

REGISTRATION REPORT

Part B

Section 8

Environmental Fate

Detailed summary of the risk assessment

Product name(s): **INTUITY PLUS**

(Mandestrobin 40 SC)

Chemical active substance:

Mandestrobin, 400 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

(authorization)

Applicant: XXXX

Submission date: February 2024

MS Finalisation date: August 2025

Version history

When	What
February 2024	Article 33 submission – Initial Applicant’s version
May 2024	- Update of the cover page with the product trade name ‘Intuity Plus’. Mandestrobin 40 SC is the internal unique name. The internal name Mandestrobin 40 SC is the one used across the dRR content. - Update of Appendix 1: studies source and owner updated
October 2024	Response to zRMS request to add PECsw step 4 calculations
May 2025	Response to zRMS request to add PECgw calculations using spring cereals as a surrogate crop for spring oilseed rape
August 2025	Final dRR by zRMS

Table of Contents

8	Fate and behaviour in the environment (KCP 9).....	4
8.1	Critical GAP and overall conclusions.....	5
8.2	Metabolites considered in the assessment.....	7
8.3	Rate of degradation in soil (KCP 9.1.1).....	8
8.3.1	Aerobic degradation in soil (KCP 9.1.1.1)	8
8.3.2	Anaerobic degradation in soil (KCP 9.1.1.1).....	11
8.4	Field studies (KCP 9.1.1.2).....	11
8.4.1	Soil dissipation testing on a range of representative soils (KCP 9.1.1.2.1). 11	
8.4.2	Soil accumulation testing (KCP 9.1.1.2.2)	11
8.5	Mobility in soil (KCP 9.1.2)	11
8.5.1	Column leaching (KCP 9.1.2.1).....	13
8.5.2	Lysimeter studies (KCP 9.1.2.2).....	13
8.5.3	Field leaching studies (KCP 9.1.2.3)	13
8.6	Degradation in the water/sediment systems (KCP 9.2, KCP 9.2.1, KCP 9.2.2, KCP 9.2.3)	13
8.7	Predicted Environmental Concentrations in soil (PEC _{soil}) (KCP 9.1.3)	14
8.7.1	Justification for new endpoints	15
8.7.2	Active substance(s) and relevant metabolite(s)	15
8.7.2.1	PEC _{soil} of Mandestrobin 40SC formulation	15
8.8	Predicted Environmental Concentrations in groundwater (PEC _{gw}) (KCP 9.2.4)	15
8.8.1	Justification for new endpoints	16
8.8.2	Active substance(s) and relevant metabolite(s) (KCP 9.2.4.1).....	17
8.9	Predicted Environmental Concentrations in surface water (PEC _{sw}) (KCP 9.2.5)	24
8.9.1	Justification for new endpoints	24
8.9.2	Active substance(s), relevant metabolite(s) and the formulation (KCP 9.2.5)	25
8.10	Fate and behaviour in air (KCP 9.3, KCP 9.3.1)	30
Appendix 1	Lists of data considered in support of the evaluation.....	31
Appendix 2	Detailed evaluation of the new Annex II studies (evaluated in previous Registration Report)	35
A 2.1	Study 1: Gilbert 2016.....	35
A 2.2	Study 2: Kodaka, 2016.....	44
A 2.3	Study 3: Lamond (2017)	49
A 2.4	Study 4: Kang, S. (2017).....	55
A 2.5	Studies 5 and 6 – Anaerobic degradation	63

8 Fate and behaviour in the environment (KCP 9)

The product Mandestrobin 40SC is a fungicide containing 400 g/L of mandestrobin. It is intended to be applied on winter and spring oilseed rape for the control of *Sclerotinia sclerotiorum* according to the GAP presented in the following table.

8.1 Critical GAP and overall conclusions

Table 8.1-1: Critical use pattern of the formulated product

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, Fn, Fpn G, Gn, Gpn or I **	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	Application				Application rate			PHI (days)	Remarks: e.g. g saf- ener/ synergist per ha	Conclusion
					Method / Kind	Timing / Growth stage of crop & season	Max. number a) per use b) per crop/ season	Min. interval between applications (days)	L product/ha a) max. rate per appl. b) max. total rate per crop/season	g as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha min/max			
Zonal uses (field or outdoor uses, certain types of protected crops)														
1	Central Zone (PL, AT, HU, RO, DE, NL, CZ, SK, SI)	Winter and Spring Oilseed rape	F	<i>Sclerotinia sclerotiorum</i>	Foliar spray	BBCH 60-69	a) 1 b) 1	-	a) 0.5 L/ha b) 0.5 L/ha	a) 200 g/ha b) 200 g/ha	100-300	-	the PHI is covered by the time remaining between application and harvest	

** F: professional field use, Fn: non-professional field use, Fpn: professional and non-professional field use, G: professional greenhouse use, Gn: non-professional greenhouse use, Gpn: professional and non-professional greenhouse use, I: indoor application

Explanation for column 15 “Conclusion”

A	Safe use
R	Further refinement and/or risk mitigation measures required
C	To be confirmed by cMS
N	No safe use

Comments of zRMS:	In acidic soils (pH ≤ 6.7) the formulation could be applied every other year in spring and winter oilseed rape. In neutral and alkaline soils (pH ≥ 7.2) the formulation could be applied every year in spring and winter oilseed rape.
-------------------	--

Table 8.1-2: Assessed (critical) uses during approval of mandestrobin concerning the Section Environmental Fate

1	2	3	4	5	6	7	8	9	10	11	12	13	14
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, Fn, Fpn G, Gn, Gpn or I **	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	Application				Application rate			PHI (days)	Remarks: e.g. g safener/ synergist per ha
					Method / Kind	Timing / Growth stage of crop & season	Max. number a) per use b) per crop/ season	Min. interval between applications (days)	L product/ha a) max. rate per appl. b) max. total rate per crop/season	g as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha min/max		
1	N & S EU	Winter oilseed rape	F	<i>Sclerotinia</i>	High volume spray, broadcast	63-67	1	--	a) 0.5 L/ha b) 0.5 L/ha	a) 200 b) 200	100/300	-†	

†pre-harvest interval is defined by the application conditions at the BBCH growth stages stated

** F: professional field use, Fn: non-professional field use, Fpn: professional and non-professional field use, G: professional greenhouse use, Gn: non-professional greenhouse use, Gpn: professional and non-professional greenhouse use, I: indoor application

8.2 Metabolites considered in the assessment

Table 8.2-1: Metabolites of mandestrobin potentially relevant for exposure assessment

Metabolite	Molar mass	Chemical structure	Maximum observed occurrence in compartments	Exposure assessment required due to
2-COOH-S-2200 (<i>RS</i>)-2-{2-[1-methoxy-1-(<i>N</i> -methylcarbamoyl)methyl]benzyloxy}-4-methylbenzoic acid	343.38		Soil/Water/Sediment: 8.7/--/--	PECsoil, PECgw, PECsw/PECsed
5-COOH-S-2200 (<i>RS</i>)-3-{2-[1-methoxy-1-(<i>N</i> -methylcarbamoyl)methyl]benzyloxy}-4-methylbenzoic acid	343.38		Soil/Water/Sediment: 18.0/3.6/--	PECsoil, PECgw, PECsw/PECsed
S-2200-OR (<i>RS</i>)-2-[2-(2-hydroxy-3,6-dimethylbenzyl)phenyl]-2-methoxy- <i>N</i> -methylacetamide	313.39		Soil/Water/Sediment: --/20.7/--	PEC _{sw/sed}
S-2200-ORC (<i>RS</i>)- <i>N</i> ,1,4-trimethyl-6,11-dihydrodibenzo[<i>b,e</i>]oxepine-6-carboxamide	281.35		Soil/Water/Sediment: --/13.7/--	PEC _{sw/sed}
DX-CA-S-2200 (<i>RS</i>)-2-(<i>N</i> -methylcarbamoyl-methoxymethyl) benzoic acid	223.2		Soil/Water/Sediment 8.3/--/--	PECsoil, PECgw, PECsw/PECsed

8.3 Rate of degradation in soil (KCP 9.1.1)

Studies on degradation in soil with the formulation were not performed, since it is possible to extrapolate from data obtained with the active substance.

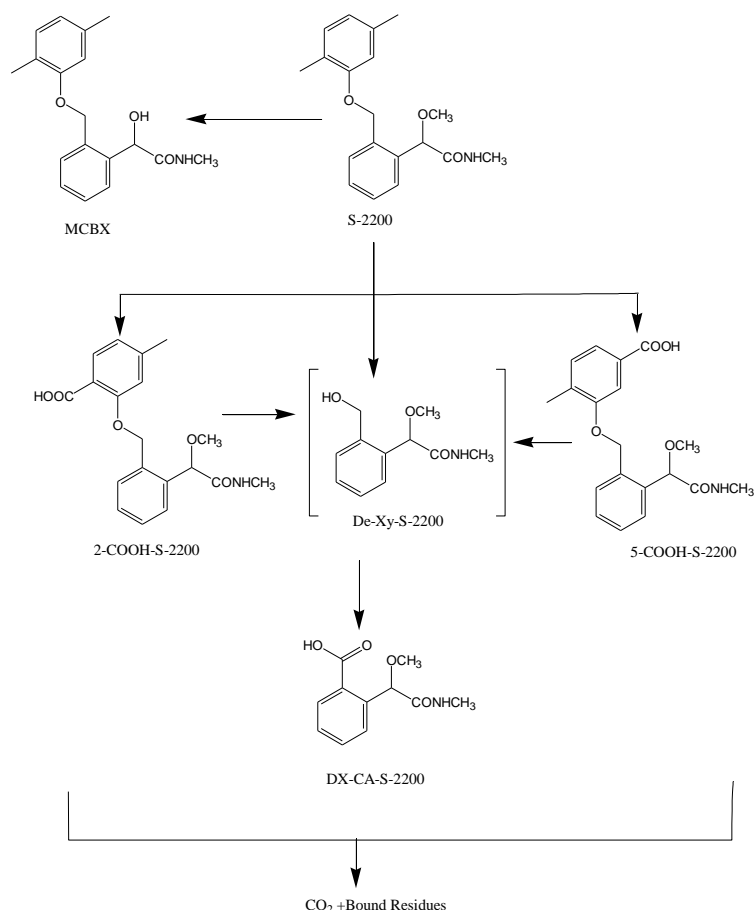


Figure 8.3-1: Proposed degradation route of mandestrobin in soil

3 major or minor non-transient metabolites are observed in laboratory aerobic degradation studies: 5-COOH-S-2200 observed up to 18.0% AR, 2-COOH-S-2200 observed up to 8.7% AR and DX-CA-S-2200, observed in a new study Gilbert (2016, see appendix 3) up to 8.3% AR (acidic conditions only).

8.3.1 Aerobic degradation in soil (KCP 9.1.1.1)

A pH dependency has been observed at EU level for degradation of mandestrobin. The initial set of data allowed to derive a modelling endpoint for soils with pH (CaCl₂) ≤ 5.9 and ≥ 7.2, with a gap in-between. An additional study (Gilbert, 2016, see appendix 2) was undertaken and accepted in previous registration Reports (RR) in Central zone (AGES, 2016) and Southern zone (ANSES, 2021). This study provides four additional DT₅₀ for mandestrobin and its metabolite 5-COOH-S-2200 (with additional formation fraction for this metabolite).

Table 8.3-1: Summary of aerobic degradation rates for mandestrobin - laboratory studies

Mandestrobin, Laboratory studies, aerobic conditions								
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	DT ₅₀ (d) 20°C pF2/10kPa	χ ² error (%)	Method of calculation	Evaluated on EU level y/n Reference
Speyer 2.2	2.1	5.5	20°C/pF2	335/- k ₁ :0.1116 k ₂ :0.00167 g:0.1247	415.1 ^a	2.4	DFOP	y/EFSA (2015)
Chelmorton	3.4	5.9	20°C/pF2	102.8/530.3 k ₁ :0.1661 k ₂ :3.765x10 ⁻³ g:0.2636	184.1 ^a	2.4	DFOP	y/EFSA (2015)
Speyer 5M	1.3	7.2	20°C/pF2	76.1/252.8	76.1	2.4	SFO	y/EFSA (2015)
Aschard	1.3	7.4	20°C/pF2	62.5/207.5	62.5	3.3	SFO	y/EFSA (2015)
SK920191	3.8	7.6	20°C/pF2	53.1/176.4	53.1	6.7	SFO	y/EFSA (2015)
Monteil	1.4	7.7	20°C/pF2	49.7/165.2	49.7	3.2	SFO	y/EFSA (2015)
Geometric mean, acidic soils (pH≤5.9) n=2					276.4			
Geometric mean, alkaline soils (pH ≥7.2) n=4					59.5			
Newhaven	3.7	5.5	20°C/pF2	103/512	176.2 ^a	1.38	DFOP	y ^b / Gilbert (2016)
Kenslow	3.2	5.3	20°C/pF2	187/822	273.7 ^a	0.53	DFOP	y ^b / Gilbert (2016)
Barrow	3.5	5.8	20°C/pF2	135/672	231.4 ^a	1.66	DFOP	y ^b / Gilbert (2016)
Quilen	3	6.7	20°C/pF2	112/528	178.9 ^a	2.06	DFOP	y ^b / Gilbert (2016)
Geometric mean, soils with pH≤ 6.7, n=6					231.2			
Geometric mean, soils with pH ≥7.2, n=4					59.5			

^a slow rate k value of DFOP kinetics

^b AGES, 2016; ANSES, 2021.

Table 8.3-2: Summary of aerobic degradation rates for 5-COOH-S-2200 - laboratory studies

5-COOH-S-2200, Laboratory studies, aerobic conditions									
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	f.f	DT ₅₀ (d) 20°C pF2/10kPa	χ ² error (%)	Method of calculation	Evaluated on EU level y/n Reference
Speyer 5M	1.3	7.2	20°C/pF2	33.6/111.5	0.5349	33.6	8.6	SFO-SFO	y/EFSA (2015)
	1.3	7.2	20°C/pF2	41.0/136.1	-	41.0	2.37	SFO	y/EFSA (2015)
Speyer 2.2	2.1	5.5	20°C/pF2	52.3/173.9	0.4092	52.3	3.0	DFOP-SFO	y/EFSA (2015)
SK920191	3.8	7.6	20°C/pF2	20.4/67.9	0.6965	20.4	6.1	SFO-SFO	y/EFSA (2015)
	3.8	7.6	20°C/pF2	30.3/100.5	-	30.3	7.95	SFO	y/EFSA (2015)
Chelmorton (SK104691)	3.4	5.9	20°C/pF2	136.8/454.4	0.2052	136.8	2.1	DFOP-SFO	y/EFSA (2015)
	3.4	5.6	20°C/pF2	21.9/72.7	-	21.9	4.5	SFO	y/EFSA (2015)
Aschard	1.3	7.4	20°C/pF2	38.3/127.1	0.6924	38.3	1.4	SFO-SFO	y/EFSA (2015)
Monteil	1.4	7.7	20°C/pF2	24.9/82.7	0.4747	24.9	2.9	SFO-SFO	y/EFSA (2015)
Geometric mean (n=9)					-	36.9			
Arithmetic mean (n=6)					0.502	-			
Newhaven	3.7	5.5	20°C/pF2	29.5/98.1	0.3585	29.5	7.9	DFOP-SFO	y ^a / Gilbert (2016)
Kenslow	3.2	5.3	20°C/pF2	60.4/200.5	0.3644	60.4	2.2	DFOP-SFO	y ^a / Gilbert (2016)
Barrow	3.5	5.8	20°C/pF2	59.0/195.9	0.3272	59.0	1.6	DFOP-SFO	y ^a / Gilbert (2016)

Quilen	3	6.7	20°C/pF2	87.5/290.8	0.1804	87.5	1.9	DFOP-SFO	y ^a / Gilbert (2016)
Geometric mean (n=14)					-	41.75			
Arithmetic mean (n=10)					0.424	-			

^a AGES, 2016; ANSES, 2021.

Table 8.3-3: Summary of aerobic degradation rates for 2-COOH-S-2200 - laboratory studies

2-COOH-S-2200, Laboratory studies, aerobic conditions									
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	f.f	DT ₅₀ (d) 20°C pF2/10kPa	χ ² error (%)	Method of calculation	Evaluated on EU level y/n Reference
Speyer 5M	1.3	7.2	20°C/pF2	20.6/68.3	0.2848	20.6	4.6	SFO-SFO	y/EFSA (2015)
	1.3	7.2	20°C/pF2	25.9/86.1	-	25.9	4.01	SFO	y/EFSA (2015)
Speyer 2.2	2.1	5.5	20°C/pF2	>1000/>1000	-	>1000*	1.8	DFOP-SFO	y/EFSA (2015)
SK920191	3.8	7.6	20°C/pF2	15.3/50.9	0.3035	15.3	9.9	SFO-SFO	y/EFSA (2015)
	3.8	7.6	20°C/pF2	20.4/67.8	-	20.4	2.03	SFO	y/EFSA (2015)
Chelmorton (SK104691)	3.4	5.9	20°C/pF2	177.3/588.8	0.1306	177.3	11.2	DFOP-SFO	y/EFSA (2015)
	3.4	5.6	20°C/pF2	18.1/60.0	-	18.1	3.36	SFO	y/EFSA (2015)
Aschard	1.3	7.4	20°C/pF2	29.5/97.9	0.3076	29.5	4.3	SFO-SFO	y/EFSA (2015)
Monteil	1.4	7.7	20°C/pF2	29.0/96.5	0.1664	29.0	3.0	SFO-SFO	y/EFSA (2015)
Geometric mean (n=8)					-	28.7			
Arithmetic mean (n=5)					0.2386	-			

* Value not used for determination of geometric mean due to low occurrence of the metabolite in this soil (≤3 % AR)

A minor non-transient metabolite, DX-CA-S-2200, was observed in acidic soil in the new aerobic study Gilbert (2016). A degradation study on De-Xy-S-2200, in which DX-CA-S-2200 is formed as a metabolite, (Lamond; 2017; See Appendix 2) was previously evaluated and accepted in the RR for a different mandestrobin product (ANSES, 2021). Acceptable DT₅₀ values from this study are summarized below. Due to the absence of any specific data, a formation fraction of 1 (from mandestrobin) is used for simulation modelling. A previous preliminary degradation study (Kodaka *et al*, 2016 See Appendix 2) had also been evaluated but considered unacceptable – even though the worst case DT₅₀ was subsequently used for soil PEC calculations.

Table 8.3-4: Summary of aerobic degradation rates for reliable DT₅₀ for DX-CA-S-2200 – Laboratory studies

DX-CA-S-2200, Laboratory studies, aerobic conditions - De-Xy-S-2200 applied study									
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	f.f	DT ₅₀ (d) 20°C pF2/10kPa	χ ² error (%)	Method of calculation	Evaluated on EU level y/n Reference
Calke	2.4	5.2	20°C/pF2	5.2 / 17.3	NR	5.2	7.3	SFO-SFO	y ^a / Lamond 2017
South Witham	2.8	7.5	20°C/pF2	3.2 / 10.5	NR	3.2	6.5	SFO-SFO	
RefeSol 02A	1.0	6.6	20°C/pF2	8.1 / 21.9	NR	8.1	2.7	SFO-SFO	
Geomean (n=3)						5.1			

^a ANSES, 2021.

NR: not relied on

8.3.2 Anaerobic degradation in soil (KCP 9.1.1.1)

Studies on anaerobic degradation in soil with the formulation were not performed, since it is possible to extrapolate from data obtained with the active substance.

An anaerobic soil degradation study for the active substance was set as a data requirement (EFSA Journal 2015; 13(5):4100). Studies of Lewis & Cooper (2015a, b) with each isomer are summarized in Appendix 2. No significant degradation of the substance was observed under anaerobic conditions.

8.4 Field studies (KCP 9.1.1.2)

8.4.1 Soil dissipation testing on a range of representative soils (KCP 9.1.1.2.1)

Table 8.4-1: Summary of field dissipation rates of mandestrobin (EFSA Journal 2015; 13(5):4100)

Mandestrobin, field studies										
Soil type (bare ground).	Location (country or USA state).	OC%	pH	Depth (cm)	DT ₅₀ (d) actual	DT ₉₀ (d) actual	χ ² error (%)	DT ₅₀ (d) Norm.	Method of calculation	Evaluated on EU level y/n Reference
Clay	France (South)	1.4	7.6	30	2.29	47.92	11.1	-	DFOP	y/EFSA (2015)
Loam	Spain	1.3	7.8	30	2.82	55.09	8.7	-	DFOP	y/EFSA (2015)
Silt Loam	Austria	2.5	7.7	30	8.28	81.63	5.7	-	DFOP	y/EFSA (2015)
Loamy sand	Germany	1.7	5.4	30	4.53	225.93	29.1	-	DFOP	y/EFSA (2015)

8.4.2 Soil accumulation testing (KCP 9.1.1.2.2)

No data required: DT_{90field} is <1 year.

8.5 Mobility in soil (KCP 9.1.2)

Studies on mobility in soil with the formulation were not performed, since it is possible to extrapolate from data obtained with the active substance. Since the EFSA conclusion reports arithmetic mean Koc values for mandestrobin, 5-COOH-S-2200 and 2-COOH-S-2200, these have been used for risk assessment, in line with Central Zone guidance (2018).

Table 8.5-1: Summary of soil adsorption for mandestrobin

Mandestrobin								
Soil Type	OC %	Soil pH (CaCl ₂)	Kd (mL/g)	Koc (mL/g)	Kf (mL/g)	Kfoc (mL/g)	1/n	Evaluated on EU level y/n Reference
SK961089 – clay loam	5.0	7.4	-	-	15	296	0.9163	y/EFSA (2015)
SK 104691 (Chelmorton) – silt loam	2.5	6.1	-	-	7	287	0.9315	y/EFSA (2015)
SK179618 – loam/silt loam	3.9	5.0	-	-	18	466	0.9615	y/EFSA (2015)
SK566696 – loamy sand	1.3	4.0	-	-	10	797	0.8981	y/EFSA (2015)

Saitama – sandy loam	3.1	5.6	-	-	12	397	0.8888	y/EFSA (2015)
Arithmetic mean						449	0.9192	
Geometric mean						416	-	
pH dependence, Yes or No				No				

Table 8.5-2: Summary of soil adsorption for 5-COOH-S-2200

5-COOH-S-2200								
Soil Type	OC %	Soil pH (CaCl ₂)	K _d (mL/g)	K _{oc} (mL/g)	K _f (mL/g)	K _{foc} (mL/g)	1/n	Evaluated on EU level y/n Reference
Acidic soil								
SK566696 – loamy sand	1.3	4.0			8.90	684	0.9259	y/EFSA (2015)
Neutral/alkaline soils								
SK 104691 (Chelmorton) – silt loam	2.5	6.0	-	-	1.35	54	0.8822	y/EFSA (2015)
Landlook 250 – sandy clay	3.3	6.1	-	-	1.80	55	0.8861	
Landlook 308 – sandy clay loam	1.4	6.4	-	-	1.04	74	0.8955	
Landlook 301 – clay	3.0	6.5	-	-	1.91	64	0.9081	
SK961089 – clay loam	5.0	7.4	-	-	1.44	29	0.8740	
Arithmetic mean						55	0.8892	
Geometric mean						52.7	-	
pH dependence, Yes or No				Yes				

Table 8.5-3: Summary of soil adsorption for 2-COOH-S-2200

2-COOH-S-2200								
Soil Type	OC %	Soil pH (CaCl ₂)	K _d (mL/g)	K _{oc} (mL/g)	K _f (mL/g)	K _{foc} (mL/g)	1/n	Evaluated on EU level y/n Reference
Acidic soil								
SK566696 – loamy sand	1.3	4.0			2.94	226	0.9225	y/EFSA (2015)
Neutral/alkaline soils								
SK 104691 (Chelmorton) – silt loam	2.5	6.0	-	-	0.27	11	0.9158	y/EFSA (2015)
Landlook 250 – sandy clay loam	3.3	6.1	-	-	0.46	14	0.8556	
Landlook 308 – sandy clay loam	1.4	6.4	-	-	0.34	24	0.8861	
Landlook 301 – clay	3.0	6.5	-	-	0.58	19	0.8603	
SK961089 – clay loam	5.0	7.4	-	-	0.28	6	0.9031	
Arithmetic mean						15	0.8841	
Geometric mean						13.3		
pH dependence, Yes or No				Yes				

For metabolite DX-CA-S-2200, a new adsorption study (Kang, 2017, see Appendix 2) was previously evaluated and accepted in the RR for a different mandestrobin product (ANSES, 2021).

