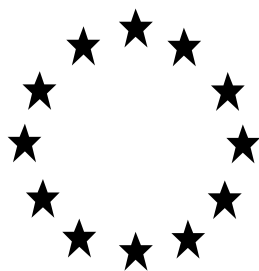


Revised Addendum following the Evaluation of Confirmatory data after approval



ISOXABEN

Summary and Conclusion **Volume 3** **Annex B.7** **Residues**

Applicant: Dow AgroSciences
Rapporteur Member State: Sweden

Feb 2014

Volume 1

Level 1: Statement of subject matter and purpose for which the monograph was prepared

Level 2: Reasoned statement of the overall conclusions drawn by the Rapporteur Member State

Appendix 1: Standard terms and abbreviations

Appendix 2: Specific terms and abbreviations

Appendix 3: List of endpoints

Level 3: Proposed decision with respect to the application for inclusion of the active substance in Annex I

Level 4: Further information to permit a decision to be made, or to support a review of the conditions and restrictions associated with the proposed inclusion in Annex 1

Volume 2

Annex A: List of the tests and studies submitted and of information available

Volume 3

Annex B: RMS summary, evaluation and assessment of the data and information

Annex B.1: Identity

Annex B.2: Phys/chem.

Annex B.3: Data application and further information.

Annex B.4: Proposal for classification and labelling

Annex B.5: Analytical method

Annex B.6: Toxicology and metabolism

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Appendix 1: Standard terms and abbreviations

Appendix 2: Specific terms and abbreviations

Volume 4

Annex C: Confidential information and summary and assessment of information relating to the collective submission of dossiers

ISOXABENAddendum to DAR following submission of confirmatory data

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Background

Commission Directive 2011/32/EU (8 March 2011) to include isoxaben in Annex I to Directive 91/414/EEC (later transferred to Commission Implementing Regulation (EU) No 540/2011 of 25 May 2011) contains a provision that Member States shall request the submission of confirmatory information on the potential risk in rotational crops. This provision relates to a concern raised in the EFSA conclusion on the peer review of isoxaben (EFSA Journal 2010; 8(9): 1714), that for rotational crop a case was presented which showed that after one year residues in soil would be so low that significant residues in rotational crops would not be expected. However, this case was not proved satisfactory with the studies submitted. Firstly, the chosen soil trials were not the ones that had the longest DT₉₀ value, since the longest DT₉₀ (sandy loam) for the parent is about 4 years. In addition, there was no consideration of the metabolites which are also persistent in the soil. As a consequence, a data gap was identified for rotational crops. To address the data gap a confined rotational crop (CRC) study was carried out. This study is evaluated below.

Summary and conclusion of the evaluation

It was concluded during the peer review of isoxaben that further evidence was required to show the relevance of isoxaben residues for rotational crops. To address this concern the applicant submitted a confined rotational crop (CRC) study to determine the amount, nature and distribution of residues in the raw agricultural commodities from three rotational crops (a cereal, a leafy vegetable and a root crop) that were planted in a sandy loam soil at 30, 120, 180 and 365 days after treatment (DAT) with a single surface application of ¹⁴C isoxaben- phenyl and isoxazole labels at 250 g ai/ha.

The study was performed according to OECD guideline 502, and the principal parts were done according to GLP. The study was well performed and reported. The question raised in the EFSA conclusion of the peer-review of isoxaben has been addressed.

It is concluded that metabolism in primary (cereals and oil seed rape) and rotational crops (cereals, lettuce and radish) is similar for the tested crops and a separate residue definition in rotational crops is not necessary provided that the GAP is within the tested conditions in this CRC study.

Residues in cereal feed were at each plant back interval at a higher level than the residues in commodities for human consumption. Calculation of the maximum dietary burden for livestock at 365 DAT shows that residues from tested rotational crops in the total diet for dairy cattle and beef cattle is about 10 times below the trigger value 0.1 mg/kg for feeding study (EU guideline 7031/VI/95 rev.4). The qualitative nature as well as the level of the terminal residue in the RAC at the time of harvest or when utilized for livestock feed is of no concern if isoxaben is used within the tested condition (2X recommended GAP of 125 g ai/ha) at 365 DAT.

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With a pre-emergence soil application of isoxaben to a sandy loam soil in amounts of 250 g ai/ha the soil metabolites 2,6 dimethoxybenzamide with DT90 of 134 days, 1-hydroxyisoxaben with DT90 of 732 days and the parent isoxaben with DT90 of 2103 days are transferred from soil to tested edible crops (leafy crop immature and mature lettuce), root crop (radish) or cereals (wheat) above 0.01 mg eq. /kg until 180 DAT. At 30 and 120 DAT phytotoxicity was observed for lettuce and radish. A plant back interval of at least 180 days is therefore recommended for the three tested rotational crops (cereal, leafy vegetable and root crops).

