	Sample 2	0.0166	0.0203	-1.78	-1.69
	Mean	0.0163	0.0199	-1.79	-1.70
0.00632	Sample 1	0.00296	0.00338	-2.53	-2.47
	Sample 2	0.00282	0.00353	-2.55	-2.45
	Mean	0.00289	0.00345	-2.54	-2.46

Table CA.B.8.1.6.2-15: Concentration and Logarithm of Concentration of bixlozone-3-OH Propanamide at Desorption Equilibrium after 24 hours in the Water Phase and bound to Soil Lufa 2.4 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
0.910	Sample 1	0.335	0.572	-0.475	-0.243
	Sample 2	0.356	0.557	-0.449	-0.254
	Mean	0.345	0.564	-0.462	-0.248
0.456	Sample 1	0.166	0.285	-0.779	-0.545
	Sample 2	0.173	0.284	-0.762	-0.546
	Mean	0.170	0.285	-0.771	-0.546
0.0889	Sample 1	0.0298	0.0606	-1.53	-1.22
	Sample 2	0.0311	0.0586	-1.51	-1.23
	Mean	0.0305	0.0596	-1.52	-1.22
0.0466	Sample 1	0.0124	0.0338	-1.91	-1.47
	Sample 2	0.0129	0.0337	-1.89	-1.47
	Mean	0.0126	0.0337	-1.90	-1.47
0.00795	Sample 1	0.00242	0.00552	-2.62	-2.26
	Sample 2	0.00205	0.00618	-2.69	-2.21
	Mean	0.00224	0.00585	-2.65	-2.23

Table CA.B.8.1.6.2-16: Concentration and Logarithm of Concentration of bixlozone-3-OH Propanamide at Desorption Equilibrium after 24 hours in the Water Phase and bound to Soil St. Bauzille 12-060 for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
0.896	Sample 1	0.345	0.560	-0.462	-0.252
	Sample 2	0.324	0.570	-0.489	-0.244
	Mean	0.335	0.565	-0.475	-0.248
0.457	Sample 1	0.166	0.291	-0.780	-0.535
	Sample 2	0.165	0.285	-0.783	-0.546
	Mean	0.165	0.288	-0.781	-0.541
0.0872	Sample 1	0.0321	0.0547	-1.49	-1.26
	Sample 2	0.0294	0.0584	-1.53	-1.23
	Mean	0.0308	0.0566	-1.51	-1.25
0.0457	Sample 1	0.0138	0.0321	-1.86	-1.49
	Sample 2	0.0136	0.0314	-1.87	-1.50
	Mean	0.0137	0.0318	-1.86	-1.50
0.00800	Sample 1	0.00233	0.00584	-2.63	-2.23
	Sample 2	0.00224	0.00586	-2.65	-2.23
	Mean	0.00224	0.00585	-2.64	-2.23

Table CA.B.8.1.6.2-17: Concentration and Logarithm of Concentration of bixlozone-3-OH Propanamide at Desorption Equilibrium after 24 hours in the Water Phase and bound to Soil Refesol 06-A for Soil to Solution Ratio 1/1

C_0 [µg/mL]		C ads aq [µg/mL]	C ads s [µg/g]	log C ads aq [µg/mL]	log C ads s [µg/g]
0.996	Sample 1	0.250	0.754	-0.603	-0.122
	Sample 2	0.257	0.742	-0.590	-0.129
	Mean	0.253	0.748	-0.596	-0.126
0.503	Sample 1	0.131	0.372	-0.884	-0.430
	Sample 2	0.121	0.383	-0.916	-0.417
	Mean	0.126	0.378	-0.900	-0.423
0.0972	Sample 1	0.0238	0.0739	-1.62	-1.13
	Sample 2	0.0222	0.0751	-1.65	-1.12
	Mean	0.0230	0.0745	-1.64	-1.13
0.0487	Sample 1	0.0110	0.0382	-1.96	-1.42
	Sample 2	0.0109	0.0378	-1.96	-1.42
	Mean	0.0109	0.0380	-1.96	-1.42
0.00867	Sample 1	0.00189	0.00675	-2.72	-2.17
	Sample 2	0.00189	0.00682	-2.72	-2.17
	Mean	0.00189	0.00678	-2.72	-2.17

DISCUSSION

Both the Freundlich Adsorption and Desorption Coefficients calculated by the Applicant are provided in Table CA.B.**8.1.6.2-18** and Table CA.B.**8.1.6.2-19**. For all included soils the CA was able to replicate evaluation figures within reason.

Table CA.B.8.1.6.2-18: Freundlich Adsorption Coefficients (Applicant Figures)

Parameters	**Lufa 2.1**	**Lufa 2.4**	**St. Bauzille 12-060**	**Refesol 06-A**
Slope (1/n)	0.924	0.908	0.916	0.951
Intercept (log K ads F)	-0.14	0.11	0.13	0.39
K ads F [$\mu g^{1-1/n}(cm^3)^{1/n}g^{-1}$]	0.73	1.3	1.4	2.5
R Squared	0.9987	0.9962	0.9991	0.9995
% oc	0.68	1.89	2.10	2.62
K ads Foc[$^{\mu g1-1/n}(cm^3)^{1/n}g^{-1}$]	107	68	65	94

Table CA.B.8.1.6.2-19: Freundlich Desorption Coefficients (Applicant Figures)

Parameters	**Lufa2.1**	**Lufa 2.4**	**St. Bauzille 12-060**	**Refesol 06-A**
Slope (1/n)	0.938	0.891	0.911	0.957
Intercept (log K ads F)	-0.03	0.16	0.17	0.44
K ads F [$\mu g^{1-1/n}(cm^3)^{1/n}g^{-1}$]	0.93	1.4	1.5	2.8
R Squared	0.9938	0.996	0.9984	0.9996
% oc	0.68	1.89	2.10	2.62
K ads Foc[$^{\mu g1-1/n}(cm^3)^{1/n}g^{-1}$]	136	76	71	106

The CA performed all relevant quality checks (OECD 106 evaluators checklist, November 2017) as part of confirming the acceptability of the study and of the reported endpoints. For all soils, the quality criteria were met and the results are summarised in Table CA.B.**8.1.6.2-20**.

Table CA.B.8.1.6.2-20: Summary Results Table (values calculated by CA, OECD 106 evaluators checklist, November 2017)

Soil	Units	LUFA 2.1	LUFA 2.4	St Bauzille	Refsol 06-A
Adsorption method (direct/ indirect)	-	indirect	indirect	indirect	indirect
Soil:solution Ratio	(g dw/ml)	1/1	1/1	1/1	1/1
Mass balance of ^{14}C (at all tested conc)	%	101.1-101.4	97.7-100.6	90.1	94.1-94.4
Adsorbed percentage	%	42.2-51.3	57.2-70.0	59.23-69.2	71.2-76.5
Kd * Soil/ Solution		0.73-1.05	1.34-2.33	1.45-2.24	2.47-3.25
adsKF (95% confidence interval [lower-upper])	L/kg dw	0.731 (0.669-0.797)	1.287 (1.095-1.513)	1.358 (1.255-1.470)	2.455 (2.297-2.624)
ads 1/n (95% confidence interval)	-	0.924 (0.896-0.951)	0.908 (0.862-0.954)	0.916 (0.862-0.954)	0.950 (0.933-0.968)
ads R^2	-	0.999	0.996	0.999	0.999
ads K_{FOC}	L/kg OC	107.4	68.1	64.7	93.7
K_{FE}/K_F	-	1	1.01	1.20	1.09

The values calculated by the CA in Table CA.B.8.1.6.2-20 are slightly different to those reported by the applicant however they are considered to be marginal and due to rounding errors. The study is considered acceptable by the CA and the applicants values will be used in the exposure modelling (see Table CA.B.8.1.6.2-21).

Table CA.B.8.1.6.2-21: Calculation of arithmetic mean and geomean for adsorption and desorption (applicant values)

Soil	KF,ads	KFoc,ads	1/n	$K_{F,des}$	$K_{Foc,des}$	1/n	% OC	pH ($CaCl_2$)
Lufa 2.1	0.73	107	0.924	0.93	136	0.938	0.68	4.84
Lufa 2.4	1.3	68	0.908	1.4	76	0.891	1.89	7.41
St Bauzille	1.4	65	0.916	1.5	71	0.911	2.10	7.53
Refsol 06-A	2.5	94	0.951	2.8	106	0.957	2.62	7.34
Arithmetic mean	-	-	0.925	-	-	0.924	-	-
Geomean	1.35	81.7	-	1.53	93.9	-	-	-

Figure CA.B.8.1.6.2-2: Freundlich Adsorption Plots: Lufa 2.1 (calculated by CA)

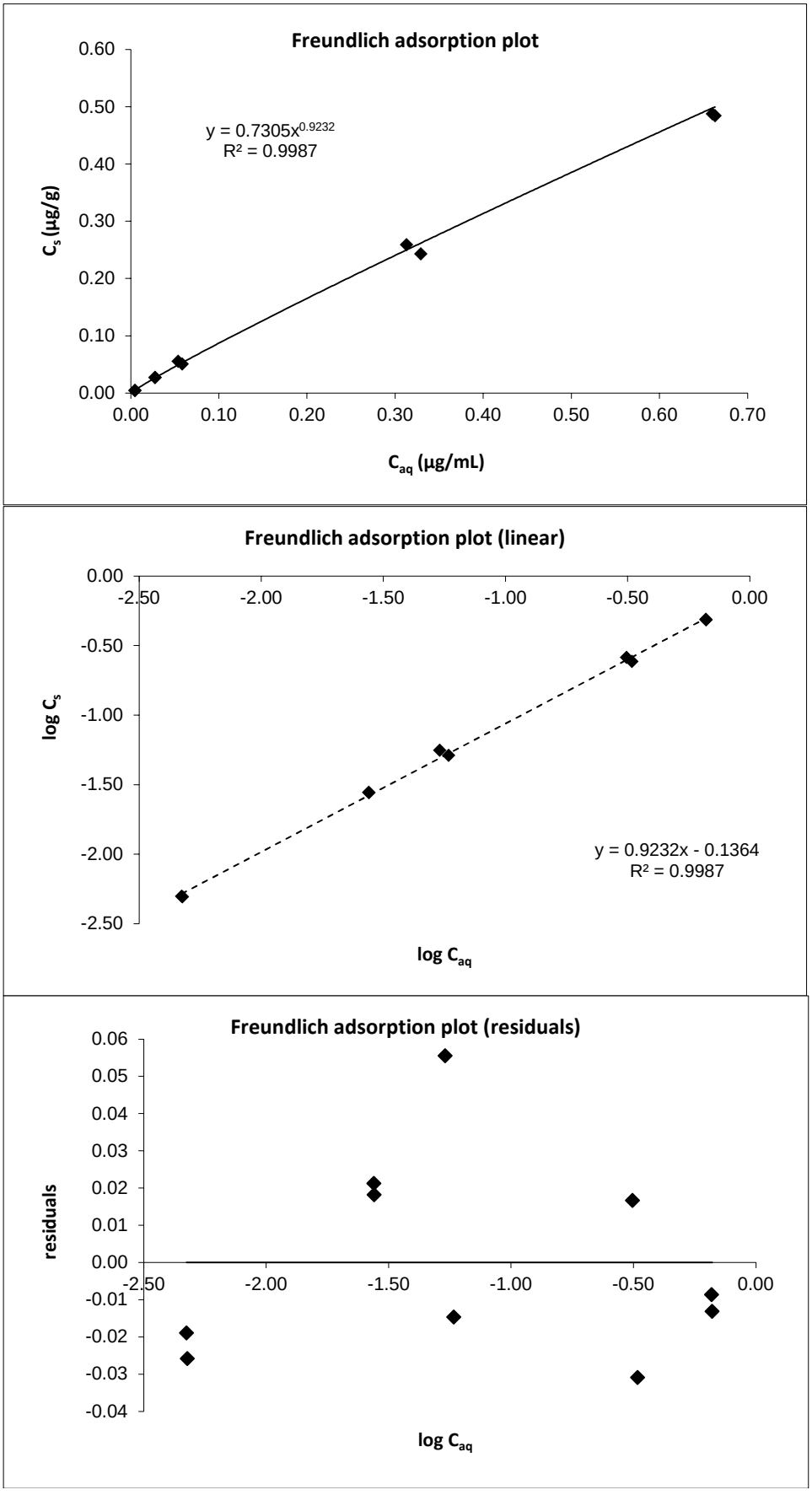
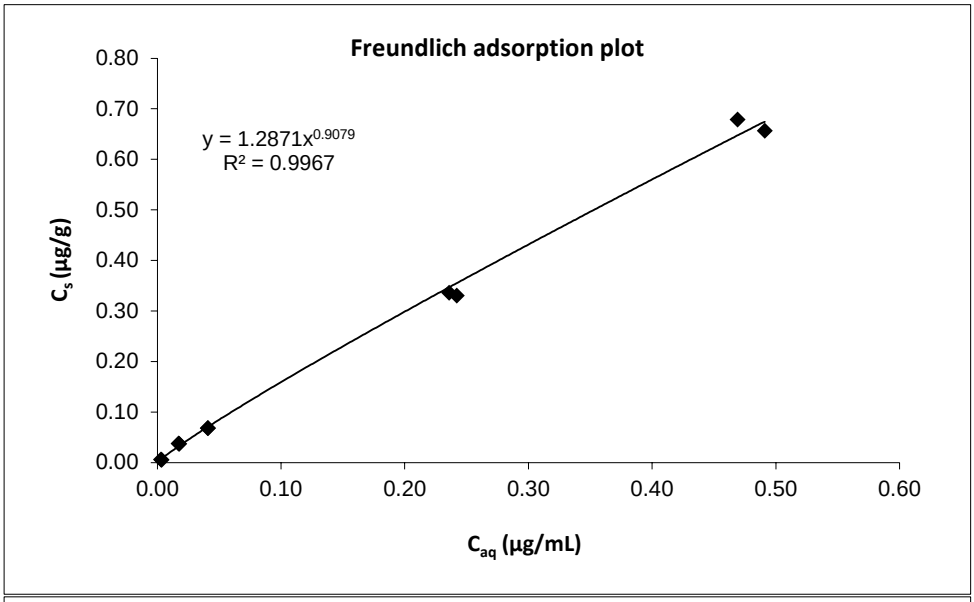
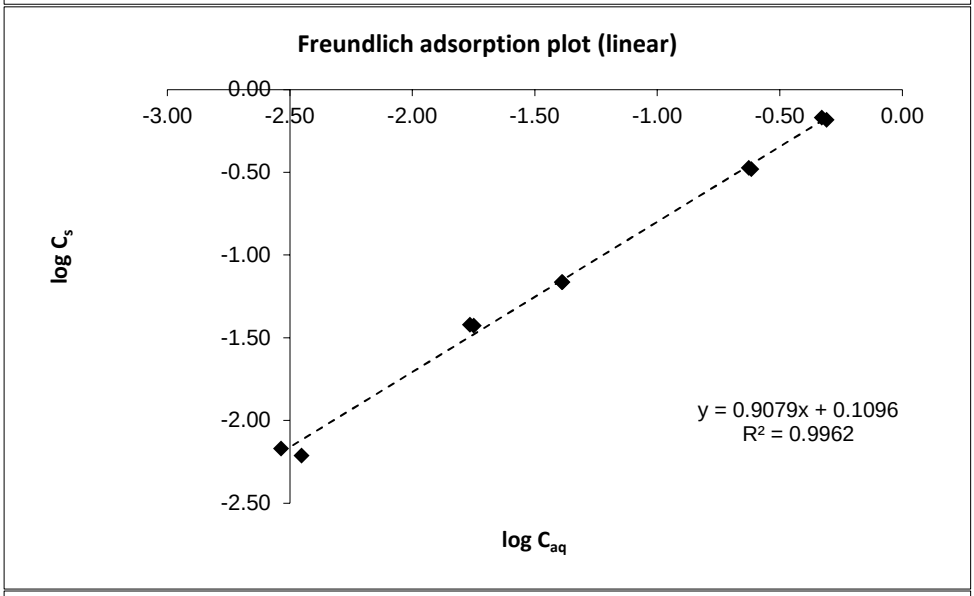


Figure CA.B.8.1.6.2-3: Freundlich Adsorption Plots Lufa 2.4 (calculated by CA)





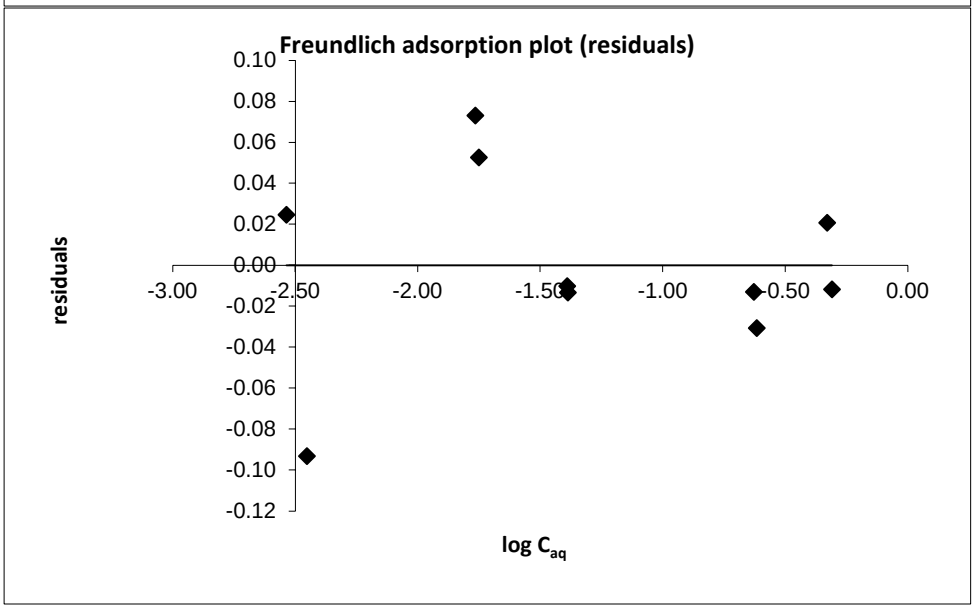


Figure CA.B.8.1.6.2-4: Freundlich Adsorption Plots St. Bauzille 12-060 (calculated by CA)

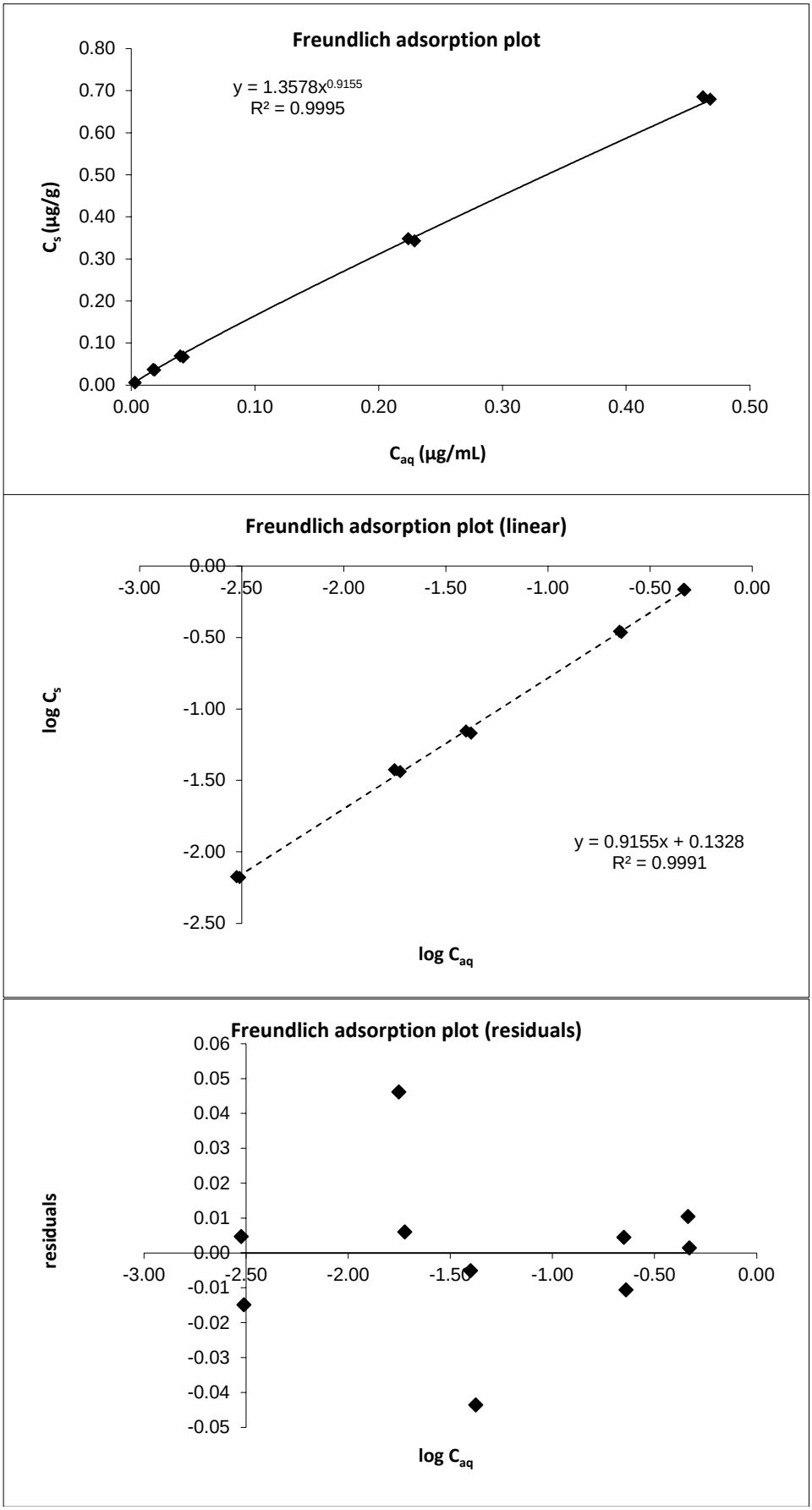
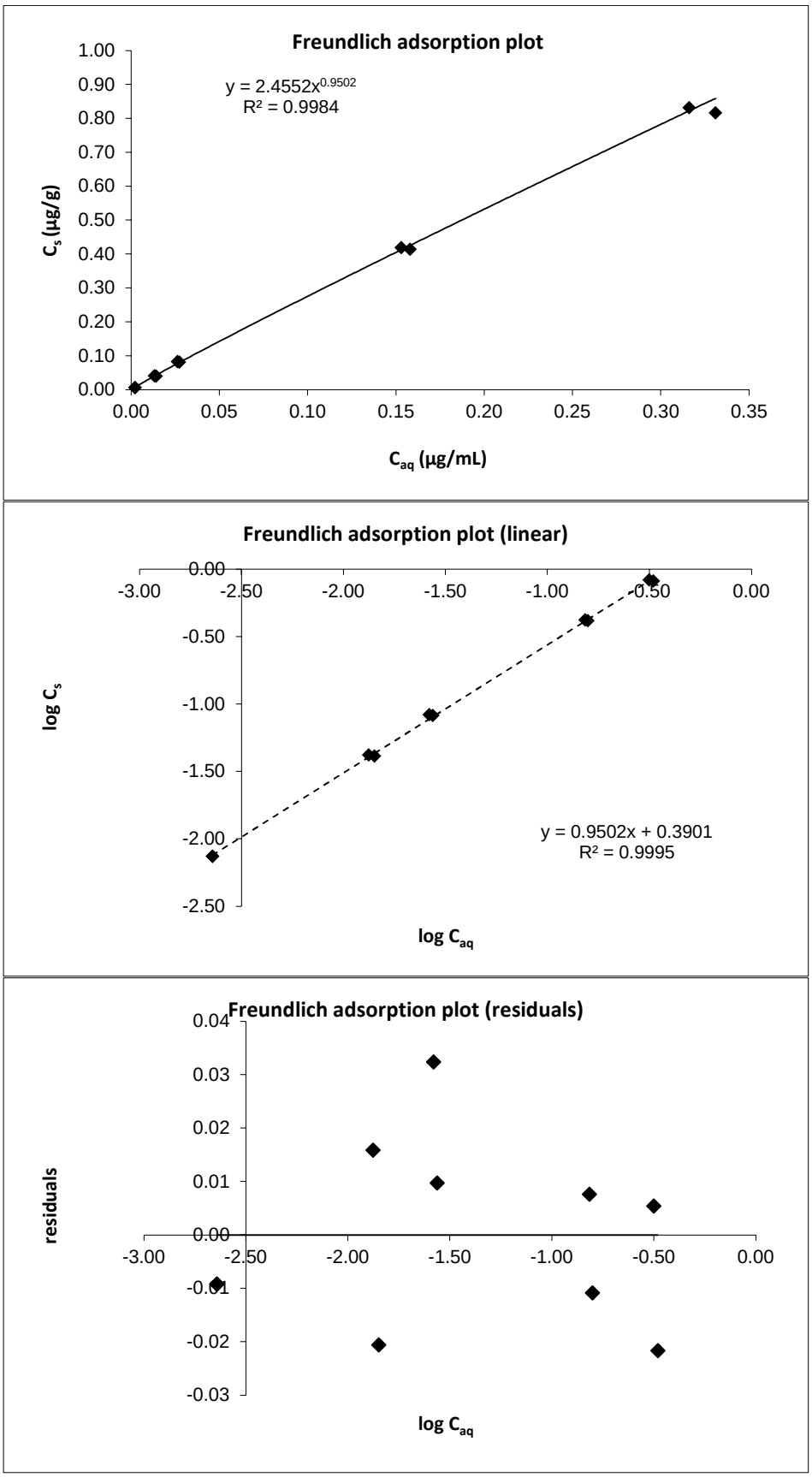
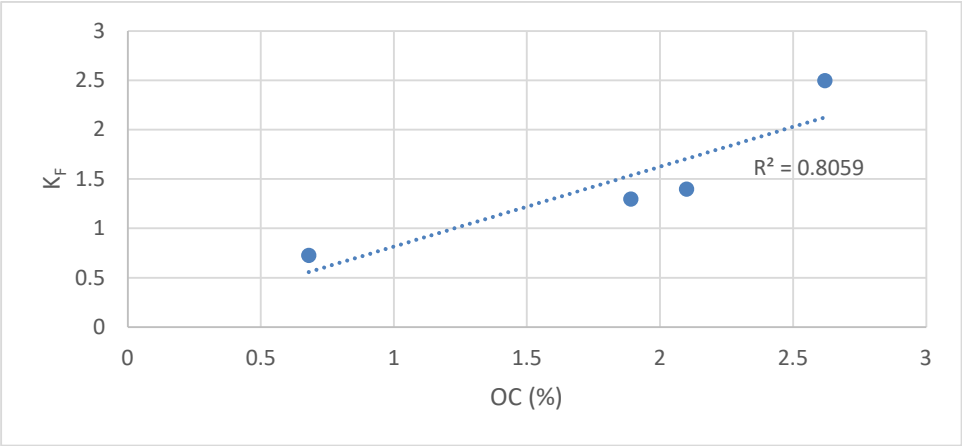


Figure CA.B.8.1.6.2-5: Freundlich Adsorption Plots: Refesol 06-A (calculated by CA)



As can be seen in Figure CA.B.**8.1.6.2-6**, K_F increased with the organic carbon content and therefore it is acceptable to calculate Kfoc values.

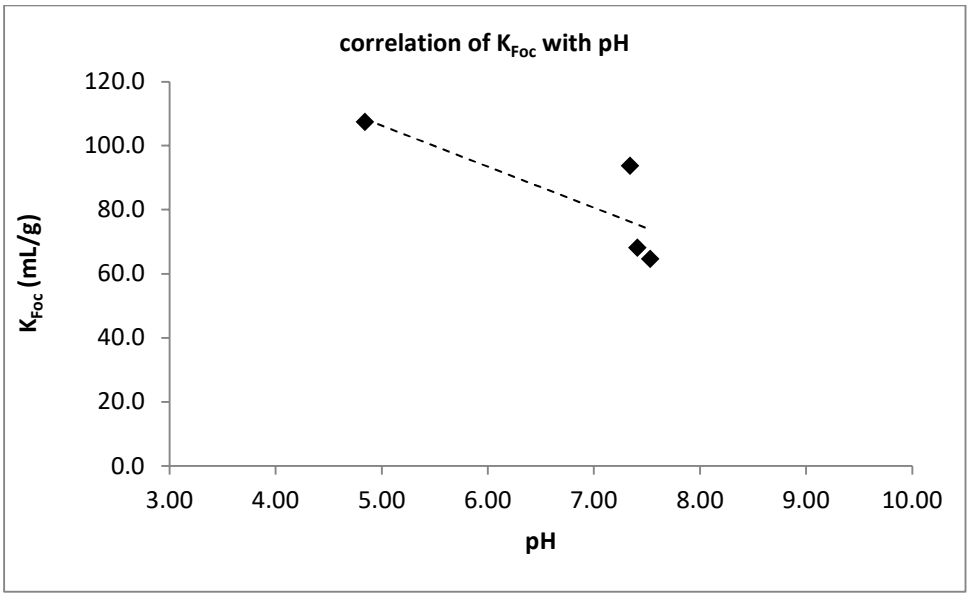
Figure CA.B.8.1.6.2-6: K_F compared with organic carbon



pH dependence

To determine whether pH dependence influenced adsorption, the CA used the Kendal rank correlation coefficient to measure the association between K_{FOC} and pH. The results confirmed there was no significant relationship (-1.000, p = 0.089). For completeness the CA also carried out a regression analysis for soil pH and K_{FOC} values (Figure CA.B.**8.1.6.2-7**). However as can be seen in the figure, the spread of soil pH data do not lend themselves to meaningful assessment of possible pH effects (i.e. where a single soil was tested with pH 4.8 and the remaining three soils varied between pH 7.3 and 7.5.

Figure CA.B.8.1.6.2-7: Correlation of K_{Foc} with pH



CONCLUSION

The adsorption/desorption properties of bixlozone-3-OH-propanamide were determined in four different soils from Europe (LUFA 2.1, LUFA 2.4, St. Bauzille 12-060 and Fraunhofer Refesol 06-A), according to OECD guideline 106 and U.S. EPA OPPTS 835.1230.

For each soil, quality criteria were met. The CA accepts the findings of this study.

Adsorption correlated with organic carbon content and K_{FOC} values for bixlozone-3-OH-propanamide were 65-107 mL/g (geometric mean 81.7 mL/g, arithmetic mean 1/n = 0.925) and desorption K_{FOC}-des values were 71-136 mL/g (geometric mean 93.9 mL/g, arithmetic mean 1/n = 0.924). There was no evidence of a relationship between sorption and soil pH.

CA.B.8.1.6.3. *2,4-Dichlorobenzoic Acid Determination of Adsorption/Desorption Behaviour in Four Soils*

Report:	KCA 7.1.3.1.2/02, Göcer, M. (2017)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	2,4-Dichlorobenzoic acid Adsorption/Desorption Behaviour in Four Soils
Testing facility:	Eurofins Agroscience Services EcoChem GmbH, Germany
Document No:	Study no. S16-01057, FMC Tracking no. 2016EFT-ISX2469
Guidelines:	OECD Guideline 106 (2000); US EPA Guideline, OPPTS 835-1230 (October 2008) SANCO/3029/99 rev.4
Deviations	None
GLP:	Yes (laboratory certified by German National Authority)

CA Comments:	In the opinion of the CA, an insufficient centrifuge speed and/or time was used to remove the aqueous solution from the soil pellet, resulting in more than half the aqueous phase remaining after centrifugation in the Lufa 2.4 soil for example. This in turn led to unacceptable errors and/or variation in the soil and aqueous concentrations. In the opinion of the CA, further efforts should have been made to reduce the volume of liquid entrained because this is a major potential source of error when conducting the study following the direct method (particularly for poorly sorbed substances). The desorption properties could not be accurately established because of the low initial adsorption values. **This study is <u>not</u> relied upon**

INTRODUCTION

The adsorption/desorption properties of 2,4-dichlorobenzoic acid, a metabolite of bixlozone, were determined in four different soils of European origin (LUFA 2.1, LUFA 2.4, St. Bauzille 12-060 and Fraunhofer Refesol 06-A), at a constant temperature between 20 –25 °C, applying the batch equilibrium method according to OECD guideline 106 (2000) and U.S. EPA OPPTS 835.1230 (2008).

Preliminary investigations were conducted to check for the adsorption to the glass test vessels, to confirm the stability of the test item in 0.01M calcium chloride, to determine the soil solution ratio to be used (1:1, 1:2.5, 1:25) at an initial aqueous concentration of 1.0 mg/L, and to determine the time required for the compound to equilibrate between soil and solvent.

For the definitive test, the sterilised soils were pre-equilibrated overnight (minimum 12 hours) in 0.01M $CaCl_2$ solution (*ca* 18 mL) in glass flasks with PTFE sealed screw caps. Bixlozone-3-OH-propanamide dissolved in 0.01M $CaCl_2$ solution was added to four different soils (20 g oven-dried equivalents) and the solutions made up to 20 mL in total, to give initial nominal concentrations of 1.0, 0.5, 0.1, 0.05 and 0.01 mg/L. Test vessels were prepared in duplicate for each soil and each test item concentration. Test vessels were shaken for 48 hours (adsorption equilibrium time) in the dark at constant temperature (20 – 25 °C).

The desorption isotherms of the four soils were not determined because of the slight adsorption demonstrated.

Soil Collection

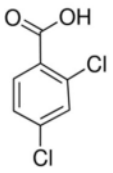
The soils 2.1, 2.4 and St. Bauzille 12-060 were delivered already sieved to a particle size ≤ 2 mm. The soil 06-A was sieved at test facility to a particle size ≤ 2 mm. The soils were airdried at the test facility at ambient temperature.

The dry substance of the soils was determined by heating three aliquots at 105 °C until there was no significant change in weight (approx. 16 hours). For all calculations the mass of soil refers to oven dry mass, *i.e.* the weight of soil corrected for moisture content.

Table CA.B.8.1.6.3-1: Soil Properties

Soil Characterisation			**LUFA 2.1**	**LUFA 2.4**	**St. Bauzille 12-060**	**RefeSol 06-A**
Sampling location			Dudenhofen, Germany	Leimersheim, Germany	Herault, France	Schmallenberg, Germany
Date Of collection			05 April 2016	05 April 2016	27 April 2016	14 March 2016
Sampling Depth (cm)			0 - 20 cm	0 - 20 cm	0 - 10 cm	0 - 25 cm
Storage conditions	Before drying		4 °C	4 °C	4 °C	4 °C
	After Drying		at ambient temperature	at ambient temperature	at ambient temperature	at ambient temperature
Particle size distribution		Sand (%)	86.0	34.5	13.7	12.5
		Silt (%)	10.5	40.6	46.0	38.8
		Clay (%)	3.5	24.9	40.4	48.7
Textural classification (USDA)			Loamy sand	Loam	Silty clay	Clay
pH ($CaCl_2$)			4.84	7.41	7.53	7.34
% Organic matter[†]			1.17	3.26	3.62	4.52
% Organic carbon			0.68	1.89	2.1	2.62
CEC (meq/100g)			4.3	32.0	19.0	29.1
Bulk density, disturbed (g/L)			1447	1220	1210	1202
Maximum water holding capacity (%)			31.65	49.16	46.33	53.05
Pesticide use history at the collection site			None for previous 5 years	None for previous 5 years	None for previous 5 years	None for previous 4 years

Table CA.B.8.1.6.3-2: Test compound details

Name:	2,4-Dichlorobenzoic acid
Structure:	
CAS Number:	50-84-0
Molecular Weight:	191.0 g/mol
Batch Number:	S34634V
Radiochemical purity:	99.9 % w/w
Storage:	ambient (≤ 30 °C), dark, dry

Preparation of test solutions

The stock solution containing 1489 mg/L 2,4-Dichlorobenzoic Acid in acetonitrile/0.01 M $CaCl_2$ (1/1, v/v) was prepared by dissolving 14.9 mg of 2,4-Dichlorobenzoic Acid in 10 mL acetonitrile/0.01 M $CaCl_2$ (1/1, v/v).

An application solution containing 100 mg/L test item was prepared by diluting 1.343 mL stock solution with 0.01 M $CaCl_2$ to a final volume of 20 mL. The application solution was used for samples containing 1 mg/L (Tier 1, Tier 2 and Tier 3 experiments) and 0.5 mg/L test item (Tier 3 experiment).

Furthermore two different application solutions were prepared for Tier 3 experiments in 0.01 M $CaCl_2$ containing 10 mg/L and 1 mg/L 2,4-Dichlorobenzoic Acid. Aliquots of these solutions were used for application of samples containing the following concentrations of the test item: 0.1, 0.05 and 0.01 mg/L 2,4-Dichlorobenzoic Acid in 0.01 M $CaCl_2$ solution.

The CA notes that the stock solution contains more than 0.1% solvent. (3.7225%). This is not considered to have affected the overall results of the experiment because the application solution was further diluted giving less than 0.01% (0.0025%) of solvent when applied to soil.

TIER 1 PRELIMINARY TEST METHOD

Soil solution ratios

The preliminary study was designed to determine the soil solution ratio for further tests (tier 2 and 3), and also to establish the adsorption of the test item to the surface of the test vessel and the stability of the test item during the test period.

Four soils 2.1, 2.4, 12-060 and 06-A and three soil/solution ratios were used.

- 20 g soil and 20 cm^3 aqueous solution of the test substance (ratio 1/1);
- 20 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/2.5);
- 2 g soil and 50 cm^3 aqueous solution of the test substance (ratio 1/25);

The Tier 1 experiments, including controls, were performed in duplicate using the serial method.

An aliquot of the application solution was added dropwise to the soil/0.01 M $CaCl_2$ suspension. With the addition of the application solution the final volume was achieved and the test vessel was sealed. The test system was then well mixed.