Table 8.5-4: Adsorption/Desorption parameters for ¹⁴C-DX-CA-S-2200 in five soils

Soil	OC (%)	pH (CaCl ₂)	K ^F _{ads,OC} (mL/g)	1/n	Evaluated on EU level y/n Reference
Hareby	2.3	7.4	3.89	0.739	y ^a / Kang 2017
Clipstone	1.1	6.1	2.48	0.918	
Brierlow	4.0	5.4	6.09	0.954	
Newhaven	3.6	5.4	7.20	0.972	
Kenslow	3.3	5.0	10.0	0.951	

Arithmetic values			-	0.907	
Geometric mean			5.32	-	

^a ANSES, 2021.

8.5.1 Column leaching (KCP 9.1.2.1)

No data, none required.

8.5.2 Lysimeter studies (KCP 9.1.2.2)

Assessed for the approval of the active substance, please refer to the EFSA journal 2015. No additional studies have been performed.

8.5.3 Field leaching studies (KCP 9.1.2.3)

No data, none required.

8.6 Degradation in the water/sediment systems (KCP 9.2, KCP 9.2.1, KCP 9.2.2, KCP 9.2.3)

Studies on degradation in water/sediment systems with the formulation were not performed, since it is possible to extrapolate from data obtained with the active substance.

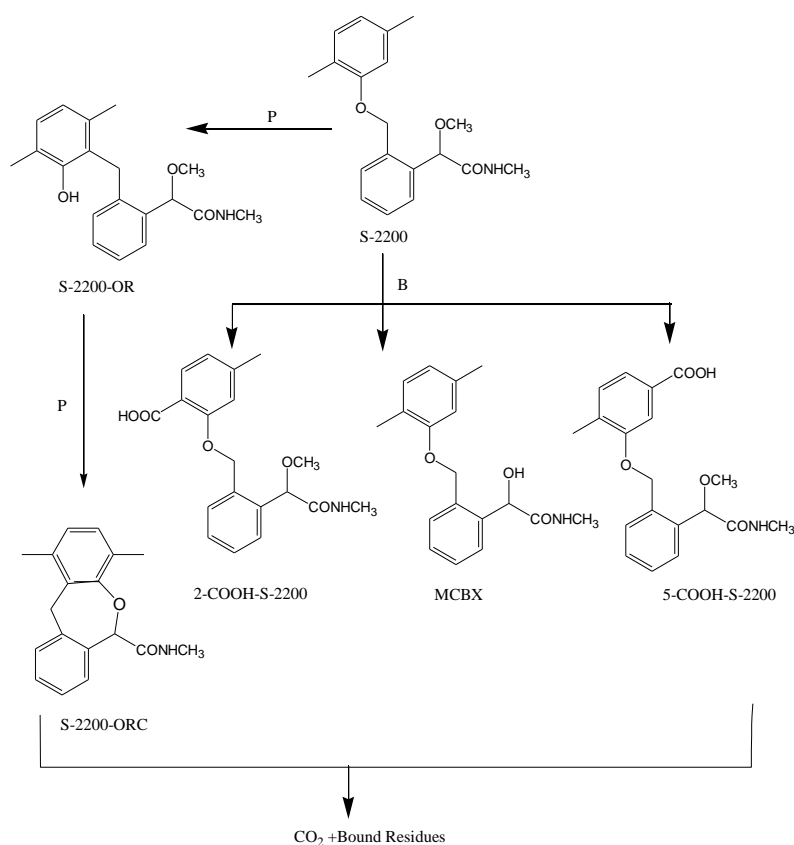


Figure 8.6-1: Proposed degradation route of mandestrobin in water (biotic and photolytic)
P = photolytic, B= biotic

Table 8.6-1: Summary of degradation in water/sediment of mandestrobin

Mandestrobin Distribution (max. sediment 69.7 % after 31 days)										
Water/sediment system	pH water/sed	DegT50 whole syst. (d)	DegT90 whole syst. (d)	Kinetic fit	DissT50 water (d)	DissT90 water (d)	Kinetic fit	DissT50 sed. (d)	Kinetic fit	Evaluated on EU level y/n Reference
Calwich Abbey	7.7/8.0	212	703	SFO	7.8	64.8	FOMC	n.c.	-	y/EFSA (2015)
Swiss Lake	6.7/5.9	519	1725	SFO	19.0	157.7	FOMC	n.c.	-	y/EFSA (2015)
Geometric mean (n=2)		332	-							

* data of *R*-isomer and *S*-isomer combined

† dissipation value

Table 8.6-2: Summary of observed metabolites

5-COOH-S-2200 Water/sediment system	Max. in water/sediment 3.6 %	Evaluated on EU level y/EFSA (2015)
S-2200-OR Water/sediment system	Max. in water/sediment 20.7 % after 7 d (photolytic degradation)	Evaluated on EU level y/EFSA (2015)
S-2200-ORC Water/sediment system	Max. in water/sediment 13.7 % after 14 d (photolytic degradation)	Evaluated on EU level y/EFSA (2015)

8.7 Predicted Environmental Concentrations in soil (PEC_{soil}) (KCP 9.1.3)

zRMS Comments:	<p>Calculations of PEC_s for active substance, its metabolite and formulation used oilseed rape were submitted and accepted.</p> <p>The relevant endpoints used for PECs assessment were agreed at the EU level or accepted at the zonal level.</p> <p>The max. DT₅₀ for metabolite DX-CA-S-2200 of 18.5 d (Kodaka, 2016) was used in PECs assessment. From the newer study, Lamond, 2017, the DT₅₀ values for DX-CA-S-2200 of 5.2, 3.2 and 8.1 days are recommended to use for risk assessment. As DT₅₀ of 18.5 d is a worse case, the submitted PECs assessment was accepted.</p> <p>The maximum PEC_s values for active substance and its metabolite are presented in following table:</p>													
	<table> <tr> <th>Crop</th><th>Winter and Spring oilseed rape</th></tr> <tr> <th>Compound</th><th>PECs mg/kg soil</th></tr> <tr> <td>Mandestrobin</td><td>0.053 0.069*</td></tr> <tr> <td>5-COOH-S-2200</td><td>0.011</td></tr> <tr> <td>2-COOH-S-2200</td><td>0.005</td></tr> <tr> <td>DX-CA-S-2200</td><td>0.003</td></tr> <tr> <td>Formulation</td><td>0.144</td></tr> </table> <p>These values will be used in further risk assessment.</p>	Crop	Winter and Spring oilseed rape	Compound	PECs mg/kg soil	Mandestrobin	0.053 0.069*	5-COOH-S-2200	0.011	2-COOH-S-2200	0.005	DX-CA-S-2200	0.003	Formulation
Crop	Winter and Spring oilseed rape													
Compound	PECs mg/kg soil													
Mandestrobin	0.053 0.069*													
5-COOH-S-2200	0.011													
2-COOH-S-2200	0.005													
DX-CA-S-2200	0.003													
Formulation	0.144													

8.7.1 Justification for new endpoints

No deviation from EFSA (2015) agreed endpoints for mandestrobin, 5-COOH-S-2200 and 2-COOH-S-2200. Metabolite DX-CA-S-2200 observed in the additional aerobic degradation study on acidic soils is taken into account for PEC_{soil} calculations. A worst case DT₅₀ value of 18.5 days from a preliminary study (Kodaka, et al, 2016), was used for DX-CA-S-2200 in the RR (ANSES, 2021). Therefore this is considered as the current EU endpoint, even though shorter DT₅₀ values were obtained in the definitive study (Lamond, 2017).

8.7.2 Active substance(s) and relevant metabolite(s)

Table 8.7-1: Input parameters related to application for PEC_{soil} calculations

Use No.	1
Crop	Oilseed rape (winter and spring)
Application rate (g a.s./ha)	Mandestrobin 200g a.s./ha
Number of applications	1
Crop interception (%)	80% (BBCH 60-69)
Depth of soil layer (relevant for plateau concentration) (cm)	20 cm (tillage) + 5 cm for the ultimate application

Table 8.7-2: Input parameter for mandestrobin and relevant metabolite for PEC_{soil} calculation

Compound	Mol. weight (g/mol)	Max. occurrence (%)	DT ₅₀ (days)	Value in accordance to EU endpoint y/n/ Reference
Mandestrobin	313.39	--	415.1 (slow rate k value of DFOP kinetics/laboratory study)	y/EFSA (2015)
5-COOH-S-2200	343.38	18.0	136.8 (worst case laboratory study)	y/EFSA (2015)
2-COOH-S-2200	343.38	8.7	177.3 (worst-case laboratory study)	y/EFSA (2015)
DX-CA-S-2200	223.2	8.3	18.5 (worst case laboratory study)	y/Kodaka <i>et al.</i> (2016) ^a

^a ANSES, 2021.

Table 8.7-3: PEC_{soil} after applications on oilseed rape

PEC _{soil} (mg/kg)	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200
	Single application			
Initial	0.053	0.011	0.005	0.003
Plateau concentration (20 cm) after year 6	0.016	<0.001	<0.001	-
PEC _{accumulation} (PEC _{act} + PEC _{soil plateau})	0.069	0.011	0.005	-

8.7.2.1 PEC_{soil} of Mandestrobin 40SC formulation

Table 8.7-4: PEC_{soil} values on oilseed rape

Preparation	Application rate (g/ha)*	PEC _{act} (mg/kg)
Mandestrobin 40SC	541	0.144

*Based on 0.5 L product/ha and a density of 1.0825 kg/L

8.8 Predicted Environmental Concentrations in groundwater (PEC_{gw}) (KCP 9.2.4)

zRMS Comments:	The submitted PEC _{gw} assessment was accepted. Calculations of PEC _{GW} for active substance and its relevant metabolites were provided in Tier 1 with PUF = 0. The new metabolite DX-CA-S-2200 was also considered – not noted in EFSA Journal 2015;13(5):4100. This metabolite was observed in new laboratory study (evaluated and
----------------	---

	<p>accepted by AT, Registration Report for S-2200 25SC (Intuity/Sisam), AGES, 2016. The Applicant used the DT₅₀ value of 5.1 d although the used DT₅₀ of 6.0 d was used in Central Zone (zRMS AT). The proposed endpoints for metabolite DX-CA-S-2200 used in PECgw assessment were accepted. The recommended FOCUS models were used: FOCUS PELMO, FOCUS PEARL and FOCUS MACRO. In accordance with all model requirements, in PECgw assessment the geometric mean of K_{foc} should be used. In modelling, the Applicant has used an arithmetic mean, the values agreed at the EU level. In this case, as both relevant values of geometric and arithmetic means are very similar, the submitted assessment was accepted.</p> <p>The application dates were accepted. The acidic and neutral/alkaline soils were taken into consideration. The annual and biennial application in acidic soils was considered. In case of missed scenarios (Châteaudun, Hamburg and Kremsmünster) for spring OSR, the PECgw values obtained for winter OSR was used. The Applicant has provided an additional PECgw assessment for spring OSR using the spring cereals as a surrogate crop. This approach will be decided at cMS level.</p> <p>In acidic soils, the formulation could be applied every other year in spring and winter oilseed rape. The maximum PEC_{GW} values for active substance and metabolites (5-COOH-S-2200, 2-COOH-S-2200, DX-CA-S-2200) were below the trigger value of 0.1 µg/L if applied every other year in acidic soils.</p> <p>In neutral and alkaline soils, the formulation could be applied every year in spring and winter oilseed rape. The maximum PEC_{GW} values for active substance were below the trigger value of 0.1 µg/L if applied every other year in acidic soils. For metabolites 5-COOH-S-2200, 2-COOH-S-2200, the PECgw values were above the trigger value: 0.138 µg/L and 0.328 µg/L, respectively, (both in Hamburg scenario, winter OSR).</p> <p>The relevant decision/restriction will be made at cMS level.</p>
--	--

8.8.1 Justification for new endpoints

A new aerobic degradation study on mandestrobin in acidic soils under laboratory conditions (Gilbert, 2016) has been performed since the EFSA conclusion. As this study identified DX-CA-S-2200 as requiring risk assessment, further studies were undertaken to obtain DT₅₀ (Lamond, 2017) and K_{oc} (Kang, 2017) values.

Compound	Parameter	EFSA conclusion (2015)	Current agreed endpoint in the Centralzone ^a	Endpoint used in this assessment ^b
mandestrobin	DT ₅₀ [d]	Soil pH CaCl ₂ ≤ 5.9 : 276.4	Soil pH CaCl ₂ ≤ 6.7 : 231.2	Soil pH CaCl ₂ ≤ 6.7 : 231.2
5-COOH-S-2200	DT ₅₀ [d]	36.91	41.75	41.75
	Formation fraction	0.502	0.424	0.424
DX-CA-S-2200	DT ₅₀ [d]	Not observed	6.0	5.1
	K _{oc} , l/n		10, 1	5.3, 0.907
	Formation fraction		1	1

^aRegistration Report for S-2200 25SC (Intuity/Sisam), AGES, 2016

^bRegistration Report for S-2200 25SC (Intuity/Sisam), ANSES, 2021

8.8.2 Active substance(s) and relevant metabolite(s) (KCP 9.2.4.1)

PEC_{gw} values were provided in the report of Jones & Jarvis (2023a) and Jones & Tallentire (2025). Input data, as well as results, are summarised in the tables below. 2-COOH-S-2200 and 5-COOH-S-2200 have already been determined to be non-relevant (see document B10).

Table 8.8-1: Input parameters related to application for PEC_{gw} calculations

Use No.	1	1	1
Crop	Winter OSR	Summer OSR	Spring cereals (as a surrogate crop for Summer OSR)
Application rate (g as/ha)	Mandestrobin: 200g a.s./ha	Mandestrobin: 200g a.s./ha	Mandestrobin: 200g a.s./ha
Number of applications/interval (d)	1	1	1
Relative application date	See below	See below	See below
Crop interception (%)	80%	80%	80 %
Frequency of application	annual, biennial	annual, biennial	annual, biennial
Models used for calculation	FOCUS PEARL v5.5.5, FOCUS PELMO v6.6.4, FOCUS MACRO v5.5.4	FOCUS PEARL v5.5.5, FOCUS PELMO v6.6.4, FOCUS MACRO v5.5.4	FOCUS PEARL v5.5.5, FOCUS PELMO v6.6.4, FOCUS MACRO v5.5.4

Table 8.8-2: Application dates used for groundwater risk assessment

Crop	Scenario	BBCH 60
Oilseed rape, winter	Châteaudun	20-Apr (110)
	Hamburg	05-May
	Jokioinen	-
	Kremsmünster	05-May
	Okehampton	30-Apr
	Piacenza	15-Apr
	Porto	20-Apr
	Sevilla	-
	Thiva	-
Oilseed rape, spring	Châteaudun	-
	Hamburg	-
	Jokioinen	05-Jul
	Kremsmünster	-
	Okehampton	15-May
	Piacenza	-
	Porto	31-May
	Sevilla	-
	Thiva	-
Spring cereals (as a surrogate for Oilseed rape, spring)	Châteaudun	11-Jun (162)
	Hamburg	08-Jun
	Jokioinen	-

	Kremsmünster	08-Jun
	Okehampton	-
	Piacenza	-
	Porto	-
	Sevilla	-
	Thiva	-

Scenario is not presented due to calculations already provided for the intended crop

Table 8.8-3: Summary of input data used in the modelling

Parameter	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200 (acidic conditions only)
Molecular weight [g/mol]	313.39	343.38	343.38	223.2
Water solubility [mg/L]	15.8 (20°C)	134 (20°C)	607 (20°C)	1000
Vapor pressure [Pa]	3.36×10^{-8} (20°C)	2.8×10^{-9} (20°C)	2.8×10^{-9} (20°C)	1.0×10^{-9}
DT ₅₀ (soil) [d]	pH ≤ 6.7: 231.2 pH ≥ 7.2: 59.5	41.75	28.7	5.1
K _{FOC} [L/kg]	449	Acidic pH: 684 Basic pH: 55	Acidic pH: 226 Basic pH: 15	5.3
K _{FOM} [L/kg]	260	Acidic pH: 397 Basic pH: 32	Acidic pH: 131 Basic pH: 8.6	3.1
1/n [-]	0.919	Acidic pH: 0.926 Alkaline pH: 0.889	Acidic pH: 0.923 Alkaline pH: 0.884	0.907
Plant uptake factor (PUF/TSCF) [-]	0	0	0	0
Formation fraction [-]	0.42 → 5-COOH-S-2200 0.24 → 2-COOH-S-2200 1.0 → DX-CA-S-2200	0.42 from parent	0.24 from parent	1.0 from parent

Table 8.8-4: PEC_{GW}, mandestrobin and its metabolites in acidic conditions

FOCUS Model	Scenario	PEC _{GW} [µg/L]				PEC _{GW} [µg/L]			
		Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200
		Winter oilseed rape (BBCH 60) 1×200 g/ha (annual), 80% int.				Winter oilseed rape (BBCH 60) 1×200 g/ha (biennial), 80% int.			
FOCUS PEARL	Châteaudun	0.027	0.002	0.002	0.013	0.010	0.001	0.001	0.007
	Hamburg	0.191	0.011	0.012	0.162	0.080	0.005	0.005	0.075
	Jokioinen	-	-	-	-	-	-	-	-
	Kremsmünster	0.129	0.007	0.008	0.048	0.054	0.003	0.004	0.021
	Okehampton	0.177	0.010	0.011	0.097	0.074	0.004	0.005	0.046
	Piacenza	0.075	0.005	0.005	0.046	0.030	0.002	0.002	0.022
	Porto	0.073	0.004	0.005	0.092	0.030	0.002	0.002	0.043
	Sevilla	-	-	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-	-	-
FOCUS PELMO	Châteaudun	0.005	<0.001	<0.001	0.017	0.003	<0.001	<0.001	0.008
	Hamburg	0.125	0.007	0.008	0.201	0.058	0.003	0.004	0.093
	Jokioinen	-	-	-	-	-	-	-	-
	Kremsmünster	0.104	0.006	0.007	0.064	0.043	0.002	0.003	0.029
	Okehampton	0.185	0.010	0.012	0.133	0.077	0.004	0.005	0.062
	Piacenza	0.078	0.004	0.005	0.093	0.030	0.002	0.002	0.042
	Porto	0.115	0.006	0.008	0.178	0.047	0.002	0.003	0.084
	Sevilla	-	-	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-	-	-
FOCUS MACRO	Châteaudun	0.021	0.001	0.001	0.014	0.008	<0.001	<0.001	0.008

Bold – Used to highlight where further refinement of the application frequency was triggered due to exceedance of a threshold.

FOCUS Model	Scenario	PEC _{GW} [µg/L]				PEC _{GW} [µg/L]			
		Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200
		Spring oilseed rape (BBCH 60) 1×200 g/ha (annual), 80% int.				Spring oilseed rape (BBCH 60) 1×200 g/ha (biennial), 80% int.			
FOCUS PEARL	Châteaudun	-	-	-	-	-	-	-	-
	Hamburg	-	-	-	-	-	-	-	-
	Jokioinen	0.001	<0.001	<0.001	0.142	0.008	<0.001	0.001	0.075
	Kremsmünster	-	-	-	-	-	-	-	-
	Okehampton	0.161	0.009	0.010	0.099	0.067	0.004	0.004	0.046
	Piacenza	-	-	-	-	-	-	-	-
	Porto	0.066	0.004	0.004	0.083	0.027	0.001	0.002	0.039
	Sevilla	-	-	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-	-	-
FOCUS PELMO	Châteaudun	-	-	-	-	-	-	-	-
	Hamburg	-	-	-	-	-	-	-	-
	Jokioinen	0.001	<0.001	<0.001	0.211	0.008	<0.001	0.001	0.113
	Kremsmünster	-	-	-	-	-	-	-	-
	Okehampton	0.169	0.009	0.011	0.131	0.070	0.004	0.005	0.060
	Piacenza	-	-	-	-	-	-	-	-
	Porto	0.092	0.005	0.006	0.141	0.038	0.002	0.003	0.066
	Sevilla	-	-	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-	-	-
FOCUS MACRO	Châteaudun	-	-	-	-	-	-	-	-

Bold – Used to highlight where further refinement of the application frequency was triggered due to exceedance of a threshold.

FOCUS Model	Scenario	PEC _{Gw} [µg/L]				PEC _{Gw} [µg/L]			
		Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200
		Spring oilseed rape (Spring cereals used as surrogate crop, BBCH 60) 1×200 g/ha (annual), 80% int.				Spring oilseed rape (Spring cereals used as surrogate crop, BBCH 60) 1×200 g/ha (biennial), 80% int.			
FOCUS PEARL	Châteaudun	0.011	0.001	0.001	0.013	0.004	<0.001	<0.001	0.006
	Hamburg	0.209	0.012	0.013	0.190	0.089	0.005	0.006	0.088
	Jokioinen	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Kremsmünster	0.129	0.007	0.008	0.050	0.055	0.003	0.004	0.023
	Okehampton	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Piacenza	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Porto	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Sevilla	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Thiva	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Thiva	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
FOCUS PELMO	Châteaudun	0.004	<0.001	<0.001	0.013	0.002	<0.001	<0.001	0.006
	Hamburg	0.138	0.008	0.009	0.228	0.066	0.004	0.004	0.100 ¹¹
	Jokioinen	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Kremsmünster	0.100	0.005	0.006	0.068	0.041	0.002	0.003	0.031
	Okehampton	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Piacenza	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Porto	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
	Sevilla	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹	- ¹
FOCUS MACRO	Châteaudun	0.020	0.001	0.001	0.015	0.008	<0.001	<0.001	0.009

Bold – Used to highlight where further refinement of the application frequency was triggered due to exceedance of a threshold.