500 µL of the test item solution were applied (corresponding to 50 µg test item) to the 1/2.5 and 1/25 samples and 200 µL (corresponding to 20 µg test item) of the test item solution were applied to the 1/1 samples for the Tier 1 experiments (duplicates were used for each soil type).

500 µL of the application solution (100 mg/L) were applied to the 1/2.5 and 1/25 samples and 200 µL of the application solution were applied to the 1/1 samples for the Tier 1 experiments. The nominal test concentration for the Tier 1 experiment was 1.0 mg/L and the actual test item concentration was determined in the control samples by LC-MS/MS analysis to be 0.9679 mg/L.

The sampling times were 0 (only control samples) and 24 hours after application.

Stability of item during test

Two control samples in 0.01 M $CaCl_2$ solution without soil were treated with the test item and were subjected to the same steps as the test systems, in order to check the stability of the test item in 0.01 M $CaCl_2$ solution and its possible adsorption on the surfaces of the test vessels.

Mass Balance

The mass balance was evaluated for all soil/solution ratios in the Tier 1 test after 24 hours. For this purpose, the aqueous and the solid phase were separated by centrifugation. The aqueous phase was recovered as completely as possible and analysed. The extraction of the soil was performed twice with each 80 mL of acetonitrile/water (80/20, v/v) at ambient temperature and shaking for 30 minutes. The

extracts were separated from the soil by centrifugation at 2300 rpm for 4 minutes. The amount of the test item in the combined soil extracts was determined by HPLC-MS/MS and the mass balance was calculated.

TIER 1 RESULTS

Soil Solution Ratios

The metabolite 2,4-Dichlorobenzoic acid exhibited poor adsorption (less than the preferred 20-80%). The table below represents the adsorption results of duplicate sampling (averages only) for the four tested soils after 24 hours of agitation. The ratio that gave the highest rate of adsorption was 1/1. The CA agrees that this was the most appropriate choice of soil: solution ratio for further tier studies.

Table CA.B.8.1.6.3-3: Average amount adsorbed to Soils 2.1, 2.4, 12-060, and 06-A after 24 hours for Soil to solution Ratio 1/1, 1/ 2.5 and 1/25 (tier 1)

	Soil/solution Ratio 1/1	Soil solution Ratio 1/ 2.5	Soil solution ratio 1/25
Lufa 2.1	5.6	0.5	-3.4
Lufa 2.4	13.4	5.9	-1.1
St. Bauzille 12-060	11.4	3.8	-1.6
Refesol 06-A	11.4	1.4	-2.5

Note: negative values arise due to analytical variance

Stability in 0.01M Calcium Chloride

In the two control samples (only 0.01 M $CaCl_2$ solution containing the test item without soil) of the Tier 1 experiment, the mean recovery was 101.5 % after 24 hours of application. Therefore, the test item was stable in solution without soil during the entire experimental period. Furthermore, no adsorption of the test item to the surface of the glass test vessel was observed after 24 hours of agitation.

Table CA.B.8.1.6.3-4: Stability of 2,4-Dichlorobenzoic acid in 0.001 M $CaCL_2$ Solution (tier 1)

	Sampling Interval [hours]	Recovery single values [% of applied]	Mean recovery [% of applied]
Control	0	99.5	100.0
		100.5	
	24	101.5	101.5
		101.5	

Mass balance

The mass balance was between 98.8 % and 107.7 % for soil 2.1, between 102.4 % and 103.5 % for soil 2.4, between 99.8 % and 103.5 % for soil 12-060 and between 97.6 % and 104.6 % for soil 06-A. The CA agrees that the test item can be considered stable for the time scale of the Tier 1 test.

Table CA.B.8.1.6.3-5: Parental mass balance after 24 hours of agitation (tier 1)

Sample	Ratio	replicate	Recovery [%]	Mean recovery [%]
Lufa 2.1	1/1	1	108.2	107.3
		2	106.4	
	1/2.5	1	93.1	98.8
		2	104.5	
	1/25	1	106.1	107.7
		2	109.3	
Lufa 2.4	1/1	1	100.1	103.5
		2	107.0	
	1/2.5	1	102.4	102.4
		2	102.4	
	1/25	1	102.9	102.9
		2	103.0	
St. Bauzille 12-060	1/1	1	99.9	99.8
		2	99.6	
	1/2.5	1	100.2	100.6
		2	101.0	
	1/25	1	103.6	103.5
		2	103.4	
Refesol 06-A	1/1	1	98.7	97.6
		2	96.5	
	1/2.5	1	101.6	101.9
		2	102.1	
	1/25	1	104.7	104.6
		2	104.5	

TIER 1 CONCLUSION

No adsorption of the test item to the surface of the test vessels was observed after 24 hours. The mass recovery of the test item was determined to be between 93.1%-108.6% for the four soils. The CA agrees that the test item was stable for the duration of the tier 1 study. The highest rates of adsorption were observed at a soil solution ratio of 1/1. While it is noted that the level of adsorption is less than the 20 % level preferred in the OECD 106 Guideline, the Guideline states that a 1:1 soil/solution ratio is recommended where low sorption occurs. The CA agrees with the applicant's choice to use this ratio in further tier studies.

TIER 2 METHOD

Based on the results of Tier 1 experiments, the Tier 2 test was performed at a soil/solution ratio of 1/1 (20 g soil and 20 cm^3 aqueous solution) for all soils (2.1, 2.4, 12-060, 06-A) using the serial method in duplicate.

200 µL of the application solution (100 mg/L) were applied to the 1/1 samples for the Tier 2 experiments. The nominal test concentration for the Tier 2 experiments was 1.0 mg/L and the actual test item concentration was determined in the control samples by LC-MS/MS analysis to be 1.0575 mg/L for the adsorption experiments and 0.8293 mg/L for the desorption experiments.

An aliquot of 0.200 mL test item solution at 100 mg/L were applied (corresponding to 20 µg test item) to each pre-equilibrated sample (duplicates were used for each soil type) and shaken at 150 rpm at a constant temperature between 20-25°C.

The sampling times for both adsorption and desorption were 0 (only control samples), 2, 4, 6, 24 and 48 hours after application. The desorption experiments were performed following a 48 hour adsorption phase.

Two control samples containing 0.01 M $CaCl_2$ solution were treated with the test item and were subjected to the same steps as the test systems, in order to check the stability of the test item in 0.01 M $CaCl_2$ solution. Furthermore, two blank systems (no test item) for each soil at a soil/solution ratio of 1/1 were prepared and treated in the same manner as the spiked soil samples. An aliquot of the aqueous phase of each blank was measured by LC-MS/MS.

TIER 2 RESULTS

Table CA.B.8.1.6.3-6: Concentration of Test Item in Supernatant for Soil to Solution Ratio 1/1 during Tier 2 Adsorption Experiment

Lufa 2.1		Concentration [µg/mL]				
Time [h]		2	4	6	24	48
Raio 1/1	Sample 1	1.0241	0.9882	1.0034	1.0405	0.8967
	Sample 2	1.0262	1.0323	1.0482	1.0259	0.9717
Lufa 2.4		Concentration [µg/mL]				
Time [h]		2	4	6	24	48
Ratio 1/1	Sample 1	0.9874	1.0066	1.0246	0.9842	0.9827
	Sample 2	1.0251	1.0377	0.9262	0.9831	0.9496
St. Bauzille 12-060		Concentration [µg/mL]				
Time [h]		2	4	6	24	48
Ratio 1/1	Sample 1	1.0127	1.031	0.9967	0.9578	0.9757
	Sample 2	0.9299	1.0797	1.1496	1.0062	0.9602
Refesol 06-A		Concentration [µg/mL]				
Time [h]		2	4	6	24	48
Ratio 1/1	Sample 1	1.0446	1.0646	1.0446	1	0.968
	Sample 2	1.0244	1.068	1.0446	1.0011	0.9779
Control		Concentration [µg/mL]				
Time [h]		2	4	6	24	48
Control	Sample 1	1.0406	1.07	1.1009	1.0619	1.0354
	Sample 2	1.0614	1.0484	1.085	1.0563	1.0376

Mass Balance

Based on the results of Tier 1 the mass balance was evaluated for Tier 2 at soil/solution ratios of 1/1 for all soils after 48 hours adsorption. The amount of the test item in the supernatant and the soil extracts was determined and the mass balance was calculated.

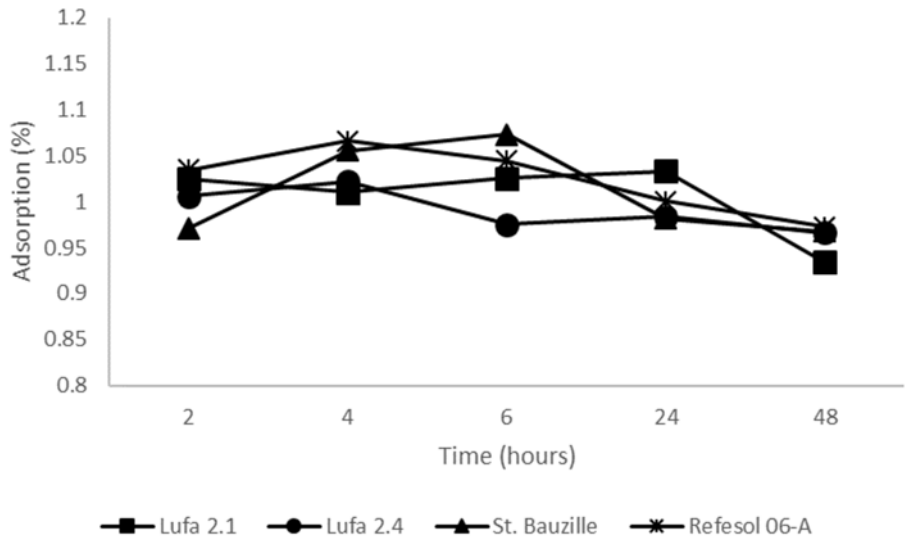
Table CA.B.8.1.6.3-7: Parental Mass Balance after 48 hours of Agitation (Tier 2 Adsorption)

Sample	Ratio	replicate	Recovery [%]	Mean recovery [%]
Lufa 2.1	1/1	1	100.1	98.9
		2	97.7	
Lufa 2.4	1/1	1	102.4	103.0
		2	103.5	
St. Bauzille 12-060	1/1	1	91.7	92.8
		2	93.9	
Refesol 06-A	1/1	1	96.7	97.5
		2	98.2	

TIER 2 CONCLUSION

The mass balance for tier 2 was between 92.8 % and 103.0 %. The test item was therefore considered to be stable for the timescale of the experiment. The CA agrees with the applicant's decision to use an equilibration time of 48 hours.

Figure CA.B.8.1.6.3-1: Adsorption equilibrium time (preliminary experiment) of 2,4-dichlorobenzoic acid



TIER 3 METHOD

Five test item concentrations (1.0, 0.5, 0.1, 0.05 and 0.01 mg/L, equivalent to 20, 10, 2, 1 and 0.2 µg test item) were used, covering two orders of magnitude, in the direct test method (appropriate due to the low sorption). In the choice of these concentrations the water solubility and the resulting aqueous equilibrium concentrations were taken into account. The Tier 3 adsorption test was performed with all four soils (2.1, 2.4, 12-060, 06-A) at a soil/solution ratio of 1/1 (20 g soil and 20 cm^3 aqueous solution) using the serial method in duplicate.

The nominal test concentrations for the Tier 3 experiment were 1.0, 0.5, 0.1, 0.05 and 0.01 mg/L and the actual test item concentrations were determined in the control samples by LC-MS/MS analysis to be 0.9302, 0.4835, 0.1146, 0.0545 and 0.0101 µg/mL, respectively.

Each experiment (one soil and five treatment levels) was done with pre-equilibrated soils in duplicate to allow estimation of the variance of the results. Furthermore, two control samples were prepared containing 0.01 M $CaCl_2$ solution without soil.

The test was performed by the serial method in 100 mL glass bottles with PTFE sealed screw caps. Agitation was performed at a constant temperature between 20 - 25°C on a flatbed shaker with a frequency of around 150 rpm to keep the soil dispersed in the aqueous phase.

After 48 h the suspensions were centrifuged at 2300 rpm for 4 minutes. The aqueous phase was recovered as completely as possible (see 'discussion' section below for further consideration of this). Then, the extraction of the soil was performed. The amount of the test item adsorbed to the soil was determined in the combined soil extracts (direct method).

After a request for further information, the CA can confirm that the concentrations reported by the applicant in the soil did attempt to account for pore water concentration. However as noted above and in the discussion section below, the volume of the liquid entrained in the soil pellet were significant (up to 50% of the total liquid volume in the Lufa 2.4 soil) and this adds considerable uncertainty to the calculated sorption values in the opinion of the CA.

RESULTS

Table CA.B.8.1.6.3-8: Concentration and Logarithm of Concentration of 2,4-Dichlorobenzoic Acid as Adsorption Equilibrium after 48 hours in the water Phase and bound to Soil 2.1 for Soil to Solution Ration 1/1 (Tier 3)

C_0 [µg/mL]		C ads aq[µg/mL]	C ads s[µg/g]	Log C ads aq[µg/mL]	Log C ads s[µg/g]
0.93	Sample 1	0.881	0.0818	-0.0552	-1.09
	Sample 2	0.849	0.0588	-0.0711	-1.23
	Mean	**0.865**	**0.0703**	**-0.0632**	**-1.16**
0.483	Sample 1	0.454	0.0601	-0.343	-1.22
	Sample 2	0.467	0.0481	-0.033	-1.32
	Mean	**0.461**	**0.0541**	**-0.337**	**-1.27**
0.115	Sample 1	0.0978	0.00865	-1.01	-2.06
	Sample 2	0.0973	0.00612	-1.01	-2.21
	Mean	**0.0976**	**0.00739**	**-1.01**	**-2.14**
0.0545	Sample 1	0.0502	0.00235	-1.3	-2.63
	Sample 2	0.0494	0.0032	-1.31	-2.49
	Mean	**0.0498**	**0.00277**	**-1.3**	**-2.56**
0.0101	Sample 1	0.0112	-0.00041	-1.95	N/A
	Sample 2	0.0106	-0.00044	-1.97	N/A
	Mean	**0.0109**	**-0.0004**	**-1.96**	**N/A**

N/A: not available

Table CA.B.8.1.6.3-9: Concentration and Logarithm of Concentration of 2,4-Dichlorobenzoic Acid at Adsorption Equilibrium after 48 hours in the Water Phase and bound to Soil 2.4 for Soil to Solution Ratio 1/1 (Tier 3)

C_0 [µg/mL]		C ads aq[µg/mL]	C ads s[µg/g]	Log C ads aq[µg/mL]	Log C ads s[µg/g]
0.93	Sample 1	0.891	0.12	-0.05	-0.92
	Sample 2	0.871	0.117	-0.06	-0.932
	Mean	**0.881**	**0.119**	**-0.055**	**-0.926**
0.483	Sample 1	0.464	0.0646	-0.333	-1.19
	Sample 2	0.473	0.0336	-0.325	-1.47
	Mean	**0.469**	**0.0491**	**-0.329**	**-1.33**
0.115	Sample 1	0.0942	0.0112	-1.03	-1.95
	Sample 2	0.093	0.00991	-1.03	-2
	Mean	**0.0936**	**0.0106**	**-1.03**	**-1.98**
0.0545	Sample 1	0.0489	0.00424	-1.31	-2.37
	Sample 2	0.0503	0.00263	-1.3	-2.58
	Mean	**0.0496**	**0.00343**	**-1.3**	**-2.48**
0.0101	Sample 1	0.0108	0.000303	-1.97	-3.52
	Sample 2	0.0095	0.00102	-2.02	-2.99
	Mean	**0.0102**	**0.000663**	**-1.99**	**-3.25**

Table CA.B.8.1.6.3-10: Concentration and Logarithm of Concentration of 2,4-Dichlorobenzoic Acid at Adsorption Equilibrium after 48 hours in the Water Phase and bound to Soil 12-060 for Soil to Solution Ratio 1/1 (Tier 3)

C_0 [µg/mL]		C ads aq[µg/mL]	C ads s[µg/g]	Log C ads aq[µg/mL]	Log C ads s[µg/g]
0.93	Sample 1	0.882	0.0987	-0.0548	-1.01
	Sample 2	0.887	0.0772	-0.0519	-1.11
	Mean	**0.884**	**0.0879**	**-0.0533**	**-1.06**
0.483	Sample 1	0.468	-0.00156	-0.33	N/A
	Sample 2	0.477	0.0497	-0.321	-1.3
	Mean	**0.473**	**0.0241**	**-0.325**	**N/A**
0.115	Sample 1	0.0992	0.0091	-1	-2.04
	Sample 2	0.0979	0.00505	-1.01	-2.3
	Mean	**0.0986**	**0.00708**	**-1.01**	**-2.17**
0.0545	Sample 1	0.0501	0.00412	-1.3	-2.39
	Sample 2	0.0478	n.d.	-1.32	N/A
	Mean	**0.049**	**N/A**	**-1.31**	**N/A**
0.0101	Sample 1	0.0101	N/A	-2	N/A
	Sample 2	0.0108	N/A	-1.97	N/A
	Mean	**0.0105**	**N/A**	**-1.98**	**N/A**

n.d.: not determined, because sample was broken
N/A: not available

Table CA.B.8.1.6.3-11: Concentration and Logarithm of Concentration of 2,4-Dichlorobenzoic Acid at Adsorption Equilibrium after 48 hours in the Water Phase and bound to Soil 06-A for Soil to Solution Ratio 1/1 (Tier 3)

C_0 [µg/mL]		C ads aq[µg/mL]	C ads s[µg/g]	Log C ads aq[µg/mL]	Log C ads s[µg/g]
0.93	Sample 1	0.884	n.d.	-0.0534	N/A
	Sample 2	0.888	0.138	-0.0514	-0.859
	Mean	**0.886**	**N/A**	**-0.0524**	**N/A**
0.483	Sample 1	0.476	0.0365	-0.322	-1.44
	Sample 2	0.472	0.0561	-0.326	-1.25
	Mean	**0.474**	**0.0463**	**-0.324**	**-1.34**
0.115	Sample 1	0.0983	0.00934	-1.01	-2.03
	Sample 2	0.0995	0.0106	-1	-1.98
	Mean	**0.0989**	**0.00995**	**-1**	**-2**
0.0545	Sample 1	0.0495	0.00378	-1.31	-2.42
	Sample 2	0.0492	0.00487	-1.31	-2.31
	Mean	**0.0494**	**0.00432**	**-1.31**	**-2.37**
0.0101	Sample 1	0.0115	-0.00146	-1.94	N/A
	Sample 2	0.0112	-0.000699	-1.95	N/A
	Mean	**0.0114**	**-0.00108**	**-1.95**	**N/A**

N/A: not available

ADSORPTION KINETICS

The Freundlich adsorption coefficients calculated by the applicant are provided in Table CA.B.**8.1.6.3-12**. The CA performed all relevant quality checks (OECD 106 evaluators checklist, November 2017) as part of confirming the acceptability of the study and the reported endpoints and found some differences between the CA values and the applicants. The applicant values are therefore not accepted by the CA. These are discussed below.

Table CA.B.8.1.6.3-12: Applicant's reported regression parameters (not accepted by CA)

Parameters	LUFA 2.1	LUFA 2.4	St. Bauzille 12-060	Refesol 06-A
Slope (1/n)	1.169	1.185	1.103	1.148
Intercept (log K_{ads} F)	-0.989	-0.877	-0.986	-0.872
K_{ads} F	0.10	0.13	0.10	0.13
R square	0.9815	0.9936	0.9925	0.9882
% oc	0.68	1.89	2.10	2.62
K_{ads} FOC	15	7.0	4.9	5.1

Desorption

The desorption isotherms of the four soils were not determined because of the very low adsorption, which was <20 % for each soil.

DISCUSSION

For Lufa 2.1, not all quality criteria were met. The EFSA guidance (2017) states that accurate determination of the distribution coefficient, Kd, is best achieved via both the indirect and direct methods when the Kd * soil/solution ratio is greater than 0.3. It may also be possible to calculate accurate values for Kd, even when Kd * soil/solution ratio is < 0.3 provided that the method is suitably rigorous in terms of minimising errors associated with the liquid entrained in the soil pellet. For the direct method, the ratio has to be derived after centrifugation, and is therefore calculated as the ratio between the soil mass divided by the residual moisture volume in the soil pellet. For Lufa 2.1, the Kd * soil/solution ratio was between 0.15 and 0.45 for all concentrations and therefore not acceptable. The CA notes that the method was not suitably rigorous in terms of minimising errors associated with the liquid entrained in the soil pellet. Approximately 50 % of the aqueous solution remained in the soil pellet and needed to be accounted for post analysis leading to errors (*i.e.* negative values for the soil compartment). The centrifuge speed and time of 2300 rpm and 4 minutes used within the study were not appropriate according to Figure 1a in the OECD 106 guidance document and could be improved to minimise errors. The adsorbed percentage was also not within the acceptable range of >20 % (3.41-15.1 %) however the CA notes that an optimum soil:solution ratio (1:1) was used by the applicant and could not be improved. The fit to the Freundlich isotherm was below the OECD 106 recommended value of >0.975, with an R^2 value of 0.969; the CA considers this was due to errors introduced post analysis to account of the large amount of residual moisture volume in the soil pellet. The K_{FOC} and 1/n endpoints derived from this soil are not considered robust enough for use in exposure modelling. The quality criteria for Lufa 2.1 for each concentration is summarised in Table CA.B.**8.1.6.3-13**.

Table CA.B.8.1.6.3-13: Quality criteria calculated by the CA for soil Lufa 2.1

Start concentration (mg/L)	Liquid entrained (mL)	kd x soil:solution ratio	% adsorbed
0.9302	6.00	0.310	5.32
0.9302	6.03	0.230	8.74
0.4835	5.88	0.454	6.93
0.4835	5.95	0.346	3.41
0.1146	5.79	0.306	14.7
0.1146	6.00	0.210	15.1
0.05445	6.39	0.147	7.81
0.05445	6.20	0.209	9.27
0.01005	5.86	N/A	N/A
0.01005	5.79	N/A	N/A

N/A Not available

For Lufa 2.4, not all quality criteria were met. The test item mass balance was > 90 % (*i.e.* 102.4-103.5 %). The Kd * soil/solution ratio was between 0.056 and 0.276, and the adsorbed percentage was not within an acceptable range >20 % (2.17-10.2 %). As for the previous soil, the CA has similar concerns over the centrifuge speed and time of 2300 rpm and 4 minutes used within the study. These parameters were not appropriate to minimise errors associated with the liquid entrained in the soil pellet. The visual fit to the Freundlich isotherm was acceptable, with relatively small and randomly distributed residuals, however the R^2 value was not acceptable (0.959). The K_{FOC} and 1/n endpoints derived from this soil are not considered robust enough for use in exposure modelling. The quality criteria for Lufa 2.4 for each concentration is summarised in Table CA.B.**8.1.6.3-14**.

Table CA.B.8.1.6.3-14: Quality criteria calculated by the CA for soil Lufa 2.4

Start concentration (mg/L)	Liquid entrained (mL)	kd x soil:solution ratio	% adsorbed
0.9302	10.12	0.266	4.21
0.9302	10.07	0.267	6.36
0.4835	10.20	0.273	4.03
0.4835	10.20	0.139	2.17
0.1146	9.86	0.241	17.8
0.1146	9.89	0.215	18.8
0.05445	9.85	0.176	10.2
0.05445	9.91	0.106	7.62
0.01005	9.95	0.056	N/A*
0.01005	9.80	0.219	5.47

N/A Not available

* concentration measured in aqueous solution after 48 hours was higher than initial concentration

For St Bauzille 12-060, low recoveries for the soil sample and a broken sample meant that only two concentrations were analysed in duplicate and therefore accurate K_{OC} and 1/n parameters could not be derived for this soil.

For Refesol 06-A, as above, because of low recoveries for the soil sample and a broken sample only three concentrations were analysed in duplicate and therefore K_{OC} and 1/n parameters could not be derived for this soil.

Table CA.B.8.1.6.3-15: Summary Results Table

Soil	**Units**	**LUFA 2.1**	**LUFA 2.4**	**St Bauzille 12-060**	**Refsol 06-A**
Adsorption method (direct/ indirect)	-	direct	direct	direct	direct
Soil:solution Ratio	(g dw/mL)	1:1	1:1	1:1	1:1
Mass balance of ^{14}C (at all tested conc)	%	97.1-100.1	102.4-103.5	91.7-93.9	96.7-98.2
Adsorbed percentage	%	3.41-15.1	2.17-10.2	n.a.	n.a.
Kd * Soil/ Solution		0.15-0.45	0.056-0.276	n.a.	n.a.
adsK_F (95% confidence interval)	L/kg dw	0.103 (0.102-0.683)	0.132 (0.077-0.225)	n.a.	n.a.
ads 1/n	-	1.169	1.181	n.a.	n.a.
ads R^2	-	0.969	0.959	n.a.	n.a.
ads K_{FOC}	L/kg OC	15.1	7.0	n.a.	n.a.

n.a insufficient data to perform quality checks

Figure CA.B.8.1.6.3-2: Freundlich Adsorption Plots: Lufa 2.1

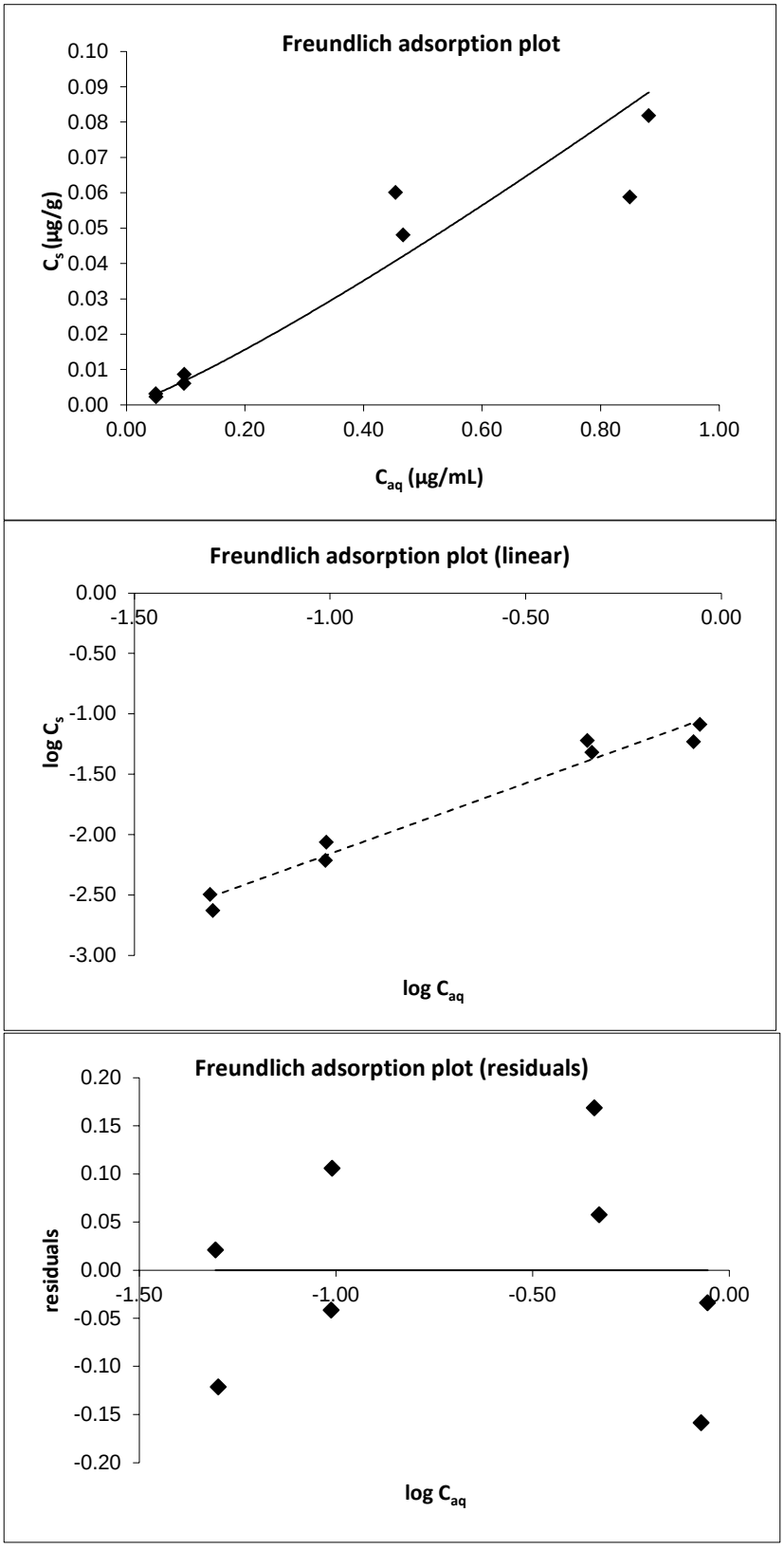
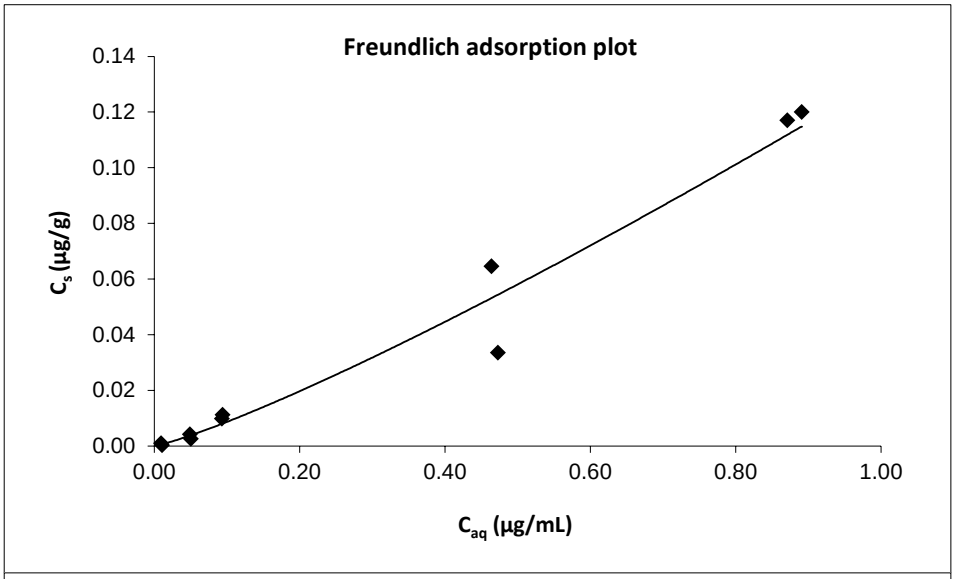
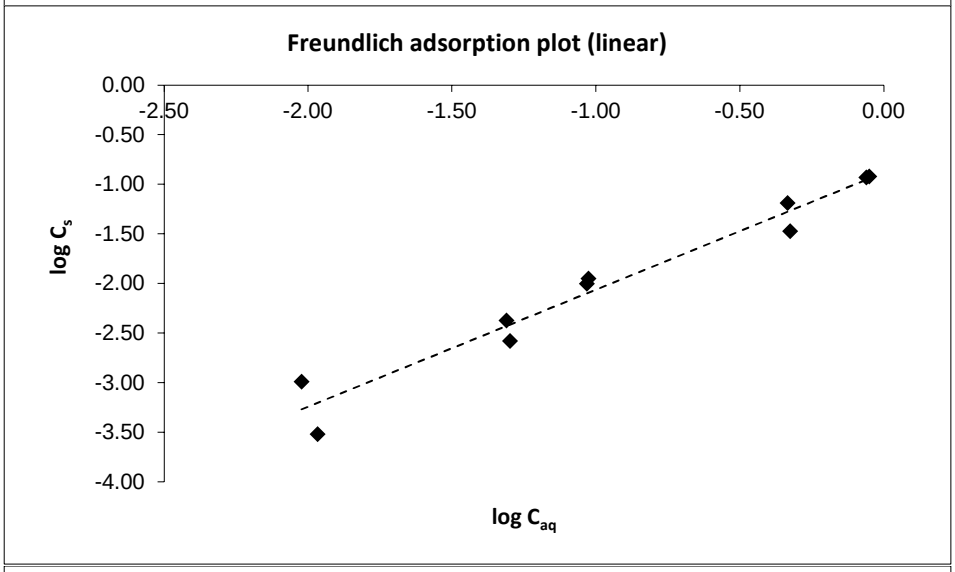
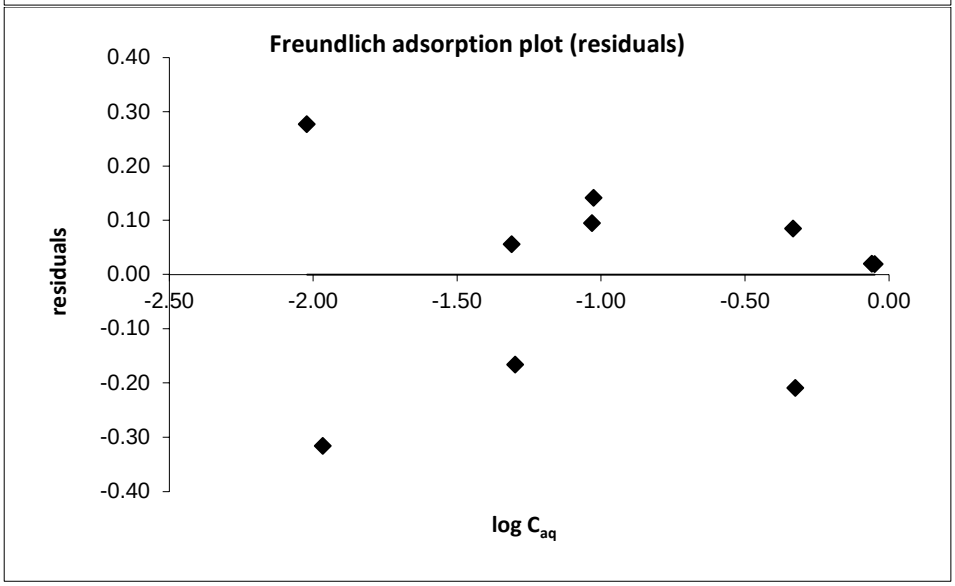


Figure CA.B.8.1.6.3-3: Freundlich Adsorption Plots: Lufa 2.4







It was not possible to establish any correlation between K_{foc} and pH because of the exclusion of two soils from the study. Furthermore, the CA notes that three of the soils are of very similar pH and therefore, even if the soils had been accepted, it would be difficult to comment on the possibility of pH dependence.

CONCLUSION

The adsorption properties of 2,4-dichlorobenzoic acid were determined in four different soils from Europe (LUFA 2.1, LUFA 2.4, St. Bauzille 12-060 and Fraunhofer Refesol 06-A), according to OECD guideline 106 and U.S. EPA OPPTS 835.1230. The desorption properties could not be accurately established because of the low initial adsorption values.

In the opinion of the CA, an insufficient centrifuge speed and/or time was used to remove the aqueous solution from the soil pellet, resulting in more than half the aqueous phase remaining after centrifugation in the Lufa 2.4 soil for example. This in turn led to unacceptable errors and/or variation in the soil and aqueous concentrations. In the opinion of the CA, further efforts should have been made to reduce the volume of liquid entrained because this is a major potential source of error when conducting the study following the direct method (particularly for poorly sorbed substances). For St Bauzille 12-060 and Refsol 06-A, because of low recoveries for the soil samples (*i.e.* negative values) and broken samples, only two or three concentrations were analysed in duplicate. Accurate and robust K_{FOC} and 1/n parameters could not be derived for these soils. As such, default K_{OC} and 1/n values of 0 mL/g and 1 respectively are to be used for this compound in the exposure assessment.

CA.B.8.1.7. **Mobility in soil**

No further studies assessing the mobility of bixlozone in soil are submitted or required.

CA.B.8.1.8. **Summary of fate and behaviour in soil**

Parent dosed studies

A laboratory aerobic degradation study was submitted in which bixlozone degradation was investigated in four European soils and three US soils (pH range 5.4 to 8.0). At study end (120 d), 24.54-75.83 % AR of bixlozone was remaining. Mineralisation resulted in CO_2 steadily increasing over the duration of the study, reaching 10.40-47.41 % AR (Phenyl-U-^{14}C] label) and 11.64-54.36 % AR [carbonyl-^{14}C] label after 120 days. Unextracted residues ranged between 3.30-11.64 % AR (Phenyl-U-^{14}C] label) and 21.8-28.48 % AR [carbonyl-^{14}C] label after 120 days. There was no significant difference between the results from the two radiolabel positions. [^{14}C]-bixlozone degraded with best-fit DT_{50} values in the range 64.1 days to >1000 days and normalised DT_{50} values for use in exposure modelling in the range 52.5 to 330 days (geomean value of 134 days).

No metabolites were observed >5 % of applied radioactivity. Metabolite 2,4-dichlorobenzoic acid peaked at day 30 reaching a mean maximum of 4.9 % AR before declining to <LOQ by study end, and 2,4-dichlorobenzyl alcohol reached a mean maximum of 2.8 % of applied radioactivity. 2,4-dichlorobenzaldehyde did not exceed 1% of applied radioactivity in any soil at any timepoint. All unknown metabolites individually accounted for less than 3.6 % of applied radioactivity.

An anaerobic degradation study was also submitted for bixlozone in two European soils and two US soils. In the aerobic phase, no metabolites were observed at concentrations >5 %AR. In the anaerobic phase, the metabolite bixlozone-3-hydroxy-propanamide was detected at >10% AR (maximum mean of 14.76 % AR, 120 d sample), and 2,4-dichlorobenzoic acid was present at ≥5% AR at a single time-point (maximum mean of 5.80 % AR at day 120 and increasing). 2,4-dichlorobenzaldehyde and 2,4-dichlorobenzyl alcohol were observed at mean maximum concentrations of 2.4 and 2.16% AR, respectively. All unknown metabolites individually accounted for less than 3.6% AR. Bixlozone degraded in soils incubated under anaerobic conditions with a DT50 values ranging from 206 to 871 days (geomean = 470 days). See metabolite summary section below for justification regarding the exclusion of the anaerobic metabolite results from the terrestrial exposure assessment.

The applicant submitted a soil photolysis study for bixlozone in which the degradation rate was assessed under irradiated and dark conditions in each of 2 soils and with 2 radiolabels. The treated soils were continuously irradiated for up to 15 days alongside dark control samples. The irradiation intensity to the soil surface per day by artificial sunlight was approximately equivalent to 34 days of natural summer sunlight at latitude 30-50°N. [Carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone degraded slowly on soil surface under irradiated conditions (geomean DT_{50} = 100 days, converted to natural summer light), with no degradates > 5% AR being observed in either irradiated or dark control samples. The largest degradate reached a maximum of 3.85 % AR in irradiated samples after 15 days continuous irradiation and was tentatively identified as 2, 4-dichlorobenzoic acid. A number of other minor degradates were also observed, none exceeding 3.57% of applied radioactivity. Degradation in the dark controls was slower over the incubation period, except for carbonyl labelled Leimersheim soil where it was almost identical.

Three field soil dissipation studies were submitted, covering 7 test sites in Europe. Generally at each site, studies were conducted using two formulation types and encompassed both soil incorporation and bare soil treatment. Two metabolites were detected in the field studies. Metabolite 3-OH-propanamide (3-OH) was detected at a maximum of 6.95% (on a mass basis; 6.90% on a molar basis) in one study at one time point (but was not increasing at study termination); due to the very limited evidence of 3-OH formation under aerobic soil conditions, the CA does not consider it necessary to consider 3-OH in the terrestrial exposure assessment. Metabolite 2,4-dichlorobenzoic acid (2,4-DBA) was detected at a maximum of 69.4% (on a mass basis; 99.53% on a molar basis) and so the CA does consider it necessary to include 2,4-DBA in the terrestrial exposure assessment. It is noted the applicant considers a worst-case 2,4-DBA formation of 100% in the PECsoil calculations which is accepted by the CA.

A storage stability study was carried out to evaluate stability of bixlozone and metabolites 2,4-DBA and 3-OH in LUFA 2.4 soil (pH 7.3, %OC 2.03). The specimens were weighed into 50 mL centrifuge tubes and then placed in a freezer set to maintain a temperature of <-18ºC. Residues of bixlozone, 2,4-DBA and 3-OH showed no

significant decrease (≤15% as compared to the zero-time value) in soil when stored deep frozen at <-18°C for up to 24 months.

A kinetic assessment was undertaken on the soil dissipation studies to determine triggering, PEC_{soil}, Persistence and modelling endpoints. The outcome of the triggering endpoint assessment was that the potential for bixlozone accumulation in soil is to be assessed as part of the PECsoil assessment. Due to the short 2,4-DBA laboratory DT_{50} values, accumulation of metabolite 2,4-DBA does not need to be considered and so only PECsoil,initial values need to be determined. The longest non-normalised bixlozone DT_{50} value was 300 d (from the CS formulation at site GE01). The longest non-normalised SC formulation bixlozone DT_{50} value was 247 d (site IT01) and is to be used in the bixlozone PECsoil calculations.

For all soil dissipation trial sites, SFO fits were considered good enough to determine modelling endpoints. Based on the results of the EFSA DegT50 tool and independent statistical advice, the SC formulation endpoints were considered most appropriate for consideration with the laboratory data. The EFSA DegT50 calculator indicated the SC field soil dissipation endpoints were shorter than the laboratory values and so it is not appropriate to combine the data. The geomean DT_{50} of the SC formulation field data, to be used in the exposure models, is 48.0 days. Modelling endpoints for 2,4-DBA could not be obtained due to insufficient data. Therefore, the CA considers the modelling endpoints from the laboratory study to be appropriate for use in the exposure calculations, with a default formation fraction of 1.0.

Bixlozone Persistence endpoints greater than the 120 d trigger were calculated at 10 trial sites. Furthermore, persistence endpoints greater than the 180 d 'very Persistent' trigger were calculated for 6 trial sites. Therefore, the CA considers it appropriate to classify bixlozone as very Persistent in soil.

The adsorption and desorption behaviour of [^{14}C]-bixlozone was studied in five European and three US soils (pH 5.4 to 8.0). Adsorption K_{Foc} values for [^{14}C]-bixlozone were 334 – 465 mL/g (geometric mean 381.5 mL/g, arithmetic mean 1/n = 0.874) and desorption $K_{Foc\text{-}des}$ values were 481 – 754 mL/g (geometric mean 564 mL/g, arithmetic mean 1/n = 0.876), indicating that there is a degree of irreversibility to [^{14}C]-bixlozone adsorption. There was no evidence of any pH dependence.

Metabolite dosed studies

As indicated above, metabolite 2,4-DBA was concluded as being a major soil metabolite under both aerobic and anaerobic soil conditions. Metabolite 3-OH was concluded as being a major anaerobic soil metabolite only. Nevertheless, the applicant submitted aerobic degradation studies for both metabolites. The aerobic degradation studies used three European soils (pH 4.84 to 7.53) and were treated with non-labelled test substances. The specimens were incubated in the dark at 20 °C. 3-OH degraded with normalised (20 °C, pF2) SFO DT_{50} values in the range 6.8 to 12.0 hours (geomean value = 9.1 hours). 2,4-DBA degraded with normalised (20 °C, pF2) SFO DT_{50} values in the range 3.5 to 8.9 days (geomean value = 5.4 days). The 2,4-DBA geomean value of 5.4 days is appropriate for use in the exposure calculations, with the default formation fraction of 1.0.

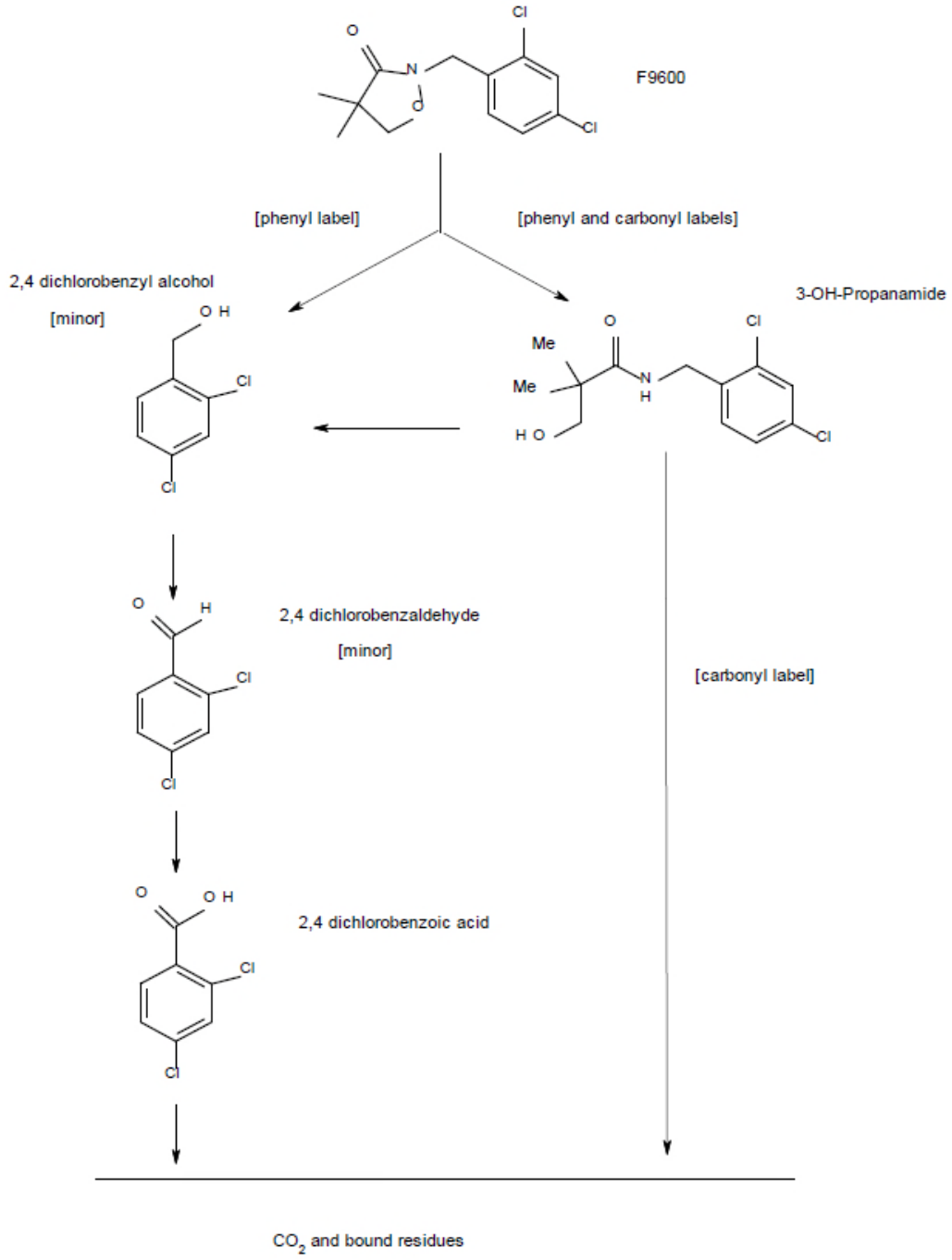
Anaerobic degradation studies for 3-OH and 2,4-DBA were carried out on one European soil (pH 7.3). For 2,4-DBA, the specimens were incubated in the dark at 20 ± 2°C prior to flooding. Due to the rapid 3-OH aerobic degradation, no incubation prior to flooding was performed in the 3-OH study. The soils were flooded with nitrogen purged de-ionised water to an approximate depth of 2 cm above the soil surface to establish anaerobic conditions which were maintained by a flow of nitrogen through the flasks for *ca* 120 days. Anaerobic modelling DT_{50} values for 3-OH and 2,4-DBA were 66.1 days and 275 days respectively. The applicant provided justification excluding 3-OH from the PECsoil calculations. The applicant states prolonged occurrence of anaerobic conditions (>90 days) are required for 3-OH to form in significant levels and that this is inconsistent with productive agriculture to assume that farmers will grow crops under conditions where prolonged presence of anaerobic conditions may be regularly expected. Furthermore, 3-OH exhibits rapid degradation (DT50 = 0.4 d) under aerobic conditions meaning, once aerobic conditions are re-established, the metabolite would degrade rapidly ensuring significant levels do not occur. This justification is accepted by the CA.

The adsorption/desorption properties of 3-OH and 2,4-DBA were determined in four different soils of European origin (pH ($CaCl_2$) 4.84-7.53, %OC 0.68-2.62) . Adsorption K_{FOC} values for 3-OH-propanamide were 65-107 mL/g (geometric mean 81.7 mL/g, arithmetic mean 1/n = 0.925) and desorption K_{FOC}-des values were 71-136 mL/g (geometric mean 93.9 mL/g, arithmetic mean 1/n = 0.924). There was no evidence of a relationship

between sorption and soil pH. However, the CA rejects the 2,4-DBA results from all four soils used within the study for use in the exposure assessment. An insufficient centrifuge speed and/or time was used to remove the aqueous solution from the soil pellet resulting in errors in the soil concentration. For two soils, because of low recoveries for the soil samples (*i.e.* negative values) and broken samples, only two or three concentrations were analysed in duplicate. Accurate and robust K_{FOC} and 1/n parameters could not be derived for these soils. Therefore, default sorption parameters (Koc = 0 mL/g, 1/n = 1) are to be used in the exposure calculations.

The metabolic pathway of bixlozone in soil is presented in Figure CA.B.**8.1.8-1**. The pathway is taken from the bixlozone anaerobic degradation study (Simmonds, R., 2015b, section CA.B.8.1.1.2.1). It is noted the schematic diagram presented in the bixlozone aerobic degradation study (Simmonds, R., 2015a, section CA.B.8.1.1.1.1) does not include metabolite 3-OH-propanamide. However, as this metabolite was detected in the soil dissipation studies (although not in quantities to fulfil the major metabolite criteria), the CA considers the anaerobic metabolic pathway to be appropriate for aerobic degradation as well. However, it is noted 3-OH is a terminal metabolite in soil and so the exact conditions in which it forms does not impact the evaluation.

Figure CA.B.8.1.8-1: Bixlozone metabolic pathway in soil



CA.B.8.2. FATE AND BEHAVIOUR IN WATER AND SEDIMENT

CA.B.8.2.1. Route and rate of degradation in aquatic systems (chemical and photochemical degradation)

CA.B.8.2.1.1. *Hydrolytic degradation*

Report:	KCA 7.2.1.1 Roohi, A.; Cooper, T., (2015)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Aqueous Hydrolysis as a Function of pH
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/004, FMC Tracking no. 2013EFT-ISX1024
Guidelines:	OECD Guideline 111 (April 2004); OPPTS Guideline 835.2120 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

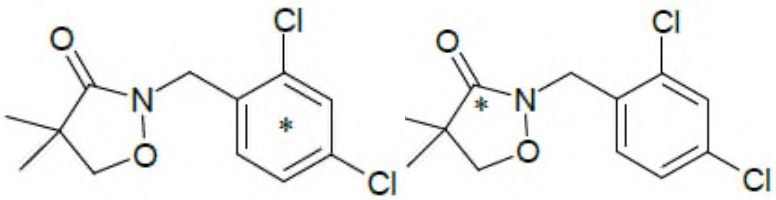
CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

The hydrolytic degradation of bixlozone was investigated following OECD 111 guidelines. A preliminary test conducted at 50 °C and pH 4 7 and 9 was undertaken, followed by a definitive study at 25 °C, 40 °C and 50 °C at pH 9.

Materials and methods

Test substance [phenyl-U-^{14}C]-bixlozone [carbonyl-^{14}C]-bixlozone



* Position of [^{14}C]-radiolabel ‘ Position of [^{14}C]-radiolabel

Active ingredient content [phenyl-U-^{14}C]-bixlozone: >98.5% in all treatment solutions (from HPLC)
[carbonyl-^{14}C]-bixlozone: >98.5% in all treatment solutions (from HPLC)

CAS No 81777-95-9

Stability of compound Radiochemical purity confirmed at application

METHOD

Preliminary test

In a preliminary study 1mg/L (nominal) [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone were added to sterile buffer solutions at pH 4, 7 and 9 in sealed, sterile brown glass vials in the dark. Twelve incubation vessels per pH were used (6 per radio label), with two additional samples for each pH treated with unlabelled bixlozone maintained at the same conditions to monitor pH and sterility. Sodium acetate (0.01 M), tris (hydroxymethyl) aminomethane hydrochloride (0.01 M), and di-sodium tetraborate (0.01 M) were used as buffer solutions for pH 4, 7, and 9, respectively. Duplicate samples were incubated at 50 ± 0.5°C for five days. Tier 1 samples were analysed directly by LSC and by HPLC within 1 day of sampling.

Sterility was checked by pipetting an aliquot of selected study samples onto nutrient agar and incubating at room temperature.

Tier 2 test

The tier 2 study was conducted with 1 mg/L (nominal) of [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone at pH 9 at 25, 40, and 50 ± 0.5°C for 30 days. As the preliminary test found bixlozone to be hydrolytically stable at 50°C at pH 4 and 7, the CA considers the temperature range appropriate. Forty incubation vessels were used (twenty for each radio label) and four additional samples for each temperature were treated with unlabelled bixlozone to monitor pH and sterility. Duplicate samples were taken for analysis at 0, 1, 3, 8, 14, 21 and 30 days. -

Description of analytical procedures

Duplicate samples of the aqueous solutions were analysed directly by LSC and HPLC with an UV detector.

Radioactivity was quantified by LSC. Radiopurity of test material was checked using HPLC and compared against a reference standard (and was ≥98.7% for Tier 1 and 2). Identity of test material was confirmed using LC-FTMS (Fourier transform-based mass spectrometry).

The limit of quantification (LOQ) is 0.06% AR for LSC and 0.2% AR for HPLC. Limit of detection (LOD) for LSC is 30 dpm (<0.01% AR).

Most tier 2 samples were analysed on the day of sampling, with a maximum delay between sampling and chromatographic analysis of 1 day. Samples were stored at <-15 °C in the dark prior to analysis.

RESULTS

Mass Balance

Mass balances were in the range 96.29%−104.37% of applied radioactivity (AR) at pH 4, 7, and 9 (50°C) in the preliminary study and were in the range 90.78%−110.32% AR at 25, 40, and 50°C (pH = 9) in the definitive study.

Preliminary test

Degradation of [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone (>10% AR) was observed at pH 9 at 50°C with *ca* 78.9-79.6 %AR remaining after 5 days, whereas minimal degradation (<1%AR) was seen at pH 4 and 7. According to the OECD guideline, the higher tier test should be performed in the appropriate buffer if there is >10 % hydrolysis in the tier 1 test. For bixlozone, this is true in a buffer of pH 9.

One isolated microbial colony was detected for the initial tier 1 sterility sample at pH 7, and a single colony on the final sterility sample of the tier 1 test at pH 4; no degradation of parent was seen at pH 4 and so this result was not considered to indicate that sterility had been compromised. These colonies are thought to be as a result of contamination during processing. This is accepted by the CA in this case.

Tier 2 test (pH 9 only)

At 25°C, minimal degradation occurred for both radiolabels, with [phenyl-U-^{14}C]-bixlozone reduced by a mean of 3.07 % AR and [carbonyl-^{14}C]-bixlozone reduced by a mean of 3.66 % AR during the study.

At 40°C (pH 9) the level of [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone declined to 70.1% and 69.6% AR, respectively. Two unidentified degradates RRT 0.89 (max. 8.9 and 8.1% AR at day 30 for the phenyl- and carbonyl-label, respectively) and RRT 0.91 (max. 21.7 and 20.4% AR at day 30) were detected. Several minor degradates (all <5% throughout study) were detected in samples from both labels.

At 50°C (pH 9), the level of [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone declined to 32.4% and 30.6% AR, respectively, with three unidentified degradates >5% AR. RRT 0.89 (max. 22.6 % and 20.3% AR for the phenyl- and carbonyl-labels, respectively), RRT 0.91 (max 34.8 and 26.5% AR for the phenyl- and carbonyl-labels, respectively), and RRT 1.08 (max. 11.5 and 15.8% AR for the phenyl- and carbonyl-labels, respectively) all reached their maximum concentration at the end of the incubation (day 30). Several minor degradates (all <5% throughout study) were detected in samples from both labels.

According to the OECD guideline, hydrolysis products representing ≥10 % of the applied dose at pH 4-9 and 25˚C should be identified and it is down to expert judgement to decide whether to identify those ≤10%AR at pH 4-9 and 25˚C. There were no hydrolysis products >10%AR at pH 9 and 25˚C (and none seen at pH 4 and 7 for 50˚C). The rate and extent of degradation increased with increasing temperature and increasing pH for both labels. Unidentified metabolites were present at >10 % AR at pH 9 in both the 40°C and 50°C tests and were just under 5% at study end at pH 9 and 25˚C. The CA considers that given the metabolites only occurred >10 % AR at pH 9 at 40 and 50°C, they are unlikely to be formed in significant amounts under environmentally relevant temperature and pH.

Sterility of all samples of the tier 2 test was confirmed.

Table CA.B.8.2.1.1-1: Product balance following hydrolytic degradation of [phenyl-U-^{14}C]-bixlozone in pH 9.0 solution incubated at 25°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.89	Unidentified RRT 0.91-0.92	Unidentified RRT 1.04-1.14	Mass balance (%AR)
0	100.11	<LOQ	<LOQ	<LOQ	100.11
0	101.53	<LOQ	<LOQ	<LOQ	101.53
Mean	**100.82**	**<LOQ**	**<LOQ**	**<LOQ**	**100.82**
1	100.15	<LOQ	<LOQ	1.44	101.58
1	100.41	0.27	0.63	0.41	101.72
Mean	**100.28**	**0.13**	**0.32**	**0.92**	**101.65**
3	102.3	0.38	0.80	<LOQ	103.48
3	102.06	<LOQ	0.99	<LOQ	103.05
Mean	**102.18**	**0.19**	**0.90**	**<LOQ**	**103.27**
8	99.94	0.56	1.06	0.30	101.86
8	99.75	0.50	1.32	0.00	101.56
Mean	**99.84**	**0.53**	**1.19**	**0.15**	**101.71**
14	98.1	0.37	2.82	0.69	101.98
14	98	0.32	2.21	0.22	100.75
Mean	**98.05**	**0.34**	**2.51**	**0.45**	**101.36**
21	96.71	1.02	3.56	<LOQ	101.29
21	94.77	1.13	3.68	0.38	99.97
Mean	**95.74**	**1.08**	**3.62**	**0.19**	**100.63**
30	98.59	1.11	4.83	0.30	104.83
30	96.91	1.17	4.90	0.17	103.15
Mean	**97.75**	**1.14**	**4.87**	**0.23**	**103.99**

Table CA.B.8.2.1.1-2: Product balance following hydrolytic degradation of [carbonyl-^{14}C]-bixlozone in pH 9.0 solution incubated at 25°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.84-0.89*	Unidentified RRT 0.91	Unidentified RRT 1.07-1.17*	Mass balance (%AR)
0	102.43	<LOQ	<LOQ	0.29	102.72
0	99.15	<LOQ	<LOQ	0.89	100.04
Mean	**100.79**	**<LOQ**	**<LOQ**	**0.59**	**101.38**
1	98.29	0.16	0.59	0.39	99.43
1	100.08	<LOQ	<LOQ	<LOQ	100.08
Mean	**99.19**	**0.08**	**0.29**	**0.19**	**99.76**
3	102.16	<LOQ	0.59	0.73	103.48
3	102.87	0.22	0.32	<LOQ	103.41
Mean	**102.51**	**0.11**	**0.45**	**0.37**	**103.44**
8	98.48	<LOQ	1.25	<LOQ	99.73
8	99.77	<LOQ	1.35	0.95	102.08
Mean	**99.12**	**<LOQ**	**1.30**	**0.48**	**100.90**
14	99.24	<LOQ	2.21	0.36	101.81
14	98.38	0.17	2.44	0.73	101.72
Mean	**98.81**	**0.09**	**2.33**	**0.54**	**101.77**
21	100.32	0.66	3.12	0.76	104.85
21	99.84	0.69	3.47	0.46	104.46
Mean	**100.08**	**0.68**	**3.29**	**0.61**	**104.66**
30	97.82	0.82	4.43	0.63	103.70
30	96.43	0.86	4.57	0.34	102.21
Mean	**97.13**	**0.84**	**4.50**	**0.48**	**102.96**

* Multiple peaks combined, no single degradate >1% AR

Table CA.B.8.2.1.1-3: Product balance following hydrolytic degradation of [phenyl-U-^{14}C]-bixlozone in pH 9.0 solution incubated at 40°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.83	Unidentified RRT 0.89	Unidentified RRT 0.91	Unidentified RRT 0.96	Unidentified RRT 1.01-1.14	Mass balance (%AR)
0	100.11	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	100.11
0	101.53	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	101.53
Mean	**100.82**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**100.82**
1	101.46	<LOQ	0.87	0.95	<LOQ	0.33	103.61
1	99.60	<LOQ	0.86	0.80	<LOQ	0.43	101.69
Mean	**100.53**	**<LOQ**	**0.86**	**0.88**	**0.00**	**0.38**	**102.65**
3	99.94	<LOQ	1.62	2.84	0.17	0.37	104.94
3	101.69	<LOQ	0.94	2.70	<LOQ	<LOQ	105.33
Mean	**100.81**	**<LOQ**	**1.28**	**2.77**	**0.09**	**0.18**	**105.13**
8	93.32	<LOQ	2.49	7.50	0.29	0.51	104.12
8	91.51	<LOQ	2.99	7.48	<LOQ	<LOQ	101.99
Mean	**92.42**	**<LOQ**	**2.74**	**7.49**	**0.15**	**0.26**	**103.05**
14	83.61	<LOQ	4.36	11.94	0.14	1.66	101.71
14	74.72	<LOQ	3.70	10.96	<LOQ	1.39	90.78
Mean	**79.17**	**<LOQ**	**4.03**	**11.45**	**0.07**	**1.53**	**96.24**
21	77.07	<LOQ	6.36	15.95	0.24	2.59	102.20
21	81.37	<LOQ	6.59	16.70	0.25	1.76	106.67
Mean	**79.22**	**<LOQ**	**6.48**	**16.32**	**0.24**	**2.17**	**104.43**
30	71.37	<LOQ	8.51	21.45	0.88	4.11	106.32
30	68.85	0.15	9.36	21.92	0.60	4.14	105.03
Mean	**70.11**	**0.08**	**8.94**	**21.69**	**0.74**	**4.12**	**105.67**

Table CA.B.8.2.1.1-4: Product balance following hydrolytic degradation of [carbonyl-^{14}C]-bixlozone in pH 9.0 solution incubated at 40°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.14-0.18	Unidentified RRT 0.89	Unidentified RRT 0.91	Unidentified RRT 0.97	Unidentified RRT 1.04-1.12	Mass balance (%AR)
0	102.43	<LOQ	<LOQ	<LOQ	<LOQ	0.29	102.72
0	99.15	<LOQ	<LOQ	<LOQ	<LOQ	0.89	100.04
Mean	**100.79**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.59**	**101.38**
1	101.96	<LOQ	<LOQ	1.35	<LOQ	0.69	104.01
1	101.40	<LOQ	0.27	1.35	<LOQ	0.68	103.71
Mean	**101.68**	**<LOQ**	**0.13**	**1.35**	**<LOQ**	**0.69**	**103.86**
3	98.69	<LOQ	1.08	3.44	<LOQ	0.74	103.95
3	101.27	<LOQ	1.01	3.44	<LOQ	0.00	105.72
Mean	**99.98**	**<LOQ**	**1.04**	**3.44**	**<LOQ**	**0.37**	**104.83**
8	89.47	<LOQ	2.44	5.91	<LOQ	0.32	98.14
8	94.01	<LOQ	2.78	6.60	<LOQ	0.00	103.38
Mean	**91.74**	**<LOQ**	**2.61**	**6.25**	**<LOQ**	**0.16**	**100.76**
14	90.67	<LOQ	4.44	11.60	<LOQ	1.38	108.09
14	87.85	<LOQ	4.18	11.32	<LOQ	1.91	105.25
Mean	**89.26**	**<LOQ**	**4.31**	**11.46**	**<LOQ**	**1.64**	**106.67**
21	75.80	<LOQ	6.19	13.84	<LOQ	2.04	97.87
21	74.90	0.78	5.06	15.82	0.09	1.60	98.25
Mean	**75.35**	**0.39**	**5.62**	**14.83**	**0.05**	**1.82**	**98.06**
30	66.95	0.57	7.42	19.06	<LOQ	4.36	98.36
30	72.34	0.42	8.70	21.65	<LOQ	2.46	105.57
Mean	**69.65**	**0.49**	**8.06**	**20.35**	**<LOQ**	**3.41**	**101.96**

Table CA.B.8.2.1.1-5: Product balance following hydrolytic degradation of [phenyl-U-^{14}C]-bixlozone in pH 9.0 solution incubated at 50°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.47	Unidentified RRT 0.61-0.86	Unidentified RRT 0.89	Unidentified RRT 0.91	Unidentified RRT 0.94	Unidentified RRT 0.96	Unidentified RRT 1.03-1.06	Unidentified RRT 1.08	Unidentified RRT 1.12-1.35	Mass balance (%AR)
0	100.11	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	100.11
0	101.53	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	101.53
Mean	**100.82**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**100.82**
1	100.07	<LOQ	<LOQ	1.94	2.49	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	104.50
1	99.96	<LOQ	<LOQ	2.36	2.86	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	105.18
Mean	**100.01**	**<LOQ**	**<LOQ**	**2.15**	**2.68**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**104.84**
3	94.15	<LOQ	<LOQ	4.01	7.17	<LOQ	<LOQ	<LOQ	0.32	<LOQ	105.65
3	93.34	<LOQ	<LOQ	3.96	8.14	<LOQ	<LOQ	<LOQ	0.37	0.37	106.17
Mean	**93.74**	**<LOQ**	**<LOQ**	**3.98**	**7.65**	**<LOQ**	**<LOQ**	**<LOQ**	**0.34**	**0.18**	**105.91**
8	75.52	<LOQ	<LOQ	10.35	17.07	<LOQ	0.44	<LOQ	1.96	<LOQ	105.35
8	74.74	<LOQ	<LOQ	10.60	17.68	<LOQ	0.63	<LOQ	2.48	<LOQ	106.13
Mean	**75.13**	**<LOQ**	**<LOQ**	**10.48**	**17.38**	**<LOQ**	**0.53**	**<LOQ**	**2.22**	**<LOQ**	**105.74**
14	58.90	<LOQ	<LOQ	12.68	24.77	<LOQ	1.48	<LOQ	4.64	0.44	102.91
14	59.48	<LOQ	<LOQ	15.79	22.72	<LOQ	1.64	<LOQ	5.76	0.23	105.62
Mean	**59.19**	**<LOQ**	**<LOQ**	**14.24**	**23.74**	**<LOQ**	**1.56**	**<LOQ**	**5.20**	**0.34**	**104.26**
21	45.68	<LOQ	<LOQ	17.30	29.94	<LOQ	3.00	1.07	7.43	0.97	105.39
21	45.86	0.13	<LOQ	19.28	31.65	0.28	2.83	0.76	9.04	0.49	110.32
Mean	**45.77**	**0.06**	**<LOQ**	**18.29**	**30.80**	**0.14**	**2.91**	**0.91**	**8.24**	**0.73**	**107.85**
30	32.22	<LOQ	0.48	22.84	34.35	0.76	4.31	1.40	11.13	0.60	108.09
30	32.57	0.12	0.17	22.38	35.20	0.60	3.87	1.27	11.77	0.63	108.60
Mean	**32.40**	**0.06**	**0.32**	**22.61**	**34.78**	**0.68**	**4.09**	**1.33**	**11.45**	**0.61**	**108.34**

Table CA.B.8.2.1.1-6: Product balance following hydrolytic degradation of [carbonyl-^{14}C]-bixlozone in pH 9.0 solution incubated at 50°C (Tier 2 study)

Time (days)	Bixlozone	Unidentified RRT 0.14-0.87	Unidentified RRT 0.89	Unidentified RRT 0.91	Unidentified RRT 0.94	Unidentified RRT 1.03-1.06	Unidentified RRT 1.08	Unidentified RRT 1.12-1.31	Mass balance (%AR)
0	102.43	<LOQ	<LOQ	<LOQ	<LOQ	0.29	<LOQ	<LOQ	102.72
0	99.15	<LOQ	<LOQ	<LOQ	<LOQ	0.89	<LOQ	<LOQ	100.04
Mean	**100.79**	**<LOQ**	**<LOQ**	**<LOQ**	**<LOQ**	**0.59**	**<LOQ**	**<LOQ**	**101.38**
1	90.32	<LOQ	1.52	2.79	<LOQ	<LOQ	<LOQ	3.54	98.18
1	95.84	<LOQ	1.02	3.30	<LOQ	<LOQ	<LOQ	0.39	100.55
Mean	**93.08**	**<LOQ**	**1.27**	**3.05**	**<LOQ**	**<LOQ**	**<LOQ**	**1.97**	**99.36**
3	95.21	<LOQ	3.93	7.82	<LOQ	<LOQ	<LOQ	<LOQ	106.96
3	87.45	<LOQ	4.08	6.30	<LOQ	<LOQ	<LOQ	0.93	98.76
Mean	**91.33**	**<LOQ**	**4.00**	**7.06**	**<LOQ**	**<LOQ**	**<LOQ**	**0.47**	**102.86**
8	70.80	<LOQ	6.96	10.35	<LOQ	0.46	7.09	0.53	98.93
8	72.38	<LOQ	8.50	15.00	<LOQ	0.00	1.79	<LOQ	97.67
Mean	**71.59**	**<LOQ**	**9.10**	**12.68**	**<LOQ**	**0.23**	**4.44**	**0.27**	**98.30**
14	61.34	1.32	13.62	22.09	<LOQ	0.19	5.09	0.53	104.17
14	58.22	1.65	14.61	21.70	<LOQ	0.19	5.31	0.72	102.40
Mean	**59.78**	**1.49**	**14.11**	**21.89**	**<LOQ**	**0.19**	**5.20**	**0.62**	**103.29**
21	45.41	3.43	18.12	26.19	0.19	0.96	7.52	0.54	102.37
21	41.24	3.61	17.19	25.01	0.13	1.04	8.81	0.68	97.73
Mean	**43.32**	**3.52**	**17.66**	**25.60**	**0.16**	**1.00**	**8.17**	**0.61**	**100.05**
30	30.92	6.07	19.90	25.43	<LOQ	1.50	15.75	0.36	99.94
30	30.34	6.49	20.63	27.46	<LOQ	1.10	15.83	0.24	102.11
Mean	**30.63**	**6.28**	**20.27**	**26.45**	**0.00**	**1.30**	**15.79**	**0.30**	**101.02**

The applicant calculated degradation rates using CAKE 2.0. For day 0 in the kinetic fit, they corrected the recovered amount for radiochemical purity (99.7 and 99.6% for phenyl and carbonyl radiolabels, respectively), then at other time-points used measured parent. The recovered amount at 25°C for day 0 for each label, was also used for day 0 at 40 and 50˚C. The CA accepted this as a best estimate of the amount of parent dosed into the system and independently verified the applicant's results.

The best fit DT_{50}- and DT_{90}-values for both labels are shown in Table CA.B.**8.2.1.1-7**. Hydrolysis increased with increasing temperature. At pH 9 no significant hydrolysis was observed at 25°C with bixlozone still accounting for > 97% AR after 30 days.

Table CA.B.8.2.1.1-7: The best fit DT_{50}- and DT_{90}-values for [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone for hydrolysis in sterile buffer solutions at pH 9

Radiolabel	Temp (°C)	Kinetic model†	DT_{50} (days)	DT_{90} (days)	χ^2 (%)	t-test	Visual fit
[phenyl-U-^{14}C]-bixlozone	25	SFO	446	>1000	1.01	p<0.001	Good
	40	SFO	53.5	178	2.33	p<0.001	Good
	50	SFO	18	59.7	1.43	p<0.001	Good
[carbonyl-^{14}C]-bixlozone	25	SFO	742	>1000	1.01	p=0.0196	Good
	40	SFO	53.6	178	1.79	p<0.001	Good
	50	SFO	17.9	59.3	2.00	p<0.001	Good

† SFO = single first order

The applicant has also calculated Arrhenius constants at 10, 20, 25, 30, 60 and 70°C from the mean of the rate constants for radiolabels at 40 and 50°C. The CA accepts the applicant's results, however, notes that due to the lack of hydrolytic degradation observed at environmentally relevant temperature, this does not change the regulatory outcome.

CONCLUSIONS

Bixlozone was hydrolytically stable at pH 4 and 7 over 5 days at 50°C. Bixlozone did not hydrolyse at pH 9 over 30 days at the environmentally relevant temperature of 25°C with expected DT_{50}-values > 1 year. Therefore, no metabolic pathway has been proposed by the applicant. The rate and extent of degradation, however, increased with increasing temperature and pH. Unidentified metabolites were formed at >10% at pH 9 and 40-50˚C, but the CA considers that these metabolites will be unlikely to form at significant levels under environmentally relevant temperature and pH conditions.

CA.B.8.2.1.2. ***Direct photochemical degradation***

Report:	KCA 7.2.1.2, O'Connell, C., (2015)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Phototransformation of Chemicals in Water - Direct Photolysis
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/14/003, FMC Tracking no. 2013EFT-ISX1023
Guidelines:	OECD Guideline 316 (October 2008); OPPTS Guideline 835.2240 (no date provided)
GLP:	Yes (laboratory certified by UK National Authority)

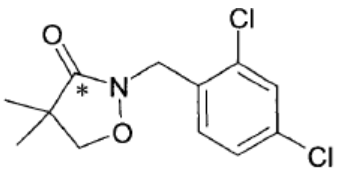
CA comments	No significant deviations from the guidelines occurred. **This study is relied upon.**

INTRODUCTION

The photolytic degradation of bixlozone in water was investigated following OECD 316 guidelines. Samples were continuously irradiated at 25°C with a xenon lamp (wavelengths <290 nm filtered out, similar spectrum to natural sunlight) for up to 13 days. The daily averaged intensity was adjusted to 64 W/m² for the 300-400 nm range (equivalent to 2.53 days of natural summer sunlight at latitude 30-50°N). The maximum incubation time corresponded to approximately 33 days of natural summer sunlight at latitude 30-50°N.

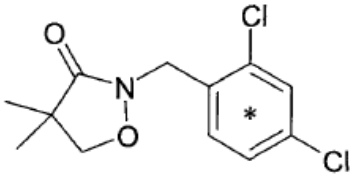
Test substances

Test substance: [carbonyl-^{14}C]-bixlozone



(* indicates position of [^{14}C]-label)

Test substance: [phenyl-U-^{14}C]-bixlozone



(* indicates position of [^{14}C]-label)

Lot/Batch no.	[carbonyl-^{14}C]-bixlozone: CFQ42018 [phenyl-U-^{14}C]-bixlozone: CFQ42017
Purity	Radiochemical Purity [carbonyl-^{14}C]-bixlozone: >97% (by HPLC) Radiochemical Purity [phenyl-U-^{14}C]-bixlozone: >97% (by HPLC)
CAS No	81777-95-9
Stability of compound	Radiochemical purity confirmed before application

METHOD

Tier 1 test (theoretical screen)

Determination of UV absorbance was performed with unlabelled bixlozone prepared in acetonitrile at concentrations of 0.005, 0.01, 1.0 and 10.0 mg/mL. Pyridine and p-nitroacetophenone (PNAP) solutions were also prepared as actinometers in acetonitrile. The spectra from 200 to 800 nm were determined for the test matrix. Determination of optical dilution was also performed with 1.0 mg/L bixlozone.