¹ Scenario is not presented due to calculations already provided for the intended crop

¹¹ Assessing the pesticide concentration per year within the PELMO files shows that the value is <0.1 µg/L (value of 0.0998 µg/L) rounded up to 0.100 µg/L

Table 8.8-5: PEC_{GW}, mandestrobin and its metabolites, in basic conditions

FOCUS Model	Scenario	PEC _{GW} [µg/L]			PEC _{GW} [µg/L]		
		Mandestrobin	5-COOH-S-2200	2-COOH-S-2200	Mandestrobin	5-COOH-S-2200	2-COOH-S-2200
		Winter oilseed rape (BBCH 60) 1×200 g/ha (annual), 80% int.			Spring oilseed rape (BBCH 60) 1×200 g/ha (annual), 80% int.		
FOCUS PEARL	Châteaudun	<0.001	0.019	0.092	-	-	-
	Hamburg	<0.001	0.138	0.328	-	-	-
	Jokioinen	-	-	-	<0.001	0.038	0.261
	Kremsmünster	<0.001	0.088	0.184	-	-	-
	Okehampton	<0.001	0.106	0.222	<0.001	0.098	0.234
	Piacenza	<0.001	0.036	0.102	-	-	-
	Porto	<0.001	0.057	0.136	<0.001	0.062	0.124
	Sevilla	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-
FOCUS PELMO	Châteaudun	<0.001	0.011	0.070	-	-	-
	Hamburg	<0.001	0.120	0.297	-	-	-
	Jokioinen	-	-	-	<0.001	0.041	0.284
	Kremsmünster	<0.001	0.078	0.179	-	-	-
	Okehampton	<0.001	0.117	0.248	<0.001	0.112	0.256
	Piacenza	<0.001	0.050	0.146	-	-	-
	Porto	<0.001	0.107	0.183	<0.001	0.082	0.160
	Sevilla	-	-	-	-	-	-
	Thiva	-	-	-	-	-	-
FOCUS MACRO	Châteaudun	<0.001	0.017	0.068	-	-	-

Bold – Used to highlight where further refinement of the application frequency was triggered due to exceedance of a threshold.

FOCUS Model	Scenario	PEC _{GW} [µg/L]		
		Mandestrobin	5-COOH-S-2200	2-COOH-S-2200
		Spring oilseed rape (Spring cereals used as surrogate crop, BBCH 60) 1×200 g/ha (annual), 80% int.		
FOCUS PEARL	Châteaudun	< 0.001	0.013	0.082
	Hamburg	< 0.001	0.159	0.403
	Jokioinen	- ¹	- ¹	- ¹
	Kremsmünster	< 0.001	0.089	0.198
	Okehampton	- ¹	- ¹	- ¹
	Piacenza	- ¹	- ¹	- ¹
	Porto	- ¹	- ¹	- ¹
	Sevilla	- ¹	- ¹	- ¹
	Thiva	- ¹	- ¹	- ¹
FOCUS PELMO	Châteaudun	< 0.001	0.008	0.061
	Hamburg	< 0.001	0.141	0.343
	Jokioinen	- ¹	- ¹	- ¹
	Kremsmünster	< 0.001	0.083	0.199
	Okehampton	- ¹	- ¹	- ¹
	Piacenza	- ¹	- ¹	- ¹
	Porto	- ¹	- ¹	- ¹
	Sevilla	- ¹	- ¹	- ¹
FOCUS MACRO	Châteaudun	<0.001	0.019	0.080

Bold – Used to highlight where further refinement of the application frequency was triggered due to exceedance of a threshold.

¹ Scenario is not presented due to calculations already provided for the intended crop

In basic conditions, mandestrobin was always < 0.1 µg/L and the non-relevant metabolites, 5-COOH-S-2200 and 2-COOH-S-2200 were always <0.75 µg/L following annual application. In acidic soil conditions, the non-relevant metabolites, 5-COOH-S-2200 and 2-COOH-S-2200 were always <0.1 µg/L following annual application. However, biennial application simulations were necessary to achieve PEC_{GW} values of < 0.1 µg/L for mandestrobin and DX-CA-S-2200 in all relevant scenarios (Jokioinen is not considered a relevant scenario in CEU conditions.)

8.9 Predicted Environmental Concentrations in surface water (PEC_{sw}) (KCP 9.2.5)

zRMS Comments:	The submitted PEC _{sw} and PEC _{sed} calculations were accepted.
	All used endpoints for active substance and its metabolites were accepted. The new metabolite DX-CA-S-2200 was also considered – not noted in EFSA Journal 2015;13(5):4100. This metabolite was observed in new laboratory study (evaluated and accepted by AT, Registration Report for S-2200 25SC (Intuity/Sisam), AGES, 2016. The Applicant used the DT ₅₀ value of 5.1 d although the used DT ₅₀ of 6.0 d was used in Central Zone (zRMS AT). The proposed endpoints for metabolite DX-CA-S-2200 used in PEC _{gw} assessment were accepted.
	D2 scenario is not relevant for Central Zone and was not taken into consideration. The recommended FOCUS models were used: FOCUS Step 1 & 2, Step 3. Additionally, the PEC _{sw} assessment was provided in Step 4 with relevant mitigation measures.
	The max PEC _{sw} for Central Zone obtained in Step 4 assessment with mitigation measures (10 m NSS + 10 m VFS and 20 m NSS + 20 m VFS) was added by the Applicant.
	The PEC drift values using the Drift Calculator in SWASH model presented in Table 8.9-11 were accepted.

Crop	Application rate g a.s./ha	No spray buffer (m)	Max PEC _{sw} (µg/l)
Winter and spring OSR	541 g [prod]/ha equivalent to 200 g a.s/ha	1	3.476

The mitigation measure will be recommended in the ecotoxicological section, if relevant.
--

8.9.1 Justification for new endpoints

A new aerobic degradation study on mandestrobin in acidic soils under laboratory conditions (Gilbert, 2016) has been performed since the EFSA conclusion. As this study identified DX-CA-S-2200 as requiring risk assessment, further studies were undertaken to obtain DT₅₀ (Lamond, 2017) and Koc (Kang, 2017) values.

Compound	Parameter	EFSA conclusion (2015)	Current agreed endpoint in the Central zone ^a	Endpoint used in this assessment ^b
mandestrobin	DT ₅₀ [d]	Soil pH CaCl ₂ ≤ 5.9 : 276.4	Soil pH CaCl ₂ ≤ 6.7 : 231.2	Soil pH CaCl ₂ ≤ 6.7 : 231.2
5-COOH-S-2200	DT ₅₀ [d]	36.91	41.75	41.75
DX-CA-S-2200	DT ₅₀ [d]	Not observed	Not considered	5.1
	Koc			5.3

	% Formation soil/water/sediment			8.3/-/-
--	------------------------------------	--	--	---------

^aRegistration Report for S-2200 25SC (Intuity/Sisam), AGES, 2016

^bRegistration Report for S-2200 25SC (Intuity/Sisam), ANSES, 2021

8.9.2 Active substance(s), relevant metabolite(s) and the formulation (KCP 9.2.5)

PEC_{sw} values were provided in the reports of Jones & Jarvis (2023b) and Jones & Hilton (2024). Results for mandestrobin were provided for Steps 1-4 (At Steps 3 & 4 only scenarios relevant to the Central Zone were presented) whilst results for the metabolites, S-2200-OR, S-2200-ORC, 5-COOH-S-2200, 2-COOH-S-2200 and DX-CA-S-2200, were provided for Steps 1-2. Additional modelling to Step 4 has been provided at the request of zonal rapporteur member state (zRMS) with a letter delivered in August 2024 (Jones & Hilton, 2024). Input data, as well as results, are summarised in the tables below.

Table 8.9-1: Input parameters related to application for PEC_{SW/SED} calculations

Plant protection product	Mandestrobin 40SC	Mandestrobin 40SC
Crop	winter oilseed rape	Summer oilseed rape
Application rate (kg as/ha)	0.200	0.200
Number of applications/interval (d)	1 / -	1 / -
Application window	Step 2: March to May Steps 3 & 4: see Table 8.9-2	Step 2: June- Sept Steps 3 & 4: see Table 8.9-2
Application method	Spray	Spray
CAM (Chemical application method)	2	2
Soil depth (cm)	4	4
Interception	Full crop cover	Full crop cover
Models used for calculation	FOCUS STEPS 1-2 v 3.2 SWASH v5.3, FOCUS PRZM v4.3.1, FOCUS MACRO v 5.5.4, FOCUS TOXSWA v4.4.3	

Table 8.9-2: FOCUS Steps 3 & 4 Scenario related input parameters for PEC_{sw/SED} calculations for the application of mandestrobin 40SC

Scenario	Crop	Appl. window	Appl. date
D2	Oil seed rape, winter	15-Jun-15-Jul	15-Jun-1986
D2	Oil seed rape, winter	15-Jun-15-Jul	15-Jun-1986
D3	Oil seed rape, winter	25-May-24-Jun	24-May-1992
D4	Oil seed rape, winter	5-Jun-5-Jul	04-Jul-1985
D5	Oil seed rape, winter	10-May-9-Jun	11-May-1978
R1	Oil seed rape, winter	25-May-24-Jun	13-Jun-1984
R3	Oil seed rape, winter	20-Apr-20-May	22-Apr-1980
D1	Oil seed rape, spring	4-Jul-3-Aug	04-Jul-1982
D1	Oil seed rape, spring	4-Jul-3-Aug	04-Jul-1982
D3	Oil seed rape, spring	15-Jun-15-Jul	23-Jun-1992
D4	Oil seed rape, spring	17-Jun-17-Jul	04-Jul-1985
D5	Oil seed rape, spring	25-May-24-Jun	27-May-1978
R1	Oil seed rape, spring	7-Jun-7-Jul	09-Jun-1978

Table 8.9-3: Input parameters related to mandestrobin and metabolite(s) for PEC_{sw/sed} calculations STEP 1/2

Compound	Mandestrobin	S-2200-OR	S-2200-ORC	5-COOH-S-2200	2-COOH-S-2200	DX-CA-S-2200
Molecular weight (g/mol)	313.9	313.39	281.35	343.38	343.38	223.2
Water solubility (mg/L)	15.8	1	1	134	607	1
K _{foc} (mL/g)	449	10	10	55.2 ^{II}	14.8 ^{II}	5.3
DT _{50,soil} (d)	Acidic pH: 231.2 ^I Basic pH: 59.5 ^{II}	1000	1000	41.75	28.7	5.1
DT _{50,water} (d)	1000	1000	1000	1000	1000	1000
DT _{50,sed} (d)	332	1000	1000	1000	1000	1000
DT _{50,whole system} (d)	332	1000	1000	1000	1000	1000
Max. occurrence (%)	-	Soil: 0 Total system: 20.7	Soil: 0 Total system: 13.7	Soil: 18.0 Total system: 3.6	Soil: 8.7 Total system: 1.0	Soil: 8.3 Total system: 0.0

^I Acidic endpoint when modelled as parent only or when modelling DX-CA-S-2200

^{II} Basic endpoint

All endpoints are considered to be the current acceptable endpoints (i.e. based on RR from ANSES, 2021). As a conservative approach in Steps 1&2, these datasets have not been separated and the worst-case value has been selected in each instance (acidic for mandestrobin, basic for 5-COOH-S-2200 and 2-COOH-S-2200). DX-CA-S-2200 is only observed under acidic conditions and therefore, the acidic pH DT_{50,soil} has been used in this instance. **At Steps 3 & 4, only scenarios relevant to the Central zone are shown (tables 8).**

Table 8.9-4: Input parameters related to mandestrobin for PEC_{sw/sed} calculations Steps 3 & 4

Parameter	Mandestrobin
Molecular weight [g/mol]	313.39
Water solubility [mg/L]	15.8 (at 20 °C)
Vapor pressure [Pa]	3.36×10 ⁻⁸ (at 20 °C)
DT ₅₀ (soil) [d]	Acidic pH: 231.2; Basic pH: 59.5
K _{FOC} [L/kg]	449
K _{FOM} [L/kg]	260
1/n [-]	0.919
DT ₅₀ water [d]	1000 ^I
DT ₅₀ sediment [d]	332 ^I
Crop uptake factor [-]	0

^I According to the K_{FOC} and relating to the FOCUS guidance (2015) For compounds with K_{FOC} between 100 and 2000mL/g, the FOCUS kinetics advice regarding running simulations with both combinations for ascribing the whole system DT₅₀ and default and selecting the results that give the highest concentrations for the risk assessment should be followed. According to the FOCUS guidance (2015), the geometric mean (n=2) total system DT₅₀ of 332 days (EFSA, 2015) was used for the degradation in the sediment phase and the FOCUS default of 1000 days in the water phase for option A, the reverse was used for option B. Following assessment of the results, option A was found to produce the highest PEC_{sw} values and therefore, only option A results have been presented.

Table 8.9-5: Overview of maximum PEC_{sw}, Step 1, mandestrobin and its metabolites

Compound	Step 1	
	PEC _{sw} [µg/L]	PEC _{sed} [µg/kg]
	Oil seed rape (winter or spring), 1 × 200 g/ha	
Mandestrobin	43.55	191.90
S-2200-OR	14.00	1.40
S-2200-ORC	8.32	0.83

5-COOH-S-2200	14.77	8.14
2-COOH-S-2200	6.97	1.03
DX-CA-S-2200	3.91	0.21

Table 8.9-6: Overview of maximum PEC_{SW}, Step 2, mandestrobin and its metabolites

Compound	North and South Europe		Scenario/ Season
	PEC _{SW} [µg/L]	PEC _{SED} [µg/kg]	
	Oil seed rape, winter; 1 × 200 g/ha; full canopy		
Mandestrobin	3.37	14.36	NEU: Mar-May
	5.43	23.58	SEU: Mar-May
S-2200-OR	1.03	0.10	NEU: Mar-May
	1.68	0.17	SEU: Mar-May
S-2200-ORC	0.61	0.06	NEU: Mar-May
	1.00	0.10	SEU: Mar-May
5-COOH-S-2200	0.76	0.42	NEU: Mar-May
	1.45	0.80	SEU: Mar-May
2-COOH-S-2200	0.34	0.05	NEU: Mar-May
	0.65	0.10	SEU: Mar-May
DX-CA-S-2200	0.11	0.01	NEU: Mar-May
	0.23	0.01	SEU: Mar-May
	Oil seed rape, spring; 1 × 200 g/ha; full canopy		
Mandestrobin	3.37	14.36	NEU: Jun-Sep
	4.40	18.97	SEU: Jun-Sep
S-2200-OR	1.03	0.10	NEU: Jun-Sep
	1.35	0.13	SEU: Jun-Sep
S-2200-ORC	0.61	0.06	NEU: Jun-Sep
	0.80	0.08	SEU: Jun-Sep
5-COOH-S-2200	0.76	0.42	NEU: Jun-Sep
	1.10	0.61	SEU: Jun-Sep
2-COOH-S-2200	0.34	0.05	NEU: Jun-Sep
	0.50	0.07	SEU: Jun-Sep
DX-CA-S-2200	0.11	0.01	NEU: Jun-Sep
	0.17	0.01	SEU: Jun-Sep

Highest values in bold for each compound

Table 8.9-7: Maximum mandestrobin PEC_{SW}, Step 3, oil seed rape, winter, 1×200 g/ha (acidic conditions)

Scenario	Appl. rate [g/ha]	Appl. Date	Step 3	
			Date of max	Global max PEC _{SW} [µg/L]
D2 Ditch	200	15-Jun-1986	31-Oct-1986	3.031
D2 Stream	200	15-Jun-1986	31-Oct-1986	1.894
D3 Ditch	200	24-May-1992	24-May-1992	1.270
D4 Pond	200	04-Jul-1985	29-Dec-1985	0.938
D4 Stream	200	04-Jul-1985	04-Jul-1985	1.096
D5 Pond	200	11-May-1978	15-Feb-1979	0.466
D5 Stream	200	11-May-1978	11-May-1978	1.182
R1 Pond	200	13-Jun-1984	21-Jun-1984	0.231
R1 Stream	200	13-Jun-1984	21-Jun-1984	1.823
R3 Stream	200	22-Apr-1980	13-May-1980	1.653

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{SW} highlighted red.

Table 8.9-8: Maximum mandestrobin PEC_{sw}, Step 3, oil seed rape, winter, 1×200 g/ha (basic conditions)

Scenario	Appl. rate [g/ha]	Step 3		
		Appl. date	Date of max	Global max PEC _{sw} [µg/L]
D2 Ditch	200	15-Jun-1986	15-Jun-1986	1.478
D2 Stream	200	15-Jun-1986	15-Jun-1986	1.274
D3 Ditch	200	24-May-1992	24-May-1992	1.270
D4 Pond	200	04-Jul-1985	30-Dec-1985	0.311
D4 Stream	200	04-Jul-1985	04-Jul-1985	1.096
D5 Pond	200	11-May-1978	11-May-1978	0.103
D5 Stream	200	11-May-1978	11-May-1978	1.182
R1 Pond	200	13-Jun-1984	21-Jun-1984	0.228
R1 Stream	200	13-Jun-1984	21-Jun-1984	1.792
R3 Stream	200	22-Apr-1980	13-May-1980	1.505

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red

Table 8.9-9: Maximum mandestrobin PEC_{sw}, Step 3, oil seed rape, spring, 1×200 g/ha (acidic conditions)

Scenario	Appl. rate [g/ha]	Step 3		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	23-Jun-1992	23-Jun-1992	1.271
D4 Pond	200	04-Jul-1985	29-Dec-1985	1.018
D4 Stream	200	04-Jul-1985	04-Jul-1985	1.096
D5 Pond	200	27-May-1978	07-Feb-1978	0.609
D5 Stream	200	27-May-1978	27-May-1978	1.182
R1 Pond	200	09-Jun-1978	09-Jul-1978	0.339
R1 Stream	200	09-Jun-1978	17-Jun-1978	1.918

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

Table 8.9-10: Maximum mandestrobin PEC_{sw}, Step 3, oil seed rape, spring, 1×200 g/ha (basic conditions)

Scenario	Appl. rate [g/ha]	Step 3		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	23-Jun-1992	23-Jun-1992	1.271
D4 Pond	200	04-Jul-1985	30-Dec-1985	0.372
D4 Stream	200	04-Jul-1985	04-Jul-1985	1.096
D5 Pond	200	27-May-1978	07-Feb-1978	0.214
D5 Stream	200	27-May-1978	27-May-1978	1.182
R1 Pond	200	09-Jun-1978	09-Jul-1978	0.322
R1 Stream	200	09-Jun-1978	17-Jun-1978	1.893

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

Table 8.9-11: Overview of maximum PEC_{sw}, Step 4, Oil seed rape, winter, 1×200 g/ha, acidic conditions

Scenario	Appl. rate [g/ha]	Step 4, 10 m db+vfs			Step 4, 20 m db+vfs		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]	Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	24-May-1992	24-May-1992	0.183	24-May-1992	24-May-1992	0.095
D4 Pond	200	04-Jul-1985	29-Dec-1985	0.934	04-Jul-1985	29-Dec-1985	0.932
D4 Stream	200	04-Jul-1985	09-Dec-1985	0.913	04-Jul-1985	09-Dec-1985	0.913
D5 Pond	200	11-May-1978	15-Feb-1979	0.464	11-May-1978	15-Feb-1979	0.462
D5 Stream	200	11-May-1978	24-Jan-1978	0.503	11-May-1978	24-Jan-1978	0.503
R1 Pond	200	13-Jun-1984	21-Jun-1984	0.102	13-Jun-1984	21-Jun-1984	0.055
R1 Stream	200	13-Jun-1984	21-Jun-1984	0.830	13-Jun-1984	21-Jun-1984	0.435
R3 Stream	200	22-Apr-1980	13-May-1980	0.740	22-Apr-1980	13-May-1980	0.385

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

Table 8.9-12: Overview of maximum PEC_{sw}, Step 4, Oil seed rape, winter, 1×200 g/ha, basic conditions

Scenario	Appl. rate [g/ha]	Step 4, 10 m db+vfs			Step 4, 20 m db+vfs		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]	Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	24-May-1992	24-May-1992	0.183	24-May-1992	24-May-1992	0.095
D4 Pond	200	04-Jul-1985	30-Dec-1985	0.307	04-Jul-1985	30-Dec-1985	0.304
D4 Stream	200	04-Jul-1985	07-Dec-1985	0.307	04-Jul-1985	07-Dec-1985	0.307
D5 Pond	200	11-May-1978	07-Feb-1978	0.092	11-May-1978	07-Feb-1978	0.092
D5 Stream	200	11-May-1978	11-May-1978	0.229	11-May-1978	24-Jan-1978	0.125
R1 Pond	200	13-Jun-1984	21-Jun-1984	0.100	13-Jun-1984	21-Jun-1984	0.054
R1 Stream	200	13-Jun-1984	21-Jun-1984	0.816	13-Jun-1984	21-Jun-1984	0.427
R3 Stream	200	22-Apr-1980	13-May-1980	0.674	22-Apr-1980	13-May-1980	0.351

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

Table 8.9-13: Overview of maximum PEC_{sw}, Step 4, Oil seed rape, spring, 1×200 g/ha, acidic conditions

Scenario	Appl. rate [g/ha]	Step 4, 10 m db+vfs			Step 4, 20 m db+vfs		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]	Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	23-Jun-1992	23-Jun-1992	0.183	23-Jun-1992	23-Jun-1992	0.095
D4 Pond	200	04-Jul-1985	29-Dec-1985	1.014	04-Jul-1985	29-Dec-1985	1.012
D4 Stream	200	04-Jul-1985	09-Dec-1985	0.993	04-Jul-1985	09-Dec-1985	0.993
D5 Pond	200	27-May-1978	07-Feb-1978	0.609	27-May-1978	07-Feb-1978	0.609
D5 Stream	200	27-May-1978	24-Jan-1978	0.759	27-May-1978	24-Jan-1978	0.759
R1 Pond	200	09-Jun-1978	09-Jul-1978	0.146	09-Jun-1978	09-Jul-1978	0.077
R1 Stream	200	09-Jun-1978	17-Jun-1978	0.874	09-Jun-1978	17-Jun-1978	0.458

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

Table 8.9-14: Overview of maximum PEC_{sw}, Step 4, Oil seed rape, spring, 1×200 g/ha, basic conditions

Scenario	Appl. rate [g/ha]	Step 4, 10 m db+vfs			Step 4, 20 m db+vfs		
		Appl. Date	Date of max	Global max PEC _{sw} [µg/L]	Appl. Date	Date of max	Global max PEC _{sw} [µg/L]
D3 Ditch	200	23-Jun-1992	23-Jun-1992	0.183	23-Jun-1992	23-Jun-1992	0.095
D4 Pond	200	04-Jul-1985	30-Dec-1985	0.368	04-Jul-1985	30-Dec-1985	0.366
D4 Stream	200	04-Jul-1985	07-Dec-1985	0.368	04-Jul-1985	07-Dec-1985	0.368
D5 Pond	200	27-May-1978	07-Feb-1978	0.214	27-May-1978	07-Feb-1978	0.214
D5 Stream	200	27-May-1978	24-Jan-1978	0.257	27-May-1978	24-Jan-1978	0.257
R1 Pond	200	09-Jun-1978	09-Jul-1978	0.139	09-Jun-1978	09-Jul-1978	0.073
R1 Stream	200	09-Jun-1978	17-Jun-1978	0.862	09-Jun-1978	17-Jun-1978	0.452

Highlighted cells indicate relevant entry route: drift (blue), runoff (green) or drainage (grey), global maximum PEC_{sw} highlighted red.

PEC_{sw}/sed of Mandestrobin 40SC

Table 8.9-15: Drift PEC values from the formulation on Winter and Spring Oilseed Rape

Crop	Application rate (g/ha)	PEC _{sw} (µg/L)*
at 1m (1.93%)		
Winter and spring oilseed rape	541	3.48

*based on 0.5 L product/ha and a density of 1.0825 kg/L g/cm³

8.10 Fate and behaviour in air (KCP 9.3, KCP 9.3.1)

Table 8.10-1 Summary of atmospheric degradation and behaviour

Compound	Mandestrobin
Direct photolysis in air	No studied – no data requested
Quantum yield of direct phototransformation	$\Phi = 0.269 - 0.283$
Photochemical oxidative degradation in air	DT ₅₀ (h): 1.332 derived by the Atkinson model (version 1.9) OH (12h) concentration assumed = $1.5 \times 10^6 \text{ cm}^{-3}$
Volatilisation	From plant surfaces; not studied From soil surfaces: not studied
Metabolites	None

The vapour pressure at 20 °C of the active substance mandestrobin is $< 10^{-5} \text{ Pa}$ ($3.36 \times 10^{-8} \text{ Pa}$). Hence the active substance mandestrobin is regarded as non-volatile. Therefore exposure of adjacent surface waters and terrestrial ecosystems by the active substance mandestrobin due to volatilization with subsequent deposition is considered negligible.

Appendix 1 Lists of data considered in support of the evaluation

Tables considered not relevant can be deleted as appropriate.

MS to blacken authors of vertebrate studies in the version made available to third parties/public.

List of data submitted by the applicant and 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	Owner
KCA 7.1.1.2/01	Lewis, C. J. & Cooper, T.	2015a	[14C]S-2354 (S-2200 S-isomer): Anaerobic Soil Metabolism and Degradation XXXX. Report ROM-0060 GLP, Unpublished	N	XXXX
KCA 7.1.1.2/02	Lewis, C. J. & Cooper, T.	2015b	[14C]S-2167 (S-2200 R-isomer): Anaerobic Soil Metabolism and Degradation XXXX. Report ROM-0061 GLP, Unpublished	N	XXXX
KCA 7.1.2.1.1/01	Gilbert, J.	2016	[14C]Mandestrobin (S-2200): Aerobic Soil Metabolism and Degradation XXXX. Report ROM-0062 GLP, Unpublished	N	XXXX
KCA 7.1.2.1.2/01	Kodaka, R., Abe, J. & Fujisawa, T	2016	Aerobic soil metabolism study of [phenyl-14C]De-Xy-S-2200 (preliminary study). XXXX. Report no.: ROM-0063 Non-GLP, unpublished	N	XXXX
KCA 7.1.2.1.2/02	Lamond, P.	2017	[14C]De-Xy-S-2200: Aerobic Soil Metabolism Study in Three Soils Smithers Viscient (ESG) Ltd. Study No. 3201768 XXXX. Report ROM-0070 GLP, Unpublished	N	XXXX
KCA 7.1.3.1/01	Kang, S.	2017	[14C]DX-CA-S-2200 – Adsorption-Desorption on Five Soils using a Batch Equilibrium Method Smithers Viscient. Study No. 13048.6948 XXXX. Report ROM-0067	N	XXXX

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	Owner
			GLP, Unpublished		
KCP 9.2.4.1/01	Jones, B. & Jarvis, T.	2023a	Mandestrobin 40SC: Predicted Environmental Concentrations (PEC) of mandestrobin and its metabolites in groundwater in the central zone of the EU using the FOCUS groundwater scenarios. Exponent International Ltd.. Report No.: 2202311.UK0- 8800 Non-GLP, unpublished	N	XXXX
KCP 9.2.4.1/02	Jones, B. & Tallentire, E.	2025	Mandestrobin 40SC: Predicted Environmental Concentrations (PEC) of mandestrobin and its metabolites in groundwater in the central zone of the EU using the FOCUS groundwater scenarios – Surrogate crops Exponent International Ltd.. Report No.: 2202311.UK0- 7285 Non-GLP, unpublished	N	XXXX
KCP 9.2.5/01	Jones, B. & Jarvis, T.	2023b	Predicted Environmental Concentrations in surface water after post-emergence application of mandestrobin to oilseed rape, spring and winter in the Central Zone of the European Union: FOCUS Step 1-3 calculations. Exponent International Ltd.. Report No.: 2202311.UK0-7229 Non-GLP, unpublished	N	XXXX
KCP 9.2.5/02	Jones, B. & Hilton, M.	2024	Predicted environmental concentrations in surface water after post-emergence application of mandestrobin to oilseed rape, spring and winter in the Central Zone of the European Union – Additional Step 4 modelling: FOCUS Step 3-4 calculations Exponent International Ltd.. Report No.: 2202311.UK0-6711 Non-GLP, unpublished	N	XXXX

* XXXX.

List of data submitted or referred to by the applicant and relied on, but already evaluated at EU peer review

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	Owner
KCP XX	Author	YYYY	Title Company Report N Source GLP/non GLP/GEP/non GEP Published/Unpublished	Y/N	Owner

The following tables are to be completed by MS

List of data submitted by the applicant and not 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	Owner
KCP XX	Author	YYYY	Title Company Report N Source GLP/non GLP/GEP/non GEP Published/Unpublished	Y/N	Owner

List of data relied on not submitted by the applicant but necessary for evaluation

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	Owner
KCP XX	Author	YYYY	Title Company Report N Source GLP/non GLP/GEP/non GEP Published/Unpublished	Y/N	Owner

Appendix 2 Detailed evaluation of the new Annex II studies (evaluated in previous Registration Report)

A 2.1 Study 1: Gilbert 2016

Previous conclusion from zRMS FR (2021)

The study was already assessed by RMS AT in a zonal assessment of the representative formulation. The RMS AT did not agree with the kinetic assessment provided by the applicant since the fits following SFO kinetics were not deemed to be acceptable and the respective residuals indicated a systematic error for all four soils. The bi-phasic kinetics provided a much better fit and lower χ^2 values and therefore, the ZRMS AT is of the opinion that the slow phase degradation rate from the DFOP kinetics should be used for modelling.

The consideration of the new study will result in a new geometric mean DT50 value to be used for modelling for the parent S-2200 and for the metabolite 5-COOH-S-2200. The formation fraction of the parent to the metabolite 5-COOH-S-2200 will also be revised.

zRMS FR agrees with AT that biphasic kinetics should be used for all four soils. AT fittings are considered as conservative and should be used for both the active substance and metabolite 5-COOH-S-2200.

In three out of four soils, the metabolite DX-CA-S-2200 did not show a plateau or decline, fits are uncertain and RMS AT proposed that they should not be used. In the ultimate soil, the metabolite was not observed in significant levels.

zRMS FR notes that the metabolite DX-CA-S-2200 seems to be observed in levels triggering a risk assessment (minor, non transient metabolite) in acidic soils, and more especially, when looking at the residues of Graham and Gilbert (2011a, b, c and d) and results of the laboratory studies on photolytic degradation in soil provided in the DAR, for soils with pH CaCl₂<6.2.

zRMS FR agrees with the endpoints recommended by AT presented in Tables A2.1-8 and A2.1-9.

Reference: KCA 7.1.2.1.1 Gilbert, J. (2016).

Report [14C]Mandestrobin (S-2200): Aerobic Soil Metabolism and Degradation.
XXXX. Report ROM-0062.
Deviations:No
GLP:Yes (laboratory certified by UK National Authority)

Acceptability Acceptable with the kinetics update of AT

Soil samples (50 g) were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2. Mandestrobin (0.8 mg/kg) was added and units were maintained in the dark, at 20 ± 2°C under aerobic conditions for up to 120 days. The rate of transformation of mandestrobin was modelled using SFO kinetics and good fits were obtained for all four soils. The DT₅₀ of mandestrobin varied between 107 and 163 days.

Material and methods

1. Test Materials: Mandestrobin; (RS)-2-methoxy-N-methyl-2-[α-(2,5-xylyloxy)-o-tolyl]acetamide
[Benzyl-¹⁴C]Mandestrobin; specific activity 4.55/4.33 MBq/mmol

Description: Not stated

Lot/Batch: CFQ40467 and CFQ42324

Purity: radiochemical purity ≥98.9%

CAS#: 394657-24-0

- Stability of compound:** Radiopurity of the test substance was checked prior to study initiation
- 2. Soil:** Topsoil was collected from a site where there had been no pesticide use in at least the last 5 years. Soil was stored at $4 \pm 2^\circ\text{C}$ in the dark for *ca* 2 months prior to the initiation of the experiment in accordance with ISO 10381-6:1993(E).

Table A2.1-1: Characteristics of the soils used

Soil Characterisation	Newhaven	Kenslow	Barrow	Quilen
Classification USDA	Silt loam	Loam	Sandy clay loam	Loam
% Sand	27	38	52	35
% Silt	58	47	22	42
% Clay	15	15	26	23
% Organic carbon	3.7	3.2	3.5	3.0
pH (H ₂ O)	6.6	6.4	6.7	7.2
pH (0.01M CaCl ₂)	5.5	5.3	5.8	6.7
Cation exchange Capacity (mEq/100g)	22.4	21.3	35.8	32.9
Water holding capacity at pF2 (%)	40.3	39.8	68.2	36.8
Microbial Biomass ($\mu\text{g C/g soil}$) ¹	-	-	-	716.5

¹ Determined before soil dispensing

Study design and methods

1. Experimental Conditions

Soil samples (50 g) were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2. Mandestrobin (0.8 mg/kg) was added and units were maintained in the dark, at $20 \pm 2^\circ\text{C}$ under aerobic conditions for up to 120 days. Air drawn over the surface of the units was passed through a series of traps (ethanediol, paraffin in xylene and two sodium hydroxide traps) to collect evolved radiolabelled volatiles.

2. Sampling

Duplicate units of each soil were removed for analysis immediately after test substance application and at 7, 14, 30, 59 and 120 days after treatment.

3. Description of analytical procedures

Soil samples were extracted twice with acetone:water (9:1 v/v) and further residues with acetone. Soils were further extracted with acetone:0.1 M hydrochloric acid (5:1, v/v) and acetone. Radioactivity was determined by LSC. Subsamples of each concentrated extract were analysed HPLC and selected samples confirmed by TLC.

Radioactivity trapped in NaOH was confirmed to be CO₂ by barium chloride precipitation. Unextractable soil residues were determined by combustion/LSC.

Results

Small amounts of radioactivity reported in unknowns (each $\leq 2.7\%$) and unresolved background have not been included in Tables below. These results in slight discrepancies between the mass balance and the sum of the components listed in the tables. However this is not considered to be of concern.

The mean recovery of applied radioactivity for the samples treated with [¹⁴C]mandestrobin, ranged from 94 to 101%.

Table A2.1-2: Percent recovery of applied radioactivity in Newhaven soil following application of [Benzyl-¹⁴C]mandestrobin under aerobic conditions

Time (day)	Mandestrobin	(R/S)-MCBX	2-COOH-S-2200	5-COOH-S-2200	De-Xy-S-2200	DX-CA-S-2200	Unextracted	CO ₂	Total
0	98.6	ND	ND	ND	0.5	0.8	0.3	-	100.3
0	100.1	ND	ND	ND	0.8	0.1	0.3	-	101.4
7	83.7	0.3	1.8	4.6	0.2	1.1	5	1.9	99.5
7	83.2	0.3	2	4.9	ND	0.8	5.3	2.6	100.5
14	73.9	0.4	2.7	6.4	ND	2.4	7.2	3.6	97.7
14	74.3	0.5	2.4	7.3	ND	2	7.7	3.9	99.7
30	68.4	0.5	3.3	6.9	1	2.9	9.8	3.9	98.3

30	68.6	0.6	3.1	7.9	ND	3.6	9.3	5.1	100.4
59	58.1	0.4	3.4	6.5	0.4	4.6	13.4	5.7	96.6
59	57.8	0.7	3.8	6.6	0.5	5.8	12.9	6.7	98.4
120	45.4	0.7	2.3	2.7	2.3	6.9	20.2	11.3	97.2
120	47.5	0.6	2.5	2.9	ND	9.6	18	10.2	98

Table A2.1-3: Percent recovery of applied radioactivity in Kenslow soil following application of [Benzyl-¹⁴C]mandestrobin under aerobic conditions

Time (day)	Mandestrobin	(R/S)-MCBX	2-COOH-S-2200	5-COOH-S-2200	De-Xy-S-2200	DX-CA-S-2200	Unextracted	CO ₂	Total
0	98.1	ND	ND	ND	0.2	0.4	0.2	-	99.2
0	99.3	ND	ND	ND	0.3	0.6	0.2	-	100.6
7	85.6	ND	1.4	4.3	0.1	0.5	4.1	1	97.9
7	88.7	ND	1.2	3.8	ND	0.7	3.5	1	99.2
14	79.4	0.2	1.8	5.7	ND	0.8	5.9	2.1	97.3
14	82.5	ND	1.5	5.3	ND	0.7	5.7	0.3	97.4
30	74.3	0.3	2.2	6.9	0.3	1.5	7.7	3.4	99.3
30	74.8	0.5	2.2	7	0.2	1.4	7.3	3.4	99.3
59	68.7	0.4	2.4	6.5	ND	3	10.4	3.3	98.4
59	66.1	0.4	2.7	7.7	0.1	3.4	10.8	4.4	98.7
120	57.2	0.5	2.8	5.9	0.7	4.7	16.4	5.7	97.6
120	60.3	0.6	2.2	5.2	ND	6.5	15.1	5.7	99.7

Table A2.1-4: Percent recovery of applied radioactivity in Barrow soil following application of [Benzyl-¹⁴C]mandestrobin under aerobic conditions

Time (day)	Mandestrobin	(R/S)-MCBX	2-COOH-S-2200	5-COOH-S-2200	De-Xy-S-2200	DX-CA-S-2200	Unextracted	CO ₂	Total
0	97.4	ND	ND	ND	0.4	0.3	0.4	NA	98.6
0	99.9	ND	ND	ND	ND	ND	0.4	NA	100.6
7	85.6	0.2	1.7	4.9	0.3	0.7	4.4	1.7	100.1
7	85.5	0.2	1.6	4.2	ND	0.8	4.4	1.6	99.2
14	76.4	0.4	2.6	6	ND	1.1	7.1	0.5	95.4
14	76	0.3	2.2	6.5	ND	1.1	7.5	3.5	97.6
30	70.7	0.5	3.1	7.8	ND	2.6	9.1	4.5	100.3
30	69.8	0.5	2.8	7.6	ND	2.3	9.2	4.8	99.2
59	60.4	0.4	3.4	7.6	0.2	3.1	13.5	7.5	98.4
59	59.2	0.4	3.4	7.1	0.3	2.4	14.1	8.7	98.3
120	52.2	0.6	2.8	5.8	ND	3.5	18.5	9.3	97.1
120	52.9	0.6	2.9	6.3	ND	3.7	18.4	11.1	99.2

Table A2.1-5: Percent recovery of applied radioactivity in Quilen soil following application of [Benzyl-¹⁴C]mandestrobin under aerobic conditions

Time (day)	Mandestrobin	(R/S)-MCBX	2-COOH-S-2200	5-COOH-S-2200	De-Xy-S-2200	DX-CA-S-2200	Unextracted	CO ₂	Total
0	97.9	ND	ND	ND	0.4	0.9	0.6	NA	100.3
0	97.5	ND	ND	ND	0.4	0.5	0.6	NA	99.6
7	83.1	0.2	1.4	2.8	0.1	0.1	11.1	1	100.6
7	82.9	0.1	1.1	2.6	0.3	0.1	11	1	99.4
14	77.5	0.3	1.6	3.6	0.1	0.3	13.8	1.3	98.8
14	77.4	0.2	1.6	3.9	ND	ND	13.7	0.7	97.9
30	68.6	0.3	1.6	4.5	ND	0.1	19.8	2.8	98.7
30	67.7	0.3	1.8	4.4	ND	0.1	19.3	2.6	97.1
59	56.7	0.4	1.8	4.8	ND	ND	25.6	3.7	93.7
59	58.1	0.3	1.4	5.2	ND	ND	26.8	4	96.3
120	48.2	0.3	1.2	5	ND	ND	30.9	8.4	94.2
120	49.8	0.3	1.1	4.6	ND	ND	30.5	6.1	93.1

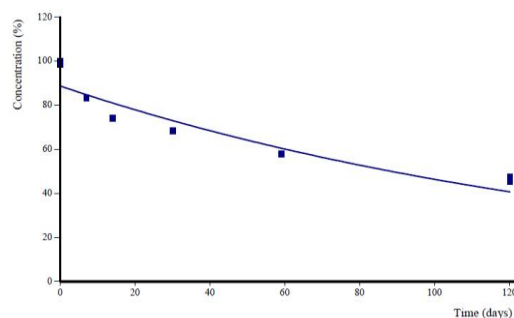
The metabolite 5-COOH-S-2200 was the most significant metabolite and reached a maximum of 7.7% before declining. As in the previous studies, the metabolites 2-COOH-S-2200, MCBX, De-Xy-S-2200

and DX-CA-S-2200 were all detected as low levels. However in this study DX-CA-S-2200 reached a maximum of 8.6% at the final timepoint.

Kinetic fittings provided by the applicant

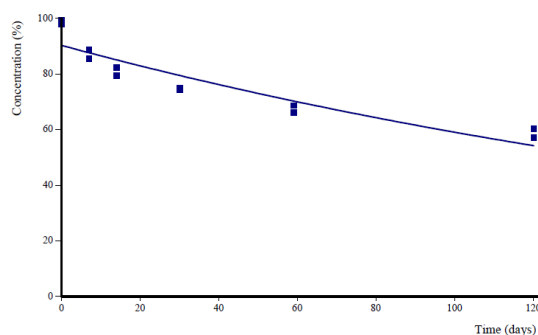
Newhaven

Observations and Fitted Model:



■ Observations — Fit

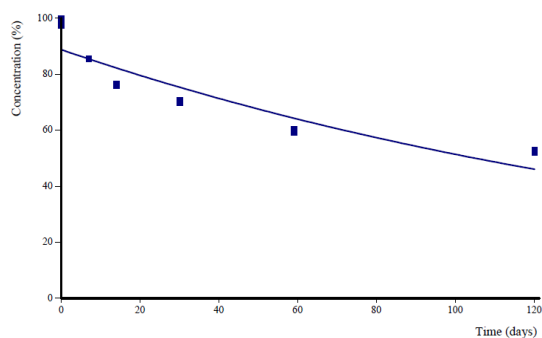
Observations and Fitted Model:



■ Observations — Fit

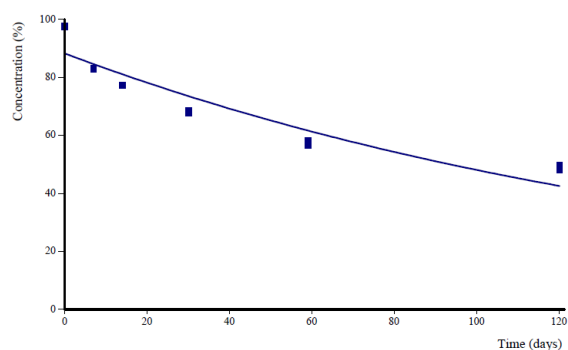
Barrow

Observations and Fitted Model:



■ Observations — Fit

Observations and Fitted Model:



■ Observations — Fit

Kenslow

Quilen

Degradation showed a good correlation with SFO kinetics and hence these were selected for modelling endpoints.

Table A2.1-6: DT₅₀ and DT₉₀ data for mandestrobin in three acidic and one neutral aerobic soils

Soil	pH (CaCl ₂)	DT ₅₀ (days)	DT ₉₀ (days)	χ^2 Err %	Prob. > t	R ²
Newhaven	5.5	107	355	6.76	1.56E-005	0.8729
Kenslow	5.3	163	542	4.95	1.87E-005	0.8539
Barrow	5.8	127	421	6.55	3.26E-005	0.8436
Quilen	6.7	114	379	6.26	1.18E-005	0.8772

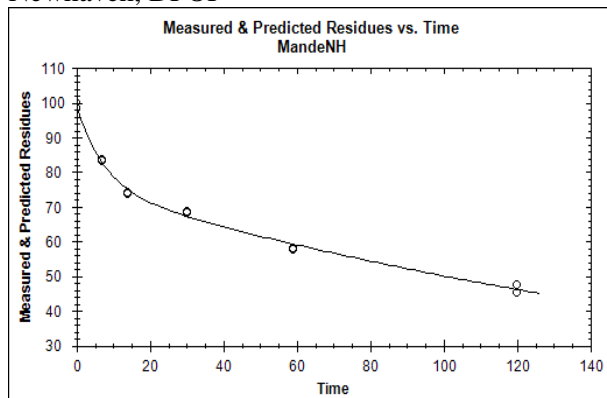
**The study has already been assessed by RMS AT in a zonal registration report of the preparation. Kinetic fittings were criticized, both by RMS AT and UK. The following kinetic fittings were therefore provided by RMS AT. Updated kinetics were performed using KinGUI v.2
Kinetic fittings provided by RMS AT**

Table A2.1-6: Parent kinetics

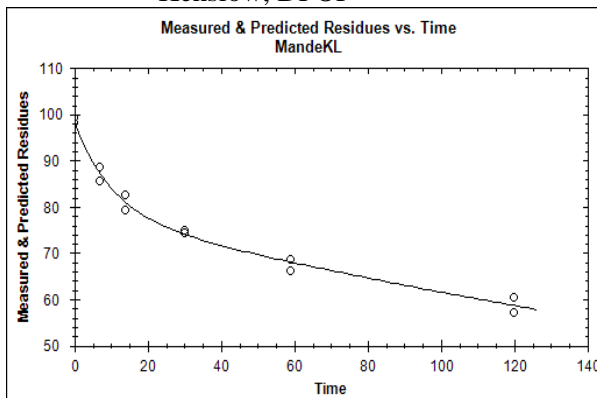
Parent only									
Soil	pH (CaCl ₂)	DT-50 (days)	DT50 modelling	DT-90 (days)	Kinetic parameters	χ^2 Err %	R ²	T-Test	kinetic
Newhaven	5.5	106.8	-	354.7	k: 6.492e-3	6.8	0.873	p <0.05	SFO
		106.6	-	>1000	α : 0.25984 β : 7.95275	1.9	0.991	α : 9.94e-7 β : 0.000864	FOMC
		101.4	168.98	493.7	k1: 1.294e-1 k2:4.102e-3 g: 2.421e-1	1.2	0.996	p k1 <0.05 p k2 <0.05	DFOP
		96.9	-	407.4	k1: 8.764e-2 k2: 5.184e-3 Tb: 2.316	3.5		p k1 <0.05 p k2 <0.05	HS
Kenslow	5.3	163	-	542	k: 0.004245	4.9	0.854	p <0.05	SFO
		362.27	-	>1000	α : 0.17638 β : 7.25928	0.6	0.989	α : 5.10e-7 β : 0.000896	FOMC
		190.93	285.6	853.99	k1: 9.678e-2 k2:2.427e-3 g: 2.052e-1	0.5	0.990	p k1 <0.05 p k2 <0.05	DFOP
		159.68	-	626.36	k1: 1.41e-1 k2: 3.449e-3 Tb: 1.036			p k1 <0.05 p k2 <0.05	HS
Barrow	5.8	126.8	-	421.3	k: 5.466e-3	6.6	0.844	p <0.05	SFO
		161.1	-	>1000	α : 0.21878 β : 7.07570	1.2	0.994	α : 2.77e-8 β : 0.000141	FOMC
		141.56	265.4	757.7	k1: 8.223e-2 k2:2.612e-3 g: 2.763e-1	1.6	0.993	p k1 <0.05 p k2 <0.05	DFOP
		118.9	-	487.8	k1: 5.963e-2 k2: 4.363e-3 Tb: 3.156	4.5	0.952	p k1:0.49 p k2:0.21	HS
Quilen	6.7	114	-	379	k: 6.079e-3	6.3	0.877	p <0.05	SFO
		124.16	-	60348	α : 0.26333 β : 9.62086 (>1000)	1.0	0.997	α : 7.69e-9 β : 2.63e-5	FOMC
		120.28	228.4	650.56	k1: 7.361e-2 k2: 3.035e-3 g: 2.798e-1	1.6	0.994	p k1 <0.05 p k2 <0.05	DFOP
		105 (140)	-	431		4.0	0.968	p k1 <0.05 p k2 <0.05	HS

DFOP kinetics were considered as best-fit for all four soils. Graphs are presented below.