Tier 2 test (definitive test)

[Carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone (1 mg a.s./L nominal) were dispensed in a sterile aqueous buffer (0.01 M, phosphate) at pH 7 in duplicate quartz photolysis vessels for irradiated samples. For dark controls, the same nominal concentration and buffer were used, applied to single quartz photolysis vessels.

The irradiated test solutions were continuously irradiated at 25±1°C with a xenon lamp (wavelengths <290 nm filtered out, similar spectrum to natural sunlight and a light intensity of 63.7 Wm^{-2}) for up to 13 days in a Heraeus Suntest (CPS+) unit. Appropriate dark controls were included, with [carbonyl-^{14}C]-bixlozone controls placed in a suntest unit with the xenon lamp switched off, and [phenyl-U-^{14}C]-bixlozone controls placed in an incubator. To ensure continuous darkness, the vessels were covered with aluminium foil.

Duplicate irradiated and single dark control samples were taken at time zero and after 1, 2, 5, 7, 9, and 13 days. The daily averaged intensity was adjusted to 64 W/m² for the 300-400 nm range (equivalent to 2.53 days of natural summer sunlight at latitude 30-50°N). The maximum incubation time corresponded to approximately 33 days of natural summer sunlight at latitude 30-50°N.

The applicant has argued that due to high mass balance values, no volatile material was lost during the preliminary investigations, and therefore no traps were required in the definitive experiments. The CA considers that volatile traps should have been used for the final study, however, judges that the results are valid for the following reasons. The mass balance derived in both the preliminary study and definitive study is within the acceptable range of 90-110 % AR. At no point during the definitive test did mass balance fall below 91.5 % for the carbonyl label, or 93.8 % for the phenyl label. The applicant accounted for recoveries not attributable to the test item, showing that there were multiple minor components produced during the study. Therefore, any unquantified volatiles or CO_2 would be lower than 10 % and not defined as significant in the guideline. The results of the soil hydrolysis study provides some reassurance that volatile compounds and CO_2 trapped and analysed in that study, accounted for <3.4 %AR. Therefore, the CA accepts that significant volatile compounds, CO_2 or metabolites would not be expected in this study.

Single samples were taken from both irradiated and non-irradiated solutions for the determination of quantum yield. A validated method was used to determine PNAP (pyridine/p-nitroacetophenone) concentrations for the quantum yield determination.

The sampling times of 0, 1, 2, 5, 7, and 13 days were used for irradiated and dark control samples (including actinometers). All samples were immediately chilled in the dark at *ca* 4°C to ensure any potential volatiles were cooled. After approximately 15 minutes samples were equilibrated to room temperature for analysis by LSC and HPLC.

Sterility of buffer solution was verified before incubation and at the end of the study by aseptically pipetting an aliquot of the solution onto sterile nutrient agar and leaving at room temperature.

Description of analytical procedures

LSC was used to quantify mass balance of [phenyl-U-^{14}C]- and [carbonyl-^{14}C]-bixlozone in irradiated and dark control samples. Radioactivity less than twice background level was considered to be below the limit of detection.

Duplicate irradiated and single dark control samples (per radiolabel) from the aqueous solutions were analysed by reverse phase HPLC using a zorbax RX C18 column, gradient elution of 0.01 % acetic acid in water and 0.01 % acetic acid in acetonitrile, ^{14}C-flow-through with UV and radio-detection (LOQ 0.13% AR; LOD not provided). Radiochemical purity was also measured via this method. Selected samples were also analysed by co-chromatography by normal phase TLC with chloroform:methanol:acetic acid (90:10:1) and dichloromethane:methanol:formic acid (98:2:1) against reference standards to confirm sample peak identification. due to a non-calibrated measuring cylinder being used, measured results from the zero hour samples were used to determine the concentration of the solution applied.

Identity of [14C]-bixlozone was performed by LC-MS using the same HPLC conditions as above and monitoring an ion transition of 100 – 500 m/z.

Actinometer analysis was performed via isocratic HPLC using acetonitrile:water:acetic acid (50:50:1, v/v/v), utilising a series of five linearity solutions (nominal concentrations of 40 – 210 ng/mL).

RESULTS

Mass Balance

Recoveries of radioactivity for the irradiated samples were 91.5-102.8% AR for [carbonyl-^{14}C]-bixlozone and 93.8-104.4% AR for [phenyl-U-^{14}C]-bixlozone. Recoveries of radioactivity for the non-irradiated (dark control) samples were 96.0-100.5% AR for [carbonyl-^{14}C]-bixlozone and 98.3-102.5% AR for [phenyl-U-^{14}C]-bixlozone.

Findings

Radiochemical purity for the dosing solution of [Carbonyl-^{14}C]-bixlozone and [Phenyl-U-^{14}C]-bixlozone was 99.2 % and 98.7 % respectively.

Tier 1 test (theoretical screen)

The maximum absorbance for bixlozone was detected at 200 nm, and absorbance within the 290 – 800 nm spectrum was low (mean of 0.0000 ± 0.0014 au at a nominal concentration of 10.0 mg/mL). However, the applicant has performed a tier 2 test, stating that the absence of detectable absorption does not exclude the possibility of photodegradation. Due to the low absorbance >290 nm, the applicant was unable to determine a quantum yield.

Optical dilution analysis showed maximum absorbance of $3x10^{-6}$au for both 290 and 295 nm. Based on this, the CA accepts that the test solution used in the Tier 2 test is optically dilute.

Tier 2 test (definitive test)

The results of analyses are shown in Table CA.B.**8.2.1.2-1**. Bixlozone was degraded to many minor photoproducts after 13 days continuous irradiation with in total 30 and 32 degradation products for [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone, respectively. All of the degradation products were < 5% AR at each sampling point. No major degradation products were found. The non-irradiated samples (dark control) confirmed that there was no degradation in darkness and indicated that bixlozone was hydrolytically stable during the incubation period for both label positions.

The sterility of the test solutions was maintained throughout the experiment, except for two solutions with a single (1 day, irradiated) and two colonies (9 days, non-irradiated) on agar plates for the [carbonyl-^{14}C]-label. The applicant states this did not invalidate these data points, as the data obtained from the replicate analysis or from other samples incubated in the dark showed similar degradation behaviour. This is accepted by the CA.

The applicant reported that the 9-day irradiated samples treated with [phenyl-U-^{I4}C]-bixlozone felt noticeably chilled compared to previous sampling, due to erroneous measurements of the temperature probe. It was discovered that the probe was monitoring air temperature not buffer temperature in error. As the air heats quicker than the buffer under the UV lamp, this prompted the cooling plate to be chilled, with samples falling from 25±1°C to *ca* 23°C for roughly 2 days. This was not considered to have adversely affected results, as the results were similar to those observed in the preliminary study with the same radiolabel and the degradation rate of the actinometer irradiated alongside the phenyl- treated sample was comparable to that of actinometer irradiated alongside the carbonyl- treated sample.

Table CA.B.8.2.1.2-1: Mass balance following the aqueous photolysis of [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone in pH 7 buffer

	[Carbonyl-^{14}C]-bixlozone				[Phenyl-U-^{14}C]-bixlozone			
Time (days)	**Bixlozone (% AR)**	**Metabolites D1-D36 (≤ max % AR per individual)**	**No. of metabo-lites**	**Mass balance (% AR)**	**Bixlozone (% AR)**	**Metabolites D1-D36 (≤ max % AR per individual)**	**No. of metabo-lites**	**Mass balance (% AR)**
Time zero								
0	98.02	≤ 1.27	4	99.7	101.1	≤ 1.06	2	103.2
0	97.10			100.3	95.27			96.8
Irradiated								
1	100.8	≤ 0.92	2	102.1	99.99	≤ 1.00	11	104.4
1	98.71			100.4	98.76			103.4
2	92.73	≤ 0.99	21	102.8	95.83	≤ 0.69	13	101.0
2	89.43			97.8	93.18			97.8
5	83.79	≤ 2.21	21	98.4	89.12	≤ 1.30	24	99.5
5	83.89			98.1	84.91			100.4
7	77.19	≤ 2.52	18	94.2	81.68	≤ 1.51	27	94.6
7	78.56			95.6	80.10			99.1
9	69.37	≤ 2.89	23	94.4	77.04	≤ 1.86	28	97.3
9	74.31			95.5	74.87			98.1
13	57.07	≤ 4.49	26	91.5	67.64	≤ 3.54	29	94.8
13	56.29			96.3	61.45			93.8
Dark control								
1	98.6	≤1.12	3	100.5	101.2	≤0.74	1	101.9
2	97.8	≤0.87	1	98.7	98.3	≤0.99	5	100.7
5	99.1	≤1.45	1	100.5	99.0	≤1.05	3	101.6
7	96.2	≤1.00	2	97.6	96.7	≤0.66	3	98.3
9	94.1	≤1.32	2	96.0	100.6	≤0.70	2	101.8
13	98.4	≤ 1.29	1	99.7	100.7	≤ 0.45	4	102.5

It was not possible to determine the quantum yield for bixlozone for both label positions due to the very low UV absorption at wavelengths > 290 nm.

In order to convert the time under continuous irradiation within the suntest unit, the applicant has used the equation below from the draft OECD guideline for phototransformation of chemicals on soil surfaces:

$$d = \frac{h \times r}{0.75 \times 12}$$

Where d = days of summer sunlight (natural sunlight)
h = hours of irradiation by the xenon lamp
r = ratio of intensity (irradiance) of the xenon radiation to that of summer sunlight
0.75 = correction for diurnal variation of natural sunlight
12 = conversion factor of hours to days

As the irradiance intensity within the suntest unit was measured as 63.7 W/m^2, the intensity of natural summer sunlight in the range 300 – 400 nm taken from the guideline of 67 W/m^2, the r value is 0.951. The CA has also performed this calculation and gets very similar answers, with differences most likely due to rounding. The CA has checked and accepts the applicant's conversion, where 24 hours of continuous irradiation is equivalent to 2.53 days of natural summer sunlight.

Transformation of the parent compound

The first order DT_{50} and DT_{90} values (SFO) for [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone were determined following FOCUS kinetics guidance in CAKE v.2 (Table CA.B.**8.2.1.2-2**). The CA re-ran the kinetic assessment and was able to reproduce, or get very similar results to, the applicant's kinetic endpoints. All SFO visual and statistical fits were good, and therefore fitting with other models was not attempted. The applicant's fittings are therefore accepted by the CA.

Table CA.B.8.2.1.2-2: Aquatic photolytic degradation rate of [^{14}C]-bixlozone

	Irradiated						**Dark control**		
	Test conditions				**Natural sunlight†**		**Test conditions**		
Radiolabel position	**DT_{50} (hours)**	**DT_{90} (hours)**	**χ^2 error (%)**	**t-test**	**DT_{50} (days)**	**DT_{90} (days)**	**DT_{50} (hours)**	**χ^2 error (%)**	**t-test**
[carbonyl-^{14}C]-bixlozone	417	>1000	2.3	p<0.001	44.0	>105.6	> 1000	1.2	p=0.25
[phenyl-U-^{14}C]-bixlozone	515	>1000	1.4	p<0.001	54.4	>105.6	> 1000	1.2	p=0.50

† Converted to equivalent days under natural summer sunlight at latitude 30-50°N

CONCLUSION

The degradation rate of both labels under irradiated conditions was slow, whilst appropriate controls confirmed that there was no degradation in darkness. Bixlozone was slowly degraded to many minor photoproducts after 13 days continuous irradiation. All of these degradation products were < 5% AR at each sampling point. The first-order DT_{50} values were 44.0 and 54.4 days for [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone, respectively, under natural summer sunlight at latitude 30-50°N. It was not possible to determine the quantum yield for bixlozone due to the very low UV absorption at wavelengths > 290 nm.

CA.B.8.2.2. **Route and rate of biological degradation in aquatic systems**

CA.B.8.2.2.1. ***"Ready biodegradability"***

Report:	KCA 7.2.2.1/01, Shannon, M (2017)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Assessment of Ready Biodegradability by Measurement of CO_2 Evolution
Testing facility:	Smithers Viscient (ESG) Ltd, Harrogate, UK
Document No:	Study no. 3201875, FMC Tracking no. 2017EFT-ISX3306
Guidelines:	OECD Guideline 301B US EPA Guideline, OPPTS 835.3110 (January, 1998)
GLP:	Yes (laboratory certified by UK National Authority)

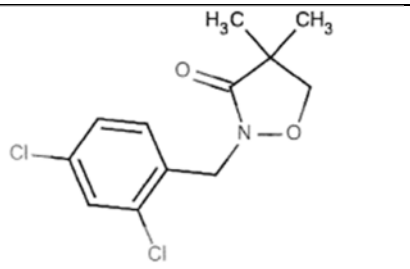
CA comments	The CA notes there is a relatively large difference between both test substance replicates, with replicate 2 exhibiting very little to no biodegradation over the study period (max 3% biodegradation). However, because replicate 1 also exhibited little biodegradation (max 13% biodegradation) over the course of the study with both replicates showing biodegradation far below the 60% required for the substance to be considered readily biodegradable, the CA is of the opinion that this difference does not impact upon the conclusions of the study. **This study is relied upon.**

INTRODUCTION

The objective of this study was to assess the 'ready' biodegradability of bixlozone by measuring the yield of carbon dioxide (CO_2) recovered under standard test conditions. The study was carried out in accordance with OECD Guideline 301B. No deviation from the guidelines was noted.

The study was undertaken on non-radiolabelled bixlozone and sodium benzoate was used as a reference substance.

Table CA.B.8.2.2.1-1: Test Material Information

Test Substance Name:	Bixlozone technical
Synonyms:	Bixlozone
Structure:	
Molecular formula:	$C_{12}H_{13}Cl_2NO_2$
Molecular Weight:	274.143
Batch Number:	PL14-0163
Purity:	99.8%

STUDY DESIGN

Test system

A sample of activated sludge was collected from one of the return lines at Burley Menston sewage treatment works (West Yorkshire, UK) which has a predominantly domestic waste-water catchment.

The sample was transported in a closed container, but with an adequate headspace, to prevent the sample becoming anaerobic. On arrival, the sample was aerated by means of a compressed air supply.

The suspended solids concentration of the activated sludge was determined by filtering a subsample (25 mL) through a pre-dried and pre-weighted glass microfibre filter (Whatman GF/C). The filtered and retained solids were then dried in an oven (nominally 105 ˚C) and re-weighed. The weight of the sludge solids was determined from the difference in the weights before and after drying. The concentration of suspended solids was calculated to be 2.76 g/L.

The activated sludge used in this study was not deliberately acclimatised or adapted to bixlozone before exposure under test conditions.

Experimental conditions

The study consisted of four treatment groups:

- Blank control: Inoculated mineral salts medium
- Test substance: Inoculated mineral salts medium and test substance
- Reference substance: Inoculated mineral salts medium and sodium benzoate
- Toxicity control: Inoculated mineral salts medium, test substance and sodium benzoate

Duplicate vessels were prepared for the test substance, reference substance and blank control groups. A single vessel was prepared for the toxicity control.

The test was conducted in an aqueous, synthetic, mineral salts medium. A test medium concentrate was prepared in reversed osmosis (RO) water containing 30 mL/L solution (a) and 3 mL/L of each of solutions (b), (c), and (d). Solutions (a) to (d) were prepared as follows:

> (a) potassium dihydrogen phosphate (8.50 g); dipotassium hydrogen phosphate (21.75 g); disodium hydrogen phosphate dihydrate (33.40 g); ammonium chloride (0.50g) all dissolved in and made up to 1 L with RO water.
>
> (b) calcium chloride dihydrate (36.40 g), dissolved in and made up to 1 L with RO water.
>
> (c) magnesium sulphate heptahydrate (22.50 g), dissolved in and made up to 1 L with RO water.
>
> (d) ferric chloride hexahydrate (0.25 g) and concentrated hydrochloric acid (1 drop), dissolved in and made up to 1 L with RO water.

On the basis of the suspended solids determined to be 2.76 g/L, the medium was inoculated with activated sludge (261 mL in a total volume of 8 L) to give a suspended solids concentration of 90 mg/L. This provided a nominal final solids concentration of 30 mg/L in each test vessel (500 mL added to a total volume of 1.5. L).

The inorganic carbon concentration of the inoculated mineral salts medium was determined using an InnovOx carbon analyser. In this analysis, inorganic carbon (IC) in the samples was released as CO_2 by acidification with hydrochloric acid. The CO_2 was then passed to a non-dispersive infra-red (NDIR) detector. The concentration of carbon dioxide was determined in the NDIR detector, by measuring the amount of infra-red energy absorbed by the sample. A calibration check was performed on each occasion by injecting a series of sodium hydrogen carbonate standards. The existing calibration curve was used to quantify the IC present in the samples. Each sample was analysed in triplicate/quadruplicate.

Bixlozone was accurately weighed (42.85 to 42.87 mg) for direct addition to the test substance and toxicity control vessels, to give a nominal test substance concentration corresponding to 15 mg carbon/L.

A reference substance stock solution (2.25 g carbon/L) was prepared by dissolving sodium benzoate (1.93g) in RO water (500ml). Reference and toxicity control vessels were treated with the stock solution (10 mL), to give a nominal sodium benzoate concentration corresponding to 15 mg carbon/L.

Measurements of pH were made in the blank control and reference substance vessels at the start of incubation and in all vessels at the end of the test prior to the addition of the hydrochloric acid. Measured pH values ranged for pH 7.50 to pH 7.51 on Day 0 and pH 7.46 to pH 7.64 on Day 28. The test vessels were incubated in the dark at 22 ± 2ºC. Air flow was delivered from a cylinder of CO_2-free air. Adjustments were made as necessary to maintain flow rate of ca 50mL per minute.

Sampling

At appropriate intervals the air supply to each vessel was interrupted and the trap bottle (containing aqueous barium hydroxide at nominally 0.0125 M) nearest to the vessel was removed from sampling. The remaining two bottles of the series were moved towards the test vessel and a fresh trap bottle placed on the end of the series. Once the series of trap bottles were connected to the test vessel the air supply was restarted. The initial barium hydroxide stock concentration and the residual concentrations in the detached trap bottles were determined by titration against hydrochloric acid (nominally 0.05 M) using 0.5% ethanolic phenolphthalein indicator matching (± 0.1 mL) titres were obtained.

Evolved CO_2 from the vessels was trapped in the barium hydroxide traps by formation of a barium carbonate precipitate. This resulted in a decrease in the concentration of barium hydroxide. Consequently, the amount of evolved CO_2 was calculated from the decrease in the barium hydroxide concentration, determined by titration against hydrochloric acid.

Following the trap analysis on Day 28, each culture vessel was opened and concentrated hydrochloric acid (1mL) added. The vessels were then reconnected to the series of trap bottles and aeration continued until the following day.

The acidification and aeration procedure drove off generated carbon dioxide remaining in solution. Final sampling and titrations were carried out on Day 29, when all of the traps in each series were sampled.

Calculations

The Theoretical CO_2 yields were calculated as below.

$TCO = D_{abs} \times P_c \times 3.667$

Where:
D_{abs} = the absolute dose i.e. the amount (mg) of the test or references substance added to the culture
P_c = the percentage carbon content of the test of reference substance
3.667 = the weight (mg) of CO_2 produced from 1 mg of carbon.

Biodegradation (D_t) of the reference substance and of bixlozone expressed in terms of percentage theoretical CO_2 yield (82.5 mg CO_2) was calculated by applying the formula:

$$D_t = \frac{cumulative\ mg\ CO_2\ produced\ at\ time\ (t)}{82.5} \times 100$$

RESULTS

Table CA.B.8.2.2.1-2: Biodegradation as a percentage of theoretical CO_2 yield

Treatment group		Biodegradation (%)												
		Day												
		2	**5**	**7**	**9**	**12**	**16**	**19**	**22**	**26**	**28**	**29****		
Reference substance	Rep 1	32	57	67	71	78	82	83	86	86	87	89	89	90
	Rep 2	28	51	64	72	78	85	88	91	91	92	92	94	94
	Mean	**30**	**54**	**65**	**71**	**78**	**83**	**86**	**88**	**89**	**89**	**91**	**91**	**92**
Test substance	Rep 1	0	3	5	6	8	9	10	11	11	12	13	13	13
	Rep 2	0	0	0	0	2	2	2	3	2	1	0	0	0
	Mean	**0**	**1**	**2**	**3**	**5**	**6**	**6**	**7**	**7**	**6**	**7**	**7**	**7**
Toxicity control*	Rep 1	0	31	54	63	69	73	76	77	78	78	77	78	78

*Toxicity control values are corrected for the mean test substance degradation

**Day 29 refers to the day that titrations of trap content from acidified vessels were performed. Actual acidification was performed on Day 28.

The CA notes there is a relatively large difference between both test substance replicates, with replicate 2 exhibiting very little to no biodegradation over the study period. The applicant does not explore the possible reasons for this difference in the study report. However, because replicate 1 also exhibited little biodegradation over the course of the study, the CA is of the opinion that this difference does not impact upon the conclusions of the study. Furthermore, the CA notes that there is a large margin between the 60% ready biodegradability threshold and the level of biodegradation detected in the study. The CA also notes that the toxicity control does not indicate that the test substance was toxic to the inoculum.

CONCLUSION

To be considered ready biodegradable, a test substance must achieve a 60% biodegradation by the end of the test. Additionally, the test substance must biodegrade by at least 60% within the 10 days once 10% biodegradation is reached. Bixlozone showed limited biodegradation with a maximum biodegradation replicate value of 13% during the study. Therefore, bixlozone cannot be considered readily biodegradable.

CA.B.8.2.2.2. *Aerobic mineralisation in surface water*

Report:	KCA 7.2.2.2/01, Simmonds, R. (2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Aerobic Mineralization in Surface Water Study (OECD 309)
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/15/007, FMC Tracking no. 2015EFT-ISX2197
Guidelines:	OECD Guideline 309 OPPTS Guideline 835.3190 (October 2008)
GLP:	Yes (laboratory certified by UK National Authority)

CA comments	The CA notes the following issues with the study: • The test water was stored for five days after sampling, prior to study commencement, at a temperature of 20 °C as opposed to the OECD stated temperature of 4 °C. However, the control samples showed the test water to still be viable during the course of the study and so this is deemed to have not significantly affected the outcomes of the study. • The volume of acetonitrile added to the solvent control flask was twice the volume present in the 100 µg/L test concentration samples. However, rapid mineralisation in the control samples was observed indicating the test system was not significantly affected. • Mass balances in the control samples were outside the OECD range of 90 – 110%. However, as significant biological activity was still exhibited in the controls, and acceptable mass balances were observed in the main test systems, this was deemed to not significantly affect the outcomes of the study. For the reasons outlines above, the CA does not consider these issues to have significantly impacted on the conclusions of the study. **This study is relied upon.**

INTRODUCTION

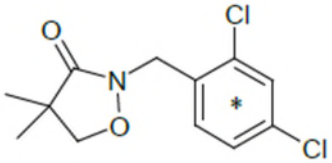
The aerobic mineralisation and degradation rate of the herbicidal active substance bixlozone in an aquatic system under dark conditions was investigated. The test was undertaken to OECD 309 guidelines.

METHOD

Test Materials

The chemical properties of the active substance used in this study are presented in Table CA.B.8.2.2.2-1.

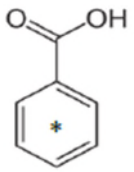
Table CA.B.8.2.2.2-1: Bixlozone Test Chemical Properties

Common Name:	Bixlozone
Chemical Name (IUPAC):	2-[(2,4-dichlorobenzyl)methyl]-4,4-dimethyl-1,2-oxazolidin-3-one
Chemical Name (CAS):	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-3-isoxazolidinone
CAS registry number:	81777-95-9
Molecular Formula	C12H13Cl2NO2
Molecular weight	274.14 (unlabelled)
Solubility:	42.0 mg/L (purified water), 42.3 mg/L (pH 4), 39.6 mg/L (pH 7), 41.9 (pH 9) [5]
Molecular structure:	 * Position of [^{14}C]-radiolabel
Source:	Supplied by Quotient Bioresearch (at sponsor's request)
Batch Number:	CFQ42475
FMC Isotope Number:	333
Physical form:	Solution in Toluene
Radiochemical purity:	99.9% (as stated on certificate of analysis)
Specific activity:	60 mCi/mmol or 8.04 MBq/mg
Storage conditions:	< -20°C in the dark

The CA notes that, in line with the OECD 309 guidelines, preferably both rings in the chemical structure would be radiolabelled. However, as acceptable mass balances were obtained with only the phenyl ring radiolabelled, the CA accepts the radiolabel position on this occasion.

The chemical properties of benzoic acid, used in the control test systems, are summarised in Table CA.B.8.2.2.2-2.

Table CA.B.8.2.2.2-2: Benzoic acid (control item) chemical properties

Source:	Supplied by Sponsor
Manufacturing Lot Number:	G3773-62
Sample Reference (Lot No.):	PL14-0163
% Concentration:	99.8 % w/w (as stated on the certificate of analysis)
Expiry date:	February 10, 2021
Molecular structure:	 * Position of [^{14}C]-radiolabel
Source:	American Radiochemicals inc.
Batch number:	121214 (ARC 0187)
Purity:	99% (as stated on the certificate of analysis)
Specific Activity:	60 mCi/mmol (18.18 MBq / mg)
Storage conditions:	< -15°C (after preparation)
Safety precautions:	Normal handling procedures for radiochemicals

Test System

Water was collected from Carsington Water, UK, an approximately 300 hectare reservoir which receives water from the River Derwent and surrounding grassland. It also receives urban discharges. Water was collected by bucket and passed through a 100 µm filter bag into a 15 litre bottle. The water was couriered at ambient temperature to the test facility and acclimatised at 20 °C before use in the study. The study commenced on 11 October; five days after collection. During this time the water was incubated at 20 °C. The CA notes that this is contrary to the OECD guidelines which state that the water should be kept at 4 °C between collection and use, however mineralisation in the positive control flasks demonstrates that the test water was still viable throughout the study. Water characteristics are summarised in Table CA.B.8.2.2.2-3.

Table CA.B.8.2.2.2-3: Physio-chemical characteristics of surface water

	Carsington Water
Batch ID	16/065
Source	Carsington Water, Millfields, UK
OS Map reference	SK 24813 49995
Sampling date:	06/10/2016
Visual quality	Turbid, green-brown colour
Depth where sampled	50-65 cm (lake edge)
Temperature at sampling depth (°C)	13.7
Oxygen at sampling depth (% sat.)	91.4
Conductivity at sampling depth (ppm / µS)	146 / 291
pH at sampling depth	7.60
5d-BOD (mg/L)	14.6*

*mean of 3 replicates

Experimental conditions

Test water samples (100 mL) were added to 250 mL glass conical flasks attached to an aeration and volatile trapping system, consisting of 1 × ethylene glycol and 2 × 2 M potassium hydroxide traps. Test systems were treated with [phenyl-U-^{14}C]-bixlozone at nominal application rates of 10 and 100 µg/L and incubated at 20 ± 2°C, in the dark; the measured application rates of the two test systems were 9.8 µg/L and 97.8 µg/L. Aerobic conditions in the water phase were maintained by the constant passage of moist air through the sample flasks, above the water surface, and out through the trap solutions, and by the stirring of the water to facilitate mass transfer across the air/water interface.

For each of the two test concentrations, 24 flasks containing the test water were prepared for treatment with bixlozone, allowing duplicate samples to be taken at each of 10 specified sampling time-points whilst leaving four flasks which could be used as spares. A further 6 flasks containing the test water were prepared and maintained on the system as untreated spares.

Sterilised control flasks (two per dose rate) were prepared and incubated under the same conditions as the test systems to determine abiotic degradation of the test item; sterilisation was done by autoclave at 121 °C for 15 minutes. Positive control flasks were treated with [phenyl-U-^{14}C]-benzoic acid at a nominal concentration of 10 µg/L. Solvent control flasks were treated with [phenyl-U-^{14}C]-benzoic acid at a nominal concentration of 10 µg/L and 50 µL of acetonitrile to determine any effect of the solvent on mineralisation. The redox potential, pH and dissolved oxygen content of the water in control flasks, treated with non-radiolabelled bixlozone (two per dose rate), were measured throughout the incubation period.

Application

A stock solution was prepared by reconstituting 1.7 mg of the test item in 4 mL of acetonitrile and 46 mL of water (final volume 50 mL). A 100µL aliquot of this stock solution was transferred to a 10 mL volumetric flask and diluted to volume with water.

For the 10 µg/L application solution, two 0.795 mL aliquots (total 1.59 mL) of the stock solution were placed in a 50 mL volumetric flask and diluted to volume with water. For the 100 µg/L application solution, three 5 mL aliquots and a 0.89 mL aliquot (total 15.89 mL) of the stock solution were placed in a 50 mL volumetric flask and diluted to volume with water.

Flasks containing the test water were each treated with 1 mL of the appropriate [14C]-bixlozone application solution using a calibrated positive displacement pipette, adding the solution dropwise onto the water surface. Sterile replicates were treated under a running laminar flow hood with the same volumes and application solution as above. Flasks for the determination of transformation products were treated with 3 x 1.059 mL aliquots (total 3.177 mL) of the stock solution.

For the positive controls [^{14}C]-Benzoic Acid application solution, the application solution was prepared as a dilution of a standard solution in water. An aliquot (5 x 1 mL aliquots) of the standard solution of [14C]-Benzoic acid was placed into a 20 mL volumetric flask and diluted to volume with water.

The positive controls were treated with 1.1 mL per flask of the [14C]-Benzoic Acid application solution, which contained no organic solvent, using a positive displacement pipette. Solvent controls were also treated with 1.1 mL of the [^{14}C]-Benzoic acid application solution using a positive displacement pipette, but had an additional 50 µL of acetonitrile added to each flask, using a positive displacement pipette, to mimic the volume of acetonitrile added to each flask treated with the [^{14}C]-bixlozone 100 µg/L application solution (representing twice the maximum volume of solvent in test flasks). The Applicant states that the solvent controls still showed rapid mineralisation of [^{14}C]-benzoic acid (see Results section below), therefore it was assumed that increased volume of solvent had no effect on the test system's mineralisation capacity. The CA accepts the applicant's justification.

For the unlabelled bixlozone application solutions, these were prepared as dilutions of a standard solution which contained bixlozone at a concentration of 1073 µg/mL. The unlabelled application solution for the 10 µg/L test concentration was prepared by diluting 9 µL of the aforementioned standard to 10 mL with water. The unlabelled application solution for the 100 µg/L test concentration was prepared by diluting 93 µL of the 1073 µg/mL standard to 10 mL with water.

The flasks for the monitoring of test conditions were treated with 1 mL per flask of the corresponding unlabelled application solution using a calibrated positive displacement pipette.

Sampling

For each of the test concentrations, duplicate flasks and their associated traps were removed at 0, 1, 3, 7, 14, 21, 28, 35, 49 and 62 days after treatment. Sterile control flasks were removed for analysis after

1 and 62 days incubation. Positive control flasks were removed for analysis after 7 and 14 days and solvent controls were analysed on day 7.

Description of analytical procedure

At the time of sampling, 10 mL acetonitrile and 2 mL formic acid were added to the test systems , and then purged to KOH traps to determine the dissolved $^{14}CO_2$. Quantitative measurement of radioactivity was carried out by LSC (Water samples LOD/LOQ: 0.20/0.68 % AR for 10 µg/L systems, 0.02/0.07 % AR for 100 µg/L systems). Water samples were analysed by high performance liquid chromatography (HPLC) coupled to a radioactivity detector (LOD/LOQ: 0.97/3.25% AR). Selected samples were also analysed by liquid chromatography coupled to mass spectrometry (LC-MS) to confirm the identity of bixlozone. The identity of the radioactivity in the KOH traps was characterised by addition of barium chloride (to confirm $^{14}CO_2$), any remaining radioactivity in the supernatant was quantified by LSC. Aliquots of the sterile control flasks were applied to nutrient agar plants, which were then incubated at 20ºC to verify the sterility of the samples.

RESULTS

For both test concentrations, the overall material balances showed mean recovery values of 97.4 % and 97.7 % AR for the 10 and 100 µg/L test concentrations respectively. Individual recoveries were all within the range 90-100 % AR.

Mean mass balances in the sterile control samples were 99.9 and 99.1 % AR for the 10 and 100 µL test concentrations respectively. The mean mass balances in the positive control and solvent control samples were 75.6 % and 75.7 % AR, respectively. The Applicant suggests that these lower recoveries are due to the presence of a significant proportion of the applied radioactivity being incorporated within the biomass where it is poorly accessible to the scintillation cocktail, leading to underestimation of the total activity present in the water phase. As these controls are intended to demonstrate sufficient biological activity within the test system, and that mass balances within the OECD recommended range were recorded for the bixlozone test systems, the CA accepts the control recoveries <90 % AR are not considered to impact the overall outcome of the study.

The pH of the water in the reference flasks averaged 8.38 (range 7.85 to 8.68). The oxygen content averaged 6.29 mg/L (range 4.99 to 8.01 mg/L), while the redox potentials averaged +391.2 mV (range +205.5 to +474.5 mV), demonstrating the test systems remained aerobic throughout the study.

The achieved concentration of [^{14}C]-bixlozone in test vessels were 9.8 and 97.8 µg/L. Mean recovery of radioactivity from the water on day zero was 98.7 % and 99.8 % AR for the 10 and 100 µg/L test concentrations. Test item mineralisation was low for both test concentrations, reaching maximum mean values of 2.0 % and 1.0 % AR (total volatiles) for the 10 and 100 µg/L test concentrations respectively. Bixlozone represented 94.1 % AR and 91.7 % AR at the end of the study in the 10 µg/L and 100 µg/L test systems respectively. Unknown degradation products were observed through the course of the study. One replicate sample (10 µg/L test system, day 7), recorded a degradation product concentration of 7.1 % AR (mean of both replicates was 4.0 % AR). However, as all other samples were <5 % AR, the degradation product does not meet the major metabolite classification criteria.

The suitability of the test system was demonstrated in the positive controls treated with [^{14}C]-benzoic acid at 10 µg/L, with a mean of 57.9 % AR observed in volatile traps by day 14. A similar level of mineralisation was observed in the solvent control systems with a mean of 63.8 % AR observed in volatile traps on day 7, demonstrating that the solvent did not impact on the mineralisation capacity of the test systems. Sterility checks confirmed the sterility of the sterile controls. In the sterile controls, bixlozone accounted for 95.4 to 97.0 % AR after 62 days incubation.