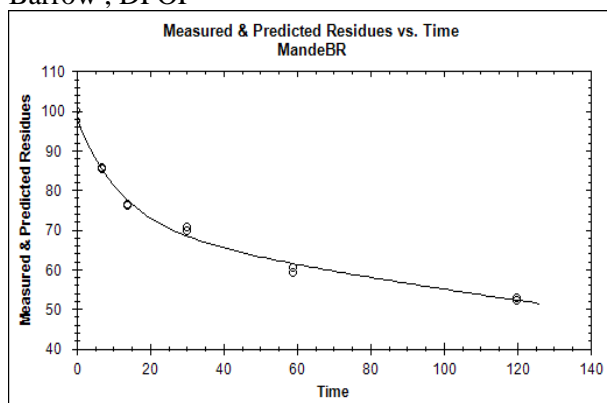
Newhaven, DFOP



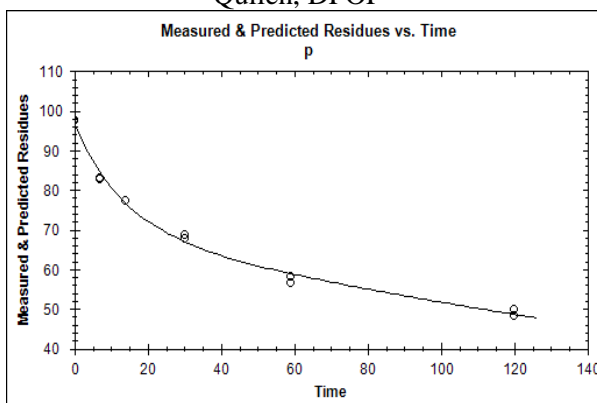
Kenslow, DFOP



Barrow , DFOP



Quilen, DFOP



RMS AT also performed kinetic fittings considering metabolite 5-COOH and DX-CA in the degradation scheme as these two metabolites were observed at significant levels results are presented below.

Table A2.1-7: Parent-metabolites kinetics

Parent and its metabolites 5-COOH-S-2200 and DX-CA-S-2200											
Soil	pH (CaCl ₂)	Substance	DT ₅₀ (d)	DT ₅₀ modelling (d)	DT ₉₀ (d)	Kinetic parameters	ff (t-test)	χ ² %	R ²	t-Test	kinetic
Newhaven	5.5	S-2200	102.4 ₉	176.24	511.6 ₈	k1: 0.1096 k2: 0.00393 g: 0.2518	p to 5-COOH: 0.3585 (2.83e-13) p to DX-CA: 0.1309 (4.21e-6)	3.02 1.38*	0.995 6	p k1 < 0.05 p k2 < 0.05 g: 1.35e14	DFO P
		5-COOH-S-2200	29.54	29.54	98.12	k: 0.02347		7.91	0.952 9	p < 0.05	DFO P-SFO
		DX-CA-S-2200	>1000	-	>>1000	k: 4.68e-9		21.11	0.857 0	p = 0.5	DFO P-SFO
Kenslow	5.3	S-2200	186.8 ₇	273.65	822.1 ₇	k1: 0.09706 k2: 2.533e-3 g: 0.1973	p to 5-COOH: 0.3644 (6.87e-13)	1.97 0.53*	0.989 9	p k1 < 0.05 p k2 < 0.05	DFO P

Parent and its metabolites 5-COOH-S-2200 and DX-CA-S-2200											
Soil	pH (CaCl ₂)	Substance	DT ₅₀ (d)	DT ₅₀ modelling (d)	DT ₉₀ (d)	Kinetic parameters	ff (t-test)	χ ² %	R ²	t-Test	kinetic
							p to DX-CA: 0.1067 (0.000402)			g: 3.72e-11	
		5-COOH-S-2200	60.36	60.36	200.5	k: 1.148e-2		2.2	0.9833	p < 0.05	DFO P-SFO
		DX-CA-S-2200	>1000	-	>1000	k: 6.127e-9		35.1	0.7627	p = 0.5	DFO P-SFO
Barrow	5.8	S-2200	134.53	231.36	671.66	k1:0.09669 k2:0.002996 g:0.2518	p to 5-COOH: 0.32721 (5.47e-15) p to DX-CA: 0.07394 (8.2e-9)	2.631.66*	0.9920	p k1 < 0.05 p k2 < 0.05 g: 1.83e-13	DFO P
		5-COOH-S-2200	58.962	58.962	195.87	k: 0.01176		1.61	0.9902	p < 0.05	DFO P-SFO
		DX-CA-S-2200	>>1000	-	>>1000	k: 3.412e-9		12.22	0.9383	p = 0.5	DFO P-SFO
Quilen	6.7	S-2200	112.45	178.94	527.94	k1:0.1219321 k2:0.0038736 g: 0.2270836	p to 5-COOH: 0.18044 (1.91e-14)	3.3772.062*	0.9907	p k1 < 0.05 p k2 < 0.05 g: 4.48e-11	DFO P
		5-COOH-S-2200	87.54	87.54	290.79	k: 0.007918		1.884	0.9910	p < 0.05	DFO P-SFO
		DX-CA-S-2200	- (not significantly observed <1% AR)								

*all data/S-2200

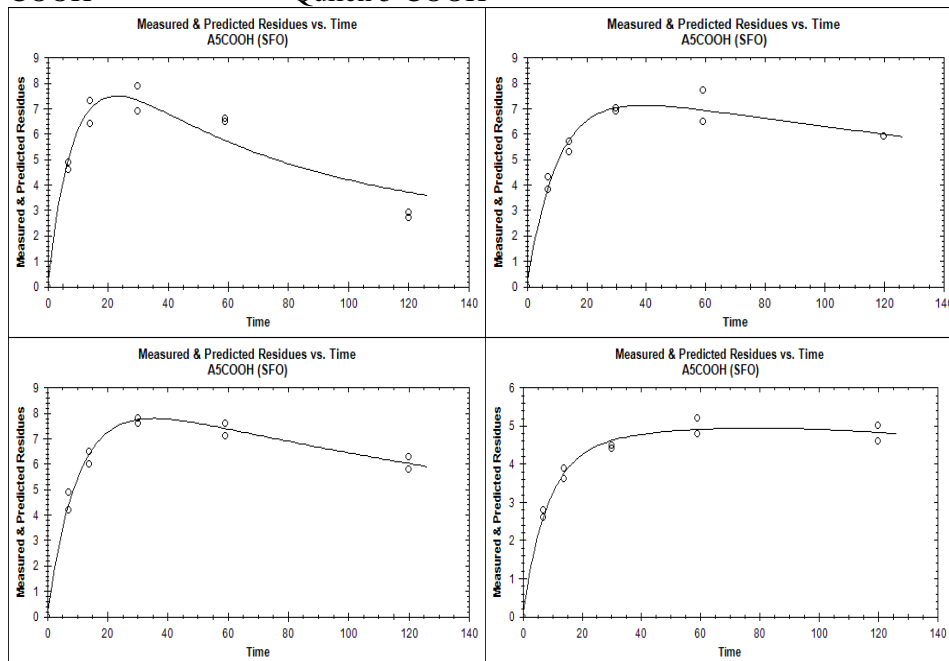
Considering parent substance degrades following DFOP kinetics, the following graphs were obtained for the metabolites. All graphs below represent DFOP (parent) -> SFO (metabolite)

Newhaven 5-COOH
 COOH

Quilen 5-COOH

Kenslow 5-COOH

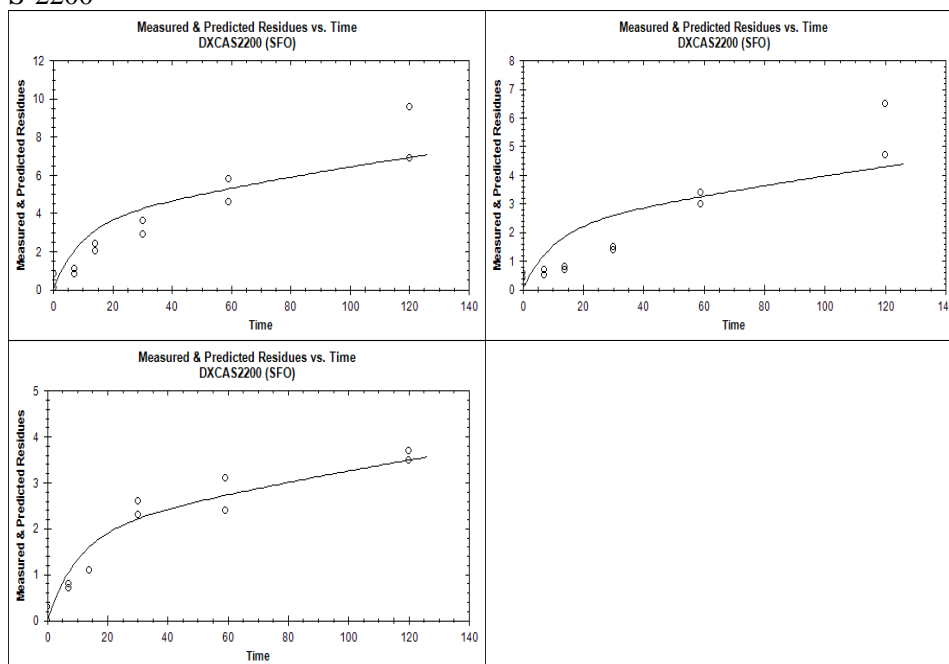
Barrow 5-



Newhaven DX-CA-S-2200
 S-2200

Kenslow DX-CA-S-2200

Barrow DX-CA-



Summary of recommended endpoints (RMS AT):

Table A2.1-8: recommended endpoints for mandestrobin from Gilbert (2016)

Parent	Aerobic conditions						
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	DT ₅₀ (d) 20°C pF2/10kPa Modelling endpoint*	χ ² error (%)	Method of calculation
Kenslow	3.2	5.3	20°C/pF2	187/822	273.7	0.53	DFOP
Newhaven	3.7	5.5	20°C/pF2	103/512	176.2	1.38	DFOP
Barrow	3.5	5.8	20°C/pF2	135/672	231.4	1.66	DFOP
Quilen	3	6.7	20°C/pF2	112/528	178.9	2.06	DFOP

*DFOP: slow rate k value

Table A2.1-9: recommended endpoints for 5-COOH-S-2200 from Gilbert (2016)

5-COOH-S-2200	Aerobic conditions							
Soil type	OC%	pH (CaCl ₂)	t. °C / % MWHC	DT ₅₀ /DT ₉₀ (d)	f.f	DT ₅₀ (d) 20°C pF2/10kPa	χ ² error (%)	Method of calculation
Newhaven	3.7	5.5	20°C/pF2	29.5/98.1	0.3585	29.5	7.9	DFOP-SFO
Kenslow	3.2	5.3	20°C/pF2	60.4/200.5	0.3644	60.4	2.2	DFOP-SFO
Barrow	3.5	5.8	20°C/pF2	59.0/195.9	0.3272	59.0	1.6	DFOP-SFO
Quilen	3	6.7	20°C/pF2	87.5/290.8	0.18044	87.5	1.9	DFOP-SFO

A 2.2 Study 2: Kodaka, 2016

zRMS Comments:	The zRMS AT comment: <i>The new degradation study (Kodaka et al, 2016) showed some deficiency, however it was considered acceptable for the PECgw calculations.</i> No further comments.
-------------------	--

Previous conclusion from zRMS FR (2021)

In previous dossiers (PAMN 2017-1546 and PMOD 2019-4604), this study was not provided by the applicant to FR. It was however provided to RMS AT in a zonal registration report of the representative formulation and FR relied on the assessment from RMS AT.

RMS AT mentioned in his report that the study showed some deficiencies, however it was considered acceptable for the PECgw calculations. With the information provided by RMS AT, FR agreed with the endpoints recommended by AT. Since it was just a preliminary study not subject to GLP, it was recommended that a new study according to GLP is provided to confirm the DT50 of the metabolite DX-CA-S-2200.

Within the current dossier (PMOD 2020-2405), this study report has been made available to FR. In addition, the applicant provided a new GLP study on the degradation of metabolite DX-CA-S-2200 (see Lamond 2017). The non-GLP preliminary study Kodaka 2016 is now considered as supportive only.

zRMS FR notes that volatile trapping was not performed at every sampling time, therefore the total mass balance is only available from DAT3 onward for California soil and German 2.2 experiments and DAT1 onward for German 2.3 soil experiment. However this has no significant impact on the results.

For information, the applicant kinetic fittings for the German 2.3 soil were not performed considering the residues provided in the study report and are therefore not correct. No update is deemed necessary since the study is only considered as supportive.

Reference: Report	KCA 7.1.2.1.2/01 Kodaka, R., Abe, J., Fujisawa, T. (2016) Aerobic Soil Metabolism Study of [phenyl- ¹⁴ C]De-Xy-S-2200 (preliminary study) XXXX. Report ROM-0063 GLP: No
Acceptability	Supportive

In the new aerobic soil degradation study of mandestrobin (Gilbert, 2016) described above the metabolite DX-CA-S-2200 exceeded 5 % AR on two consecutive sampling points in one of the four soils and reached its maximum at the end of the study (8.3 % AR, 5.6 % AR, 3.6 % AR) in three of the four soils. Therefore, according to the GD on the assessment of the relevance of GW-Metabolites (SANCO/221/2000-rev.10), its potential for leaching to groundwater has to be assessed. One of the input parameters required for FOCUS PECgw models is the rate of degradation in soil. Thus, a respective study was provided by the applicant. A new aerobic soil metabolism study investigating the fate and behaviour of DX-CA-S-2200 in soil was submitted by the applicant and is summarised below.

Soil samples (20 g) were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2.0 (2 German soils) and pF 2.5 (1 US soil). De-Xy-S-2000 (0.03 mg/kg) was added and units were maintained in the dark, at 20°C (2 German soils) or 25°C (1 US soil) under aerobic conditions for up to one month. The rate of transformation of De-Xy-S-2200 and DX-CA-S-2200 was modelled using SFO kinetics. The DT₅₀ of De-Xy-S-2200 was below 1 day and the DT₅₀ of DX-CA-S-2200 varied between 1.74 and 18.5 days.

Material and methods

- Test Materials:** [phenyl-¹⁴C]De-Xy-S-2200, (R/S)-2-(2-hydroxymethylphenyl)-2-methoxy-N-methylacetamid
specific activity 4.22 GBq/mmol (20.2 MBq/mg) ,
radiochemical purity 98.9 % (HPLC),
chemical purity 100 %

- Reference substances:** De-Xy-S-2200, chemical purity 100 %
DX-CA-S-2200, chemical purity 99.6 %
- 2. Soil:** US California soil and the two German standard soils were supplied by Valent USA and landwirtschaftliche Untersuchungs- und Forschungsanstalt Speyer in Germany. Soils were air-dried and passed through a 2 mm sieve before use.

Table A2.2-1: Characteristics of the soils used

Soil Characterisation	US California soil	German 2.2 soil	German 2.3 soil
Classification USDA	Loamy Sand	Loamy Sand	Sandy Loam
% Sand	81	79.1	61.0
% Silt	13	13.5	29.8
% Clay	6	7.9	9.2
% Organic carbon	0.3	2.36	1.02
pH (0.01M CaCl ₂)	7.5 ^a	5.6	6.2
Cation exchange Capacity (mEq/100g)	5	11	9

^a based on pH_{H2O} 8.0 as mentioned in the study report and with the conversion $\text{pH}_{\text{H}_2\text{O}} = 0.982\text{pH}_{\text{CaCl}_2} + 0.648$ (EFSA Guidance for predicting environmental concentrations in soil; 2017 – from Boesten *et al.*, 2012)

Soil samples (20 g) were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2 (2 German soils) or pF 2.5 (US soil). Soil samples were incubated for 2 weeks at 20 °C (2 German soils) or 25°C (US soil) in the dark. Moisture level was maintained up to every two weeks. [phenyl-¹⁴C]De-Xy-S-2200 (0.03 mg/kg) was added and units were maintained in the dark, at 20 ± 2°C or 25°C under aerobic conditions for up to one month. This corresponds to an application of 22.5 g/ha of De-Xy-S-2200 based on a default bulk density of 1.2 g/cm³ and a depth of 5cm.

Air drawn over the surface of the units was passed through a series of traps (ethylene glycol, sodium hydroxide) to collect evolved radiolabelled volatiles.

Single (US soil, German 2.2 soil) or duplicate (German 2.3 soil) units of each soil were removed for analysis at 0, 1/6 (4 h), 1, 3, 8 and 30 days after treatment for German 2.2 soil and at 0, 1/6 (4 h), 1, 3, 8, 17 and 30 days after treatment for US and German 2.3 soil.

Soil samples were extracted three times with acetone:water (9:1 v/v) twice with acetone/0.1 M HCl (5:1, v/v) followed by wash with acetone once for all sampling intervals. Radioactivity was determined by LSC. Subsamples of each concentrated extract were analysed by HPLC.

Radioactivity trapped in alkaline trap was confirmed to be CO₂ by barium chloride precipitation. Unextractable soil residues were determined by combustion/LSC.

Results

The mean recovery of applied radioactivity for the samples treated with [¹⁴C]De-Xy-S-2200, ranged from 88.2 to 100.5 %.

Table A2.2-2: Percent recovery of applied radioactivity in US California soil following application of [phenyl-¹⁴C]De-Xy-S-2200 under aerobic conditions

Time (day)	Aceton/H ₂ O		Aceton/HCL		Both extracts			Unextracted	CO ₂	Total
	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	Others			
0	97.6	nd	na	na	97.6	nd	nd	<0.1	na	97.6
1/6	74.8	18.5	nd	2.9	74.8	21.4	0.7	0.5	na	97.8
1	nd	34.2	nd	2.6	nd	36.8	50.5	8.0	na	95.3
3	nd	51.7	nd	4.0	nd	55.7	29.6	7.2	8.0	100.5
8	nd	21.9	nd	1.3	nd	23.2	32.8	19.3	22.9	98.2
17	nd	0.3	nd	nd	nd	0.3	16.3	35.3	36.3	88.2
30	nd	na	nd	nd	nd	nd	32.2	32.2	42.8	89.9

nd...not detected

na...not analysed

Table A2.2-3: Percent recovery of applied radioactivity in German 2.2 soil following application of [phenyl]-¹⁴C]De-Xy-S-2200 under aerobic conditions

Time (day)	Aceton/H ₂ O		Aceton/HCL		Both extracts			Unextracted	CO ₂	Total
	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	Others			
0	95.0	nd	na	na	95.0	nd	nd	0.2	na	95.2
1/6	5.7	88.1	nd	4.3	5.7	92.4	nd	0.8	na	98.8
1	nd	86.4	nd	6.5	nd	92.9	nd	4.8	na	97.7
3	nd	77.6	nd	7.8	nd	85.4	2.7	5.5	1.3	95.0
8	nd	69.2	nd	9.4	nd	78.6	nd	11.0	4.7	94.4
30	nd	19.6	nd	6.0	nd	25.6	1.1	49.7	23.5	99.8

nd...not detected

na...not analysed

Table A2.2-4: Percent recovery of applied radioactivity in German 2.3 soil following application of [phenyl]-¹⁴C]De-Xy-S-2200 under aerobic conditions

Time (day)	Aceton/H ₂ O		Aceton/HCL		Both extracts			Unextracted	CO ₂	Total
	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	De-Xy-S-2200	DX-CA-S-2200	Others			
0	99.8	nd	na	na	99.8	nd	nd	0.2	na	100.0
0	99.8	na	na	na	99.8	nd	na	0.2	na	100.0
0.1667	56.4	26.0	nd	7.1	56.4	33.1	4.5	2.9	na	96.9
0.1667	59.9	28.1	nd	6.9	59.9	35.0	3.1	2.6	na	100.6
1	24.0	19.5	1.3	8.6	25.3	28.1	23.8	15.6	5.0	97.7
1	24.5	8.8	0.7	8.5	25.2	17.3	34.0	15.4	4.3	96.4
3	2.0	16.5	nd	7.3	2.0	23.8	22.3	32.1	17.0	97.3
3	3.1	26.9	nd	9.1	3.1	36.0	26.3	21.3	14.3	101.0
8	nd	6.6	nd	8.7	nd	15.3	12.7	29.6	31.1	88.8
8	nd	14.5	nd	5.1	nd	19.6	12.7	31.2	28	91.5
17	nd	nd	nd	nd	nd	nd	8.3	46.8	39.1	94.2
17	nd	nd	nd	nd	nd	nd	10.8	37.6	38.1	86.3
30	na	na	na	na	na	na	na	41.4	43.6	92.7
30	na	na	na	na	na	na	na	39.3	43	91.2

nd...not detected

na...not analysed

Several unknown metabolites were observed, single metabolites represent a maximum of 27.5 % AR, 2.0 % AR and 16.8 % AR in US California soil, German 2.2 soil and German 2.3 soil. These unknowns were not considered as major metabolites and were not further analysed. These metabolites are not expected to exceed trigger values considering they are to be produced from the parent material via the production of De-Xy-S-2200.

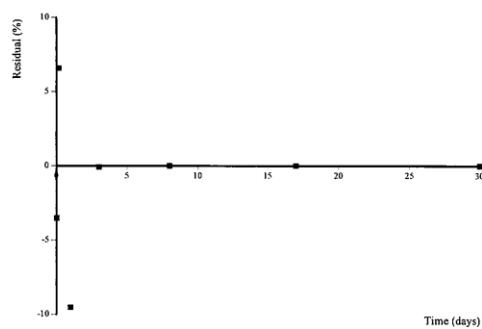
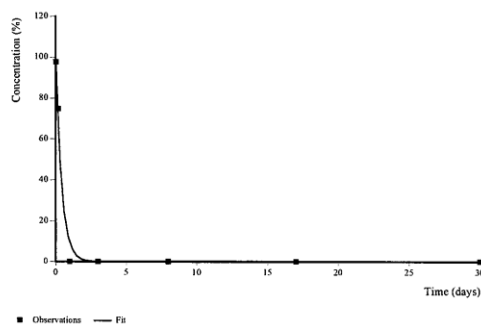
Degradation showed an acceptable correlation with SFO kinetics and hence these were selected for modelling endpoints. Table 9.1.1./01/6 shows the DT50 values obtained in each soil.

Table A2.2-5: DT₅₀ and DT₉₀ data for De-Xy-S-2200 and DX-CA-S-2200 in three aerobic soils

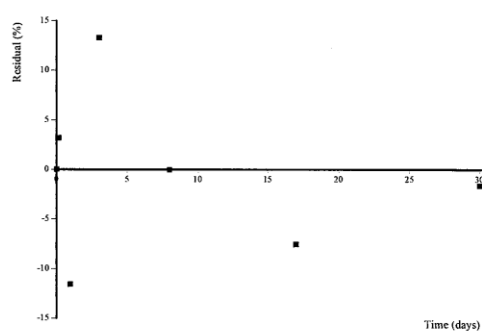
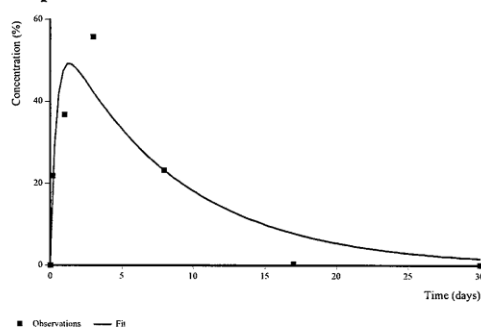
Soil	pH	DT ₅₀ (days)	DT ₉₀ (days)	χ ² Err %	R ²	ff
De-Xy-S-2200						
US California	8.0 (H ₂ O)	0.294	0.976	14.8	0.9873	-
German 2.2	5.6 (CaCl ₂)	0.0411	0.136	1.44E-5	1	-
German 2.3	6.2 (CaCl ₂)	0.404	1.34	16.9	0.9693	-
DX-CA-S-2200						
US California	8.0 (H ₂ O)	5.75	19.1	27.6	0.8641	0.5721
German 2.2	5.6 (CaCl ₂)	18.5	61.6	5.09	0.9901	0.9971
German 2.3	6.2 (CaCl ₂)	1.74	5.76	27.2	0.4138	0.6163
Geomean		5.70		Arithmetic mean		0.7285

Visual fittings :
US California, SFO-SFO fit

Compartment Parent:

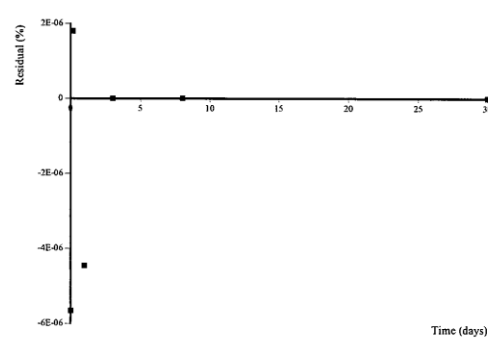
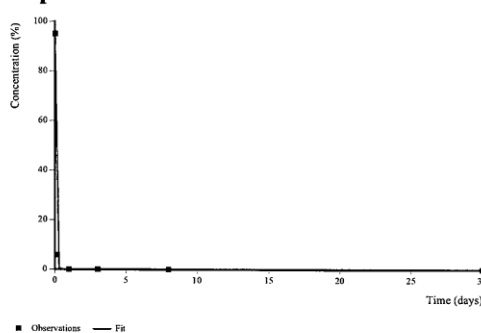


Compartment A1:

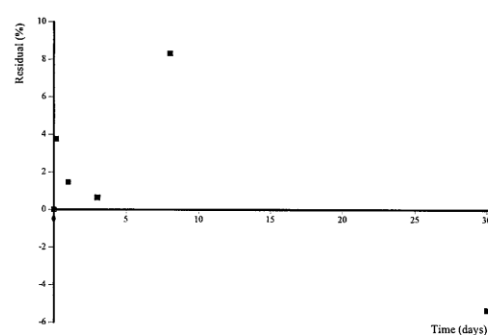
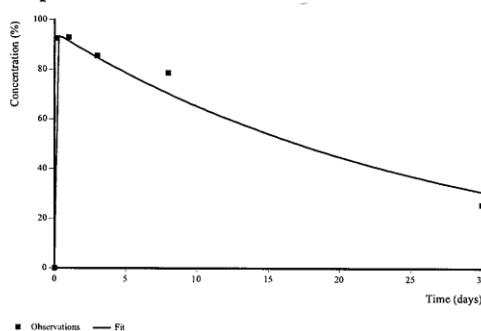


German 2.2 SFO-SFO fit

Compartment Parent:

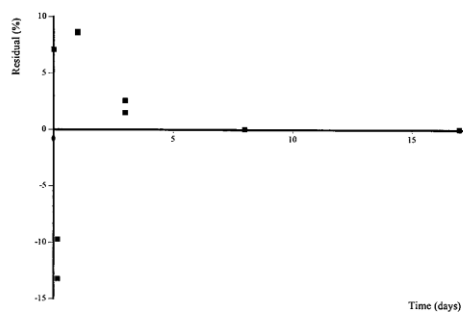
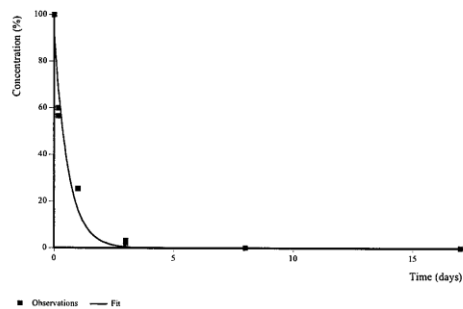


Compartment A1:

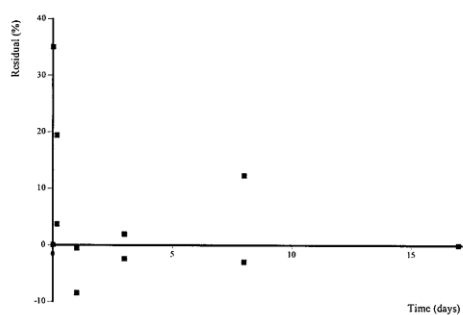
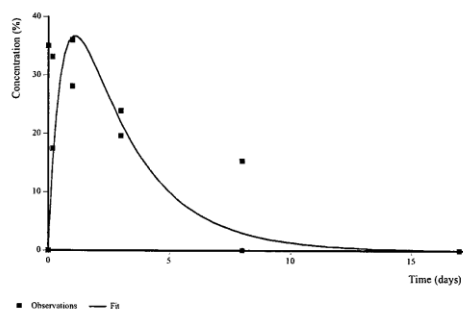


German 2.3 SFO-SFO fit

Compartment Parent:



Compartment A1:



A 2.3 Study 3: Lamond (2017)

zRMS Comments:	The endpoints from laboratory tests, recommended by zRMS-AT/FR from the degradation study are considered as appropriate input parameters for the modelling.
----------------	---

Previous conclusion from zRMS FR (2021)

No major deviation to the OECD 307 guideline was observed. For soil South Witham, it is noted that mass balance at day 7 for one of the replicate is <90% AR. This is explained by “an apparent failure to trap all evolved volatiles from this incubation unit”. This has no impact on the reliability of the concentrations obtained for De-Xy-S-2200 and DX-CA-S-2200.

Before kinetic fitting, the data were not pre-processed as recommended in FOCUS Kinetic guidance (initial value for parent and metabolite, values below LOD). However in this case, zRMS considers that this has no significant impact on the obtained DT50 values.

Contrary to the opinion of the applicant, zRMS estimates that all data should be considered for the kinetic fitting of results from soil RefeSol 02-A, since there is no reliable reason to exclude one of the replicates. This is also in agreement with the conclusions from RMS AT.

The following persistence and modelling DT50 values for DX-CA-S-2200 can be used for risk assessment: 5.2, 3.2 and 8.1 days.

Reference:	KCA 7.1.2.1.2/02
Report	Lamond, P. (2017). [¹⁴ C]De-Xy-S-2200: Aerobic Soil Metabolism Study in Three Soils. Smithers Viscient (ESG) Ltd. Study No. 3201768; Report ROM-0070
Guideline(s):	OECD 307 (April, 2002)
Deviations:	None
GLP:	Yes
Acceptability:	Yes

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Materials: [Benzyl-14C]De-Xy-S-2200; specific activity 4.55 GBq/mmol
 - Description: Not stated
 - Lot/Batch: RIS2017-001
 - Purity: Radiochemical purity 99.7 %
 - CAS#: Not stated
 - Stability of compound: The test substance had adequate stability under reflux conditions with 5:1 (v/v) acetone : 0.01M HCl (HPLC analysis)
2. Soil: Topsoil (0-20 cm) was collected from a site where there had been no pesticide use in the last 5 years. Sieved soils (2 mm) was stored at 4 ± 2°C in the dark prior to use in accordance with ISO 10381-6:1993(E). Soil characteristics are shown below.

Soil Characterisation	Calke	South Witham	RefeSol 02-A
Particle size distribution			
Classification ¹	Sandy clay loam	Clay	Silt loam
% Sand	59	35	4
% Silt	20	21	80
% Clay	21	44	16

% Organic carbon	2.4	2.8	1.0
pH (0.01M CaCl ₂)	5.2	7.5	6.6
Cation exchange Capacity (mEq/100g)	17.5	26.9	53.4
Water holding capacity at pF2 (%)	23.4	32.4	34.5
Microbial Biomass (µg C/g soil) ²			
Start (0 DAT)	605 (2.5)	816 (2.9)	269 (2.7)
End (28 DAT)	636 (2.7)	843 (3.0)	252 (2.5)

¹ USDA Particle Size Distribution and Classification

² value in bracket is expressed as % Organic Carbon; the biomass determination at 28 DAT was performed in the control units.

B. STUDY DESIGN AND METHODS

1. Experimental Conditions

Soil samples (50 g dry weight equivalent) were weighed into individual incubation vessels, and adjusted to pF 2 and maintained in the dark at 20 ± 2°C for a pre-incubation period of 10 days.

0.107 mg De-Xy-S-2200/kg soil was added to each vessel. The incubation vessels were connected to a series of liquid traps (ethanediol and NaOH) to collect evolved radiolabelled volatiles. The samples were incubated for up to 28 days in the dark at 20 ± 2°C and pF2.

2. Sampling

Duplicate samples were taken immediately after application and at 4 hr, 1, 2, 4, 7, 12, and 20 DAT; for the Calke soil an additional sample was taken at 28 DAT.

3. Description of analytical procedures

The soil samples were extracted twice with acetone: 0.1 M HCl (5:1 v/v) followed by an acetone rinse and then three extractions with acetone: 0.1 M (NH₄)₂CO₃ (4:1 v/v). The acidic extractions and the acetone rinse were combined and quantified by LSC. The basic extractions were combined and quantified by LSC. The extracts were concentrated and analysed by HPLC to quantify De-Xy-S-2200 and degradation products. Selected samples were analysed using TLC to confirm the identity of parent and degradation products.

Reflux extraction and bound residue fractionation analyses were required as unextracted residues were present at > 10% applied radioactivity for all soil types.

Reflux extraction (5/1 v/v acetone/0.01M HCl, 4 hours) was performed on one soil residue sample from each soil, from the final timepoints, following stability evaluation of the test substance.

Bound residue fractionation was performed on one soil residue sample from each soil, from the final timepoints. Following the reflux extraction analysis, the soil residue samples were shaken with NaOH solution (0.5 M, 24 hours). The residue (humic fraction) and supernatant were separated by centrifugation. The humic fraction was further washed twice with 0.5 M NaOH solution and the washings added to the original supernatant. The supernatant was acidified to *ca* pH 1 with HCl (5 M) and the precipitate formed was washed with HCl (0.1 M). The suspension was centrifuged to separate the precipitate from the wash. The supernatant and wash were combined (fulvic acid fraction) and the washed precipitate (humic acid fraction) was reconstituted in 0.5 M NaOH solution.

Radioactivity in the fulvic acid and reconstituted humic acid fractions was determined by LSC. Radioactivity in the humic fraction was determined by combustion followed by LSC.

Radioactivity in the trapping solutions was quantified by LSC. At the end of the incubation period, representative samples were analysed using BaCl₂ solution to confirm ¹⁴CO₂ by precipitatin.

II. RESULTS AND DISCUSSION

Table A.2.3-1: Calke soil treated with [¹⁴C]De-Xy-S-2200

Sampling interval (DAT)	0		0.167		1		2		4		7		12		20		28	
Vessel code	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12	A13	A14	A15	A16	A17	A18
Acidic extract	98	97.2	94.4	94.6	89.5	86.8	78.3	78.4	57	60.1	47.5	40.7	20.2	24.9	16.7	22.5	12.8	9.2
Basic extract	2.8	2.7	3.5	3.8	5.6	5.7	7.4	7.5	5.7	6.3	5.1	4.8	2.9	3.4	2.4	3.1	2.1	1.8
De-Xy-S-2200	92.5	90.5	38.2	36.7	2.1	2.1	1.4	1.7	0.5	ND								
DX-CA-S-2200	0.3	0.9	48.6	50.1	84.1	82.2	74.8	73.4	48.9	53.5	41.2	32.4	13.8	17.2	10.2	14.7	8.4	5.1
Unknown AE1	2.2	2.7	1.1	0.8	ND	ND	ND	ND	0.5	0.6	ND	ND	ND	ND	ND	ND	ND	ND
Other Unknowns	2.3	2.7	6.2	6.6	8.7	8.0	9.5	10.4	11.7	11.9	10.9	8.3	6.2	7.5	6.4	7.8	4.3	4
Unresolved Backgrounds	0.6	0.4	0.3	0.6	0.3	0.2	0.1	0.4	1.2	0.5	0.5	0.1	0.2	0.2	0.1	0.0	0.1	0.1
Total extractibles	98	97.2	94.4	94.6	95.1	92.5	85.7	85.9	62.7	66.4	52.6	40.7	20.2	24.9	16.7	22.5	12.8	9.2
Ethanediol trap	ND																	
NaOH trap	NA	NA	0.2	0.1	1.4	1.9	4.0	4.2	10	6.3	19.8	24.2	35.0	33.4	40.6	37.3	44.6	47.8
Residues	NA	0.1	0.9	0.9	4.9	5.3	9.4	9.0	19.5	18.8	25.5	28.9	37.3	35.8	37.6	34.2	33.5	37.3
Mass balance	100.8	100	99	99.4	101.4	99.7	99.1	99.1	92.2	91.5	97.9	98.6	95.4	97.5	97.3	97.1	93.0	96.1

NA = Not applicable, ND = Not detected (or < 0.1% AR)

Table A.2.3-2: South Witham soil treated with [¹⁴C]De-Xy-S-2200

Sampling interval (DAT)	0		0.167		1		2		4		7		12		20	
Vessel code	B1	B2	B3	B4	B5	B6	B7	B8	B9	B10	B11	B12 ^a	B13	B14	B19	B16
Acidic extract	96.1	97.2	80.0	83.1	47.4	47.8	32.1	31.0	24.2	22.2	11.9	17	4.9	5.8	3.6	3.8
Basic extract	3.6	3.8	9.6	8.8	11.4	10.9	11.3	10.0	7.2	6.7	4.4	7.4	1.9	2.2	1.4	1.5
De-Xy-S-2200	95	95.7	28.9	34.0	ND										^{-b}	^{-b}
DX-CA-S-2200	ND	ND	30.6	25.6	40.0	40.4	30.1	26.7	20.9	18.9	6.8	15.5	1.7	2.2	^{-b}	^{-b}
Unknown AE1	ND	ND	1.0	2.0	9.4	9.6	6.5	8.7	3.7	2.8	1.8	3.5	1.0	1.4	^{-b}	^{-b}
Other Unknowns	0.8	1.4	28.5	29.6	9.0	8.6	6.5	5.3	6.6	7.2	3.1	5.2	2.3	2.2	^{-b}	^{-b}
Unresolved Backgrounds	0.3	0.1	0.7	0.7	0.4	0.1	0.3	0.3	0.2	0.1	0.2	0.2	0.0	0.0	^{-b}	^{-b}
Total extractibles	96.1	97.2	89.6	91.9	58.8	58.7	43.4	41.0	31.4	28.9	11.9	24.4	4.9	5.8	^{-b}	^{-b}
Ethanediol trap	ND															
NaOH trap	NA	NA	0.2	0.2	5.5	4.5	14.0	13.1	24.2	16.6	33.3	3.2	41.5	39.3	44.4	41.2
Residues	0.3	0.4	10.2	8.8	33.9	31.9	36.1	37.4	39.6	39.9	46.5	43.6	49.5	49.4	47.1	43.2
Mass balance	100.0	101.4	100.0	100.9	98.2	95.1	93.5	91.5	95.2	85.4	96.1	71.2	97.8	96.7	96.5	89.7

NA = Not applicable, ND= Not detected (or < 0.1% AR)

^a There was an apparent failure to trap all evolved volatiles from this incubation unit

^b Acidic and basic extracts at 20 DAT contained < 5% applied radioactivity so HPLC analysis was not required

Table A.2.3-3: RefeSol 02-A soil treated with [¹⁴C]De-Xy-S-2200

Sampling interval (DAT)	0		0.167		1		2		4		7		12		20	
Vessel code	C1	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C15	C16
Acidic extract	97.8	99.0	98.9	96.1	79.3	81.4	67.5	68.8	52.9	53.0	62.0	33.5	19.7	22.4	13.6	10.3
Basic extract	2.0	2.0	1.9	1.9	3.2	3.0	4.0	3.8	4.4	3.5	5.3	2.7	1.9	2.2	1.6	1.5
De-Xy-S-2200	93	92.3	49.0	45.8	ND											
DX-CA-S-2200	0.5	0.4	16.9	17.6	35.3	35.7	35.8	32.8	30.2	29.3	41.0	18.8	8.1	14.0	5.0	2.3
Unknown AE1	1.6	2.0	0.8	0.9	11.2	10.2	11.6	16.1	7.5	11.7	16.8	10.6	4.6	3.7	3.3	2.0
Other Unknowns	2.5	3.6	31.5	31.2	32.8	35.5	20.1	19.7	14.9	11.8	9.3	4.0	6.9	4.6	5.2	5.9
Unresolved Backgrounds	0.2	0.7	0.6	0.5	0.0	0.0	0.0	0.3	0.3	0.2	0.3	0.2	0.1	0.2	0.0	0.1
Total extractibles	97.8	99.0	98.9	96.1	79.3	81.4	67.5	68.8	52.9	53.0	67.3	33.5	19.7	22.4	13.6	10.3
Ethanediol trap	ND															
NaOH trap	NA	NA	0.2	0.2	3.9	2.6	6.3	6.1	11.7	14.6	8.4	23.1	34.6	30.0	41.2	42.3
Residues	ND	ND	1.3	1.4	14.3	13.3	19.6	18.8	26.0	26.1	22.4	36.1	41.6	38.2	39.4	42.0
Mass balance	99.8	101.0	102.3	99.6	100.7	100.3	97.4	97.5	95.0	97.2	98.1	95.4	97.8	92.8	95.8	96.1

NA = Not applicable, ND = Not detected (or < 0.1% AR)

MASS BALANCE

The recovery of applied radioactivity for the samples treated with [¹⁴C]De-Xy-S-2200, ranged from 91.5 to 101.4 % in the Calke soil, from 85.4 to 101.4 % in the South Witham soil and from 92.8 to 102.3 % in the RefeSol 02-A soil.

BOUND RESIDUES

Reflux extraction recovered up to 1.7% AR (South Witham). Bound residue fractionation of the residues showed that radioactivity was present in all fractions but was greatest in the humin fractions.

For the Calke soil, the greatest amount of radioactivity present in the residues was found in the humic acids fraction (13.2% AR).

VOLATILISATION

¹⁴CO₂ was observed with mean values of 46.2 %AR , 42.8 % AR and 41.8 % AR in the Calke, South Witham and RefeSol 02-A, respectively at the end of the study.

METABOLITES

DX-CA-S-2200 is the main metabolite of De-Xy-S-2200. The levels of DX-CA-S-2200 reached a maximum of 84.1 % AR (Calke), 40.4 % AR (South Witham) and 35.7 % AR (RefeSol 02-A) at 1 DAT.

TRANSFORMATION

CAKE Software version 2 was used to determine the degradation rate and kinetic parameters. IRLS optimisation method was used. Fits for parent-metabolite were performed with SFO-SFO kinetics. All parameter values were left free for optimisation.

From visual assessment of the kinetic plots, data for C11 (RefeSol 02-A) of 7 DAT timepoint, clearly differed from the replicate datapoint and the trend of the other datapoints. The outlier was removed from the dataset and the SFO kinetic calculations were repeated.