Table CA.B.8.2.2.2-4: Distribution of radioactivity in natural water systems treated with [^{14}C]-bixlozone at 10 µg/L (as % applied radioactivity)

% Applied Radioactivity								
Time (day)	**Water phase**			**Volatile Traps**				**Mass balance**
	Total	**bixlozone**	**Unknown***	**Total**	**Ethylene glycol**	**KOH 1**	**KOH 2**	
0	99.0	96.1	2.9	0.6	0.0	0.3	0.3	99.5
0	98.5	98.5	-	0.5	0.1	0.3	0.2	99.0
Mean	**98.7**	**97.3**	**1.4**	**0.6**	**0.0**	**0.3**	**0.3**	**99.3**
1	98.7	97.5	1.2	0.6	0.0	0.3	0.3	99.3
1	98.9	97.3	1.5	0.7	0.0	0.3	0.3	99.5
Mean	**98.8**	**97.4**	**1.4**	**0.6**	**0.0**	**0.3**	**0.3**	**99.4**
3	96.8	96.8	-	0.5	0.0	0.3	0.2	97.3
3	97.6	95.4	2.3	0.5	0.0	0.3	0.3	98.2
Mean	**97.2**	**96.1**	**1.1**	**0.5**	**0.0**	**0.3**	**0.3**	**97.7**
7	98.0	97.1	0.9	0.7	0.0	0.4	0.3	98.7
7	98.1	91.0	7.1	0.8	0.0	0.5	0.3	98.9
Mean	**98.0**	**94.1**	**4.0**	**0.8**	**0.0**	**0.5**	**0.3**	**98.8**
14	96.5	96.5	-	0.8	0.0	0.5	0.3	97.2
14	97.4	96.2	1.2	0.8	0.0	0.5	0.3	98.3
Mean	**97.0**	**96.3**	**0.6**	**0.8**	**0.0**	**0.5**	**0.3**	**97.7**
21	97.3	95.6	1.8	1.2	0.1	0.6	0.4	98.5
21	95.8	95.8	-	1.0	0.0	0.7	0.4	96.9
Mean	**96.6**	**95.7**	**0.9**	**1.1**	**0.1**	**0.7**	**0.4**	**97.7**
28	96.3	96.3	-	1.1	0.0	0.7	0.4	97.4
28	93.5	92.7	0.8	1.0	0.0	0.4	0.6	94.6
Mean	**94.9**	**94.5**	**0.4**	**1.0**	**0.0**	**0.5**	**0.5**	**96.0**
35	94.5	91.8	2.7	1.8	0.2	1.1	0.5	96.3
35	94.4	92.6	1.8	1.5	0.0	1.0	0.4	95.9
Mean	**94.4**	**92.2**	**2.2**	**1.7**	**0.1**	**1.1**	**0.5**	**96.1**
49	93.8	93.8	-	1.7	0.0	1.4	0.3	95.5
49	91.8	91.1	0.7	2.2	0.1	1.6	0.5	94.0
Mean	**92.8**	**92.5**	**0.3**	**2.0**	**0.1**	**1.5**	**0.4**	**94.8**
62	96.8	96.0	0.8	1.2	0.0	0.8	0.3	98.0
62	92.1	92.1	-	2.2	0.1	1.5	0.6	94.2
Mean	**94.4**	**94.1**	**0.4**	**1.7**	**0.0**	**1.2**	**0.5**	**96.1**

*maximum individual value of 7.1 % AR

Table CA.B.8.2.2.2-5: Distribution of radioactivity in natural water systems treated with [^{14}C]-bixlozone at 100 µg/L (as % applied radioactivity)

Time (day)	Water phase			Volatile Traps				Mass balance
	Total	bixlozone	Unknown*	Total	Ethylene glycol	KOH 1	KOH 2	
0	99.5	97.2	2.3	0.1	0.0	0.0	0.0	99.6
0	100.1	96.8	3.2	0.1	0.0	0.0	0.0	100.1
Mean	**99.8**	**97.0**	**2.8**	**0.1**	**0.0**	**0.0**	**0.0**	**99.8**
1	99.1	95.4	3.7	0.1	0.0	0.0	0.0	99.2
1	100.0	97.9	2.0	0.1	0.0	0.1	0.0	100.1
Mean	**99.6**	**96.7**	**2.9**	**0.1**	**0.0**	**0.1**	**0.0**	**99.6**
3	98.5	95.7	2.8	0.1	0.0	0.1	0.0	98.6
3	98.7	96.6	2.1	0.1	0.0	0.1	0.0	98.9
Mean	**98.6**	**96.2**	**2.5**	**0.1**	**0.0**	**0.1**	**0.0**	**98.8**
7	99.2	96.2	3.0	0.1	0.0	0.1	0.0	99.4
7	98.3	96.7	1.6	0.2	0.0	0.2	0.0	98.4
Mean	**98.8**	**96.5**	**2.3**	**0.2**	**0.0**	**0.1**	**0.0**	**98.9**
14	97.7	95.4	2.4	0.4	0.0	0.4	0.0	98.2
14	98.2	96.1	2.0	0.2	0.0	0.2	0.0	98.4
Mean	**97.9**	**95.8**	**2.2**	**0.3**	**0.0**	**0.3**	**0.0**	**98.3**
21	98.7	94.6	4.2	0.2	0.0	0.2	0.0	99.0
21	97.4	95.3	2.1	0.3	0.0	0.2	0.1	99.7
Mean	**98.1**	**94.9**	**3.1**	**0.3**	**0.0**	**0.2**	**0.1**	**98.3**
28	96.7	94.8	1.9	0.4	0.0	0.3	0.2	97.1
28	96.1	93.9	2.3	0.5	0.0	0.3	0.2	96.6
Mean	**96.4**	**94.3**	**2.1**	**0.5**	**0.0**	**0.2**	**0.2**	**96.9**
35	94.6	91.9	2.7	0.7	0.0	0.5	0.2	95.2
35	93.9	91.9	2.0	0.7	0.0	0.4	0.2	94.5
Mean	**94.2**	**91.9**	**2.3**	**0.7**	**0.0**	**0.5**	**0.2**	**94.9**
49	96.7	95.4	1.3	0.6	0.0	0.6	0.0	97.3
49	95.4	93.9	1.5	0.6	0.0	0.5	0.1	96.0
Mean	**96.1**	**94.7**	**1.4**	**0.6**	**0.0**	**0.5**	**0.1**	**96.6**
62	93.7	91.6	2.1	1.1	0.0	0.9	0.2	94.9
62	94.9	91.8	3.2	0.8	0.0	0.4	0.4	95.7
Mean	**94.3**	**91.7**	**2.6**	**1.0**	**0.0**	**0.7**	**0.3**	**95.3**

*maximum individual value of 4.2 % AR

Table CA.B.8.2.2.2-6: Distribution of radioactivity in control systems (as % applied radioactivity)

Time (day)	Water phase			Volatile Traps				Mass balance
	Total	bixlozone	Unknown	Total	Ethylene glycol	KOH 1	KOH 2	
Sterile control: 10 µg/L								
1	100.0	99.4	0.6	0.7	0.0	0.3	0.4	100.7
1	99.3	96.3	3.0	0.6	0.0	0.3	0.3	99.9
Mean	**99.6**	**97.8**	**1.8**	**0.7**	**0.0**	**0.3**	**0.3**	**100.3**
62	98.6	95.2	3.5	0.9	0.2	0.4	0.3	99.5
62	98.7	95.5	3.2	0.7	0.0	0.3	0.3	99.4
Mean	**98.7**	**95.4**	**3.3**	**0.8**	**0.1**	**0.3**	**0.3**	**99.4**
Sterile control: 100 µg/L								
1	99.1	97.1	2.0	0.1	0.0	0.0	0.0	99.2
1	99.5	96.5	3.1	0.1	0.0	0.0	0.0	99.6
Mean	**99.3**	**96.8**	**2.6**	**0.1**	**0.0**	**0.0**	**0.0**	**99.4**
62	98.8	97.8	1.1	0.1	0.0	0.1	0.0	98.9
62	98.7	96.1	2.6	0.1	0.0	0.0	0.0	98.8
Mean	**98.7**	**97.0**	**1.8**	**0.1**	**0.0**	**0.0**	**0.0**	**98.8**
Positive control: 10 µg/L [^{14}C]-benzoic acid								
7	15.8	n.a	n.a	62.2	0.0	61.5	0.7	78.0
7	24.1	n.a	n.a	64.1	0.0	61.1	3.0	88.1
Mean	**20.0**	**n.a**	**n.a**	**63.1**	**0.0**	**61.3**	**1.8**	**83.1**
14	9.4	n.a	n.a	57.2	0.0	54.3	2.8	66.6
14	11.4	n.a	n.a	58.5	0.0	51.3	7.2	69.9
Mean	**10.4**	**n.a**	**n.a**	**57.9**	**0.0**	**52.8**	**5.0**	**68.2**
Solvent control: 10 µg/L [^{14}C]-benzoic acid								
7	10.6	n.a	n.a	65.2	0.1	62.7	2.4	75.8
7	13.2	n.a	n.a	62.4	0.0	59.7	2.6	75.6
Mean	**11.9**	**n.a**	**n.a**	**63.8**	**0.1**	**61.2**	**2.5**	**75.7**

The applicant has calculated DT_{50} values of 1040 days and 818 days for the 10 µg/L and 100 µg/L test systems respectively (and are therefore highly uncertain given they are extrapolated far beyond the study end). However, because <10 % degradation was observed in the study period for both test systems, the CA does not consider it necessary to conduct a kinetic evaluation of the results. Therefore, the applicant's DT_{50} values have not been validated and are not considered further in the risk assessment.

CONCLUSION

After 62 days, >90 % of the test substance was recovered in both the 10 µg/L and 100 µg/L test systems. Only one replicate at one timepoint recorded an unknown degradation product at a concentration >5 % AR and so no major metabolites were detected in the study. No kinetic analysis was performed due to little degradation over the study period.

CA.B.8.2.2.3. *Aerobic water/sediment*

Report:	KCA 7.2.2.3/01, Cooper, J.; Challis, P., (2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	[^{14}C]- F9600: Aerobic Aquatic Metabolism in Two Water/Sediment Systems at 20 ± 2 °C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/15/005, FMC Tracking no. 2015EFT-ISX2195
Guidelines:	OECD Guideline 308; U.S. EPA Guideline OPPTS 835.4300
GLP:	Yes (laboratory certified by UK National Authority)

CA Comments:	The CA notes the following issues were identified with the study, which are discussed further in the main text. Despite these, the CA considered the study sufficient to determine suitable regulatory endpoints. • Limited sample site history provided. No reference to agricultural, industrial or domestic inputs to catchment area. • A number of sediment and water samples were frozen at -15ºC. Guidance recommends 4ºC ± 2. • Acclimatisation of water and sediment lasted 18 days. Longer than the 2 week period recommended in the guidance. • Total recovery of %AR dips below 90% in 3 samples (88.87%, 89.22% and 89.97%). **This study is relied upon**

INTRODUCTION

The degradation of [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone was investigated in two water-sediment systems at 20 ± 2°C for a period of 100 days.

MATERIALS AND METHODS

Test and Reference Items

Table CA.B.8.2.2.3-1: Reference Item: bixlozone

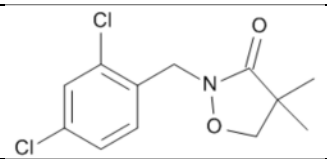
Common name:	Bixlozone
Structure:	
Name (IUPAC):	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-1,2-oxazolidin-3-one
Chemical name:	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-3-isoxazolidinone
CAS registry number:	81777-95-9
Molecular Formula:	C12H13Cl2NO2
Molecular weight:	274.14 g/mol
Solubility (in purified water):	42.0±0.3 mg/L
K_{ow} logP:	3.39 or 3.36

Table CA.B.8.2.2.3-2: Test Item: Radiolabelled [phenyl-U-^{14}C]-bixlozone

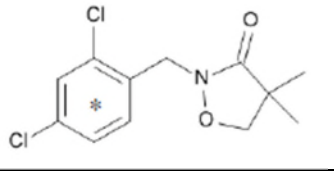
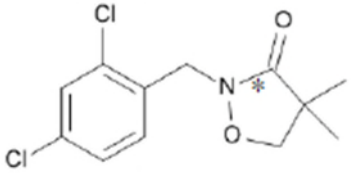
Name:	Radiolabelled [phenyl-U-^{14}C]-bixlozone
Structure and position of ^{14}C radiolabel:	
Physical Form:	Solution in Toluene
Specific Activity:	8.04 MBq/mg or 60 mCi/mmol (9.25 MBq/mL)
Radiochemical Purity:	>99.9%
Storage Conditions:	-20ºC in the dark

Table CA.B.8.2.2.3-3: Radiolabelled [carbonyl-^{14}C]-bixlozone

Name:	Radiolabelled [carbonyl-^{14}C]-bixlozone
Structure and position of ^{14}C radiolabel:	
Physical Form:	Solution in Toluene
Specific Activity:	7.91 MBq/mg 59 mCi/mmol (9.25 MBq/mL
Radiochemical Purity:	>99.9%
Storage Conditions:	-20ºC in the dark

Test System

Two natural water-sediment systems from sites in the UK were used for the study. The sediments and associated waters were collected from Calwich Abbey Lake, Calwich, Staffordshire on 17th May 2016 and from Swiss Lake, Chatsworth, Derbyshire on 23rd May 2016. At the time of collection, the temperature, oxygen content, redox potential, pH and conductivity of the surface water and the redox potential of the sediment were measured. The sediment was scooped from the top 5 cm of sediment onto the bank and allowed to drain. The sediments were passed through a 2 mm sieve during collection and were fully characterised with respect to particle size distribution, organic carbon, pH, cation exchange capacity, total nitrogen and total phosphorous.

The CA notes that the applicant does not provide any information about agricultural, industrial or domestic inputs to the catchment area.

The water samples were passed through a 212 µm sieve during collection and were characterised with respect to pH, hardness, dissolved and total organic carbon, total nitrogen and total phosphorus. The samples were transported at ambient temperature to the test facility and stored at *ca*.5ºC in the dark until use. The physico-chemical data is summarised in Table CA.B.**8.2.2.3-4**.

Table CA.B.8.2.2.3-4: Physico-chemical properties

Soil Reference (Batch ID)	Calwich Abbey	Swiss Lake
Grid Reference	OSGB-SK127431	SK 27177 69993
Textural Classification (USDA)	Loam	Sand
Sand % (50-2000 µm)	46	96
Silt % (2-50 µm)	46	4
Clay % (<2 µm)	8	0
Sediment pH in water	7.2	6.3
Sediment pH in 1 N KCl	7	6.4
Sediment pH in 0.01 M $CaCl_2$	7.1	6.1
Organic Carbon %	4.4	0.7
Organic Matter %	7.6	1.2
Cation Exchange Capacity (meq/100g)	9.8	3.1
% Sediment Moisture on receipt	152.04	37.86
Microbial Biomass (µg C/g soil) Initial Final	 887 ± 15 663 ± 4	 56.1 ± 6 64.3 ± 8

EXPERIMENTAL SETUP

The sediment and associated water were added to specially adapted individual glass incubation flasks with a screw top and straight sides of approximately 600 mL capacity (6.0 cm diameter). Each had an associated air-tight flask head with side-arm fittings to permit the passage of air through the flask. Each flask was connected to a series of trap vessels.

For both water-sediment systems, and both radiolabels, sixteen flasks were prepared for treatment with bixlozone, allowing duplicate samples to be taken at each of six specified sampling time-points, whilst leaving four flasks which could be used as spares. A further four flasks remained untreated for each water-sediment system, for both radiolabels.

Approximately 50 g oven-dried equivalent (ODE) of Calwich Abbey sediment or 84 g ODE of Swiss Lake sediment (each sieved to 2 mm) to give a layer of *ca* 3 cm depth, with *ca* 340-360 mL of the associated water, was dispensed into the glass flasks. The samples were allowed to acclimatise under study conditions for a maximum of 18 days prior to application of the test item. The CA notes that this is longer than the recommended maximum of two weeks stated in the guidance, however the CA does not consider this to have had a significant impact on the outcomes of the study. Ratios of approximately 1:4 (based upon soil: water depth) were obtained for all samples of both systems. A depth of *ca* 3 cm was achieved for the sediment layer. Water was added to give a depth of *ca* 12 cm above the sediment.

All flasks were attached to an incubation system through which moistened air was bubbled, at a rate that allowed aeration of the water without disturbance of the sediment-water interface. The passage of air was controlled by the use of small, glass, cylindrical flow restrictors containing a narrow bore. These ensured a uniform flow rate into each flask and allowed individual flasks to be disconnected without disrupting the flow to those remaining. Each flask was connected to a series of four traps, the first being a polyurethane foam bung placed in the head of the test vessel, with the remaining traps being liquids. The liquid traps consisted of one ethylene glycol trap, followed by two traps containing 2M potassium hydroxide.

The water-sediment systems were incubated at 20 ± 2°C in the dark until there was complete phase separation and to allow the oxygen levels, pH and redox potentials to establish. The flasks were maintained at 20 ± 2°C throughout the course of the study. The applicant highlights one of the temperature probes reported an increase in temperature (to *ca* 25.5°C) for approximately one day

around the 15th September 2016 (*ca* 94 days after treatment); exact measurements, dates and timings were not reported. However, the applicant states this is believed to be a faulty reading as another temperature probe in the room reported no significant fluctuations in temperature. The CA accepts the applicant's justification and so this is not expected to have impacted on the outcomes of the study.

Eight flasks were prepared for each sediment type to be used for determination of sediment biomass (four each for initial and final biomass samples). The four flasks for the final biomass determination were left untreated and remained in the system throughout the study. These were also used to measure the pH, oxygen and redox potential throughout the duration of the study.

Preparation of treatment solutions

For both [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone, the treatment solutions were prepared by transferring 1.2 mL of the supplied toluene solution to a 10 mL volumetric flask. The toluene solution was evaporated under air and then reconstituted with acetonitrile. A 100 µL aliquot of the treatment solution was transferred into a 20 mL volumetric flask and diluted to volume with acetonitrile. Triplicate 100 µL aliquots of this dilution were counted by LSC to determine the exact concentration of the solution. In addition, the purity of the test items were determined by HPLC analysis of this dilution.

Application Procedures

The water-sediment systems were each treated with the appropriate [^{14}C]-labelled treatment solution using a positive displacement pipette, adding the solution dropwise onto the water surface. All sample flasks were treated on 13th June 2016. Treatment checks were made before, during and after treatment by dispensing an equal volume of treatment solution directly into a 20 mL volumetric flask. The treatment checks were diluted to volume with acetonitrile and the radioactivity determined by LSC to obtain the treatment rate achieved.

The phenyl-labelled water-sediment flasks were each treated with 98 µL of the [^{14}C]-treatment solution, containing an average of 14.0 µg of bixlozone (6,745,733 dpm, 102.8% of target). The carbonyl labelled water-sediment flasks were each treated with 93 µL of the [^{14}C]- treatment solution, containing an average of 13.7 µg of bixlozone (6,515,967 dpm, 101.0% of target).

All sample and biomass flasks (other than the zero-time samples) were incubated in the dark at 20 ± 2°C. Aerobic conditions in the water phase were maintained by the constant passage of moist air through the sample flasks and out through the trap solutions. Duplicate flasks and their associated traps were removed at each time point.

Calculation of Target Application Rate
Water sediment samples were treated with radiolabelled bixlozone at a rate equivalent to 400 g ha-1 equivalent to an initial water concentration of 0.040 mg L^{-1} (based upon the following calculation).

Cylinder internal diameter	=	6 cm
Water Column depth not including sediment	=	12 cm
Surface area (π × (6/2)2)	=	28.3 cm^3
Volume of water column (π × (6/2)2 × 12)	=	339.3 cm^3
Target application rate	=	400.0 g ha^{-1}
equivalent to		4.00 µg cm^{-2}
Total µg per flask	=	113.1 µg
Reference water column depth	=	100.0 cm
Adjusted total µg per flask (113.1 × (12/100.0)	=	13.6 µg
Concentration in water (13.6/339.3)	=	0.040 µg cm^{-3}
	=	0.040 mg L^{-1}

Measurement and Redox Potential, Dissolved Oxygen and pH of Sediment and Water

The redox potential of the sediment and water was measured by using a Mettler Toledo InLab combi probe. The standard solution for the redox potential measurements was purchased commercially. This consisted of a solution containing ammonium iron (II) sulphate and ammonium iron (III) sulphate acidified with concentrated sulphuric acid, giving a redox potential of +470 mV using a platinum half-cell and an Ag/AgCl 4M KCl reference electrode. For the conversion to the standard redox potential +200 mV should be added to the redox values obtained.

To measure the redox potential in water, the electrode was suspended in the water through the neck of the flask, ensuring that it was not in contact with the sediment surface. For measurement of the sediment redox potential, the electrode was inserted directly into the sediment through the neck of the flask.

The pH electrode was calibrated using commercially prepared buffer solutions. The pH of the water was measured by inserting the electrode through the neck of the flask, ensuring that it was not in contact with the sediment surface, and recording the value obtained.

Prior to use, the oxygen meter was calibrated to zero in a commercially prepared zero oxygen solution and to 100% in air. The oxygen electrode was then inserted into the water phase through the neck of the flask and the reading taken.

Each flask was connected to a series of four traps. The first was a polyurethane foam bung which was placed in the heads of the test vessels, with the remaining traps being liquids. The liquid trapping solutions consisted of three graduated 50 mL tubes containing one ethylene glycol trap, followed by two traps containing 2M potassium hydroxide. These were connected in series to the incubation flasks. Moistened air was bubbled constantly, at a consistent rate, through the flasks and trap vessels during the course of the study, with interruption to the flow only occurring during flask maintenance.

Duplicate flasks and their associated traps were removed at each sampling interval. Samples were taken at zero time, 7, 14, 30, 63 and 100 days incubation.

METHOD

Quantitative measurement of radioactivity was carried out by LSC following solubilisation of the samples in a LSC cocktail. All samples were counted for 5 minutes. Sample counts were automatically corrected for background from the scintillant and solvents present and for quenching using pre calibrated quench correction curves to convert cpm to quench-corrected dpm. Background samples were counted for 10 minutes.

The limits of detection (LOD) and quantification (LOQ) for LSC analysis are set at 3 × and 10 × the standard deviation in background measurements for a series of blank vials. For both phenyl-bixlozone and carbonyl-bixlozone, the LOD corresponded to 0.04% AR and the LOQ to 0.15% AR in water.
With the exception of the zero time samples, the liquid trap solutions were removed for analysis at each sampling time. The weight of each trap solution was recorded, and the radioactivity present was determined by LSC by taking single weighed aliquots, and solubilising them with scintillant. A carbon dioxide determination of the potassium hydroxide traps was carried out on selected samples to determine the nature of activity present.

Following extraction, the sediment samples were air-dried, weighed and ground to a fine powder. Triplicate aliquots (*ca* 0.2 g) were accurately weighed and combusted. The combustion products were absorbed in Carbosorb E and mixed with Permafluor E+ prior to quantification of the radioactivity by LSC.

Sample Analysis

For water sample analysis, the water was decanted from the sediment directly into a pre-weighed plastic bottle, taking care not to disturb the sediment. The total weight was recorded, and the radioactive content determined by taking an appropriate weighed aliquot for LSC.

Following removal of the overlying water, the sediment was transferred to a suitably sized container and extracted with *ca* 150 mL of acetonitrile. The sample was placed on a wrist action shaker and shaken for *2*0 minutes. After extraction, the sample was centrifuged at 2500 rpm for 10 minutes and the supernatant decanted into a suitably sized pre-weighed plastic bottle. The sediment was then further extracted by repeating the process with two 150 mL portions (one for select samples) of acetonitrile: water (80:20 v/v) and one 150 mL portion of acetonitrile: water: formic acid (50:50:1 v/v/v). All extracts were combined, the total quantity of radioactivity was determined by measuring the total weight and taking an appropriately sized weighed aliquot for LSC. If necessary, the sediment residue was then further extracted by soxhlet. The sediment was placed into a cellulose soxhlet thimble and extracted for 6 hours with acetonitrile: water (80:20 v/v), after cooling the extract was weighed, and a single weighed aliquot taken for LSC analysis. The sediment residue was left to air dry prior to quantification of the levels of radioactivity by combustion.

Selected potassium hydroxide traps containing significant levels of trapped volatile activity were analysed by a barium chloride precipitation method, to determine the nature of the activity present. Duplicate 2 mL aliquots of each sample were taken. 1M sodium carbonate (1 mL) and 1M barium chloride (2 mL) were added to each sample and this was shaken on a wrist action shaker for 10 minutes, followed by centrifugation at 2500 rpm for 10 minutes. The supernatant was decanted, and weight recorded. A single weighed aliquot was taken for LSC radioassay. The procedure was repeated up to twice more. More than 99% of the activity was precipitated by this method, confirming the activity to be carbon dioxide. Ethylene glycol traps did not require analysis.

The polyurethane foam bungs were removed from the head of the test vessel at the time of sampling. From day 14 onwards, the bung was extracted in a plastic bottle by shaking for 20 minutes on a wrist action shaker with 100 mL of acetonitrile. The extract was decanted into a plastic bottle, and the process repeated once more. The total weight of the combined extract was recorded, and a weighed aliquot taken for LSC. Holding the bung to a Geiger counter confirmed all the radioactivity had been removed.

To determine the concentration, sample extracts were centrifuged at 3500 rpm for 10 minutes to remove very fine particulates. Recovery was checked by taking a weighed aliquot, and comparing the results to the original extract. The sediment extracts were concentrated by volume reduction using a Buchi Syncore evaporator. Approximately 100 g was weighed out into a Syncore tube and evaporated to *ca* 1-2 mL under the vacuum at 55°C. The evaporated residue was then reconstituted by thoroughly rinsing the Syncore tube with separate portions of deionised water and acetonitrile (with the aid of an ultrasonic bath). The washings were transferred into a pre-weighed volumetric flask (10 mL) and the total weight recorded. Suitably sized weighed aliquots were removed and counted by LSC.

For one sample (the applicant does not state which) a different methodology was used. The sediment extract was concentrated by volume reduction using a Turbovap. Approximately 150 g was weighed out into a Turbovap tube, and evaporated to *ca* 1-2 mL using air, with the tube being warmed to 40°C. The evaporated residue was reconstituted by thoroughly rinsing the Turbovap tube with separate portions of acetonitrile and de-ionised water (with the aid of an ultrasonic bath). The washings were transferred into a volumetric flask (10 mL). The concentrate was transferred to a pre-weighed glass vial and the total weight recorded. Suitably sized weighed aliquots were removed and counted by LSC. The applicant states that the Syncore method described in the previous paragraph was used instead of this method because a decrease in radioactivity recovery was noted using this methodology. As the mass balances recorded were all within the OECD recommended range, this approach is accepted by the CA.

RESULTS

The mass balances are presented in Table CA.B.**8.2.2.3-5** and Table CA.B.8.2.2.3-6. The applicant reports that only samples with >90% total AR were deemed acceptable, however the CA notes that on three occasions recovery dipped below 90%. Because the recoveries were only narrowly <90% (88.87%, 89.22% and 89.97%), and the corresponding replicate sample recorded recoveries were within the OECD acceptable range (resulting in mean sample point values >90%), the CA does not consider this deviation from the guidelines to significantly impact the outcomes of the study (on this occasion).

The HPLC results are presented in Table CA.B.**8.2.2.3-7** to Table CA.B.**8.2.2.3-14**. Bixlozone (mean of both labels) declined to 5.0% AR and 20.6% AR in the total system, in the Calwich Abbey and Swiss Lake systems, respectively, after 100 days. Bixlozone was observed in sediment at mean maxima of 20.99% AR (phenyl label, mean day 30) and 23.07% AR (carbonyl label, mean day 30) in the Calwich Abbey and Swiss Lake systems, respectively. The CA does not consider either compartment to be the major degradation compartment due to similar levels being recorded in both compartments, in both test systems, at each sampling point.

The CA notes four major metabolites were observed:

- 2,4-dichlorobenzoic acid was observed in both systems with the phenyl-label only at a maximum overall mean value of 40.87% AR in the total system (Calwich Abbey, day 100). It reached a maximum overall mean value of 30.36% AR in the water phase (Calwich Abbey, day 100) and 10.51% AR in the sediment phase (Calwich Abbey, day 100).
- Bixlozone-3-OH-propanamide was observed in both water-sediment systems, with both radiolabels, at a maximum overall mean value of 10.31% AR in the total system (Calwich Abbey, day 7). It reached a maximum overall mean value of 3.59% AR in the water phase (Calwich Abbey, day 7) and 9.92% AR in the sediment phase (Calwich Abbey, day 63).
- Bixlozone-dimethyl malonamide was observed in both systems, with both radiolabels, at a maximum overall mean value of 16.72% AR in the total system (Calwich Abbey, day 30). It reached a maximum overall mean value of 12.36% AR in the water phase (Calwich Abbey, day 30) and 5.70% AR in the sediment phase (Swiss Lake, day 63).
- 4-carboxy-bixlozone was observed in the Swiss Lake system only, with both radiolabels, at a maximum overall mean value of 24.45% AR (day 100) in the total system. It reached a maximum overall mean value of 17.60% AR in the water phase (Swiss Lake, day 100) and 6.85% AR in the sediment phase (Swiss Lake, day 100).

The CA notes the metabolite 4-hydroxymethyl was detected in one total-system sample (Swiss Lake, carbonyl ring, day 63) at levels >5% AR (max 5.94% AR, mean 5.08% AR). As it was only detected at one time point at levels <10% AR and was not increasing at study termination, it does not meet the major metabolite criteria. Other minor (predominately unknown) metabolites were also detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study and so further consideration is not required.

The applicant's water/sediment metabolic pathway is provided in Figure CA.B.**8.2.2.3-1**.

Table CA.B.8.2.2.3-5.: Distribution of radioactivity in the Calwich Abbey system (as % of applied radioactivity)

Day	% Applied Radioactivity						
	Water	Sediment	Volatiles**			NER	Mass balance
	Total	Total Extracted*	Ethylene Glycol	$^{14}CO_2$**	Polyurethane bung		
[Phenyl-U-^{14}C]-bixlozone							
0	94.72	1.78	n.a	n.a	n.a	0.03	96.53
0	94.30	2.78	n.a	n.a	n.a	0.04	97.12
Mean	**94.51**	**2.28**	**n.a**	**n.a**	**n.a**	**0.04**	**96.83**
7	66.34	24.88	<0.01	0.07	n.a	5.65	96.94
7	65.63	28.10	<0.01	0.05	n.a	4.51	98.29
Mean	**65.99**	**26.49**	**<0.01**	**0.06**	**n.a**	**5.08**	**97.62**
14	68.29	21.39	<0.01	0.25	0.42	5.82	96.16
14	59.28	28.73	<0.01	0.28	0.52	7.77	96.58
Mean	**63.79**	**25.06**	**<0.01**	**0.27**	**0.47**	**6.79**	**96.37**
30	41.12	42.85	<0.01	0.91	0.60	8.33	93.82
30	51.27	33.73	<0.01	0.64	1.15	8.51	95.30
Mean	**46.19**	**38.29**	**<0.01**	**0.78**	**0.88**	**8.42**	**94.56**
63	31.05	48.76	<0.01	2.82	0.65	10.25	93.52
63	31.52	48.75	<0.01	2.86	0.85	8.45	92.43
Mean	**31.28**	**48.75**	**<0.01**	**2.84**	**0.75**	**9.35**	**92.98**
100	36.85	35.47	<0.01	6.12	0.90	9.54	88.87
100	31.74	35.27	<0.01	7.22	0.58	18.88	93.69
Mean	**34.29**	**35.37**	**<0.01**	**6.67**	**0.74**	**14.21**	**91.28**
						Overall Mean	94.94
						Standard deviation	2.61
[Carbonyl-^{14}C]-bixlozone							
0	96.96	1.78	n.a	n.a	n.a	0.06	98.80
0	97.76	1.82	n.a	n.a	n.a	0.06	99.63
Mean	**97.36**	**1.80**	**n.a**	**n.a**	**n.a**	**0.06**	**99.22**
7	79.75	13.47	<0.01	<0.01	n.a	4.28	97.51
7	83.60	11.39	0.01	0.10	n.a	2.99	98.09
Mean	**81.68**	**12.43**	**0.01**	**0.05**	**n.a**	**3.64**	**97.80**
14	49.68	34.01	<0.01	4.40	0.96	8.50	97.55
14	54.33	29.91	0.01	3.94	0.46	6.37	95.002
Mean	**52.01**	**31.96**	**<0.01**	**4.17**	**0.71**	**7.43**	**96.28**
30	40.33	26.81	<0.01	12.75	1.12	12.14	93.15
30	25.08	42.00	<0.01	17.34	0.58	7.97	92.97
Mean	**32.71**	**34.41**	**<0.01**	**15.05**	**0.85**	**10.06**	**93.06**
63	7.80	34.39	<0.01	38.32	0.74	11.62	92.87
63	16.98	31.47	<0.01	36.41	0.53	7.02	92.41
Mean	**12.39**	**32.93**	**<0.01**	**37.36**	**0.64**	**9.32**	**92.64**
100	3.73	24.57	<0.01	50.39	0.79	12.86	92.35
100	4.93	19.98	<0.01	52.69	0.89	10.73	89.22
Mean	**4.33**	**22.28**	**<0.01**	**51.55**	**0.84**	**11.80**	**90.78**
						Overall Mean	94.96
						Standard deviation	3.27

*Total extracted = sum of sediment extracts 1-7: 1 - acetonitrile, 2+3 - acetonitrile: water (80:20 v/v), 4 - acetonitrile: water: formic acid (50:50:1 v/v/v), only 1-3 were performed for 0, 7 and 14 days, 5 = soxhlet with acetonitrile, 6 = THF, 7 = cyclohexane

** $^{14}CO_2$ = KOH traps 1 and 2 + trap changes

NER = Non-extracted residues

n.a not analysed

Table CA.B.8.2.2.3-6: Distribution of radioactivity in the Swiss Lake system (as % of applied radioactivity)

Day	% Applied Radioactivity						
	Water	Sediment	Volatiles			NER	Mass balance
	Total	Total Extracted*	Ethylene Glycol	$^{14}CO_2$**	Polyurethane bung		
[Phenyl-U-^{14}C]-bixlozone							
0	95.95	1.99	n.a	n.a	n.a	0.04	97.98
0	96.52	1.58	n.a	n.a	n.a	0.04	98.14
Mean	**96.23**	**1.79**	**n.a**	**n.a**	**n.a**	**0.04**	**98.06**
7	74.28	21.12	<0.01	0.08	n.a	1.63	97.12
7	74.25	18.99	<0.01	<0.01	n.a	2.93	96.17
Mean	**74.26**	**20.06**	**<0.01**	**0.04**	**n.a**	**2.28**	**96.64**
14	61.34	27.92	<0.01	0.34	0.28	6.53	96.41
14	71.79	17.67	<0.01	0.24	0.32	7.06	97.08
Mean	**66.56**	**22.79**	**<0.01**	**0.29**	**0.30**	**6.80**	**96.74**
30	56.24	31.35	<0.01	1.04	1.45	6.94	97.02
30	61.86	21.66	0.01	1.93	0.59	10.63	96.67
Mean	**59.05**	**26.50**	**<0.01**	**1.49**	**1.02**	**8.78**	**96.85**
63	47.95	39.85	<0.01	1.84	0.94	6.72	97.28
63	50.42	33.38	<0.01	3.32	1.01	8.76	96.91
Mean	**49.18**	**36.61**	**<0.01**	**2.58**	**0.97**	**7.74**	**97.09**
100	31.74	40.31	<0.01	6.60	2.69	8.63	89.97
100	33.01	41.23	<0.01	10.86	1.49	7.86	94.45
Mean	**32.37**	**40.77**	**<0.01**	**8.73**	**2.09**	**8.24**	**92.21**
						Overall Mean	**96.27**
						Standard deviation	**2.20**
[Carbonyl-^{14}C]-bixlozone							
0	96.84	3.48	n.a	n.a	n.a	0.20	100.53
0	97.38	1.86	n.a	n.a	n.a	0.01	99.25
Mean	**97.11**	**2.67**	**n.a**	**n.a**	**n.a**	**0.11**	**99.89**
7	66.34	29.09	<0.01	0.40	n.a	3.29	99.12
7	72.40	22.91	<0.01	1.05	n.a	3.69	100.05
Mean	**69.37**	**26.00**	**<0.01**	**0.73**	**n.a**	**3.49**	**99.58**
14	53.93	32.58	0.02	4.52	0.22	4.96	96.22
14	56.82	26.69	0.01	6.25	0.21	7.50	97.48
Mean	**55.37**	**29.64**	**0.01**	**5.39**	**0.21**	**6.23**	**96.85**
30	38.64	39.06	<0.01	10.63	1.17	7.02	96.53
30	33.67	24.40	<0.01	24.20	1.11	12.25	95.63
Mean	**36.15**	**31.73**	**<0.01**	**17.42**	**1.14**	**9.64**	**96.08**
63	33.37	33.90	<0.01	18.91	1.13	7.76	95.07
63	29.87	44.29	<0.01	17.36	0.90	6.76	99.18
Mean	**31.62**	**39.10**	**<0.01**	**18.14**	**1.01**	**7.26**	**97.12**
100	14.91	35.76	<0.01	33.53	1.06	7.76	93.01
100	18.61	35.22	<0.01	26.86	1.63	7.69	90.01
Mean	**16.76**	**35.49**	**<0.01**	**30.2**	**1.34**	**7.73**	**91.51**
						Overall Mean	96.84
						Standard deviation	3.12

*Total extracted = sum of sediment extracts 1-7: 1 - acetonitrile, 2+3 - acetonitrile: water (80:20 v/v), 4 - acetonitrile: water: formic acid (50:50:1 v/v/v), only 1-3 were performed for 0, 7 and 14 days, 5 = soxhlet with acetonitrile, 6 = THF, 7 = cyclohexane

** $^{14}CO_2$ = KOH traps 1 and 2 + trap changes

NER = Non-extracted residues

n.a not analysed

Table CA.B.8.2.2.3-7: Characterisation of radioactivity in the water phase and sediment extracts of the [Phenyl-U-^{14}C]-bixlozone Calwich Abbey system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity							
	Water				Sediment**			
	2,4-DBA	Bixlozone -3-OH-Prop.	Bixlozone -DMM	Bixlozone	2,4-DBA	Bixlozone -3-OH-Prop.	Bixlozone -DMM	Bixlozone
0	-	-	-	94.72	n.a	n.a	n.a	n.a
0	-	-	-	94.30	n.a	n.a	n.a	n.a
Mean	**-**	**-**	**-**	**94.51**	**n.a**	**n.a**	**n.a**	**n.a**
7	-	-	4.05	58.30	-	7.45	1.74	13.70
7	-	-	<0.01	65.63	-	5.52	1.69	17.67
Mean	**-**	**-**	**2.02**	**61.97**	**-**	**6.48**	**1.72**	**15.68**
14	<0.01	2.78	3.79	61.73	-	6.87	1.94	10.46
14	3.58	2.89	14.30	38.51	-	5.81	3.22	17.03
Mean	**1.79**	**2.84**	**9.04**	**50.12**	**-**	**6.34**	**2.58**	**13.75**
30	15.41	<0.01	0.71	20.77	1.74	3.25	2.71	23.32
30	10.92	<0.01	8.84	29.72	0.75	4.30	3.72	18.66
Mean	**13.16**	**<0.01**	**4.78**	**25.24**	**1.24**	**3.77**	**3.21**	**20.99**
63	25.62	<0.01	0.79	1.27	10.13	11.23	2.04	10.64
63	24.01	<0.01	<0.01	1.65	7.80	8.61	1.86	12.06
Mean	**24.82**	**<0.01**	**0.39**	**1.46**	**8.97**	**9.92**	**1.95**	**11.35**
100	32.21	<0.01	<0.01	1.55	12.02	1.55	4.11	5.30
100	28.52	<0.01	<0.01	<0.01	9.00	1.03	3.41	4.08
Mean	**30.36**	**<0.01**	**<0.01**	**0.77**	**10.51**	**1.30**	**3.76**	**4.68**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
** Sediment extracts (1-3/4) and soxhlet extract (5). Extracts 6 and 7 represented ≤1.5% AR so were not further characterised.
n.a not analysed
- <LOD
2,4-DBA: 2,4-dichlorobenzoic acid
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide

Table CA.B.8.2.2.3-8: Characterisation of radioactivity in the water phase and sediment extracts of the [Carbonyl-^{14}C]-bixlozone Calwich Abbey system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity					
	Water			Sediment**		
	Bixlozone -3-OH-Prop.	Bixlozone - DMM	Bixlozone	Bixlozone -3-OH-Prop.	Bixlozone - DMM	Bixlozone
0	-	-	96.96	n.a	n.a	n.a
0	-	-	97.76	n.a	n.a	n.a
Mean	**-**	**-**	**97.36**	**n.a**	**n.a**	**n.a**
7	3.51	-	76.24	8.57	1.08	3.14
7	3.66	-	79.94	4.89	0.54	4.76
Mean	**3.59**	**-**	**78.09**	**6.73**	**0.81**	**3.95**
14	<0.01	8.06	41.62	4.72	3.41	21.58
14	<0.01	16.34	38.00	4.57	4.20	16.90
Mean	**<0.01**	**12.20**	**39.81**	**4.64**	**3.81**	**19.24**
30	<0.01	21.96	16.12	4.83	4.64	10.65
30	<0.01	2.76	17.82	4.03	4.10	23.38
Mean	**<0.01**	**12.36**	**16.97**	**4.43**	**4.37**	**17.02**
63	<0.01	<0.01	2.24	6.88	1.46	9.33
63	<0.01	<0.01	9.01	3.55	0.97	11.14
Mean	**<0.01**	**<0.01**	**5.62**	**5.22**	**1.21**	**10.24**
100	<0.01	<0.01	<0.01	0.72	6.50	4.24
100	<0.01	1.24	2.12	0.65	4.81	2.75
Mean	**<0.01**	**0.62**	**1.06**	**0.68**	**5.66**	**3.50**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
** Sediment extracts (1-3/4) and soxhlet extract (5). Extracts 6 and 7 represented ≤1.5% AR so were not futher characterised.
n.a not analysed
- <LOD
2,4-DBA: 2,4-dichlorobenzoic acid
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide

Table CA.B.8.2.2.3-9: Characterisation of radioactivity in the water phase and sediment extracts of the [Phenyl-U-^{14}C]-bixlozone Swiss Lake system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity								
	Water				Sediment**				
	2,4-DBA	Bixlozone-DMM	4-COOH-bixlozone	Bixlozone	2,4-DBA	Bixlozone-3-OH-Prop.	Bixlozone-DMM	4-COOH-bixlozone	Bixlozone
0	-	-	-	95.95	n.a	n.a	n.a	n.a	n.a
0	-	-	-	96.52	n.a	n.a	n.a	n.a	n.a
Mean	**-**	**-**	**-**	**96.23**	**n.a**	**n.a**	**n.a**	**n.a**	**n.a**
7	-	-	-	74.28	-	1.76	-	-	18.13
7	-	-	-	68.56	-	3.94	-	-	12.99
Mean	**-**	**-**	**-**	**71.42**	**-**	**2.85**	**-**	**-**	**15.56**
14	3.93	11.53	-	43.10	<0.01	6.31	0.56	-	17.99
14	8.40	6.80	-	5<0.01	0.69	7.38	1.43	-	6.10
Mean	**6.17**	**9.16**	**-**	**46.55**	**0.34**	**6.84**	**1.00**	**-**	**12.04**
30	14.64	5.34	2.07	33.07	1.62	2.54	2.91	0.35	20.03
30	27.70	8.12	6.14	18.87	4.12	5.36	1.99	<0.01	7.76
Mean	**21.17**	**6.73**	**4.11**	**25.97**	**2.87**	**3.95**	**2.45**	**0.17**	**13.90**
63	11.20	14.11	1.51	16.88	2.38	3.94	7.41	<0.01	20.57
63	29.36	4.43	2.60	12.45	8.68	3.41	3.98	<0.01	13.11
Mean	**20.28**	**9.27**	**2.05**	**14.66**	**5.53**	**3.68**	**5.70**	**<0.01**	**16.84**
100	7.99	4.08	19.67	<0.01	3.15	0.67	4.27	8.82	14.68
100	5.80	2.71	15.53	8.22	1.41	2.08	3.11	4.88	16.73
Mean	**6.90**	**3.40**	**17.60**	**4.11**	**2.28**	**1.38**	**3.68**	**6.85**	**15.70**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
** Sediment extracts (1-3/4) and soxhlet extract (5). Extracts 6 and 7 represented ≤1.5% AR so were not futher characterised.
n.a not analysed
- <LOD
2,4-DBA: 2,4-dichlorobenzoic acid
Bixlozone-DMM: bixlozone-dimethyl malonamide
4-COOH-bixlozone: 4-carboxy-bixlozone
Bixlozone-3-OH-Prop: bixlozone-3-OH-propanamide

Table CA.B.8.2.2.3-10: Characterisation of radioactivity in the water phase and sediment extracts of the [Carbonyl-^{14}C]-bixlozone Swiss Lake system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity							
	Water				Sediment**			
	Bixlozone -3-OH-Prop.	Bixlozone -DMM	4-COOH-bixlozone	Bixlozone	Bixlozone -3-OH-Prop.	Bixlozone -DMM	4-COOH-bixlozone	Bixlozone
0	-	-	-	96.84	n.a	n.a	n.a	n.a
0	-	-	-	97.38	n.a	n.a	n.a	n.a
Mean	**-**	**-**	**-**	**97.11**	**n.a**	**n.a**	**n.a**	**n.a**
7	1.93	<0.01	<0.01	64.41	4.58	-	-	20.34
7	<0.01	1.56	2.64	68.19	4.89	-	-	16.88
Mean	**0.97**	**0.78**	**1.32**	**66.30**	**4.73**	**-**	**-**	**18.61**
14	<0.01	<0.01	<0.01	52.85	3.65	0.76	0.32	23.58
14	<0.01	3.86	5.32	43.85	5.46	1.01	0.75	15.98
Mean	**<0.01**	**1.93**	**2.66**	**48.35**	**4.56**	**0.89**	**0.54**	**19.78**
30	<0.01	4.44	2.77	28.74	1.38	2.93	<0.01	31.64
30	<0.01	5.50	6.45	18.74	2.90	2.91	1.01	14.49
Mean	**<0.01**	**4.97**	**4.61**	**23.74**	**2.14**	**2.92**	**0.51**	**23.07**
63	<0.01	13.69	3.00	10.55	0.99	5.60	<0.01	16.16
63	<0.01	7.80	4.64	14.25	<0.01	5.56	0.97	29.55
Mean	**<0.01**	**10.74**	**3.82**	**12.40**	**0.50**	**5.58**	**0.48**	**22.85**
100	<0.01	<0.01	10.43	3.01	0.28	3.58	5.58	21.11
100	<0.01	3.50	14.43	0.68	0.72	5.45	5.94	18.11
Mean	**<0.01**	**1.75**	**12.43**	**1.84**	**0.50**	**4.51**	**5.77**	**19.62**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
** Sediment extracts (1-3/4) and soxhlet extract (5). Extracts 6 and 7 represented ≤1.5% AR so were not further characterised.
n.a not analysed
- <LOD
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide
4-COOH-bixlozone: 4-carboxy-bixlozone

Table CA.B.8.2.2.3-11: Characterisation of radioactivity in the [Phenyl-U-^{14}C]-bixlozone Calwich Abbey total water-sediment system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity			
	2,4-DBA	**Bixlozone -3-OH-Prop.**	**Bixlozone -DMM**	**Bixlozone**
0	-	-	-	94.72
0	-	-	-	94.30
Mean	**-**	**-**	**-**	**94.51**
7	-	7.45	5.79	72.00
7	-	5.52	1.69	83.30
Mean	**-**	**6.48**	**3.74**	**77.65**
14	<0.01	9.65	5.73	72.19
14	3.58	8.71	17.52	55.54
Mean	**1.79**	**9.18**	**11.62**	**63.87**
30	17.15	3.25	3.42	44.09
30	11.66	4.30	12.56	48.38
Mean	**14.41**	**3.77**	**7.99**	**46.23**
63	35.75	11.23	2.82	11.91
63	31.82	8.61	1.86	13.71
Mean	**33.78**	**9.92**	**2.34**	**12.81**
100	44.23	1.56	4.11	6.85
100	37.52	1.03	3.41	4.08
Mean	**40.87**	**1.29**	**3.76**	**5.46**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
- <LOD
2,4-DBA: 2,4-dichlorobenzoic acid
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide

Table CA.B.8.2.2.3-12: Characterisation of radioactivity in the [Carbonyl-^{14}C]-bixlozone Calwich Abbey total water-sediment system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity		
	Bixlozone -3-OH-Prop.	**Bixlozone -DMM**	**Bixlozone**
0	-	-	96.96
0	-	-	97.76
Mean	**-**	**-**	**97.36**
7	12.08	1.08	79.38
7	8.55	0.54	84.70
Mean	**10.31**	**0.81**	**82.04**
14	4.72	11.47	63.21
14	4.57	20.54	54.89
Mean	**4.64**	**16.01**	**59.05**
30	4.83	26.59	26.78
30	4.03	6.86	41.20
Mean	**4.43**	**16.72**	**33.99**
63	6.88	1.46	11.57
63	3.55	0.97	20.15
Mean	**5.22**	**1.21**	**15.86**
100	0.72	6.50	4.24
100	0.65	6.06	4.88
Mean	**0.68**	**6.28**	**4.56**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
- <LOD
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide

Table CA.B.8.2.2.3-13: Characterisation of radioactivity in the [Phenyl-U-^{14}C]-bixlozone Swiss Lake total water-sediment system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity				
	2,4-DBA	**Bixlozone -3-OH-Prop.**	**Bixlozone - DMM**	**4-COOH-bixlozone**	**Bixlozone**
0	-	-	-	-	95.95
0	-	-	-	-	96.52
Mean	**-**	**-**	**-**	**-**	**96.23**
7	-	1.76	-	-	92.41
7	-	3.94	-	-	81.55
Mean	**-**	**2.85**	**-**	**-**	**86.98**
14	3.93	6.31	12.09	-	61.08
14	9.09	7.38	8.23	-	56.10
Mean	**6.51**	**6.84**	**10.16**	**-**	**58.59**
30	16.26	2.54	8.26	2.41	53.11
30	31.82	5.36	10.12	6.14	26.63
Mean	**24.04**	**3.95**	**9.19**	**4.28**	**39.87**
63	13.57	3.95	21.52	1.51	37.44
63	38.04	3.40	8.41	2.60	25.57
Mean	**25.81**	**3.68**	**14.96**	**2.05**	**31.50**
100	11.13	0.67	8.35	28.49	14.68
100	7.20	2.09	5.82	20.41	24.94
Mean	**9.17**	**1.38**	**7.08**	**24.45**	**19.81**

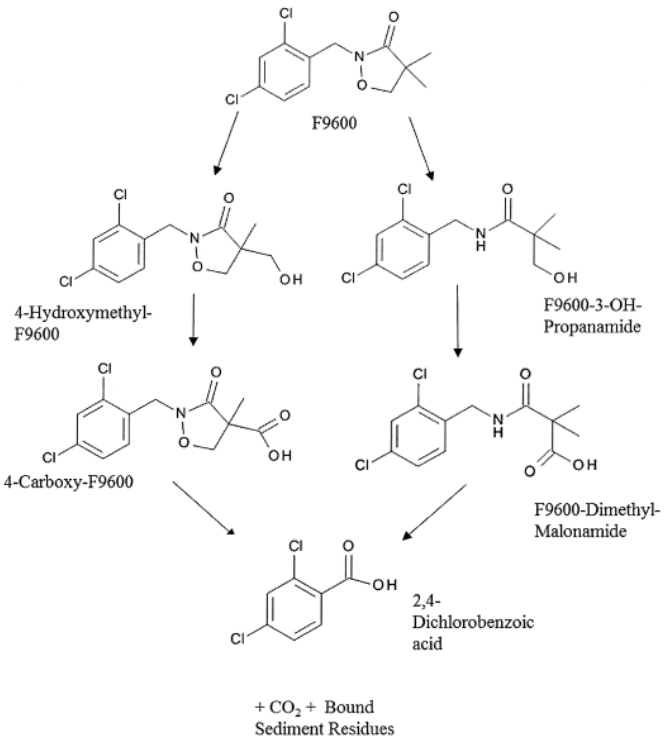
* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
- <LOD
2,4-DBA: 2,4-dichlorobenzoic acid
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide
4-COOH-bixlozone: 4-carboxy-bixlozone

Table CA.B.8.2.2.3-14: Characterisation of radioactivity in the [Carbonyl-^{14}C]-bixlozone Swiss Lake total water-sediment system (as % applied radioactivity by HPLC)*

Day	% Applied Radioactivity			
	Bixlozone-3-OH-Prop.	**Bixlozone -DMM**	**4-COOH-bixlozone**	**Bixlozone**
0	-	-	-	96.84
0	-	-	-	97.38
Mean	**-**	**-**	**-**	**97.11**
7	6.51	<0.01	<0.01	84.75
7	4.89	1.56	2.64	85.07
Mean	**5.70**	**0.78**	**1.32**	**84.91**
14	3.65	0.76	0.32	76.42
14	5.46	4.88	6.07	59.83
Mean	**4.56**	**2.82**	**3.19**	**68.13**
30	1.38	7.38	2.77	60.39
30	2.90	8.41	7.47	33.23
Mean	**2.14**	**7.89**	**5.12**	**46.81**
63	0.99	19.28	3.00	26.71
63	<0.01	13.36	5.61	43.80
Mean	**0.50**	**16.32**	**4.30**	**35.26**
100	0.28	3.57	16.01	24.12
100	0.72	8.95	20.37	18.80
Mean	**0.50**	**6.26**	**18.19**	**21.46**

* Other minor metabolites were detected but none were >5% AR at two consecutive time-points or >5% and increasing at the end of the study, therefore have been omitted from this table for simplicity.
- <LOD
Bixlozone -3-OH-Prop: bixlozone-3-OH-propanamide
Bixlozone -DMM: bixlozone-dimethyl malonamide
4-COOH-bixlozone: 4-carboxy-bixlozone

Figure CA.B.8.2.2.3-1.: Metabolic pathway in water/sediment



KINETICS

The Applicant undertook a kinetic evaluation in order to derive persistence and modelling endpoints. Kinetic analysis was performed following the recommendations of the FOCUS Kinetics workgroup [*FOCUS (2006)*]. The applicant carried out the kinetic evaluation using CAKE v3.3 with IRLS as selected optimisation. For completeness, the CA has repeated the modelling using KinGUI v2 with NLLS selected. The goodness-of-fit was evaluated by visual assessment, χ^2 minimum error, and type-I-error rate (t-test). Modelling endpoints were derived preferably from the SFO model. The initial concentration of the test substance in the total system and in water was set to the material balance recovered at 0 DAT. The individual label results were combined in the kinetic assessment (i.e. 4 replicates per sample point were modelled) and 'less than' results were treated as ½ LOD.

For UK surface water assessments, the longest non-normalised water and sediment DissT50 values are appropriate for use. The applicant did not calculate bixlozone sediment DissT50 values and so these have been calculated by the CA. The CA modelled the sediment data adjusting the time zero to the peak concentration of bixlozone in sediment. For Calwich Abbey, peak bixlozone concentration occurred at 30 DAT (determined via the mean value of all four replicates). It is noted this only leaves two data points in the decline phase and so the resulting DissT50 values should be viewed with caution; further consideration of the endpoints is provided at the end of the kinetic section. For Swiss Lake, no clear decline phase could be observed in the sediment data and so no kinetic assessment has been performed for this test system.

The CA also notes the applicant has not undertaken kinetic analysis on the metabolites, using default values for the $PEC_{SW/sed}$ calculations instead. As the applicant's approach results in a more conservative assessment, and that metabolite water/sediment DT50 values are not typically used in UK surface water assessments (as a total dose method is usually undertaken), the CA accepts the applicant's approach and so no kinetic analysis on the metabolites has been undertaken in this evaluation.

Calwich Abbey, phenyl and carbonyl labels

Modelling endpoints

Water phase:

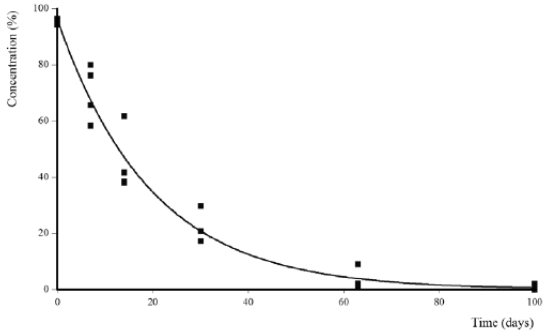
For the combined label Calwich Abbey water compartment, SFO was initially run and resulted in a good visual and statistical fit. Therefore, the SFO model was considered appropriate to determine the modelling endpoint. As the CA obtained a similar DissT50 to the applicant, the applicant's fit is accepted on this occasion. The applicant's SFO fit is summarised in Table CA.B.**8.2.2.3-15** and Figure CA.B.**8.2.2.3-2**.

Table CA.B.8.2.2.3-15: Applicant's Calwich Abbey water compartment modelling endpoint results

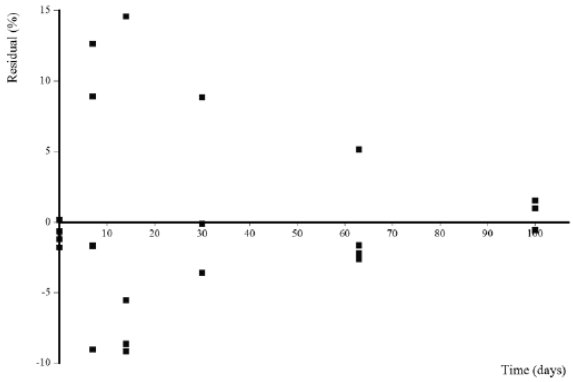
Location	Kinetic Model	Visual Fit	χ^2	k	t-test	DissT50[d]	DissT90[d]
Calwich Abbey	SFO	Good	3.0	0.0509	k: <0.001	13.6	45.3

Figure CA.B.8.2.2.3-2: Applicant's Calwich Abbey water compartment modelling endpoint graphs

SFO:



SFO:



Sediment phase:

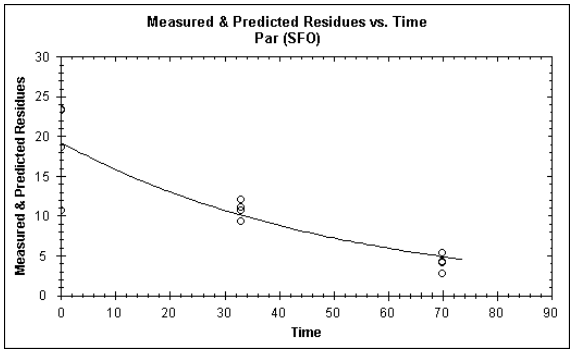
The CA plotted the sediment data from the peak occurrence (30 DAT, adjusting the subsequent time points accordingly) with a SFO fit initially. This resulted in a good visual and statistical fit of the data and so no further fits were explored. The CA's SFO fit is summarised in and Table CA.B.**8.2.2.3-16** and Figure CA.B.**8.2.2.3-3**.

Table CA.B.8.2.2.3-16: CA's Calwich Abbey sediment compartment modelling endpoint results

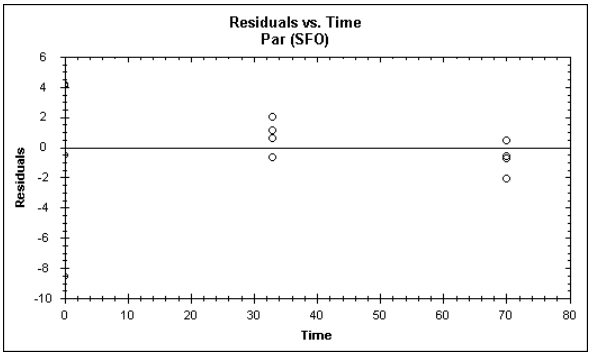
Location	Kinetic Model	Visual Fit	χ^2	k	t-test	DissT50[d]	DissT90[d]
Calwich Abbey	SFO	Good	4.9	0.01970	k: <0.001	35.2	117

Figure CA.B.8.2.2.3-3: CA's Calwich Abbey sediment compartment modelling endpoint graphs

SFO:



SFO:



Persistence endpoints

In line with the SANCO PBT guidance (Brussels, 25.09.212 – rev. 3), a total system DegT50 has been calculated for comparison against the persistence criteria. As an initial step, SFO and FOMC models were run for the total system and their results compared. Both models resulted in good visual and statistical fits. As the FOMC fit was no better than the SFO fit, the SFO fit was considered appropriate for determining the persistence endpoint. The CA obtained similar results to the applicant and so the applicant's results are accepted on this occasion and are presented in Table CA.B.**8.2.2.3-17** and

Figure CA.B.**8.2.2.3-4**.

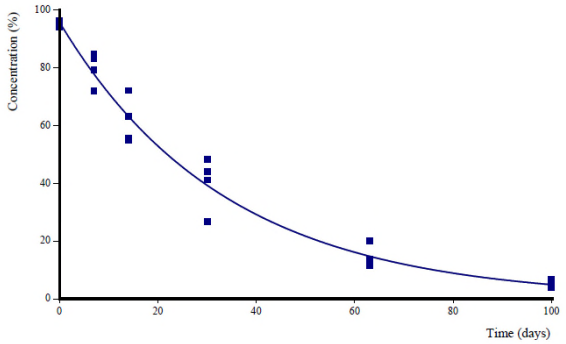
Table CA.B.8.2.2.3-17: Applicant's Calwich Abbey total system persistence endpoint results

Location	Kinetic Model	Visual Fit	χ^2	Kinetic Parameters	t-test	DegT50[d]	DegT90[d]
Calwich Abbey	**SFO**	**Good**	**1.9**	**k: 0.00177**	**<0.001**	**23.3**	**77.6**
	FOMC	Good	2.09	alpha: 575.1 beta: 1.66E+4	n/a	20	66.6

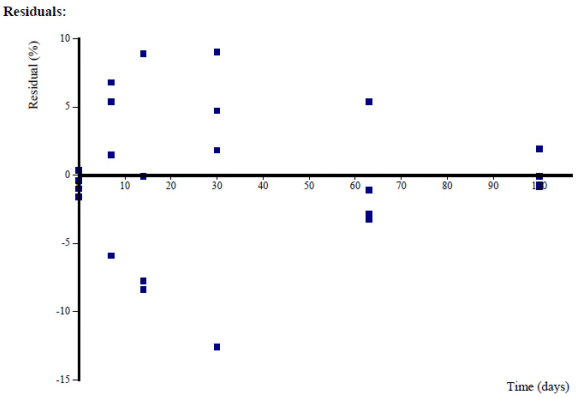
Best fit model highlighted in bold

Figure CA.B.8.2.2.3-4: Applicant's Calwich Abbey total system persistence endpoint graphs

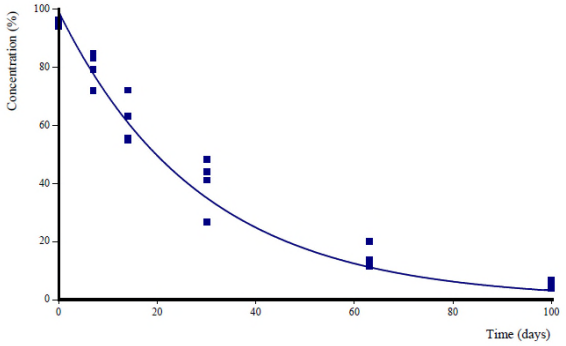
SFO:



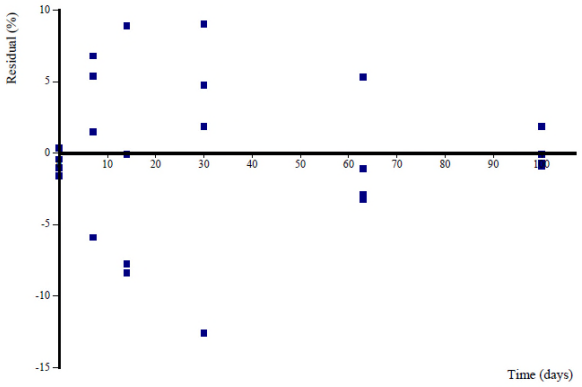
SFO:



FOMC:



FOMC:



Swiss Lake, phenyl and carbonyl labels

Modelling endpoints

Water phase:

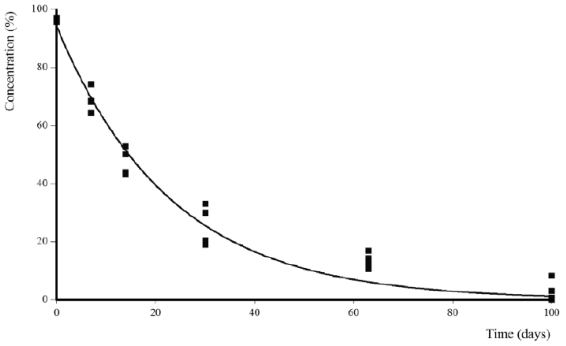
For the combined label Swiss Lake water compartment, SFO was initially run and resulted in a good visual and statistical fit. Therefore, the SFO model was considered appropriate to determine the modelling endpoint. As the CA obtained a similar DissT50 to the applicant, the applicant's fit is accepted on this occasion. The applicant's SFO fit is summarised in Table CA.B.**8.2.2.3-18** and Figure CA.B.**8.2.2.3-5**.

Location	Kinetic Model	Visual Fit	χ^2	k	t-test	DissT50[d]	DissT90[d]
Swiss Lake	SFO	Good	6.73	0.0434	<0.001	16.0	53.1

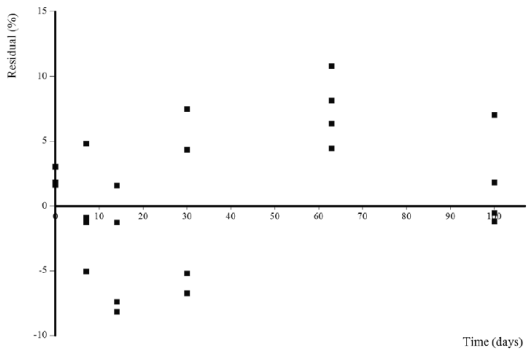
Table CA.B.8.2.2.3-18: Applicant's Swiss Lake water compartment modelling endpoint results

Figure CA.B.8.2.2.3-5: Applicant's Swiss Lake water compartment modelling endpoint graphs

SFO:



SFO:



Sediment phase:

No sediment phase kinetics have been performed due to no clear decline phase being present in the data.

Persistence endpoints

To determine the persistence endpoints, the applicant ran SFO and FOMC models initially for the total-system data. The applicant concluded the FOMC fit resulted in a slightly better visual and statistical fit than SFO and so proceeded to run DFOP and HS models. The applicant concluded the HS model provided the best visual and statistical fit of the data. The CA has repeated the applicant's modelling and agrees with the applicant's assessment. The CA also obtained similar results to the applicant and so the applicant's results are accepted on this occasion. The applicant's results are summarised in Table CA.B.**8.2.2.3-19** and

Figure CA.B.**8.2.2.3-6**.

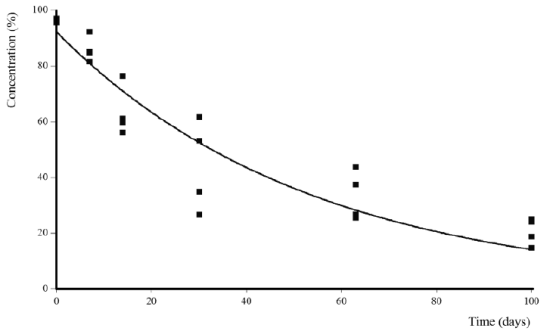
Table CA.B.8.2.2.3-19: Swiss Lake Total system modelling endpoints

Location	Kinetic Model	Visual Fit	χ^2	Kinetic Parameters	t-test	DegT50[d]	DegT90[d]
Swiss Lake	SFO	Good	8.76	k: 0.0188	<0.001	36.9	123
	FOMC	Good	5.69	alpha: 1.089 beta: 32.06	n/a	28.5	234
	DFOP	Good	6.07	k1: 0.0510 k2: 0.0067 g: 0.5697	k1: 0.099 k2: 0.269 g: n/a	27.3	218
	HS	**Good**	**4.75**	**k1: 0.0279** **k2: 0.0103** **tb: 27.22**	**k1: <0.001** **k2: 0.001** **tb: n/a**	**24.8**	**177**

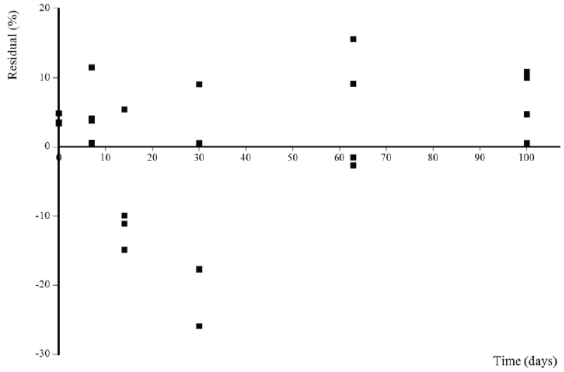
Best fit model highlighted in bold

Figure CA.B.8.2.2.3-6: Applicant's Swiss Lake total system persistence endpoint graphs

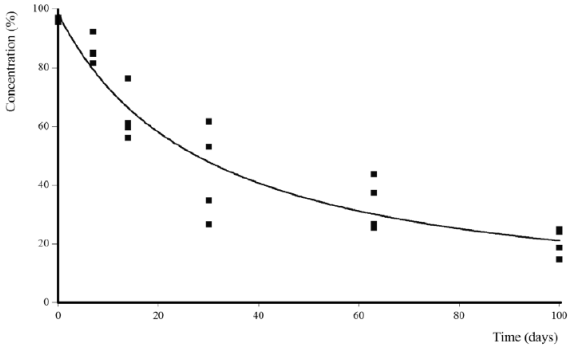
SFO:



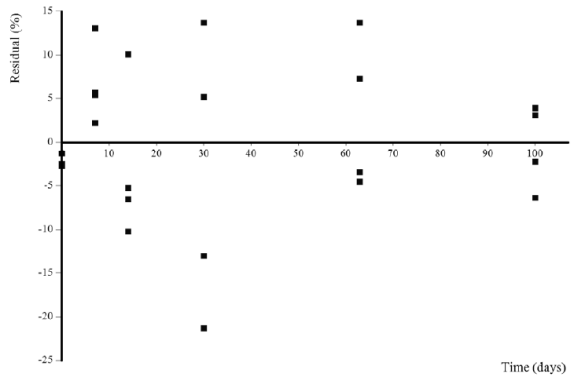
SFO:



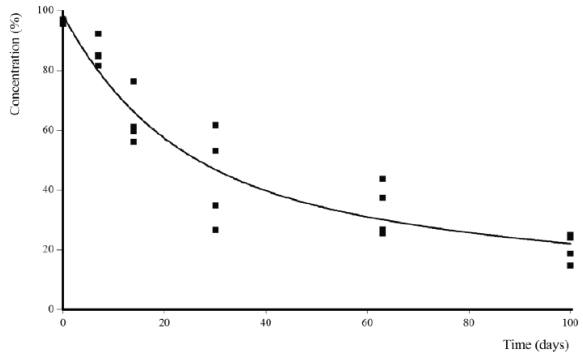
FOMC:



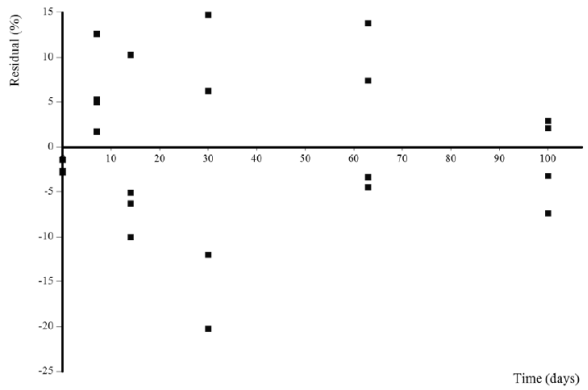
FOMC:



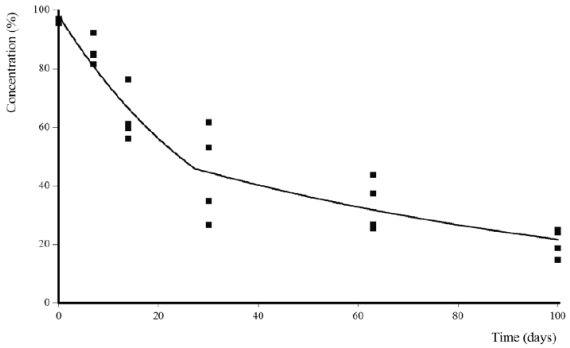
DFOP:



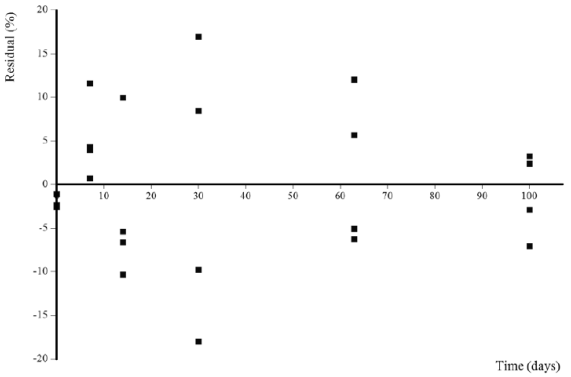
DFOP:



HS:



HS:



A summary of the modelling and persistence endpoints is presented in Table CA.B.**8.2.2.3-20**.

Regarding the longest non-normalised sediment DissT50 to be used in the UK spray drift assessment, as explained above, a kinetic assessment could only be performed on the Calwich Abbey sediment phase. However, as the Calwich Abbey data only consisted of two time points in the decline phase, the results should be viewed with caution. The CA notes that in UK assessments, where a sediment DissT50 is not available, the standard approach is to use the longest total-system DegT50 instead. The CA considers that, because the Calwich Abbey sediment DissT50 is greater than the total-system DegT50 values, the sediment DissT50 is acceptable for use in the UK spray drift calculations.
Table CA.B.. Normalisation of the endpoints is not required as the UK spray drift assessment and assessment of persistence both consider non-normalised endpoints. The longest non-normalised water DissT50 to use in the UK spray drift assessment is 16 days, obtained from the Swiss Lake test system. An assessment of the persistence of bixlozone in water/sediment is presented in section CA.B.8.2.5.

Regarding the longest non-normalised sediment DissT50 to be used in the UK spray drift assessment, as explained above, a kinetic assessment could only be performed on the Calwich Abbey sediment phase. However, as the Calwich Abbey data only consisted of two time points in the decline phase, the results should be viewed with caution. The CA notes that in UK assessments, where a sediment DissT50 is not available, the standard approach is to use the longest total-system DegT50 instead. The CA considers that, because the Calwich Abbey sediment DissT50 is greater than the total-system DegT50 values, the sediment DissT50 is acceptable for use in the UK spray drift calculations.

Table CA.B.8.2.2.3-20: Summary of modelling and persistence endpoints

Location	Compartment	Kinetic Model	DT50 (d)	DT90 (d)
Modelling endpoints				
Calwich Abbey	Water (diss)	SFO	13.6	45.3
	Sediment (diss)	SFO	35.2	117
Swiss Lake	Water (diss)	SFO	16.0	53.1
	Sediment (diss)	n/a	n/a	n/a
Persistence endpoints				
Calwich Abbey	Total system (deg)	SFO	23.3	77.6
Swiss Lake	Total system (deg)	HS	24.8	177

Conclusion

The route and rate of degradation of bixlozone in water and sediment was investigated in two water/sediment systems. Bixlozone was found to degrade into 4 major metabolites; no kinetic analysis was undertaken on the metabolites with default values to be used in the $PEC_{SW/sed}$ calculations instead. The longest non-normalised bixlozone water DissT50, to be used in the UK spray drift calculations, is 16 days, derived from the Swiss Lake test system. The longest non-normalised bixlozone sediment DissT50, to be used in the UK spray drift calculations, is 35.2 days, derived from the Calwich Abbey test system.

CA.B.8.2.2.4. *Anaerobic water/sediment*

Report:	KCA 7.2.2.3 /02 Simmonds, R. (2018)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	^{14}C- F9600Anaerobic Aquatic Metabolism in Two Water/Sediment Systems at 20 ± 2 °C
Testing facility:	Battelle UK Ltd., Springfield, UK
Document No:	Study no. KW/15/006, FMC Tracking no. 2015EFT-ISX2196
Guidelines:	OECD Guideline 309
GLP:	Yes (laboratory certified by UK National Authority)

CA Comments:	The applicant has submitted an anaerobic water/sediment study. However, as this study does not form part of the data requirements, the CA has not evaluated this study. To avoid potential confusion, no further consideration, or information pertaining to this study, is provided in the DAR. It is noted the applicant considers a novel metabolite (bixlozone-isobutyramide) arising from this study in the CP assessment. Similarly, the applicant considers a higher maximum occurrence of 3-OH from this study than was recorded in the aerobic water/sediment study. However, for the above reasons, the CA does not consider it necessary to consider the results from this study further in the evaluation. Therefore, only the substances/occurrences arising from the aerobic water/sediment study have been considered further in the DAR. **This study is <u>not</u> relied upon**

CA.B.8.2.2.5. *Irradiated water/sediment study*

The applicant has not submitted an irradiated water/sediment study, stating photochemical degradation is not a significant route of degradation for bixlozone. The CA considers the submitted aquatic degradation studies to adequately assess the behaviour of bixlozone in water and so does not consider an irradiated water/sediment study necessary.

CA.B.8.2.3. **Degradation in the saturated zone**

The applicant has not provided any specific information regarding bixlozone degradation in the saturated zone, stating no data is available. The CA considers the submitted aquatic degradation studies to adequately assess the behaviour of bixlozone in water and so does not consider further information necessary.