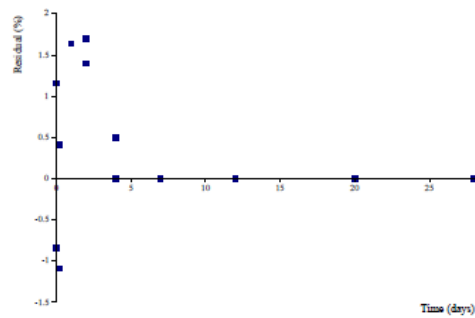
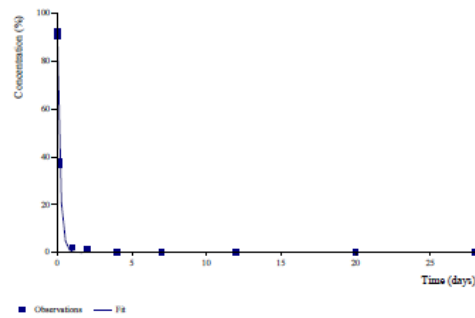
Table A2.3-4: Degradation rates and kinetic parameters for De-Xy-S-2200 and DX-CA-S-2200 SFO-SFO fits

Soil		DT ₅₀ (d)	T test (p<0.05)	DT ₉₀ (d)	χ ² Err %	r ²	ff
Calke	De-Xy-S-2200	0.13	Yes	0.44	4.2	0.9993	-
	DX-CA-S-2200	5.2	Yes	17.3	7.3	0.9822	0.9947
South Witham	De-Xy-S-2200	0.11	Yes	0.35	1.0*	0.9991	-
	DX-CA-S-2200	3.2	Yes	10.5	6.5	0.9673	0.4762
RefeSol 02-A	De-Xy-S-2200	0.17	Yes	0.57	2.63	0.999	
	DX-CA-S-2200	8.1	Yes	26.7	13.2	0.850	0.433
RefeSol 02-A Excluding C11	De-Xy-S-2200	0.17	Yes	0.57	2.7	0.9994	-
	DX-CA-S-2200	6.6	Yes	21.9	6.2	0.9777	0.4335

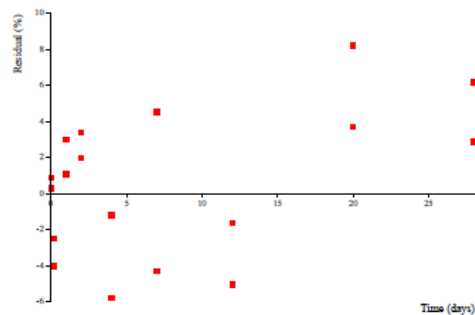
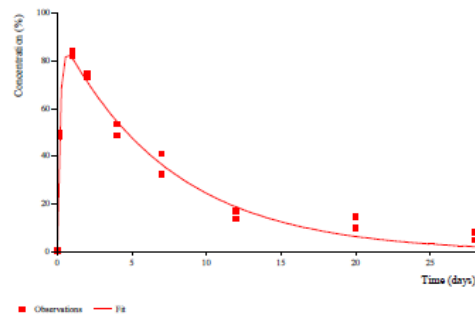
* = limit of quantification

Calke

Compartment Parent:

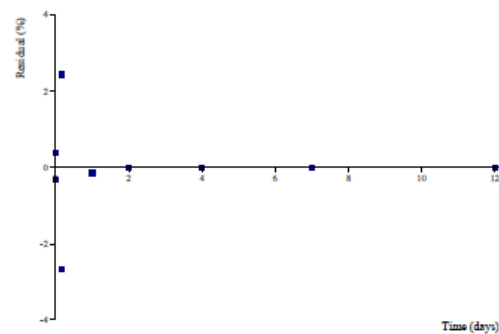
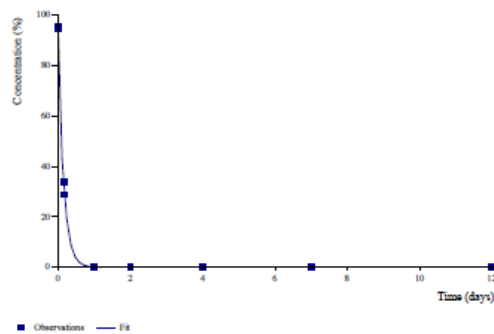


Compartment A1:

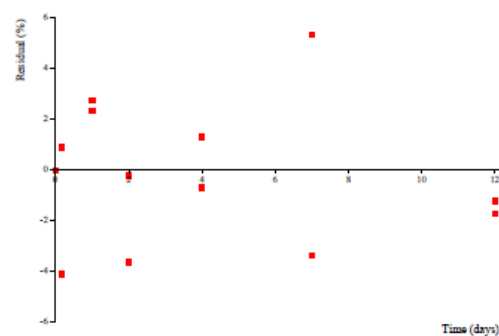
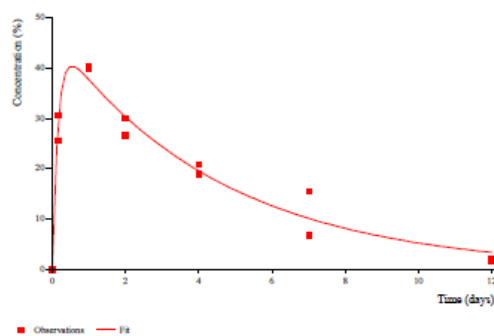


South Witham

Compartment Parent:

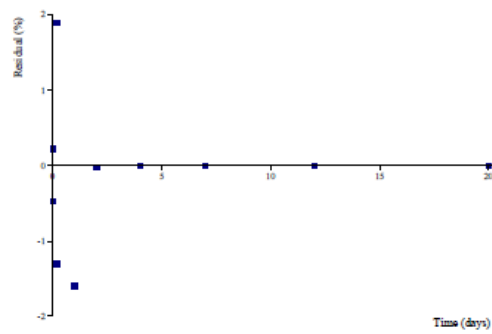
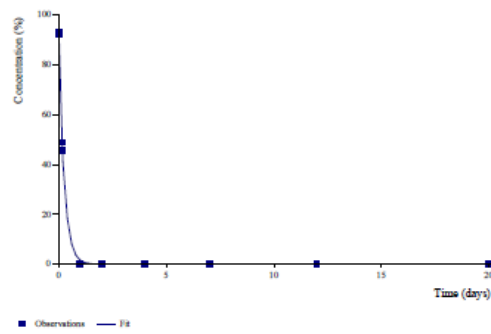


Compartment A1:

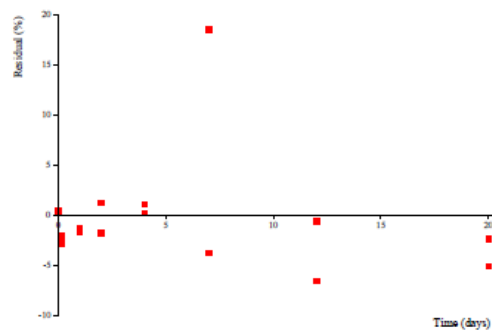
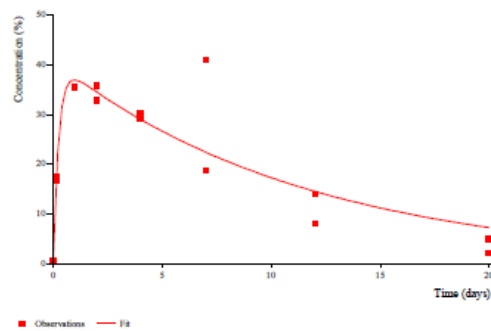


RefeSol 02-A (including all data)

Compartment Parent:

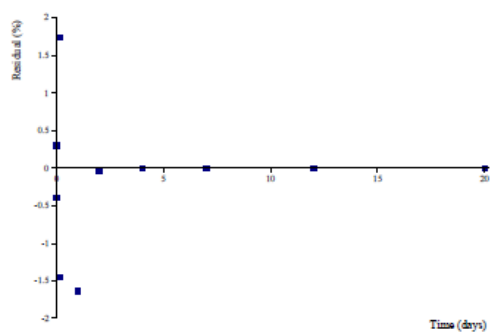
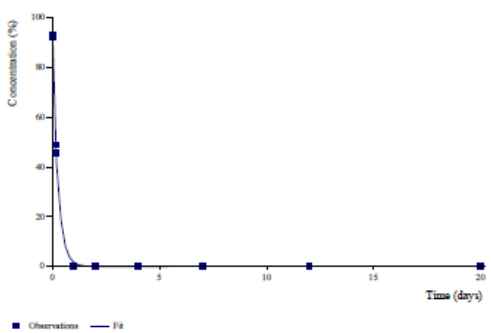


Compartment A1:

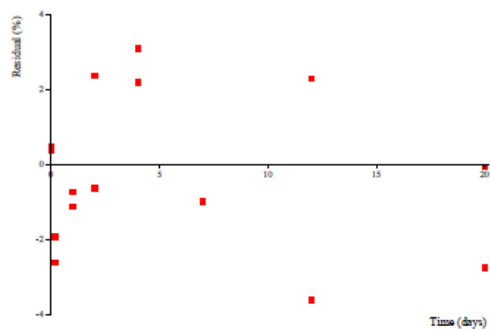
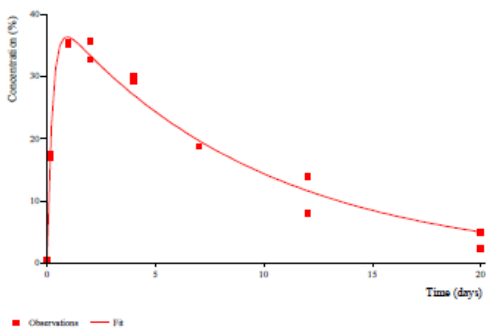


RefeSol 02-A (excluding C11)

Compartment Parent:



Compartment A1:



A 2.4 Study 4: Kang, S. (2017)

zRMS Comments:	The endpoints from laboratory tests, recommended by zRMS-FR from the adsorption study are considered as appropriate input parameters for the modelling.
-------------------	---

Previous conclusion from zRMS FR (2021)

The OECD 106 Checklist was used by zRMS, in addition to OECD 106, to check the reliability of the study.

Results from the test material stability are reported for “representative samples” for each soil type. It should be clarified from which tests (kinetics or isotherm test) these samples were taken. Results indicated that the test item was stable in all soils with more than 90% AR corresponding to test item, except in Hareby soil (88.7% AR). However it is noted that at the end of the isotherm test (after adsorption and desorption phases), up to 18.4% AR was identified as non extractable residues. It is therefore uncertain whether parental mass balance during the isotherm test after the adsorption phase is above 90% AR.

The preliminary test indicated that soil/solution ratio of 1:1 was most appropriate, although percentage of adsorption was still below 20% for 2 of the 3 tested soils (Clipstone and Brierlow). The selected ratio is considered appropriate for highly mobile substances. However, the use of direct method would have been preferred due to low adsorption percentage. In the isotherm test, it is noted that $K_D \times \text{soil:solution}$ is < 0.3 for 4 concentrations for Hareby and Clipstone soils, 3 concentrations in Newhaven soil and 2 concentrations in Brierlow soil. For Clipstone soil, the adsorption percentage is very low, ranging between 1.54 to 5.97%, no adsorption was measured at the lowest nominal concentration (the isotherm is therefore based on 4 concentrations only).

The LOQ is not reported in the study report and should be provided by the applicant, in order to check that concentration in supernatant was sufficiently accurate.

It seems that the isotherms were derived based on mean concentrations instead of using replicate values. The adsorption isotherms are visually acceptable, except for Clipstone soil.

Despite the deficiencies identified above, the study at least indicates that DX-CA-S-2200 is highly mobile. zRMS proposes that all values (including results from Clipstone soil) are kept as a conservative approach, since direct method should have been used.

Reference:	KCA 7.1.3.1/01
Report	Kang, S. (2017). [^{14}C]DX-CA-S-2200 – Adsorption-Desorption on Five Soils using a Batch Equilibrium Method Smithers Viscient. Study No. 13048.6948; Report ROM-0067
Guideline(s):	OECD 106
Deviations:	Several deviations, see above
GLP:	Yes
Acceptability:	Yes

I. MATERIAL AND METHODS

A. MATERIALS

1. Test Materials:	[Benzyl- ^{14}C]DX-CA-S-2200; Specific Activity: 4.55 GBq/mmol
Description:	Not stated
Lot/Batch:	RIS2016-003
Purity:	Radiochemical purity: 100 %

CAS#: Not stated

Stability of compound: Stable under tested conditions

2. Soil: Sieved soils (2 mm) were characterised and equilibrated overnight with 0.01 M CaCl₂ solution, prior to adsorption testing.

Soil Characterisation	Hareby soil	Clipstone soil	Brierlow soil	Newhaven soil	Kenslow soil
Particle size distribution					
% Sand	40	90	44	38	52
% Silt	24	8	50	58	40
% Clay	36	2	6	4	8
Classification ¹	Clay loam	Sand	Sandy loam	Silt loam	Sandy loam
% Organic carbon	2.3	1.1	4.0	3.6	3.3
pH (0.01M CaCl ₂)	7.4	6.1	5.4	5.4	5.0
Cation exchange Capacity (mEq/100g)	14.3	6.2	11.6	11.8	10.0
Field moisture capacity at 1/3 bar (%)	21.3	6.5	26.3	24.7	25.1

¹ USDA Particle Size Distribution and Classification

B. STUDY DESIGN AND METHODS

1. Experimental Conditions

Preliminary test:

Three soils (Hareby, Clipstone and Brierlow soils) and three soil:solution ratios (1:1, 1:2 and 1:5 w/v for each soil in duplicate) were tested. 6, 15 or 20 g dry weight soil and 20 or 30 mL of 0.01M CaCl₂ solution were used. Additionally, blank controls (no test item) and soil-less controls (with test item) were also incubated and analysed by duplicate.

The test item (¹⁴C-DX-CA-S-2200) was applied at nominal concentration of 1.00 mg/L and the samples shaken for the incubation time at 20 ± 1°C in the dark for up to 24 hours.

At the sampling points the samples were centrifuged and the supernatants were analysed by Liquid Scintillation Counting (LSC).

Adsorption and Desorption Kinetics:

The adsorption and desorption kinetics for DX-CA-S-2200 were performed in five soils (Hareby, Clipstone, Brierlow, Newhaven, and Kenslow) with the optimum soil:solution ratio (1:1 w/v) determined in the preliminary test for all five soils. A nominal concentration of 1.00 mg ¹⁴C-DX-CA-S-2200/L was used. The samples were shaken for the incubation time at 20 ± 1°C in the dark for 3, 6 and 24 hours.

Blank controls (no test item) and soil-less controls (with test item) were also incubated and analysed by duplicate.

At the 24 h adsorption equilibrium, after decanting the adsorption solution, an equivalent amount of fresh 0.01M CaCl₂ was added and the vessels were shaken in the dark at 20 ± 1°C for a further 3 h. This procedure was repeated for 6 and 24 h. At each sampling point the samples were analysed as in the preliminary test. Following the 24hr adsorption, soils were also extracted with acidified organic solvents. Organic extracts and aqueous phases were analysed by HPLC and 100% of extracted radioactivity was confirmed to be DX-CA-S-2200.

Adsorption and Desorption Isotherms (definitive test):

The test was carried out in the five soils with five nominal concentrations of ¹⁴C-DX-CA-S-2200: 0.01, 0.05, 0.10, 0.50, and 1.00 mg/L. Two additional replicates for each of the five concentrations were prepared without soil (soil-less control). Duplicate blank controls that contained soil and 0.01 M CaCl₂ solution, without test substance, were prepared for each test system.

Following the pre-equilibration period of the five soil systems with 0.01M CaCl₂ at soil:solution ratio of 1:1 w/v, the test item was applied at the five rates. The vessels were shaken in the dark at

20 ± 1°C for a 24 h adsorption equilibrium phase prior to analysis. Samples were analysed by LSC. A 24 h desorption was used for the desorption isotherm.

2. Description of analytical procedures

All aqueous samples were analysed by LSC.

Selected soils (1 mg/l treatment after 24hr adsorption) were extracted with 5:1 acetonitrile:0.1 M HCl, or 5:1 acetonitrile:0.1 M HCl, followed by 4:1 acetonitrile:0.1 M (NH₄)₂CO₃. The radioactivity in the extracts was analysed in duplicate aliquots by LSC. The extracts were combined and then a portion concentrated and analysed by HPLC/RAM. The corresponding water phase was also analysed by HPLC/RAM.

Combustion of the soil was conducted after the preliminary, kinetics, and isotherms tests were concluded to determine total residual radioactivity in the soil.

3. Calculations

Adsorption isotherms were calculated by linear regression analysis of the adsorption data according to the Freundlich equation. Calculation of the Freundlich constant and related K_{FOC} was performed by application of the indirect method. The radioactivity contents in the supernatants after adsorption was used to calculate the adsorption isotherms as well as the related distribution coefficient referenced to organic carbon content K_{FOC} values.

The substance was considered stable under the test conditions with no need for correction of the adsorption results.

II. RESULTS AND DISCUSSION

Adsorption to containers was not observed.

The calculated percent of DX-CA-S-2200 adsorbed for each soil type is summarized below.

Nominal Concentration (mg/L)	Soil Type (% Adsorbed)				
	Hareby Soil	Clipstone Soil	Brierlow Soil	Newhaven Soil	Kenslow Soil
0.0100	23.2	NA	19.5	20.4	28.0
0.0500	17.2	3.17	26.4	25.2	29.9
0.100	15.3	2.75	22.7	23.0	27.7
0.500	11.5	5.97	23.5	21.2	27.6
1.00	7.15	1.54	15.8	19.1	23.0

NA = Not Applicable, due to a negative adsorption value.

Table 22. Adsorption isotherms test results for Hareby soil.

Initial Concentration (mg/L)	C_{aq}^{ads} (mg/L)	C_s^{ads} (µg/g)	Adsorption (%)	K_d (mL/g)	K_{OC} (mL/g)
0.0106	0.00761	0.00220	22.4	0.289	12.6
0.0106	0.00746	0.00235	23.9	0.315	13.7
Mean:	0.00754	0.00228	23.2	0.302	13.1
0.0513	0.0418	0.00790	15.9	0.189	8.21
0.0513	0.0405	0.00922	18.5	0.227	9.89
Mean:	0.0412	0.00856	17.2	0.208	9.05
0.103	0.0836	0.0155	15.7	0.186	8.08
0.103	0.0844	0.0147	14.9	0.175	7.60
Mean:	0.0840	0.0151	15.3	0.180	7.84
0.515	0.444	0.0633	12.5	0.143	6.20
0.515	0.454	0.0534	10.5	0.118	5.12
Mean:	0.449	0.0584	11.5	0.130	5.66
1.03	0.931	0.0655	6.58	0.0704	3.06
1.03	0.919	0.0769	7.72	0.0836	3.64
Mean:	0.925	0.0712	7.15	0.0770	3.35

OC = organic carbon as percentage of soil, 2.3%.

C_{aq}^{ads} = concentration of the test substance in the aqueous phase.

C_s^{ads} = concentration of the test substance remaining adsorbed on soil.

K_d = adsorption coefficient.

K_{OC} = organic carbon normalized adsorption coefficient.

NOTE: Calculations are performed using unrounded numbers and not the rounded values presented in this table.

Figure 7. Adsorption isotherms test results for Hareby soil.

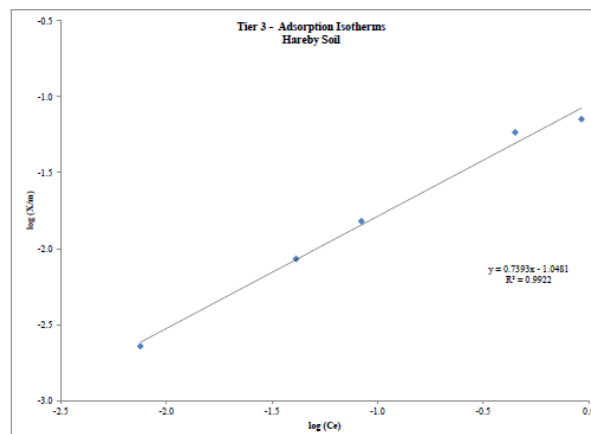


Table 23. Adsorption isotherms test results for Clipstone soil.

Initial Concentration (mg/L)	C_{aq}^{ads} (mg/L)	C_s^{ads} (µg/g)	Adsorption (%)	K_d (mL/g)	K_{OC} (mL/g)
0.0106	0.0107	NA	NA	NA	NA
0.0106	0.0122	NA	NA	NA	NA
Mean:	0.0114	NA	NA	NA	NA
0.0513	0.0473	0.00247	4.97	0.0523	4.76
0.0513	0.0491	0.000683	1.37	0.0139	1.26
Mean:	0.0482	0.00158	3.17	0.0331	3.01
0.103	0.095	0.00437	4.41	0.0461	4.19
0.103	0.098	0.00108	1.09	0.0110	1.00
Mean:	0.096	0.00272	2.75	0.0285	2.59
0.515	0.477	0.0296	5.84	0.0620	5.64
0.515	0.476	0.0309	6.10	0.0649	5.90
Mean:	0.477	0.0302	5.97	0.0635	5.77
1.03	0.966	0.0303	3.04	0.0313	2.85
1.03	1.00	0.000436	0.0438	0.000438	0.0398
Mean:	0.981	0.0153	1.54	0.0159	1.44

OC = organic carbon as percentage of soil, 1.1%.

C_{aq}^{ads} = concentration of the test substance in the aqueous phase.

C_s^{ads} = concentration of the test substance remaining adsorbed on soil.

K_d = adsorption coefficient.

K_{OC} = organic carbon normalized adsorption coefficient.

NA = Not Applicable, due to no adsorption.

NOTE: Calculations are performed using unrounded numbers and not the rounded values presented in this table.

Figure 8. Adsorption isotherms test results for Clipstone soil.

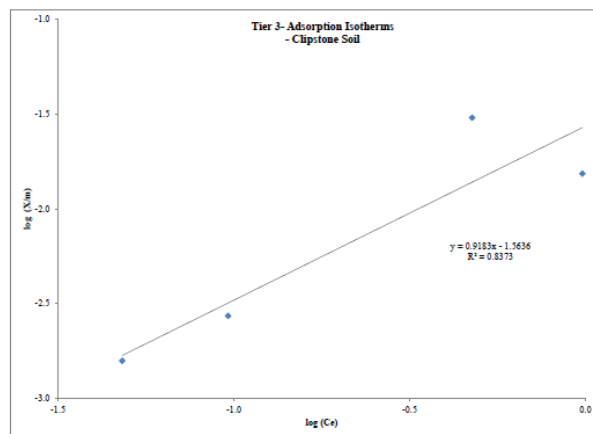


Table 24. Adsorption isotherms test results for Brierlow soil.

Initial Concentration (mg/L)	C_{aq}^{ads} (mg/L)	C_s^{ads} (µg/g)	Adsorption (%)	K_d (mL/g)	K_{OC} (mL/g)
0.0106	0.00802	0.00179	18.3	0.224	5.59
0.0106	0.00777	0.00204	20.8	0.263	6.57
Mean:	0.00790	0.00192	19.5	0.243	6.08
0.0513	0.0357	0.0141	28.3	0.394	9.86
0.0513	0.0375	0.0122	24.6	0.326	8.15
Mean:	0.0366	0.0131	26.4	0.360	9.00
0.103	0.0767	0.0224	22.6	0.292	7.29
0.103	0.0764	0.0227	22.9	0.297	7.41
Mean:	0.0766	0.0225	22.7	0.294	7.35
0.515	0.395	0.112	22.1	0.283	7.08
0.515	0.380	0.127	25.0	0.333	8.32
Mean:	0.388	0.119	23.5	0.308	7.70
1.03	0.836	0.161	16.1	0.192	4.80
1.03	0.842	0.154	15.5	0.183	4.57
Mean:	0.839	0.157	15.8	0.187	4.69

OC = organic carbon as percentage of soil, 4.0%.

C_{aq}^{ads} = concentration of the test substance in the aqueous phase.

C_s^{ads} = concentration of the test substance remaining adsorbed on soil.

K_d = adsorption coefficient.

K_{OC} = organic carbon normalized adsorption coefficient.

NOTE: Calculations are performed using unrounded numbers and not the rounded values presented in this table.

Figure 10. Adsorption isotherms test results for Brierlow soil.

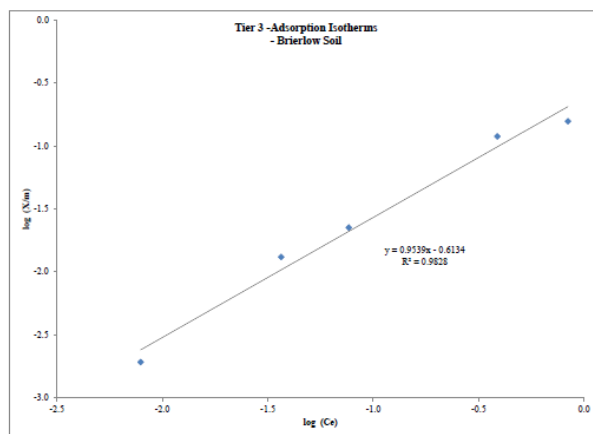


Table 25. Adsorption isotherms test results for Newhaven soil.

Initial Concentration (mg/L)	C_{aq}^{ads} (mg/L)	C_s^{ads} (µg/g)	Adsorption (%)	K_d (mL/g)	K_{OC} (mL/g)
0.0106	0.00753	0.00228	23.3	0.303	8.42
0.0106	0.00809	0.00172	17.6	0.213	5.92
Mean:	0.00781	0.00200	20.4	0.258	7.17
0.0513	0.0369	0.0129	25.9	0.349	9.68
0.0513	0.0376	0.0122	24.5	0.324	9.01
Mean:	0.0372	0.0125	25.2	0.336	9.34
0.103	0.0765	0.0225	22.8	0.295	8.18
0.103	0.0762	0.0229	23.2	0.301	8.37
Mean:	0.0764	0.0227	23.0	0.298	8.27
0.515	0.396	0.110	21.8	0.279	7.74
0.515	0.403	0.104	20.5	0.259	7.18
Mean:	0.400	0.107	21.2	0.269	7.46
1.03 ^a	NA	NA	NA	NA	NA
1.03	0.806	0.191	19.1	0.237	6.57
Mean:	0.806	0.191	19.1	0.237	6.57

^a Data not used due to high variation.

OC = organic carbon as percentage of soil, 3.6%.

C_{aq}^{ads} = concentration of the test substance in the aqueous phase.

C_s^{ads} = concentration of the test substance remaining adsorbed on soil.

K_d = adsorption coefficient.

K_{OC} = organic carbon normalized adsorption coefficient.

NA = Not Applicable due to high variation between replicates.

NOTE: Calculations are performed using unrounded numbers and not the rounded values presented in this table.

Figure 12. Adsorption isotherms test results for Newhaven soil.

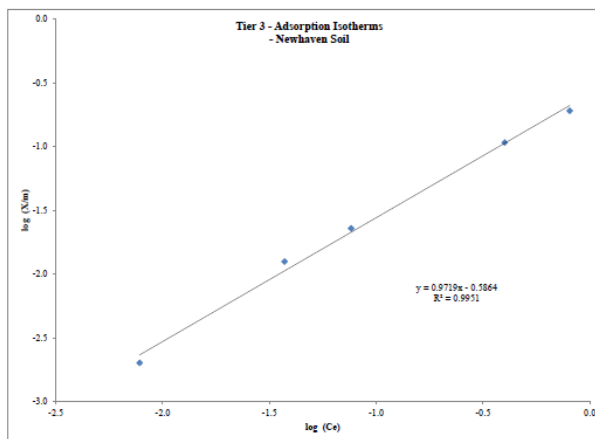


Table 26. Adsorption isotherms test results for Kenslow soil.

Initial Concentration (mg/L)	C_{aq}^{ads} (mg/L)	C_s^{ads} (μg/g)	Adsorption (%)	K_d (mL/g)	K_{OC} (mL/g)
0.0106	0.00694	0.00287	29.3	0.414	12.6
0.0106	0.00719	0.00262	26.7	0.365	11.1
Mean:	0.00706	0.00275	28.0	0.390	11.8
0.0513	0.0342	0.0155	31.2	0.453	13.7
0.0513	0.0355	0.0142	28.6	0.400	12.1
Mean:	0.0349	0.0149	29.9	0.427	12.9
0.103	0.0708	0.0283	28.6	0.400	12.1
0.103	0.0724	0.0267	26.9	0.368	11.1
Mean:	0.0716	0.0275	27.7	0.384	11.6
0.515	0.362	0.145	28.6	0.401	12.2
0.515	0.372	0.135	26.7	0.363	11.0
Mean:	0.367	0.140	27.6	0.382	11.6
1.03	0.794	0.202	20.3	0.254	7.71
1.03	0.741	0.255	25.6	0.344	10.4
Mean:	0.768	0.229	23.0	0.299	9.07

OC = organic carbon as percentage of soil, 3.3%.

C_{aq}^{ads} = concentration of the test substance in the aqueous phase.

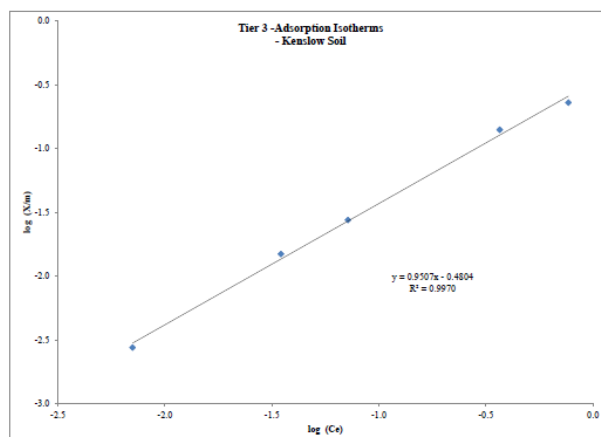
C_s^{ads} = concentration of the test substance remaining adsorbed on soil.

K_d = adsorption coefficient.

K_{OC} = organic carbon normalized adsorption coefficient.

NOTE: Calculations are performed using unrounded numbers and not the rounded values presented in this table.

Figure 14. Adsorption isotherms test results for Kenslow soil.



The adsorption and desorption Freundlich isotherm coefficients obtained from the definitive study are shown in the following table.

Table A2.4-1: Adsorption/Desorption parameters for ^{14}C -DX-CA-S-2200 in five soils

Soil	OC (%)	pH (CaCl ₂)	Adsorption			Desorption		
			1/n	$K^F_{ads,OC}$ (mL/g)	r^2	1/n	$K^F_{des,OC}$ (mL/g)	r^2
Hareby	2.3	7.4	0.739	3.89	0.9922	NA	NA	NA
Clipstone	1.1	6.1	0.918	2.48	0.8373	0.933	5.98	0.8919
Brierlow	4.0	5.4	0.954	6.09	0.9828	0.953	8.32	0.9781
Newhaven	3.6	5.4	0.972	7.20	0.9951	0.937	10.8	0.9940
Kenslow	3.3	5.0	0.951	10.0	0.9970	0.902	10.1	0.9938
Arithmetic values			0.907	-		0.931	-	
Geometric mean			-	5.32			8.58	

NA = Not Applicable

B. MASS BALANCE

The total radioactivity recovery at the end of the adsorption and desorption isotherm test ranged from 96.8 to 107 % AR. At that time, bound residues represented 3.27 to 18.4% AR.

Following 24hr adsorption at the highest dose rate, analysis of the aqueous and soil extracts from all soils confirmed that all extractable radioactivity was DX-CA-S-2200. Parental mass balance was 88.7, 98.1, 96.6, 92.8 and 98.9% AR in soils Hareby, Clipstone, Brielow, Newhaven and Kenslow, respectively.

CONCLUSION

The adsorption and desorption behaviour of DX-CA-S-2200 was investigated using five soil types covering a range of different properties. Five test concentrations were used for both adsorption and desorption covering two orders of magnitude.

The geometric mean value for the adsorption Freundlich isotherm coefficient was 5.32 mL/g. According to the McCall classification scale, DX-CA-S-2200 can be classified as having ‘very high mobility’.

Kang, 2017

A 2.5 Studies 5 and 6 – Anaerobic degradation

zRMS Comments:	The submitted study was required in accordance with data requirement. The data gap was fulfilled. The study has no effect on risk assessment.
-------------------	--

Previous conclusion from zRMS FR (2021)

The two anaerobic studies presented below are well performed and are sufficient to answer to the data gap from EFSA (2015).

Both isomers of the substance degraded during the aerobic phase but after flooding the soil, degradation ceased. The two metabolites produced during the aerobic phase, 2-COOH-S-2200 and 5-COOH-S-2200 did not decrease in concentration under flooded conditions. No unique metabolites were observed under anaerobic conditions. No isomerisation occurred

Reference: Report	KCA 7.1.1.2/01 Lewis, C. J. & Cooper, T. (2015a). [¹⁴ C]S-2354 (S-2200 S-isomer): Anaerobic Soil Metabolism and Degradation. XXXX. Report ROM-0060. Deviations: No GLP: Yes (laboratory certified by UK National Authority)
Acceptability	Acceptable

Executive Summary

Soil samples (100 g) of Speyer 5M soil were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2. Soil units were maintained in the dark, at 20 ± 2°C under aerobic conditions for 30 days, then flooded (*ca* 3cm water overlying the soil) and maintained under anaerobic conditions for a further 120 days.

S-2200 *R*-isomer degraded during the aerobic phase but after flooding the soil, degradation ceased. The two metabolites produced during the aerobic phase, 2-COOH-S-2200 and 5-COOH-S-2200 did not decrease in concentration under flooded conditions. No unique metabolites were observed under anaerobic conditions. No isomerisation of the S-2200 *S*-isomer occurred.

A. MATERIALS

1. Test Materials:	S-2200 <i>S</i> -isomer; (<i>S</i>)-2-methoxy- <i>N</i> -methyl-2-[α-(2,5-xylyloxy)- <i>o</i> -tolyl]acetamide [Benzyl- ¹⁴ C]S-2200 <i>S</i> -isomer; specific activity 4.22 MBq/mmol
Description:	Not stated
Lot/Batch:	[Benzyl- ¹⁴ C]S-2200 <i>S</i> -isomer; RIS2008-009
Purity:	[Benzyl- ¹⁴ C]S-2200 <i>S</i> -isomer; radiochemical purity ≥98%
CAS#:	394657-24-0
Stability of compound:	Radiopurity of the test substance was checked prior to study initiation
2. Soil:	Topsoil was collected from a site where there had been no pesticide use in the last 5 years. Soil was stored at 4 ± 2°C in the dark for <i>ca</i> 2 months prior to the initiation of the experiment in accordance with ISO 10381-6:1993(E).

Table A2.5-1: Characteristics of the soil used

Soil Characterisation	Speyer 5M
Particle size distribution	
% Sand	58
% Silt	31
% Clay	11
Classification ¹	Sandy loam
% Organic carbon	1.0
pH (0.01M CaCl ₂)	7.3
Cation exchange Capacity (mEq/100g)	16.1

Water holding capacity at pF2 (%)	25.5
Microbial Biomass ($\mu\text{g C/g soil}$) ²	396.4

¹ USDA Particle Size Distribution and Classification

² Determined before soil dispensing

1. Experimental Conditions

Soil samples (100 g) of Speyer 5M soil were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2. Soil units were maintained in the dark, at $20 \pm 2^\circ\text{C}$ under aerobic conditions for 30 days, then flooded (*ca* 3cm water overlying the soil) and maintained under anaerobic conditions (nitrogen atmosphere) for a further 120 days. Air drawn over the surface of the units was passed through a series of traps (ethanediol, paraffin in xylene and two sodium hydroxide traps) to collect evolved radiolabelled volatiles. The application rate of [Benzyl-¹⁴C]S-2200 *S*-isomer was 0.27 $\mu\text{g/g}$ dry weight.

2. Sampling

Duplicate units of each soil were removed for analysis immediately after test substance application, at 30 days (prior to flooding), and at 37, 44, 59, 90 and 150 days after treatment.

3. Description of analytical procedures

Where applicable the overlying water was aspirated from the incubation unit to a pre-weighed glass jar. Soil samples were extracted twice with acetone:water (9:1 v/v, *ca* 100 mL) and further residues with acetone (100mL). Soils were further extracted with acetone:0.1 M hydrochloric acid (5:1, v/v, 2 x 100 mL) and acetone (1 x 100 mL). Radioactive concentrations in the extracts, before and after concentration, were determined by LSC. Subsamples of each concentrated extract were analysed chromatographically.

After concentration, extracts were analysed by HPLC (including a chiral column) with confirmation by TLC. Radioactivity trapped in NaOH was confirmed to be CO₂ by barium carbonate precipitation. Unextractable soil residues were determined by combustion/LSC.

Bound residue fractionation was performed on a 150 DAT representative sample. Soil sample were shaken with NaOH solution. The residue (humins fraction) and supernatant were separated. The humins fraction was further washed twice with sodium hydroxide solution, centrifuged and the washings added to the original supernatant.

The supernatant was acidified to *ca* pH 1 and precipitate was washed twice with 0.1 M HCl to separate fulvic acid. The washed precipitate (humic acid fraction) was reconstituted in 0.5 M sodium hydroxide (*ca* 100 mL). Radioactivity in the fulvic acid and reconstituted humic acid fractions was determined by LSC. Radioactivity in the humins fraction was determined by combustion followed by LSC.

Results

Small amounts of radioactivity reported in unknowns and unresolved background has not been included in the Table A2.5-2 below. These results in slight discrepancies between the mass balance and the sum of the components listed in the tables. However this is not considered to be of concern. No isomerisation of S-2200 *S*-isomer occurred.

The mean recovery of applied radioactivity for the samples treated with [¹⁴C]S-2200 *S*-isomer, ranged from 94.2 to 99.1%.

Table A2.5-2: Percent recovery of applied radioactivity in Speyer 5M soil following application of [Benzyl-¹⁴C]S-2200 *S*-isomer and its metabolites under anaerobic conditions

Unit	Time (day)	S-2200 <i>S</i> -isomer	5-COOH-S-2200	2-COOH-S-2200	MCBX	DX-CA-S-2200	Un-extracted from soil	CO ₂	Mass Balance
A25	0	95.8	ND	ND	ND	ND	0.3	NA	98.9
A26	0	95.4	ND	ND	ND	ND	0.3	NA	99.2
Mean		95.6	ND	ND	ND	ND	0.3	NA	99.1

A3	30	65.0	10.2	2.3	0.4	ND	15.7	2.7	90.2
A4	30	61.8	9.6	2.4	0.4	ND	14.7	7.5	98.1
Mean		58.9	9.9	2.3	0.4	ND	15.2	5.1	94.2
A5	37	62.3	8.4	2.6	0.4	0.3	11.6	6.4	97.6
A6	37	59.3	8.4	2.6	0.3	0.4	12.8	6.3	96.4
Mean		60.8	8.4	2.6	0.3	0.4	12.2	6.4	97.0
A7	44	59.1	11.3	3.9	0.4	0.5	11.6	4.7	97.3
A8	44	60.5	6.7	3.9	0.3	0.1	11.5	4.2	96.1
Mean		59.8	9.0	3.9	0.3	0.3	11.6	4.5	96.7
A9	59	60.4	8.8	3.2	0.4	0.8	12.4	5.2	97.3
A10	59	52.1	10.2	2.6	0.5	0.5	15.6	7.0	96.2
Mean		56.3	9.5	2.9	0.4	0.6	14.0	6.1	96.8
A11	90	58.8	8.9	3.1	0.2	0.5	13.1	5.8	96.7
A12	90	61.1	8.1	3.0	0.2	0.7	12.4	6.9	97.6
Mean		59.9	8.5	3.1	0.2	0.6	12.8	6.4	97.2
A13	150	55.9	8.7	2.5	0.3	0.5	13.3	7.6	92.8
A14	150	61.9	8.9	2.8	ND	0.6	13.7	7.4	99.5
Mean		58.9	8.8	2.6	0.2	0.5	13.5	7.5	96.2

NA = not analysed, ND = not detected or <0.1%

Transformation

Degradation occurred during the aerobic phase but no further significant degradation occurred under anaerobic conditions.

Reference:	KCA 7.1.1.2/02 Lewis, C. J. & Cooper, T. (2015b).
Report	[¹⁴ C]S-2167 (S-2200 <i>S</i> -isomer): Anaerobic Soil Metabolism and Degradation. XXXX. Report ROM-0061. Deviations: No GLP: Yes (laboratory certified by UK National Authority)
Acceptability	Acceptable

Soil samples (100 g) of Speyer 5M soil were weighed into individual incubation flasks and adjusted to the water holding capacity value at pH 2. Soil units were maintained in the dark, at 20 ± 2°C under aerobic conditions for 30 days, then flooded (*ca* 3cm water overlying the soil) and maintained under anaerobic conditions for a further 122 days.

S-2200 *R*-isomer degraded during the aerobic phase but after flooding the soil degradation ceased. The two main metabolites produced during the aerobic phase, 2-COOH-S-2200 and 5-COOH-S-2200 did not decrease in concentration under flooded conditions. No unique metabolites were observed under anaerobic conditions. No isomerisation of the S-2200 *R*-isomer occurred.

1. Test Materials:	S-2200 <i>R</i> -isomer; (<i>R</i>)-2-methoxy- <i>N</i> -methyl-2-[α -(2,5-xylyloxy)- <i>o</i> -tolyl]acetamide [Benzyl- ¹⁴ C]S-2200 <i>R</i> -isomer; specific activity 4.22 MBq/mmol [Phenoxy- ¹⁴ C]S-2200 <i>R</i> -isomer; specific activity 4.44 MBq/mmol
Description:	Not stated
Lot/Batch:	[Benzyl- ¹⁴ C]S-2200 <i>R</i> -isomer; RIS2008-010 [Phenoxy- ¹⁴ C]S-2200 <i>R</i> -isomer; RIS2009-002
Purity:	[Benzyl- ¹⁴ C]S-2200 <i>R</i> -isomer; radiochemical purity ≥98% [Phenoxy- ¹⁴ C]S-2200 <i>R</i> -isomer; radiochemical purity ≥98%
CAS#:	394657-24-0
Stability of compound:	Radiopurity of the test substance was checked prior to study initiation

2. Soil:	Topsoil was collected from a site where there had been no pesticide use in the last 5 years. Soil was stored at 4 ± 2°C in the dark for <i>ca</i> 2 months prior to the initiation of the experiment in accordance with ISO 10381-6:1993(E). Soil characteristics are shown in Table A2.1-1
-----------------	---

1. Experimental Conditions

Soil samples (100 g) of Speyer 5M soil were weighed into individual incubation flasks and adjusted to the water holding capacity value at pF 2. Soil units were maintained in the dark, at $20 \pm 2^\circ\text{C}$ under aerobic conditions for 30 days, then flooded (*ca* 3cm water overlying the soil) and maintained under anaerobic conditions (nitrogen atmosphere) for a further 120 days. Air drawn over the surface of the units was passed through a series of traps (ethanediol, paraffin in xylene and two sodium hydroxide traps) to collect evolved radiolabelled volatiles. The application rate of [Benzyl- ^{14}C]S-2200 *R*-isomer or [Phenoxy- ^{14}C]S-2200 *R*-isomer was 0.27 $\mu\text{g/g}$ dry weight.

2. Sampling

Duplicate units of each soil were removed for analysis immediately after test substance application, at 30 days (prior to flooding), and at 37, 44, 61, 90 and 152 days after treatment.

3. Description of analytical procedures

Where applicable the overlying water was aspirated from the incubation unit to a pre-weighed glass jar. Soil samples were extracted twice with acetone:water (9:1 v/v, *ca* 100 mL) and further residues with acetone (100mL). Soils were further extracted with acetone:0.1 M hydrochloric acid (5:1, v/v, 2 x 100 mL) and acetone (1 x 100 mL). Radioactive concentrations in the extracts, before and after concentration, were determined by LSC. Subsamples of each concentrated extract were analysed chromatographically.

After concentration, extracts were analysed by HPLC (including a chiral column) with confirmation by TLC. Radioactivity trapped in NaOH was confirmed to be CO_2 by barium carbonate precipitation. Unextractable soil residues were determined by combustion/LSC.

Bound residue fractionation was performed on a 152 DAT representative sample. Soil sample were shaken with NaOH solution. The residue (humic fraction) and supernatant were separated. The humic fraction was further washed twice with sodium hydroxide solution, centrifuged and the washings added to the original supernatant.

The supernatant was acidified to *ca* pH 1 and precipitate was washed twice with 0.1 M HCl to separate fulvic acid. The washed precipitate (humic acid fraction) was reconstituted in 0.5 M sodium hydroxide (*ca* 100 mL). Radioactivity in the fulvic acid and reconstituted humic acid fractions was determined by LSC. Radioactivity in the humic fraction was determined by combustion followed by LSC.

Results

Small amounts of radioactivity reported in unknowns and unresolved background has not been included in the Tables A.2.6-1 and A.2.6-2 below. These results in slight discrepancies between the mass balance and the sum of the components listed in the tables. However this is not considered to be of concern. No isomerisation of S-2200 *R*-isomer occurred.

The mean recovery of applied radioactivity for the samples treated with [^{14}C]S-2200 *R*-isomer, ranged from 93.9 to 99.2%.

Table A2.6-1: Percent recovery of applied radioactivity in Speyer 5M soil following application of [Benzyl- ^{14}C]S-2200 *R*-isomer and its metabolites under anaerobic conditions

Unit	Time (day)	S-2200 <i>R</i> -isomer	5-COOH-S-2200	2-COOH-S-2200	MCBX	DX-CA-S-2200	Un-extracted from soil	CO_2	Mass Balance
A1	0	90.2	ND	ND	ND	ND	0.2	NA	98.6
A2	0	92.9	ND	ND	ND	ND	0.3	NA	97.7
Mean		91.5	ND	ND	ND	ND	0.3	NA	98.2
A3	30	56.8	12.1	5.2	0.4	ND	12.8	6.3	97.2

A4	30	53.6	13.4	5.0	0.4	ND	14.4	7.2	97.0
Mean		55.2	12.7	5.1	0.4	ND	13.6	6.8	97.1
A5	37	51.7	12.4	5.0	0.3	0.3	14.1	7.7	97.6
A6	37	50.3	11.8	5.3	0.4	0.2	14.9	7.3	97.1
Mean		51.0	12.1	5.1	0.4	0.3	14.5	7.5	97.4
A7	44	56.2	10.4	4.3	0.2	0.3	12.9	6.9	96.1
A8	44	55.6	8.1	3.2	0.3	0.4	12.1	6.1	92.1
Mean		55.9	9.2	3.8	0.3	0.4	12.5	6.5	94.1
A9	61	53.6	14.1	4.7	ND	0.4	14.2	6.9	97.1
A10	61	52.1	13.6	5.6	0.4	0.4	13.6	6.9	97.7
Mean		52.8	13.8	5.2	0.2	0.4	13.9	6.9	97.4
A11	90	53.0	12.6	6.2	ND	0.7	12.9	6.9	98.3
A12	90	49.7	13.5	5.8	ND	1.0	13.1	7.4	97.2
Mean		51.3	13.0	6.0	ND	0.9	13.0	7.2	97.8
A13	152	55.2	11.1	4.8	ND	0.6	13.4	3.7	94.3
A14	152	54.3	12.6	5.2	0.4	0.4	13.6	5.2	96.0
Mean		54.7	11.8	5.0	0.2	0.5	13.5	4.5	95.2

NA = not analysed, ND = not detected or <0.1%

Table A2.6-2: Percent recovery of applied radioactivity in Speyer 5M soil following application of [Phenoxy-¹⁴C]S-2200 R-isomer and its metabolites under anaerobic conditions

Unit	Time (day)	S-2200 R-isomer	5-COOH-S-2200	2-COOH-S-2200	MCBX	Unextracted from soil	CO ₂	Mass Balance
B1	0	93.9	ND	ND	ND	0.3	NA	98.4
B2	0	96.3	ND	ND	ND	0.3	NA	99.9
Mean		95.1	ND	ND	ND	0.3	NA	99.2
B3	30	57.0	11.5	5.0	0.3	12.9	6.4	94.9
B4	30	54.1	11.4	4.5	0.4	13.6	6.4	92.9
Mean		55.6	11.4	4.7	0.4	13.3	6.4	93.9
B5	37	54.5	11.5	4.8	ND	13.2	6.1	96.2
B6	37	54.9	11.3	4.6	0.3	12.5	6.3	95.8
Mean		54.7	11.4	4.7	0.2	12.9	6.2	96.0
B7	44	50.6	12.5	5.1	0.3	14.0	9.1	96.6
B8	44	55.0	11.7	4.9	0.4	10.9	8.4	95.7
Mean		52.8	12.1	5.0	0.4	12.5	8.8	96.2
B9	61	52.3	13.3	5.3	0.4	12.9	7.6	96.7
B10	61	56.2	11.6	5.6	0.4	12.4	5.9	97.2
Mean		54.2	12.5	5.4	0.4	12.7	6.8	97.0
B11	90	50.6	14.3	5.0	0.3	13.1	10.4	98.4
B12	90	54.3	12.7	7.2	0.2	11.8	7.4	98.6
Mean		52.6	13.5	6.1	0.3	12.5	8.9	98.5
B13	152	54.8	11.7	5.2	0.3	14.0	6.0	96.4
B14	152	52.2	11.1	4.8	0.3	14.2	5.7	93.1
Mean		53.5	11.4	5.0	0.3	14.1	5.9	94.8

NA = not analysed, ND = not detected or <0.1%

Transformation

Degradation occurred during the aerobic phase but no further significant degradation occurred under anaerobic conditions.