CA.B.8.2.4. **Potential effects of water treatment processes**

Article 4 Section 3 (b) refers to the impact of water treatment processes on water-borne residues of active substances and metabolites, i.e. the capability of water treatment processes to form potentially harmful substances when degrading the water-borne residue.

At present there is no definitive approach to consider the above. The applicant has submitted the following in regards to the potential impact of water treatments works on bixlozone:

According to Paragraph 3, Article 4 of Regulation (EC) No 1107/2009 a plant protection product "shall have no immediate or delayed harmful effect on human health, including that of vulnerable groups, or animal health, directly or through drinking water (taking into account substances resulting from water treatment)". The fate of bixlozone and the soil metabolites 2,4-dichlorobenzoic acid and bixlozone 3OH-propanamide are assessed for risk to human health when considering water treatment procedures below.

bixlozone has an isoxazole ring in its structure. This necessitates further investigation of the possibility of formation of N-Nitrosodimethylamine (NDMA) under advanced oxidative processes (AOP). Sgroi et.al. (2018) summarise the latest research into NDMA formation in water and wastewater treatment. In the review, the formation of NDMA is predominantly attributed to a nucleophilic substitution reaction between an N,N-dimethylamino (DMA) group and dichloroamine. Other functional groups are discussed, for example amides, however the DMA functional group remains important as the precursor molecule of NDMA.

bixlozone does not contain a DMA functional group. This immediately negates the risk of formation of NDMA under these oxidative processes. It is therefore considered that the probability of formation of nitrosamines from bixlozone as a result of AOPs is low.

bixlozone 3-OH propenamide is a ring-opened structure of bixlozone, which yields a secondary amine and a hydroxyl group. Commensurate, with the parent molecule, this metabolite does not contain a DMA function group and is also therefore incapable of forming NDMA and associated nitrosamines.

2,4-dichlorobenzoic acid does not contain a nitrogen atom, and therefore the formation of nitrosamines will not occur under any circumstances.

The conclusion reached is that the risk of harmful effects on human health, taking into account substances resulting from water treatment, is acceptable and no further assessment is necessary.

The CA fate specialist has consulted with the CA chemistry specialist who has confirmed that nitrosamines are unlikely to be formed. The CA chemistry specialist notes that compounds of known toxicological concern such as, anilines, hexachlorobenzene, polychlorinated biphenyls, hydrazine, phenols, etc, have not been considered by the applicant. However, these are not expected to be formed given the starting structures and so there is little potential for formation of such metabolites.

Furthermore, ozone, chlorine and ammonia (which goes on to form chloramine) are used during generic drinking water treatment processes. Again, the CA chemistry specialist notes in the case of bixlozone that these are not likely to lead to formation of potential harmful metabolites.

In the absence of any definitive guidance, the CA considers that the applicant has provided a sufficient case to demonstrate minimal risk of significant levels of harmful degradation products forming as a result of water treatment processes on bixlozone and its metabolites.

CA.B.8.2.5. **Summary of persistent assessment in water and sediment**

The criteria for a pesticide to be classed as 'Persistent' or 'very Persistent' is outlined within Annex II of EC Regulation 1107/2009. For the water and sediment compartments, these are as follows:

An active substance, safener or synergist fulfils the persistence criterion where:

- The half-life in marine water is higher than 60 days,
- The half-life in fresh or estuarine water is higher than 40 days,
- The half-life in marine sediment is higher than 180 days,
- The half-life in fresh or estuarine water sediment is higher than 120 days

An active substance, safener or synergist fulfils the 'very persistent' criterion where:

- the half-life in marine, fresh- or estuarine water is higher than 60 days,
- the half-life in marine, fresh- or estuarine water sediment is higher than 180 days

As indicated in section CA.B.8.2.1.1, bixlozone was found to be hydrolytically stable at pH 9, 25°C. Similarly, bixlozone was concluded as being not readily biodegradable (section CA.B.8.2.2.1) and very little mineralisation was observed over the duration of the aerobic mineralisation study (section CA.B.8.2.2.2).

However, bixlozone degraded quickly in both water/sediment test systems in the aerobic water/sediment study (section CA.B.8.2.2.3). The CA does not consider either compartment to be the major degradation compartment due to similar levels being recorded in both compartments, in both test systems, at each sampling point. The total system bixlozone DegT50 values of the Calwich Abbey and Swiss Lake test systems were 23.3 days and 24.8 days, respectively.

Of these aquatic studies, the CA considers the results of the water/sediment study to be the most realistic in real-world conditions. Therefore, because the water/sediment DT50 values were <40 days (the criterion for a 'P' assessment in fresh water), the CA does not consider bixlozone to be Persistent in water or sediment.

CA.B.8.2.6. **Summary of fate and behaviour in water and sediment**

The applicant submitted an aqueous hydrolysis study for bixlozone. In a preliminary test [phenyl-U-^{14}C]-bixlozone and [carbonyl-^{14}C]-bixlozone were added to sterile buffer solutions (7.5 mL) at pH 4, 7 and 9. Bixlozone was shown to be hydrolytically stable at pH 4 and 7 over 5 days at 50°C. Since both labels of bixlozone degraded only at pH 9 (>10% AR), a definitive study was conducted at 25, 40, and 50°C for 30 days at pH 9. Bixlozone did not hydrolyse at pH 9 over 30 days at the environmentally relevant temperature of 25°C with expected DT_{50}-values > 1 year. Therefore, no metabolic pathway has been proposed by the applicant. The rate and extent of degradation, however, increased with increasing temperature and pH. Unidentified metabolites were formed at >10% at pH 9 and 40-50˚C, but the CA considers that these metabolites will be unlikely to form at significant levels under environmentally relevant temperature and pH conditions at which hydrolysis is unlikely to be a major route of degradation for bixlozone.

A direct photolysis study was submitted by the applicant using [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone. Bixlozone was slowly degraded to multiple minor photoproducts after 13 days continuous irradiation. All degradation products were < 5% AR at each sampling point. The first-order DT_{50} values were 44.0 and 54.4 days for [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone, respectively, under natural summer sunlight at latitude 30-50°N. It was not possible to determine the quantum yield for bixlozone due to the very low UV absorption at wavelengths > 290 nm.

The applicant submitted a ready biodegradability study in accordance with OECD Guideline 301B (CO_2 Evolution (Modified Sturm Test)). The study was undertaken on non-radiolabelled bixlozone and sodium benzoate was used as a reference substance. Bixlozone showed limited biodegradation with a maximum replicate biodegradation of 13% during the study. Therefore, bixlozone cannot be considered readily biodegradable.

A study of aerobic mineralisation in surface water was carried out. A single water sample was collected from Carsington Reservoir UK, and treated with [phenyl-U-^{14}C]-bixlozone at nominal application rates of 10 and 100 µg/L and incubated at 20 ± 2ºC, in the dark. After 62 days, >90 % of the test substance was recovered in both the 10 µg/L and 100 µg/L test systems. Only one sample recorded an unknown degradation product at a concentration >5 % AR and so no major metabolites were detected in the study.

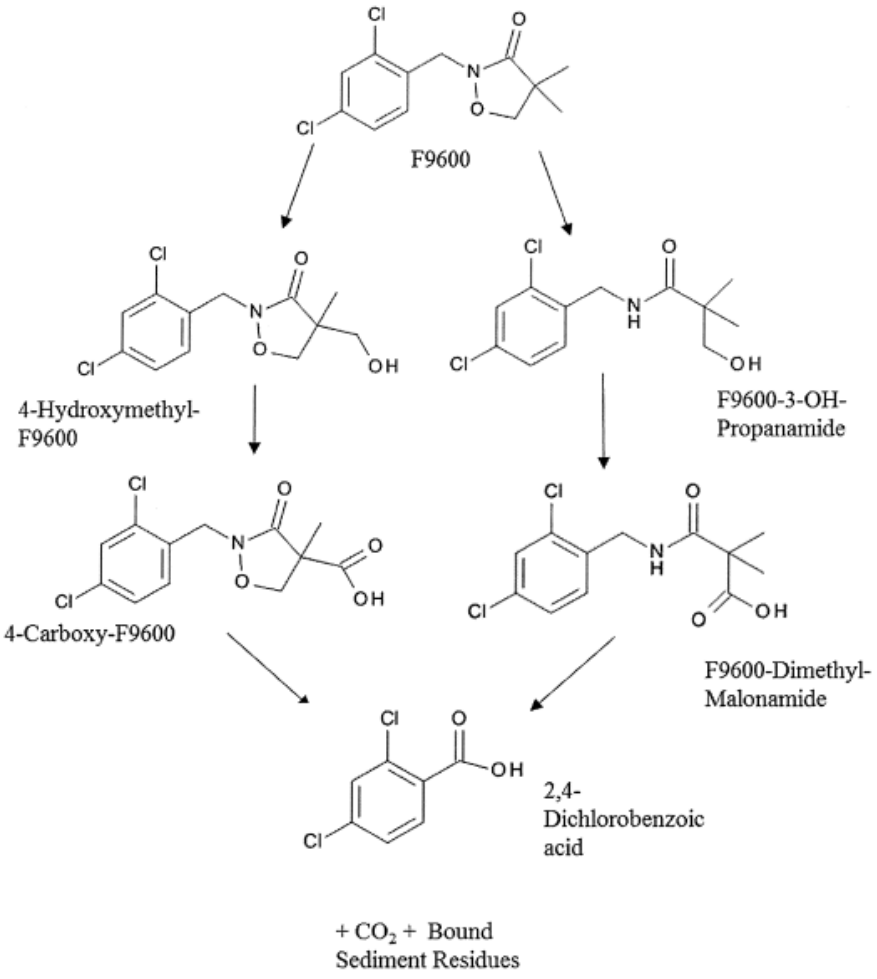
A study of aerobic aquatic mineralisation in two UK water/sediment systems was carried out. The water-sediment systems were incubated at 20 ± 2°C in the dark until there was complete phase separation and to allow the oxygen levels, pH and redox potentials to establish. The samples were treated with [carbonyl-^{14}C]- and [phenyl-U-^{14}C]-bixlozone and were maintained at 20 ± 2°C throughout the course of the study. Bixlozone (mean of both labels) declined to 5.0% AR and 20.6% AR in the total system, in the Calwich Abbey and Swiss Lake systems, respectively, after 100 days. Bixlozone was observed in sediment at mean maxima of 20.99% AR (phenyl label, mean day 30) and 23.07% AR (carbonyl label, mean day 30) in the Calwich Abbey and Swiss Lake systems, respectively. The longest non-normalised water DissT50 value to be used in the exposure assessment was 16 days, derived from Swiss Lake system. The longest non-normalised sediment DissT50, to be used in the UK spray drift calculations, is 35.2 days, derived from the Calwich Abbey test system.

Four major metabolites were observed in the water/sediment study: 2,4-dichlorbenzoic acid (max mean total system formation = 40.9% AR), 3-OH-propanamide (max mean total system formation = 10.3% AR), dimethyl malonamide (max mean total system formation = 16.7% AR) and 4-carboxy-bixlozone (max mean total system formation = 24.5% AR). These metabolites are therefore to be considered in the exposure assessment. No kinetic analysis has been performed on the metabolites by the applicant and so default water DT50 values of 1000 days are appropriate for use in the exposure assessment.

Due to total system DegT50 values being <40 days for both test systems, bixlozone was concluded as not being persistent in water/sediment.

The metabolic pathway of bixlozone in water/sediment is summarised in Figure CA.B.**8.2.6-1**.

Figure CA.B.8.2.6-1: Bixlozone metabolic pathway in water/sediment



CA.B.8.3. FATE AND BEHAVIOUR IN AIR

CA.B.8.3.1. Route and rate of degradation in air

CA.B.8.3.1.1. *Route and rate of degradation in air*

CA Comments	No individual study report was supplied by the applicant assessing the route and rate of degradation in air of bixlozone, with the applicant instead providing this information directly in the MCA document. The CA has therefore evaluated the information presented in the MCA and has summarised it below. **This summary is relied upon.**

INTRODUCTION

The degradation rates for reactions of bixlozone with OH radicals and ozone in the atmosphere were calculated by the applicant using the AOPWIN program based on ATKINSON's increment method.

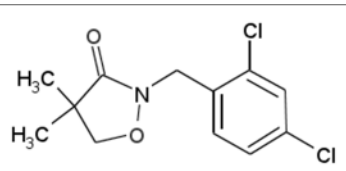
METHOD

The degradation rate resulting from the attack of OH-radicals was calculated with the AOPWIN Program (Version 1.92) based on ATKINSON's increment method [Atkinson, R (1987): *A structure-Activity Relationship for the Estimation of Rate Constants for the Gas-Phase Reactions of OH Radicals with Organic Compounds, Int.J. Chem. Kin. 19, 799*]

The degradation rate resulting from attack of ozone was calculated according to an OECD method [Anonymous (1992): *The rate of photochemical transformation of gaseous organic compounds in air under tropospheric conditions. OECD Environmental Monographs No. 61, OECD, Paris*)

The degradation rate of bixlozone with OH-radicals was estimated based on the structural formula. The structural formula and SMILE notation used for bixlozone in AOPWIN are summarised in Table CA.B.**8.3.1.1-1**:

Table CA.B.8.3.1.1-1: bixlozone structure and SMILES notation

Chemical structure	
SMILES	O=C2C(C)(C)CON2Cc1c(CL)cc(CL)cc1

RESULTS

Assuming a pseudo-first order reaction, the degradation half-life was calculated by taking into account the diurnally and seasonally averaged concentration of hydroxyl-radicals in the troposphere. The total rate constant was estimated to be k_{OH} =21.4854 $E^{-12}/cm^3/molecule^{-1}$ s^{-1}.

Equation 1: Estimation of the atmospheric degradation half-life(t1/2) of bixlozone

$t_{1/2}$ = ln2 / (21.4854 E^{-12} day x 1.5 x 10^6) s

= 5.974 h

= 0.498 d (12 h day)

CONCLUSION

Based on the results of the atmospheric degradation half-life of bixlozone ($t_{1/2}$ = 0.498 d), it can be concluded that the substance will be degraded by photochemical processes in the troposphere. Hence, due to its degradation in air, it can be concluded that there is low risk of long-range transport of bixlozone.

CA.B.8.3.2. **Transport via air**

Report:	KCA 7.3.2/01 Staffa, C. (2016)
Evaluation status:	New data, submitted for purpose of first approval in GB
Title:	Large outdoor wind tunnel study to evaluate the short range transport and deposition of volatilised F9600 (300 g a.s/ha) applied as F9600-4 formulation including the assessment of bleaching effects on a sensitive test plant (*Stellaria media*) as a function of distance from the treated area (0-20 m)
Testing facility:	RLP AgroScience GmbH, Neustadt, Germany
Document No:	Study No. AS442, FMC Tracking no. 2016EFT-ISX2732
Guidelines:	BBA Report No. 110 (2002) Siebers, J & Fent, G. (2012)
GLP:	Yes

CA Comments:	The CA notes the wind tunnel, soil management and meteorological parameters were non-GLP. Furthermore, sampling of the water in steel trays was altered from the original study plan. The resulting sampling dates were 12, 24, 48, 72 and 96 hours after treatment. The CA does not consider these to have significantly impacted on the outcomes of the study. **This study is relied upon.**

INTRODUCTION

The aim of this study was to determine realistic worst-case aqueous deposition values of volatilised bixlozone after re-entry into surface water bodies.

STUDY DESIGN

The test system allows the determination of deposition of pesticides after volatilisation adjacent to the treated areas under realistic climatic outdoor conditions.

Aqueous specimens from a non-target area, downwind of a 100 m^2 plot, which had previously been treated with the test item bixlozone-4 (SC) and lindane (SC), were analysed for deposition of bixlozone and reference item lindane.

In order to exclude drastic variations of natural weather conditions the experiments were carried out under controlled conditions in a wind tunnel. The wind tunnel had a length of approximately 55 m, a width of 6.5 m and a height of 3.1 m. At one end of the tunnel a wind engine with 26 synchronic working fans was installed. The opposite end of the tunnel was left open. Between the wind engine and the target area there was a 5 m air equilibrium distance. The target area had a width of 4 m and a length of 25 m (100 m^2). The distance from the side edges of the field to the wind tunnel was 1.25 m on each side. the sampling points were placed at defined distances on the non-target area. A sketch of the test system is shown in figure 1.

On the non-target area (white in Figure CA.B.**8.3.2-1**) green fallow was grown. Vegetation grown in this area was cut to a length of a few centimetres just before the experiment using a lawn mower.

Stainless steel trays (length 100 cm, width 50 cm, depth 12 cm) were set up on the non-target area (1, 3, 5, 10, 15 and 20 m downwind direction) and about 2.5 m in front of the target area as background control.

To get information about possible bleaching effects on plants caused by volatilised bixlozone, sensitive indicator plants (common chickweed, *Stellaria media*) were placed at various distances in downwind direction (1, 5, 10 and 20 m) and symptoms of phytotoxicity were assessed 1, 2, 3 and 4 days after application in the wind tunnel and later during further cultivation under greenhouse condition (until 21 days after application).

Figure CA.B.8.3.2-1: Sketch of test system

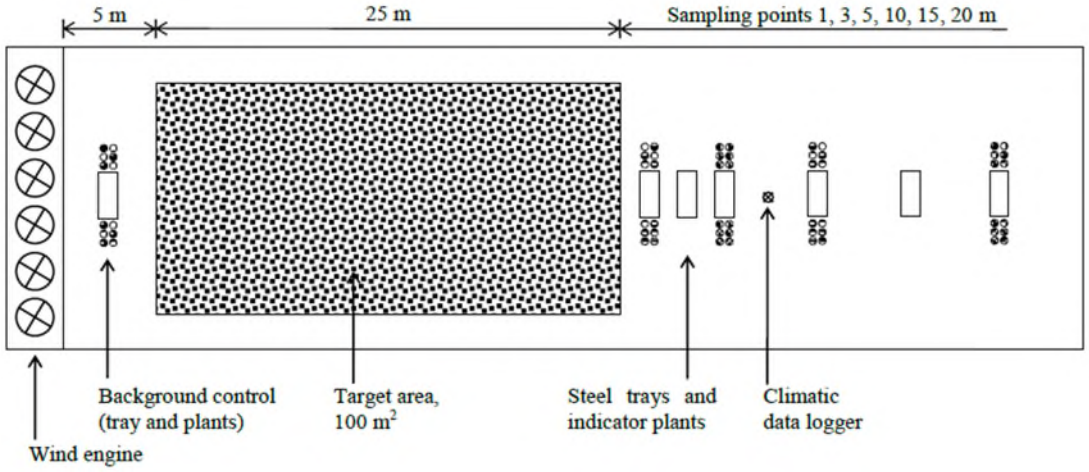


Table CA.B.8.3.2-1: Test item

Product:	bixlozone-4-CS
Lot No:	PL15-0138
Physical Description:	Tan Liquid
Active ingredient:	bixlozone
Concentration:	36.4%
Storage:	Room temperature

Table CA.B.8.3.2-2: Reference item

Trade name:	Lindane 800 SC
Lot No:	0201
Physical Description:	Light-beige dispersion
Common name:	Lindane, γ-HCH
Content:	799 g/L
Storage:	Ambient temperature

Application

Before use the stainless-steel trays were cleaned with water and then with ethanol. The boom sprayer nozzles were cleaned four days before application, and the correct functionality of the boom sprayer was checked. The output of each nozzle was determined by spraying tap water for 20 seconds at a pressure of 2 bar. The water was collected for each nozzle in 1L graduated measuring cylinders. A "practice run" application was carried out using only water before the main trial began.

The applications took place in accordance to good agricultural practice. Each application was carried out with a portable 4 m carbon boom sprayer with eight 90% drift reducing nozzles (company Lechler, 6 × ID 120-05 (symmetrical) and 2 × IS 80-05 asymmetrical edge nozzles) at a pressure of 2.0 bar. The duration of the applications was adapted depending on the results of the boom sprayer check. The target amount of water used for the application corresponded to 300 L/ha. A nominal amount of 3L of each application solution were applied to the target plot. The exact amount applied to the 100 m² target plot was determined by weighing the spray tank before and after the application procedure.

The average wind speed during the 96 hours volatilisation period was 2.04 m/s and the mean air temperature was 20.4 °C. Water sampling was carried out 12, 24, 48, 72 and 96 hours after spray application. Specimens were stored at ≤-18 °C in the dark until further analysis at the analytical test site.

In order to determine if any hydrolysis or photolysis of the test or reference item active ingredient occurred, a reference solution of water was fortified with analytical standard of bixlozone and lindane, 1 µg/L, each. This solution was incubated in a quartz glass vessel for the same time interval (96 hours) and meteorological conditions as the sampling water in the stainless steel trays. Aliquots taken prior and at the 96 hour sampling were analysed.

Sampling

Five minutes after test item application, the stainless-steel trays were carried into the wind tunnel and placed on the prescribed sampling points. The sampling points were in 1, 3, 5, 10, 15, and 20 m downwind distance from the target area. Each of the steel trays was filled with 25 L tap water. The filling procedure took less than three minutes. When all steel trays were filled with water, the wind engine was started.

The indicator plants were carried into the wind tunnel and placed on the sampling points in 1, 5, 10 and 20 m downwind distance about 30 seconds after starting the wind engine. The background control was set up some time after this process, however the CA notes that no time frame was given between application and control set up.

The control set up was carried out by placing one additional stainless steel tray in the background control area to determine the background concentration of the test item and the reference item (upwind direction 2.5 m behind the wind engine and 2.5 m before the target area) and filled with 25 L tap water. A set of control plants in gas-tight plastic bags was brought to the background control area. The plastic bags were removed, and the plants were placed on the control area.

Sampling intervals were 12, 24, 48, 72 and 96 hours after treatment. The applicant reports that these intervals are contrary to stated guidelines, however the CA does not consider this to have substantial effect on the outcome of the study, but instead acts to provide further information about the test item. At all sampling distances two 0.5 L specimens were taken out of the stainless-steel trays and filled in 1 L Nalgene bottles. The exact weight of the specimens was determined by weighing the empty and the filled bottles. Before sampling the water in the steel trays was sufficiently homogenised by stirring. After the 96 hours sampling, the total remaining volume of the water in the steel trays was determined by weighing (assumption was that 1 g water was equivalent to 1 mL) to quantify evaporation losses

Assessment

After the 4-day exposure in the wind tunnel, the *Stellaria media* indicator plants were transferred to the greenhouse for further cultivation. Symptoms of phytotoxicity were assessed 1, 2, 3 and 4 days after treatment (wind tunnel) as well as 7, 14 and 21 days after application (greenhouse). The observed bleaching was recorded and documented by photo images. The degree of the observed phytotoxicity per replicate was given in % affected plant leaf surface of the whole plant compared to the control. On the assessment dates, the effects were

estimated per pot in 5% steps (i.e. 0%, 5%, 10%, etc.). After the last assessment (DAT 21), the plants were cut directly above the ground, and the plant fresh weight of each pot was determined. The last watering was done no later than two days before harvest. Mean values and standard deviations were calculated for phytotoxicity ratings and shoot fresh weight (DAT 21).

Storage and Analysis

Directly after sampling, the aqueous specimens were stored under frozen conditions at the test facility.
The specimens were taken between June 27 and July 01, 2016 and were stored frozen directly after sampling. The specimens were shipped to the analytical test site in isolated containers on July 04, 2016 and were received on the same date. At the analytical test site, the specimens were also stored under frozen conditions until further analysis.

At the analytical test site, the sample preparation (extraction) was performed between July 11 and July 13, analysis was undertaken from July 11 to July 15, 2016. Due to high differences between two replicates (two samples, each), these samples were re-extracted and analysed on July 20, 2016.

Lindane was determined in water by GC-MS/MS after liquid/liquid partition into dichloromethane, evaporation and dilution in toluene. Bixlozone was determined by adding acetonitrile containing 0.1% formic acid to aliquots of the water samples and subsequent LC-MS/MS analysis. The analytical method was validated concurrently at 0.10 µg/L (limit of quantitation, LOQ) for both analytes and at 10 µg/L for bixlozone and 50 µg/L for lindane.

RESULTS

At the 1 m distance, deposition increased from 0.13% of the applied amount (12 h sampling) to 0.42% of the applied amount (48 h and 72 h). This was the highest deposition of bixlozone measured within this study and corresponded to about 135 µg/m^2. For the last sampling 96 hours after treatment, deposition had slightly decreased to 0.39% of the applied amount (1 m). For all sampling intervals, deposition decreased with increasing distance from the treated area and was equal or below 0.04% of applied for the 15 m and 20 m downwind distance. In the background control, no bixlozone was detected for all samplings.

Table CA.B.8.3.2-3: Absolute deposition of test item active ingredient bixlozone

Sampling Dist.	12 h [µg/m^2]	24 [µg/m^2]	48 [µg/m^2]	72 [µg/m^2]	96 [µg/m^2]
1 M	41.09	82.81	133.99	134.56	124.13
3 M	18.25	36.68	62.4	64.06	59.11
5 M	11.61	24.91	43.58	45.14	42
10 M	5.8	12.96	23.14	24.09	22.69
15 M	<LOQ	8.19	13.54	14.07	13.62
20 M	<LOQ	6.01	10.02	10.11	9.42
Back	<LOQ	<LOD	<LOD	<LOD	<LOD

Table CA.B.8.3.2-4: Relative deposition of test item active ingredient bixlozone

Distance	12 H (% a.)	24 h (% a.)	48 h (% a.)	72 h (% a.)	96 h (5 a.)
1 m	0.13	0.26	0.42	0.42	0.39
3 m	0.06	0.11	0.19	0.20	0.18
5 m	0.04	0.08	0.14	0.14	0.13
10 m	0.02	0.04	0.07	0.08	0.07
15 m	<LOQ	0.03	0.04	0.04	0.04
20 m	<LOQ	0.02	0.03	0.03	0.03
BACK	<LOD	<LOD	<LOD	<LOD	<LOD

Lindane was detected in all water specimens taken in downwind direction. At the first sampling 12 hours after treatment, the lindane concentration already occurred at a relatively high level and accounted for about 1.1% of the applied amount (1 m distance), decreasing with increasing sampling distance to 0.12% of applied at the 20 m sampling point. The highest cumulative lindane deposition was measured 48 hours after treatment and corresponded to about 2.2% of applied at the 1 m distance, decreasing to about 0.2% at the 20 m sampling point. On the two subsequent sampling intervals, the measured concentrations of lindane decreased due to declining deposition. At the last sampling performed 96 hours after treatment, a deposition of about 1.0% of applied was measured at the 1 m sampling point and decreased to about 0.1% on the 20 m sampling distance. The background control level of lindane deposition measured at the sampling point located between wind engine and target area was always below limit of detection (0.004% of the applied amount).

Table CA.B.8.3.2-5: Absolute deposition of reference item lindane

Sampling Dist.	12 h [µg/m²]	24 [µg/m²]	48 [µg/m²]	72 [µg/m²]	96 [µg/m²]
1 M	218.8	371.43	454.28	333.75	200.56
3 M	107.32	191.76	244.26	173.34	99.34
5 M	108.14	159.47	101.81	124.17	76.46
10 M	48.43	71.73	93.21	70.76	42.05
15 M	31.14	50.38	55.46	39.31	24.18
20 M	25.26	44.46	42.48	29.41	15.56
Back	<LOD	<LOD	<LOD	<LOD	<LOD

LOD Calculated limit of detection, corresponding to 0.75 µg/m²

Table CA.B.8.3.2-6: Relative deposition of reference item lindane

Sampling Dist.	12 h [µg/m²]	24 [µg/m²]	48 [µg/m²]	72 [µg/m²]	96 [µg/m²]
1 M	1.06	1.81	2.21	1.62	0.98
3 M	0.52	0.93	1.19	0.84	0.48
5 M	0.53	0.78	0.49	0.6	0.37
10 M	0.24	0.35	0.45	0.34	0.2
15 M	0.15	0.24	0.27	0.19	0.12
20 M	0.12	0.22	0.21	0.14	0.08
Back	<LOD	<LOD	<LOD	<LOD	<LOD

At day 1 to day 4 assessment dates, no bleaching of the indicator plants was observed. On the assessment date, 7 days after application, first bleaching effects were observed and accounted for 7% and 4% of the total leaf surface of the plants exposed at the 1 m and 5 m distance, respectively. For the following assessment dates, bleaching effects slightly increased. At the last assessment 21 days after application, bleaching of 13%, 7% and 1% of the total leaf surface was observed for the 1 m, 5 m and 10 m indicator plants, respectively (see Table CA.B.**8.3.2-7**). For all assessment dates, no bleaching was observed for the plants exposed at the 20 m downwind distance and on the background control area.

Table CA.B.8.3.2-7: Results of bleaching test assessment

Distance (m)	DAT 1 (%)	DAT 2 (%)	DAT 3 (%)	DAT 4 (%)	DAT 7 (%)	DAT 14 (%)	DAT 21 (%)
1	0	0	0	0	7 (±6)	9 (±6)	13 (±7)
5	0	0	0	0	4 (±5)	4 (±5)	7 (±8)
10	0	0	0	0	0	0	1 (±2)
20	0	0	0	0	0	0	0
Back	0	0	0	0	0	0	0

At test termination (DAT21), the plant fresh weight in the control was 32.1 g per replicate. For the plants exposed in downwind direction, no statistically significantly reduction of plant fresh weight over the distances was observed (see Table CA.B.**8.3.2-8**).

Table CA.B.8.3.2-8: Plant fresh weight per replicate (pot), DAT 21

Distance (m)	weight (g)	% of Control
1	34.3 (±4.2)	106.9 (±12.2)
5	33.8 (±3.4)	105.3 (±10.1)
10	33.8 (±4.0)	105.3 (±11.8)
20	35.9 (±4.2)	111.8 (±11.7)
Back	32.1 (±7.4)	100 (±23.1)

CONCLUSION

The deposition of test item active ingredient bixlozone took place at a relatively low level. Highest deposition was measured at the 48 h and 72 h sampling at the 1 m distance and corresponded to 0.42% of applied or about 135 µg/m^2. For lindane the maximum deposition corresponded to about 2.2% of the applied amount (1 m, 48 hours), which was about 5 times higher compared to the relative bixlozone deposition. The indicator plants *Stellaria media* were assessed for bleaching effects during the exposure in the wind tunnel (day 1, 2, 3 and 4). First bleaching of the indicator plants was observed 7 days after the treatment of the target area in the wind tunnel and accounted for 7% of the total leaf surface (mean, n=12) at the 1 m distance and 4% at 5 m. Bleaching effects slightly increased over time. At the last assessment on day 21 after exposure, bleaching of 13%, 7% and 1% of the total leaf surface was observed for the 1 m, 5 m and 10 m indicator plants, respectively. For all assessment dates, no bleaching was observed for the plants exposed at the 20 m downwind distance and on the background control area. No significant fresh mass reduction in relation to the non-exposed control plants was observed at the end of the test 21 days after application.

CA.B.8.3.3. **Local and global effects**

The applicant states bixlozone will not be applied in large enough quantities or persist in the atmosphere for sufficient time (DT50 air < 2 days) to cause local or global effects. This is accepted by the CA and no further information is required.

CA.B.8.3.4. **Summary of fate and behaviour in air**

The degradation rates for reactions of bixlozone with OH radicals and ozone in the atmosphere were calculated by the applicant using the AOPWIN program based on ATKINSON's increment method. A rate constant of 21.4854 x 10^{-12}/ cm^3/molecule/s was calculated for reaction with OH radicals. The atmospheric degradation half life of bixlozone was calculated to be 0.498 d (12 hour days) based on an OH radical concentration of 1.5 x 10^6 cm^3 on a 12-hour day basis. Due to its degradation in air, it was considered to have a low risk of long-range transport.

The vapour pressure of bixlozone is 1.1 x 10^{-3} (20 °C) and so meets the FOCUSair trigger of 10^{-4} for the potential of short range transport from application to soil. The Henry's Law constant is 7.2 x 10^{-3} (20 °C). The potential for transport of bixlozone in air was therefore investigated in a wind tunnel study. The amount of deposition of bixlozone was measured at varying distances from the area of application and following set time intervals after the application event. Highest aqueous deposition occurred at 48 h and 72 h at 1m distance from application and represented 0.42% of applied amount. First bleaching of the indicator plants was observed 7 days after treatment and accounted for 7% of total leaf surface area at 1 m distance form application, and 4% of total leaf surface area at 5 m. Bleaching increased over time. At the last assessment on day 21 after exposure, bleaching of 13%, 7% and 1% of the total leaf surface was observed for the 1 m, 5 m and 10 m indicator plants, respectively. Further information is provided in the Vol. 3 CP, B8.

CA.B.8.4. MONITORING DATA CONCERNING FATE AND BEHAVIOUR OF THE ACTIVE SUBSTANCE, METABOLITES, DEGRADATION AND REACTION PRODUCTS

The applicant states that no monitoring is available or required. As this is a new active substance, the CA accepts there being no data available.

CA.B.8.5. REFERENCES RELIED ON

CA.B.8.5.1. **Literature Review**

Report:	KCA Section 9 - Exponent International Ltd. (2018)
Evaluation status:	None
Title:	Submission of scientific peer-reviewed open literature under regulation (EC) No 1107/2009
Testing facility:	Exponent International Ltd. The Lenz, Hornbeam Park Harrogate North Yorkshire HG2 8RE United Kingdom
Document No:	Exponent QAID: 1508442.UK0 – 5012 FMC Tracking Number: 2018WHP-ISX4339
Guidelines:	Article 8 (5) of Regulation (EC) No. 1107/2009
GLP	No

CA comments	The CA has presented its evaluation in the main body of text below. No significant issues were identified. **This study is relied upon**

The applicant conducted a review to comply with current EFSA guidance (EFSA Journal 2011;9(2):2092) for identifying scientific peer-reviewed open literature on the active substance bixlozone and relevant metabolites, which may affect the assessment of human health, animal health and/or the environment.

The search strategy was based on a single-concept search using the STN and the Dialog platforms. The literature search was performed to cover the 10 years prior to the expected submission of the dossier for the new active substance F9600; i.e. for studies published in or after 2008 up to present. Patents and conference papers were excluded as these were not expected to contain information that was both relevant and reliable. Since bixlozone is a new active substance and no formulations have yet been commercialised, the search did not include any product names.

The compounds listed in Table CA.B.**8.5**.1-**1** were searched by the applicant.

Table CA.B.8.5.1-1: Active substance Bixlozone and its metabolites

Active substance	
ISO common names of active substance	Not available
Active substance synonyms	F9600
Chemical name (CA):	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-3-isoxazolidinone
Chemical name (IUPAC):	2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-1,2-oxazolidin-3-one
CAS numbers	81777-95-9
Development Codes:	F9600
CIPAC No.	Not assigned
Metabolite 1	
Code:	3-hydroxypropanamide-F9600
Chemical name (IUPAC):	N-(2,4-dichlorobenzyl)-3-hydroxy-2,2-

	dimethylpropanamide
CAS numbers	Not assigned
Metabolite 2	
Code:	2,4-Dichlorobenzoic acid
Chemical name (IUPAC):	2,4-dichlorobenzoic acid
CAS numbers	50-84-0
Metabolite 3	
Code:	F9600 Dimethyl Malonamide
Chemical name (IUPAC):	3-((2,4-dichlorobenzyl)amino)-2,2-dimethyl-3-oxopropanoic acid
CAS numbers	Not assigned
Metabolite 4	
Code:	5-Hydroxy-F9600
Chemical name (IUPAC):	2-(2,4-dichlorobenzyl)-5-hydroxy-4,4-dimethylisoxazolidin-3-one
CAS numbers	Not assigned
Metabolite 5	
Code:	5'-Hydroxy-F9600
Chemical name (IUPAC):	2-(2,4-dichloro-5-hydroxy benzyl)-4,4-dimethylisoxazolidin-3-one
CAS numbers	Not assigned
Metabolite 6	
Code:	6'-Hydroxy-F9600
Chemical name (IUPAC):	2-(2,4-dichloro-6-hydroxy benzyl)-4,4-dimethylisoxazolidin-3-one
CAS numbers	Not assigned
Metabolite 7	
Code:	4-Hydroxy-methyl-F9600
Chemical name (IUPAC):	2-(2,4-dichlorobenzyl)-4-(hydroxymethyl)-4-methylisoxazolidin-3-one
CAS numbers	
Metabolite 8	
Code:	Dimethyl malonamide- F9600
Chemical name (IUPAC):	3-((2,4-dichlorobenzyl)amino)-2,2-dimethyl-3-oxopropanoic acid
CAS numbers	
Metabolite 9	
Code / Trivial name:	Dimethyl malonic acid
Chemical name (IUPAC):	2,2-Dimethylmalonic acid
CAS numbers	595-46-0
Metabolite 10	
Code:	F9600-isobutyramide
Chemical name (IUPAC):	N-(2,4-dichlorobenzyl)isobutyramide
CAS numbers	Not assigned
Metabolite 11	
Code / Trivial name:	Hydroxy-Isobutyramide
Chemical name (IUPAC):	N-(2,4-dichlorobenzyl)-2-hydroxy-2-methylpropanamide
CAS numbers	Not assigned
Metabolite 12	
Code/ Trivial name:	3-hydroxypivalic acid
Chemical name	2,2-Dimethyl-3-hydroxy propionic acid
Chemical name (IUPAC):	3-Hydroxy-2,2-dimethylpropanoic acid
CAS numbers	4835-90-9
Metabolite 13	
Code:	5-OH, 5'-OH Di-Hydroxy-F9600
Chemical name (IUPAC):	2-(2,4-dichloro-5-hydroxy benzyl)- 5-hydroxy-4,4-dimethylisoxazolidin-3-one

CAS numbers	Not assigned
Metabolite 14	
Code:	4-carboxy-F9600
Chemical name (IUPAC):	2-(2,4-dichlorobenzyl)-4-methyl-3-oxoisoxazolidine-4-carboxylic acid
CAS numbers	
Metabolite 15	
Code:	2,4-Dichloroippuric acid
Chemical name (IUPAC):	N-(2,4-dichlorobenzoyl)glycine
CAS numbers	Not assigned (2,5 analogue has CAS number)

The table below shows the databases used by the applicant to perform the search. The CA considers that a reasonable number of databases have been used.

Table CA.B.8.5.1-2: Databases used/search engines

Database	Provider	Justification
ASFA (Aquatic Sciences and Fisheries Abstracts)	Dialog	ASFA (Aquatic Sciences and Fisheries Abstracts) series is the premier international reference in the field of aquatic resources. Since 1966 input to ASFA has been provided by a growing international network of information centers monitoring more than 5,000 serial publications, books, reports, conference proceedings, translations and limited distribution literature. ASFA is a component of the Aquatic Sciences and Fisheries Information System (ASFIS), formed by four United Nations agency sponsors of ASFA and a network of international and national partners.
AGRICOLA (AGRICultural OnLine Access)	Dialog	AGRICOLA (AGRICultural OnLine Access) is an extensive international bibliographic database consisting of records for literature citations of journal articles, monographs, theses, patents, translations, microforms, audiovisuals, software and technical reports. Available since 1970, AGRICOLA serves as a document locator and bibliographic access and control system for the U.S. National Agricultural Library (NAL) collection, but since 1984 the database has also included some records produced by cooperating institutions for documents not held by NAL.
AGRIS International	Dialog	AGRIS International is the international information system for agricultural sciences and technology. The AGRIS International database has served since 1974 as a comprehensive inventory of worldwide agricultural literature which reflects research results, food production, and rural development to help users identify problems involved in all aspects of world food supply. Emphasis in AGRIS International is non-U.S. This file corresponds in part to the printed publication, Agrindex, published monthly by the Food and Agriculture Organization (FAO) of the United Nations. AGRIS is a cooperative, decentralised system in which over 100 national and multinational centers take part. It collects and makes available current information on the agricultural literature of the world appearing in journals, books, reports, and conference papers. Each country which participates in AGRIS does so by submitting information about documents published within its own territories. All contributing sources are of non-U.S. origin. FAO acts as a coordinating agency within this global information system, facilitating the exchange of agricultural information to its member countries.

Database	Provider	Justification
ANABSTR	STN	The Analytical Abstracts database covers worldwide literature on analytical chemistry. The ANABSTR file contains bibliographic records with abstracts (since 1984) for documents reported in printed Analytical Abstracts. Sources for ANABSTR include journals, books, conference proceedings, reports, and standards. Bibliographic information, indexing terms, abstracts, chemical names, and CAS Registry Numbers are all searchable
Aqualine	Dialog	Aqualine contains abstracts and bibliographic citations from approximately 300 journals as well as from conference proceedings, scientific reports, books and theses. Major subjects of coverage include water resources and supplies management, water legislation, water quality, potable water distribution, wastewater collection, water treatment technologies, wastewater and sewage treatment, and ecological and environmental effects of water pollution. Previously published by the wellknown and respected WRc in England, Aqualine is now produced in joint cooperation with WRc and CSA.
BIOSIS	STN	BIOSIS Previews® is the largest and most comprehensive life science database in the world. Amongst other subject coverage includes Agriculture, Biochemistry, Biophysics, Botany, Environmental Biology, Physiology, Toxicology. Sources include periodicals, journals, conference proceedings, reviews, reports, patents and short communications. Nearly 6,000 life science journals, 1,500 international meetings as well as review articles, books, and monographs are reviewed for inclusion. Bibliographic information, indexing terms, abstracts, and CAS Registry Numbers are all searchable.
BIOTECHNO	STN	Elsevier BIOTECHNOBASE provides comprehensive international coverage of scientific, technological, and professional biotechnology literature - from fundamental research to industrial applications. The database includes both modern biotechnology (genetic engineering, bioreactors, industrial processes, etc.) and traditional biotechnology (breeding, fermentation, etc.). Special emphasis is placed on drug development, medicine and health care, microbial biotechnology, agriculture, food industry, environmental science, forensic science and textiles. BIOTECHNO draws on a core list of 280 journals relevant to biotechnology, plus a selection of other relevant journals from related disciplines.
CAB Abstracts	STN	The CAB Abstracts database covers worldwide literature from all areas of agriculture and related sciences including biotechnology, forestry, and veterinary medicine. Sources for CABA include journals, books, reports, published theses, conference proceedings, and patents. Bibliographic information, indexing terms, abstracts and CAS Registry Numbers are searchable.

Database	Provider	Justification
CAPLUS	STN	The Chemical Abstracts (CA) database covers all areas of Biochemistry, Chemistry and Chemical engineering, and related sciences. Sources include over 8,000 journals, patents from 38 national patent offices and two international patent organizations, technical reports, books, conference proceedings, and dissertations. Electronic only journals and Web preprints are also covered. Bibliographic terms, indexing terms, roles, CAS Registry Numbers, International Patent Classification and abstracts are searchable.
Chemical Abstracts REGISTRY	STN	The Chemical Abstracts REGISTRY covers all types of inorganic and organic substances, including alloys, coordination compounds, minerals, mixtures, polymers, salts, high throughput screening (HTS) compounds as well as nucleic acid and protein sequences. Substances included in REGISTRY meet the following criteria: - Identified by CAS as coming from a reputable source, including but not limited to patents, journals, chemical catalogues and selected substance collections on the web. - Described in largely unambiguous terms. - Characterised by physical methods or described in a patent document example or claim. - Consistent with the laws of atomic covalent organisation. Experimental and predicted property data and tags and spectra data.
Ecology Abstracts	Dialog	Ecologists will find in this journal the essence of current ecology research across a wide range of disciplines, reflecting recent advances in light of growing evidence regarding global environmental change and destruction. Ecology Abstracts focuses on how organisms of all kinds - microbes, plants, and animals - interact with their environments and with other organisms. Included are relevant papers on evolutionary biology, economics, and systems analysis as they relate to ecosystems or the environment. With coverage ranging from habitats to food chains, from erosion to land reclamation, the journal provides an important cross-section of current findings in target research areas. Detailed information on resource and ecosystems management and modeling contributes to the journal's practical value, as does material on the impact of climate, water resources, soil, and man or growing environmental problems such as depletion, erosion, and pollution all topics which are covered in depth. Comprehensive, yet carefully focused coverage makes this an essential resource for scientists concerned with preserving the environment.
EMBASE	STN	The Excerpta Medica database covers worldwide literature in the biomedical and pharmaceutical fields, including biological science, biochemistry, human medicine, forensic science, paediatrics, pharmacy, pharmacology and drug therapy, pharmacoeconomics, psychiatry, public health, biomedical engineering and instrumentation and environmental science. Sources for EMBASE include more than 4,000 journals from approximately 70 countries, monographs, conference proceedings, dissertations and reports.

Database	Provider	Justification
Enviroline®	Dialog	Enviroline® covered the world's environmental related information from 1975-2008. It provided indexing and abstracting coverage of more than 1,000 international primary and secondary publications reporting on all aspects of the environment highlighting such fields as management, technology, planning, law, political science, economics, geology, biology and chemistry as they relate to environmental issues. Published by Congressional Information Service it corresponds to the print Environment Abstracts.
Environment Abstracts	Dialog	Environment Abstracts (formerly Environment Abstracts published by LexisNexis) encompasses all aspects of the impact of people and technology on the environment and the effectiveness of remedial policies and technologies. As of 1994, the database also provides expanded coverage of energy-related issues. Environment Abstracts provides access to more than 950 journals published in the U.S. and abroad. The database also covers conference papers and proceedings, special reports from international agencies, non-governmental organizations, universities, associations and private corporations. Other materials selectively indexed include significant monographs, government studies and newsletters. Environment Abstracts customers will also receive access to Sustainability Science Abstracts and EIS: Digests of Environmental Impact Statements. Environment Abstracts also includes a special collection of over 4,000 full text government reports.
ESBIOBASE	STN	Elsevier BIOBASE is a bibliographic current awareness database providing comprehensive coverage of the entire spectrum of biological research worldwide. Coverage includes the following areas: applied microbiology, biotechnology, cancer research, cell & developmental biology, clinical chemistry, ecological & environmental sciences, endocrinology, genetics, immunology, infectious diseases, metabolism, molecular biology, neuroscience, plant and crop science, protein biochemistry and toxicology. Records are selected from over 1,700 international scientific journals, books and conference proceedings.
Foodline®: SCIENCE	Dialog	Foodline®: SCIENCE is a vital resource for keeping up-todate with published information on food science and technology worldwide. All aspects of the food and drink industry are covered, including ingredients and process technology, microbiology, packaging, food chemistry, biotechnology, food safety and nutrition. A key strength of the database is its currency, key journals being abstracted and available online within two weeks of delivery. More than 250 current periodicals are scanned extensively for FoodlineScience. In total, more than 1,800 records are added to FoodlineScience each month, including scientific journals, trade journals, books, book chapters, standards, technical reports and PCT, European, UK, US and Japanese patents. Produced by the Leatherhead Food Research since 1972.

Database	Provider	Justification
FSTA®	Dialog	FSTA® is produced by IFIS (UK) - core food information, an independent, not-for-profit organisation whose primary objective is to provide quality information products and services designed to meet the needs of all those working in the food sector. FSTA® is the largest and most respected collection of food science, food technology and food related human nutrition abstracts, providing content since 1969. It is compiled by a team of specialist scientists dedicated to producing a database of consistent high quality and timeliness. Continual development of coverage allows FSTA® to maintain its position as the marketleading food science database. There are more than 109,000 patent records including more than 11,000 Japanese patents. FSTA® covers journal articles (approximately 80%), patents, theses, standards, legislation, books, reviews and conference proceedings.
GeoArchive	Dialog	GeoArchive is a comprehensive database covering all types of information sources in geoscience, hydroscience, and environmental science since 1974 to current. The criteria for inclusion in GeoArchive are that the source should be publicly available and have relevant information content, even if the reference is to a small news item in a magazine. GeoArchive, produced by Geosystems, provides international coverage of over 5,000 serials, books from over a 2,000 publishers, geological maps, and doctoral dissertations. Published by Geosystems, U.K.
GEOBASE	Dialog	GEOBASE is a unique bibliographic database covering worldwide research literature since 1980 in physical and human geography, earth and environmental sciences, ecology, and related disciplines. In addition to providing comprehensive coverage of the core scientific and technical periodicals, Geobase has a unique coverage of non-English language and less readily available publications. Over 2,000 journals are fully covered with an additional 3,000 having partial coverage. Over 2,000 books, monographs, conference proceedings, and reports are also included.
MEDLINE (Medical Literature, Analysis, and Retrieval System Online)	STN Dialog	MEDLINE is produced by the U.S. National Library of Medicine (NLM) and is the U.S. National Library of Medicine's premier bibliographic database that contains more than 15 million references to journal articles in life sciences with a concentration on biomedicine. The broad coverage of the database includes basic biomedical research and the clinical sciences since 1950 including nursing, dentistry, veterinary medicine, pharmacy, allied health and pre-clinical sciences. MEDLINE also covers life sciences that are vital to biomedical practitioners, researchers and educators, including some aspects of biology, environmental science, marine biology, plant and animal science as well as biophysics and chemistry. Increased coverage of life sciences began in 2000. MEDLINE is indexed using NLM's controlled vocabulary, MeSH® (Medical Subject Headings). Approximately 400,000 records are added per year, of which more than 76% are in English.

Database	Provider	Justification
Meteorological and Geoastrophysical Abstracts	Dialog	Meteorological and Geoastrophysical Abstracts provides current citations in English for the most important meteorological and geoastrophysical research published in worldwide literature sources since 1966 to the present. Over 200 sources, including technical journals, monographs, proceedings, reviews and annual publications are scanned for relevant literature. Subject coverage includes meteorology (weather and climate), astrophysics, physical oceanography, hydrosphere and hydrology, environmental sciences, and glaciology. Content from American Meteorological Society, published by CSA.
Oceanic Abstracts	Dialog	*Oceanic Abstracts* covers the worldwide technical literature pertaining to the marine and brackish-water environment. The database focuses on marine biology and physical oceanography, fisheries, aquaculture, non-living resources, meteorology and geology, plus environmental, technological, and legislative topics. Major areas of coverage include biological oceanography, ecology, physical and chemical oceanography, marine geology, geophysics, geochemistry, marine pollution, nonliving marine resources, navigation and communications, maritime law, desalination, ships, shipping, and marine biology.
Pollution Abstracts	Dialog	Pollution Abstracts provides fast access to the environmental information necessary to ensure ongoing compliance and handle emergency situations more effectively. Pollution Abstracts combines information on scientific research and government policies in a single resource. Topics of growing concern are extensively covered from the standpoints of atmosphere, emissions, mathematical models, effects on people and animals, toxicology and health and environmental action in response to global pollution issues. To ensure comprehensive coverage, material from conference proceedings and hard-to-find documents has been summarised along with information from primary journals in the field. Published since 1966 by CSA (Cambridge Scientific Abstracts).
RTECS	STN	Registry of Toxic Effects of Chemical Substances contains factual toxicity data for commercially important substances from research and government reports. Coverage includes irritation data, federal standards and regulations, mutagenicity, tumoregenic effects, acute toxicity and multiple dose toxicity data, carcinogenicity reviews, NIOSH-recommended human exposure limits, reproductive effects, and information on activities by NIOSH, US EPA (Environmental Protection Agency), NTP (National Toxicology Program) and OSHA (Occupational Safety and Health Administration). Sources include journal articles, government reports and unpublished EPA test submissions (TSCATS). Molecular formulas, RTECS Numbers, CAS Registry Numbers, chemical names and toxic values are searchable.

Database	Provider	Justification
SCISEARCH	STN	Science Citation Index, one of the largest multidisciplinary scientific databases, is an international index to the literature covering virtually every subject area within the broad fields of science, technology and biomedicine. Records include references from over 5,600 scientific, technical and medical journals are contained in the database.
TOXCENTER	STN	Toxicology Center covers the pharmacological, biochemical, physiological, and toxicological effects of drugs and other chemicals. TOXCENTER is composed of the following subfiles: BIOSIS (1969 to date), CAplus (1907 to date), IPA (1970 to date), and MEDLINE (1953 to date). Sources include abstracts, books and book chapters, bulletins, conference proceedings, journal articles, letters, meetings, monographs, notes, papers, patents, presentations, research and project summaries, reviews, technical reports, theses, translations, unpublished material, web reprints. Records contain bibliographic data, abstracts, indexing terms, chemical names and CAS Registry Numbers
ToxFile	Dialog	ToxFile covers 1965 to the present of the toxicological, pharmacological, biochemical and physiological effects of drugs and other chemicals: adverse drug reactions, chemically induced diseases, carcinogenesis, mutagenesis, teratogenesis, environmental pollution, pesticides, waste disposal, radiation, and food contamination. ToxFile includes toxicology records derived from MEDLINE and also includes citations referred to as TOXNET records from the following organizations and data repositories: Aneuploidy File (ANEUPL), International Labor Office (CIS), Toxicology Research Projects (CRISP), Developmental and Reproductive Toxicology (DART), Environmental Mutagen Information Center File (EMIC), Epidemiology Information System (EPIDEM), Environmental Teratology Information Center File (ETICBACK), Federal Research in Progress (FEDRIP), Health Aspects of Pesticides Abstract Bulletin (HAPAB), Toxicological Aspects of Environmental Health (HEEP), Hazardous Materials Technical Center File (HMTC), National Institute for Occupational Safety and Health (NIOSH), Toxicology Document and Data Repository (NTIS), Pesticides Abstracts (PESTAB), Poisonous Plants Bibliography (PPBIB), Swedish National Chemicals Inspectorate (RISKLINE), and Toxic Substances Control Act Test Submissions (TSCATS).

Database	Provider	Justification
Toxicology Abstracts	Dialog	Toxicology Abstracts is the only comprehensive print resource for professionals in this field who must be aware of every new finding. Specifically focused to meet the needs of toxicologists, Toxicology Abstracts covers issues from social poisons and substance abuse to natural toxins, from legislation and recommended standards to environmental issues. Surveying the literature for toxicology studies of industrial and agricultural chemicals, household products, pharmaceuticals, and myriad other substances, each issue publishes information concerning the in vivo effects of toxic substances. Topics of current concern such as the effects of alcohol and smoking, drug abuse, hydrocarbon studies, nitrosamines, radiation and radioactive materials, and much more are extensively examined. Toxicity testing methodology and analytical procedures for toxic substances are also covered. Through many years of delivering crucial information on the tough, far-reaching issues of toxicology, Toxicology Abstracts has become the single most widely-used journal in this field.
TOXLINE	Dialog	Bibliographic citations to toxicological, pharmacological, biochemical and physiological effects of drugs and other chemicals. Coverage is international but contains primarily English language items; Updates are monthly, with about 9,300 new citations added each month; the file contains over 2.4 million records. The records are derived from about 16 secondary sources.
Water Resources Abstracts	Dialog	Water Resources Abstracts offers a comprehensive range of water-related topics summarising the world's technical and scientific literature on water-related topics covering the characteristics, conservation, control, pollution, treatment, use and management of water resources in the life and physical sciences, as well as the engineering and legal aspects of the conservation, control, use, and management of water. The database was originally produced by the U.S. Geological Survey starting in 1968 when it was generally known as Selected Water Resources Abstracts. Since 1994, Water Resources Abstracts has been produced by CSA (Cambridge Scientific Abstracts), which broadened the scope by including more material published outside the U.S.A. This database, which concentrates on water supply and water treatment, complements the Aquatic Sciences & Fisheries Abstracts database, ASFA, where there is greater coverage of the marine environment and biological material.

Search strategy and terms

The selection process resulted in two categories of publication:

- Studies considered to be non-relevant after initial (rapid) review.
- Potentially relevant articles requiring more detailed consideration of abstracts and / or full-text documents to assess relevance;

Table CA.B.8.5.1-3: Search terms used for the single concept search strategy

	STN Toxicology Database Cluster	**Dialog**
Date of the search:	15 February 2018	15 February 2018
Date span of the search:	2008 to 2018	2008 to 2018
Search strategies	Search Question: RN: 81777-95-9 RN: 81777-95-9 RN: 50-84-0 RN: 595-46-0 RN: 4835-90-9 NOT Document Type: conference NOT Document Type: patent	(“2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-3isoxazolidinone” OR “2-[(2,4-dichlorophenyl)methyl]-4,4dimethyl-1,2-oxazolidin-3-one” OR “F9600” OR “3hydroxypropanamide-F960” OR “N-(2,4-dichlorobenzyl)-3hydroxy-2,2-dimethylpropanamide” OR “2,4-dichlorobenzoic acid” OR “F9600 Dimethyl Malonamide” OR “3-((2,4dichlorobenzyl)amino)-2,2-dimethyl-3-oxopropanoic acid” OR “5-Hydroxy-F9600” OR “2-(2,4-dichlorobenzyl)-5-hydroxy-4,4-dimethylisoxazolidin-3-one” OR “5’-Hydroxy-F9600” OR “2-(2,4-dichloro-5-hydroxy benzyl)-4,4-dimethylisoxazolidin-3-one” OR “6’-Hydroxy-F9600” OR “2-(2,4-dichloro-6hydroxy benzyl)-4,4-dimethylisoxazolidin-3-one” OR “4Hydroxy-methyl-F9600” OR “2-(2,4-dichlorobenzyl)-4(hydroxymethyl)-4-methylisoxazolidin-3-one” OR “Dimethyl malonamide- F9600” OR “Dimethyl malonic acid” OR “F9600-isobutyramide” OR “N-(2,4dichlorobenzyl)isobutyramide” OR “Hydroxy-Isobutyramide” OR “N-(2,4-dichlorobenzyl)-2-hydroxy-2-methylpropanamide” OR “2,2-Dimethyl-3-hydroxy propionic acid” OR “5-OH, 5’OH Di-Hydroxy-F9600” OR “2-(2,4-dichloro-5-hydroxy benzyl)- 5-hydroxy-4,4-dimethylisoxazolidin-3-one” OR “4carboxy-F9600” OR “2-(2,4-dichlorobenzyl)-4-methyl-3oxoisoxazolidine-4-carboxylic acid” OR “2,4-Dichloroippuric acid” OR “N-(2,4-dichlorobenzoyl)glycine” OR “3-Isoxazolidinone, 2-[(2,4-dichlorophenyl)methyl]-4,4-dimethyl-” OR “Benzoic acid, 2,4-dichloro-” OR “Propanedioic acid, 2,2-dimethyl-” OR “2,2-Dimethylpropanedioic acid” OR “Malonic acid, dimethyl-” OR “Propanedioic acid, dimethyl-” OR “2,2-Propanedicarboxylic acid” OR “Dimethylmalonic acid” OR “Dimethylpropanedioic acid” OR “Propanoic acid, 3-hydroxy-2,2-dimethyl-, methyl ester” OR “Hydracrylic acid, 2,2-dimethyl-, methyl ester” OR “2,2-Dimethyl-3-hydroxypropanoic acid methyl ester” OR “2-Methoxycarbonyl-2-methylpropan-1-ol” OR “3-Hydroxy-2,2-dimethylpropanoic acid methyl ester” OR “3-Hydroxy-2,2-dimethylpropionic acid methyl ester” OR “Hydroxypivalic acid methyl ester” OR “Methyl .beta.-hydroxypivalate” OR “Methyl 2,2-dimethyl-3-hydroxypropanoate” OR “Methyl 2,2-dimethyl-3-hydroxypropionate” OR “Methyl 3-hydroxy-2,2-dimethylpropanoate” OR “Methyl 3-hydroxy-2,2-dimethylpropionate” OR “Methyl hydroxypivalate”) AND (at.exact("Article" OR "Book" OR "Book Chapter" OR "Government & Official Document" OR "Case Study" OR "Technical Report" OR "Report"))
Number of summary records retrieved after removing duplicates	13	24
Total number of summary records retrieved after removing duplicates	37	

Criteria of relevance and reliability
Assessment of studies for relevance was carried out by reference to their titles and if necessary abstracts. Those studies that were considered to meet the relevance criteria, following review of their abstracts were obtained. The full-text of these documents was assessed further to determine whether the information contained in the study could impact on the endpoints and risk assessment parameters related to the active substance. Reviews of the relevance and reliability of the articles brought up in the literature search were carried out by experts in the relevant technical disciplines.

The reliability assessment for any relevant studies was carried out according to Klimisch *et al.* (1997). The CA considers this reliability criteria acceptable.

Table CA.B.8.5.1-4: Reliability scores used to assess relevant environmental fate and behaviour studies

Reliability indicator	Description	Definition
1	Reliable without restriction	This includes studies or data from the literature or reports which were carried out or generated according to generally valid and/or internationally accepted testing guidelines (preferably performed to GLP) or in which the test parameters documented are based on a specific (national) testing guideline (preferably performed according to GLP) or in which all parameters described are closely related/comparable to a guideline method.
2	Reliable with restrictions	This includes studies or data from the literature, reports (mostly not performed according to GLP), in which the test parameters documented do not totally comply with the specific testing guideline, but are sufficient to accept the data or in which investigations are described which cannot be subsumed under a testing guideline, but which are nevertheless well documented and scientifically acceptable.
3	Not reliable	This includes studies or data from the literature/reports in which there are interferences between the measuring system and the test substance or in which organisms/test systems were used which are not relevant in relation to the exposure (e.g. unphysiologic pathways of application) or which were carried out or generated according to a method which is not acceptable, the documentation of which is not sufficient for an assessment and which is not convincing for an expert judgement.
4	Not assignable	This includes studies or data from the literature, which do not give sufficient experimental details and which are only listed in short abstracts or secondary literature (books, reviews, etc.).

As part of the determination of relevancy, the applicant stated that the following criteria are considered to be fundamental when considering the relevance of an open-literature study:

- Generally, degradation studies are considered relevant if they are carried out with the active substance only, and not with mixtures, since this may significantly influence the degradation behaviour. For laboratory soil degradation studies, the substrate used needs to be considered; in order to realistically reflect agro-ecosystems, it is crucial that the study is conducted with soil and that the soil is not contaminated and is representative of European agricultural soils. Temperature and moisture should be considered as reliability criteria. For field studies, relevance is based on (pedo-)climatic conditions being representative for European agriculture.
- The application of the test material needs to be considered because studies are not considered relevant if the application rates are significantly outside the representative use or the active substance is applied as a by-product (e.g. as a component of organic soil amendments).
- For adsorption studies, the substrate used needs to be considered.
- Relevance criteria for the aquatic compartment are analogous to those of soil-related data requirements.
- Monitoring studies, including those for air, may be considered relevant if the areas investigated are representative for Europe. Studies which are purely analytical, i.e. they determine levels of the active substance in certain environmental compartments, are not considered as relevant

The criteria considered for relevancy of studies relating to individual environmental fate data requirements were provided by the applicant and are detailed in Table CA.B.**8.5**.1-**5** below. The CA considers these criteria acceptable.

Table CA.B.8.5.1-5– Details of relevancy criteria specific to fate and behaviour

Data requirement (data point)	**Relevancy criteria considered**
Active substance	
Fate and behaviour in soil (KCA 7.1)	1. Well-defined test material applied as active substance or plant protection product (not as a by-product or ingredient of a soil amendment). 2. Substrate is a representative soil for agricultural uses with well-defined soil properties (e.g. pH, organic carbon content, microbial biomass etc). This is also relevant for field studies. 3. No previous contamination of the soil. 4. Active substance is not applied as a mixture with other active substances.
Fate and behaviour in water and sediment (KCA 7.2)	1. Well-defined test material applied as active substance or plant protection product. 2. Test samples used are samples from representative European aquatic resources with no contamination 3. Active substance is not applied as a mixture with other active substances.
Fate and behaviour in air (KCA 7.3)	1. Well-defined test material. 2. Areas investigated are relevant for Europe.

Findings

Articles of potential relevance to the regulatory data package for the active substance were investigated in further detail by examining the abstracts. Where articles were considered to meet the criteria for relevance, an assessment of the reliability of the study was carried out based on the approach described in Klimisch *et al.*, (1997). The applicant states this process did not identify any relevant studies with suitable reliability to inform on F9600 and its metabolites dealing with the side-effects on health, the environment and non-target species. The CA has checked the abstracts submitted by the applicant (see Table CA.B.**8.5**.1-**7** below) and agrees none are relevant for environmental fate and behaviour.

Table CA.B.8.5.1-6– Results of the study selection process for bixlozone (includes Environmental fate, ecotoxicology, residues, metabolism and toxicology)

Summary of the review	n
Total number of summary records retrieved after removing duplicates from all database searches	37
Number of summary records excluded after rapid assessment for relevance (by title/abstract)	28
Number of summary records of potential/unclear relevance assessed in further detail (by abstract/full-text)	9
Number of studies excluded from further consideration after detailed assessment for relevance (by abstract/full-text)	9
Number of studies not excluded for relevance after detailed assessment (i.e. relevant studies and studies of unclear relevance)	0
Number of relevant and reliable studies (Klimisch criteria 1-2) identified by the literature search and appraisal process	0

Table CA.B.8.5.1-7: Applicant's report of all potentially relevant studies and studies of unclear relevance after detailed assessments of relevance

Number	Author	Year	Title	Reference	Meet relevance criteria Y or N	Meet Reliability Criteria Score	Basis for relevance/ reliability decision (title, abstract or full article)	Comments	EU data point
1.	Adebusoye, S.A; Miletto, M.	2011	Characterization of multiple chlorobenzoic acid-degrading organisms from pristine and contaminated systems: mineralization of 2,4-dichlorobenzoic acid	Bioresource technology 102.3 (Feb 2011): 3041-8.	N	N/A	Abstract	A study of microbial characterisation using phylogenetic methodologies. No new information presented to inform data requirements, endpoints or risk assessments.	N/A
2.	Field, J.A.; Sierraalvarez, R.	2008	Microbial transformation of chlorinated benzoates	Reviews in Environmental Science and Biotechnology 7.3 (Sep 2008): 191-210.	N	N/A	Abstract	A review of biodegradation of chlorinated benzoates. No new information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A

Number	Author	Year	Title	Reference	Meet relevance criteria Y or N	Meet Reliability Criteria Score	Basis for relevance/ reliability decision (title, abstract or full article)	Comments	EU data point
3.	Hang Yong-tao, Zhang Li, Zuo Hai-ying, Gui Jian-ye, Li Xiao-ya, Li Gui-xiang	2010	Detection of 17 Acid Herbicide Residues in Groundwater by Gas Chromatography-Mass Spectrometry with Diazomethane Derivation	Rock and Mineral Analysis,2010,29(4):345-349	N	N/A	Abstract	Method development for detection of herbicide residues in groundwater. No new information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A
4.	Křesinová Z, *et al*.	2014	Sensitive GC/MS determination of 15 isomers of chlorobenzoic acids in accelerated solvent extracts of soils historically contaminated with PCBs and validation of the entire method	International Journal of Environmental Analytical Chemistry, 94:8, 822-836, DOI: 10.1080/03067319.2014.900677	N	N/A	Abstract	Method development for detection of chlorobenzoic acid isomers in soil. No new information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A

Number	Author	Year	Title	Reference	Meet relevance criteria Y or N	Meet Reliability Criteria Score	Basis for relevance/ reliability decision (title, abstract or full article)	Comments	EU data point
5.	Křesinová Z, Muzikář M, Olšovská J, Cajthaml T.	2011	Determination of 15 isomers of chlorobenzoic acid in soil samples using accelerated sample extraction followed by liquid chromatography	Talanta. 2011 May 30;84(4):1141-7. doi: 10.1016/j.talanta.2011.03.013. Epub 2011 Mar 16	N	N/A	Abstract	Method development for detection of chlorobenzoic acid isomers in soil. No new information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A
6.	Muzikář M. *et al.*	2011	Biodegradation of chlorobenzoic acids by ligninolytic fungi	Journal of Hazardous Materials, Volume 196, 30 November 2011, Pages 386-394	N	N/A	Abstract	Study of fungal biodegradation of chlorobenzoic acid. No suitable information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A
7.	Oliveira, BR; Penetra, A; Cardoso, VV; Benoliel, MJ; Barreto Crespo, MT; Samson, RA; Pereira, VJ	2015	Biodegradation of pesticides using fungi species found in the aquatic environment	Environmental Science and Pollution Research International 22.15 (Aug 2015): 11781-11791	N	N/A	Abstract	Study of pesticide bioremediation potential of fungal species. No new information presented to inform data requirements, endpoints or risk assessments.	N/A

Number	Author	Year	Title	Reference	Meet relevance criteria Y or N	Meet Reliability Criteria Score	Basis for relevance/ reliability decision (title, abstract or full article)	Comments	EU data point
8.	Praveen S.; Kaliwal B.	2016	Biodegradation of the fungicide propiconazole by Pseudomonas aeruginosa PS-4 strain isolated from a paddy soil.	Annals of Microbiology 66.4 (2016): 1355-1365	N	N/A	Abstract	Study of fungal biodegradation of propiconazole. Not relevant to bixlozone or its metabolites.	N/A
9.	Svobodová K, Placková M, Novotná V, Cajthaml T.	2009	Estrogenic and androgenic activity of PCBs, their chlorinated metabolites and other endocrine disruptors estimated with two in vitro yeast assays.	Sci Total Environ. 2009 Nov 1;407(22):5921-5. doi: 10.1016/j.scitotenv.2009.08.011. Epub 2009 Aug 28.	N	N/A	Abstract	Evaluation of assays for screening environmental pollutants for endocrine activity. Chlorobenzoic acids are mentioned though only in the context of having no/limited activity. No new information presented to inform data requirements, endpoints or risk assessments. The article is therefore not relevant.	N/A

CA.B.8.5.2. **References relied on**

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KCA 7.1.1.1/01, KCA 7.1.2.1.1/01	Simmonds, R	2015a (Amended 2018)	[^{14}C]-bixlozone: Route and Rate of Aerobic Degradation in Seven Soils at 20°C Battelle UK Ltd., Springfield, UK, Study No.: KW/14/001 FMC Tracking No.: 2013EFT-ISX1021 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.1.2/01, KCA 7.1.2.1.3/01	Simmonds, R	2015b (Amended 2018)	[^{14}C]-bixlozone: Route and Rate of Anaerobic Degradation in Four Soils at 20°C Battelle UK Ltd., Springfield, UK, Study No.: KW/14/002, FMC Tracking No.: 2013EFT-ISX1022 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.1.3/01	Tuffnail, W	2016	Bixlozone: Phototransformation of [^{14}C]-bixlozone on Soil Surfaces under Laboratory Conditions Quotient Bioresearch Ltd., UK, Study No.: FCC/02 FMC Tracking No.: 2015EFT-ISX2045 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.1.1/02	Kong, L	2017b	Bixlozone: Normalisation of Laboratory DT50 for Temperature (20°C) and Moisture (pF 2.0) FMC Tracking No.: 2017WHP-ISX3143 Non-GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.1.2/01	Göcer, M	2016a	Bixlozone-3-OH-Propanamide Aerobic Degradation in Three Soils at 20°C in the Dark Eurofins Agroscience Services EcoChem GmbH, Germany, Study No.: S16-01058 FMC Tracking No.: 2016EFT-ISX2465 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KCA 7.1.2.1.2/02	Göcer, M	2016b	2,4-Dichlorobenzoic Acid Aerobic Degradation in Three Soils at 20°C in the Dark Eurofins Agroscience Services EcoChem GmbH, Germany, Study No.: S16-01059 FMC Tracking No.: 2016EFT-ISX2468 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.2.1/01	Gemrot, F	2018a	Soil dissipation study after one application of F9600-4 SC or F9600-21 CS in Southern Europe (Southern France and Italy) – 2015 and 2017 SGS AGRI MIN, Brugières, France, Study No.: 15SGS088, FMC Tracking No.: 2015EFT-ISX1947 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.2.1/02	Gemrot, F	2018b	Soil dissipation study after one application of F9600-4 SC or F900-21 CS in Northern Europe (Germany) and Southern Europe (Southern France) – 2015 and 2016 SGS AGRI MIN, Brugières, France, Study No.: 15SGS111, FMC Tracking No.: 2015EFT-ISX2156 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.2.1/03	Gezahegne, W	2018	Field soil dissipation with bare soil application of F9600-4 SC at two sites in North EU (Germany and UK) in 2016-2017 Eurofins Agroscience Services EcoChem GmbH, Germany, Study No.: S16-02441, FMC Tracking No.: 2016EFT-ISX2539 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KCA 7.1.2.2.1/04	Rawle, N	2017	Storage stability study of bixlozone and its metabolites (2,4-dichlorobenzoic acid and bixlozone-3-OH-propanamide) in soil samples stored under frozen conditions. CEM Analytical Services Ltd. (CEMAS), UK, Study No.: CEMS-7213, FMC Tracking No.: 2015RES-ISX2038 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.2.2.1/05	Montesano, V.; Jarvis, T.	2018	Normalisation of the field dissipation data for F9600-4 SC or F9600-21 CS from four locations in Europe and the determination of the normalised field DT_{50} values. Exponent International Ltd, UK, Report No.: 1508442.UK0-9677 FMC Tracking No.: 2018EFT-ISX4194 Non-GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.3.1.1/01	Simmonds, M.; Hawkins, T	2016	[^{14}C]-bixlozone: Adsorption to and Desorption from Eight Soils (Amended Final Report) Battelle UK Ltd., Springfield, UK, Study No.: KW/14/005 FMC Tracking No.: 2013EFT-ISX1025 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.1.3.1.2/01	Gahm, F.; Kirchherr, M.	2017 (Amended 2018)	bixlozone-3-OH Propanamide Adsorption/Desorption Behaviour in Four Soils. Eurofins Agroscience Services EcoChem GmbH, Germany, Study No.: S16-01056 FMC Tracking No.: 2016EFT-ISX2464 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KCA 7.2.1.1/01	Roohi, A.; Cooper, T. J.	2015	[^{14}C]-bixlozone: Aqueous Hydrolysis as a Function of pH Battelle UK Ltd., Springfield, UK, Study No.: KW/14/004 FMC Tracking No.: 2013EFT-ISX1024 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.2.1.2/01	O'Connell, C	2015	[^{14}C]-bixlozone: Phototransformation of Chemicals in Water - Direct Photolysis Battelle UK Ltd., Springfield, UK, Study No.: KW/14/003 FMC Tracking No.: 2013EFT-ISX1023 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.2.2.1/01	Shannon, M	2017	Assessment of Ready Biodegradability by Measurement of CO_2 Evolution Smithers Viscient (ESG) Ltd, Harrogate, UK, Study No.: 3201875 FMC Tracking No.: 2017EFT-ISX3306 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.2.2.2/01	Simmonds, R	2018	[^{14}C] bixlozone: Aerobic Mineralisation in Surface Water (OECD 309) Battelle, UK Ltd., Springfield, UK, Study No.: KW/15/007 FMC Tracking No.: 2015EFT-ISX2197 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC
KCA 7.2.2.3/01	Cooper, J.; Challis, P.	2018	[^{14}C]-bixlozone: Aerobic Aquatic Metabolism in Two Water/Sediment Systems at 20 ± 2 °C Battelle, UK Ltd., Springfield, UK, Study No.: KW/15/007 FMC Tracking No.: 2015EFT-ISX2195 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KCA 7.3.2/01	Staffa, C.	2016	Large outdoor wind tunnel study to evaluate the short range transport and deposition of volatilised bixlozone (300 g a.s/ha) applied as bixlozone-4 formulation including the assessment of bleaching effects on a sensitive test plant (*Stellaria media*) as a function of distance from the treated area (0-20 m) RLP AgroScience GmbH, Neustadt, Germany, Study No.: AS442 FMC Tracking No.: 2016EFT-ISX2732 GLP, Unpublished	N	Y	Study to support new active approval in GB	FMC