B.7 Residues**B.7.9 Rotational crops**

To determine the amount, nature and distribution of residues in the raw agricultural commodities from three rotational crops (a cereal, a leafy vegetable and a root crop) were planted in a sandy loam at 30, 120, 180 and 365 days after treatment (DAT) with a single surface application of ¹⁴C isoxaben (phenyl and isoxazole labels) at a dose level of 250 g ai./ha.

Reference:	Graper L.K, Y. Adelfinskaya. A confined Rotational Crop Study with ¹⁴ C-Isoxaben, Phenyl and Isoxazole Labels, 2013		
Dow AgroScience LLC, (DAS)	Study ID: 110534		
Guideline:	OECD Guideline 502 for Metabolism in Rotational Crops (Issued 8 January 2007)		
GLP:	The study was conducted in accordance with the requirements for Good Laboratory Practice (GLP) Standards, 40 CFR 160 with the following exceptions; Weather data and crop/pesticide history were not collected according to GLP. The equipment used for plot maintenance was calibrated prior to use but not GLP verified. The soil characterization for the plots was not conducted according to GLP. The HPLC system used for fraction collection of some of the sample extracts was not officially GLP validated. The reference substances identified by reference number (common name): TSN102562 (2-hydroxy Isoxaben) and TSN100330 (3-hydroxy Isoxaben) were out of certification prior to or at some point during the study, respectively, although the purities had been determined prior to the study.		
Previous evaluation:	Submitted for the purpose to address requirement for confirmatory information (Regulation (EU) No 540/2011)		
Material and methods:			
Test material:	Isoxaben		
Lot/Batch/No	¹⁴ C-Isoxaben Phenyl; INV 301291, ¹⁴ C-Isoxaben Isoxazole INV 301328		
Purity	The average purity of the PH- and IZ-labelled Isoxaben prior to specific activity adjustment was 99.3 and 98.5%, respectively		
Soil	Texture (Sandy Loam)	76% sand-17% silt-7% clay	
	PH	7.8	
	Bulk Density, disturbed (gm/cc)	1.13	
	Field water holding capacity (% @1/3 bar)	12.7	
	Organic matter (%) Walkley Black	1.6	
Crop type	• Cereal (wheat), Ultra Wheat 365 DAT only and Hard Red VNS Wheat • Leafy vegetable (lettuce) Butter Crunch Lettuce • Root crop (radish), Cherry Belle Radish		

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Crop sampling:	Plants were collected by cutting off the appropriate portion or pulling from the soil prior to cutting. Samples were collected into tared bags and stored frozen.
Test concentration:	250 g ai/ha (soil application), as suspension concentrate (SC), see Table 7.9.1-1
Test system:	Four boxes were treated per radiolabel, one each for lettuce and radishes (planted at all four plant-back intervals), one for wheat (planted at 30 and 365 DAT plant-back intervals), and an additional one for wheat (planted 120 DAT). Three control boxes were planted for each crop and samples collected only for the 30 DAT, see Table 7.9.1-2
Timing of application:	Each of the four application containers per radiolabel was covered with aluminium foil to exclude light, and the contents applied to the bare soil surface in each plot using a CO ₂ pressurized sprayer. Each spray solution container was rinsed with water and sprayed evenly onto its respective plot
Test conditions:	Climatic data from weather station 6.22 miles southeast of the test site. Rainfall was monitored on site. Temperature varied from -3°C - + 36°C, humidity from 22-92% and precipitation from 0-80 mm.
Analysis	The samples from frozen storage were, pre-weighed, chopped into smaller pieces if needed, and milled with dry ice, to maintain a frozen state during milling, returned to frozen storage to allow for sublimation of the dry ice, post weighed, and returned to frozen storage pending combustion analysis. All samples, except for the 365 DAT wheat hay, straw and grain, were combusted at the test site to determine homogeneity. Homogeneous samples were shipped to DAS on dry ice using an overnight courier. See Fig. 7.9.1-1 and 7.9.1-2 for the different steps in sample extraction, analysis, characterization and identification of soluble and bound residues.

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Table 7.9.1-1 Experimental design

Parameter	Description				
Test Site	Research For Hire (RFH), 1696 South Leggett Street, Porterville, California 93257, USA				
Soil type	sandy loam				
Crop type	lettuce, radishes, wheat				
Application formulation	A suspension concentrate (SC)				
Application timing and planting	<u>Application:</u> 20 OCT 2011, 250 g ai/ha <u>Plant-backs:</u> 30 DAT, 19 NOV 2011 120 DAT, 17 FEB 2012 180 DAT, 17 APR 2012 (only lettuce and radish) 365 DAT, 19 OCT 2012				
		BBCH stage of growth			
	Crop	30 DAT	120 DAT	180 DAT	365 DAT
Immature harvests	immature lettuce	43-45	41-43	45	44
	wheat forage	34	34	- ^a	34
	wheat hay	83	83	- ^a	83
Mature harvests	mature lettuce	- ^b	- ^c	49	49
	radish tops & roots	42-49 ^d	42-43 ^d	49	49
	wheat straw & grain	89	89	- ^a	89

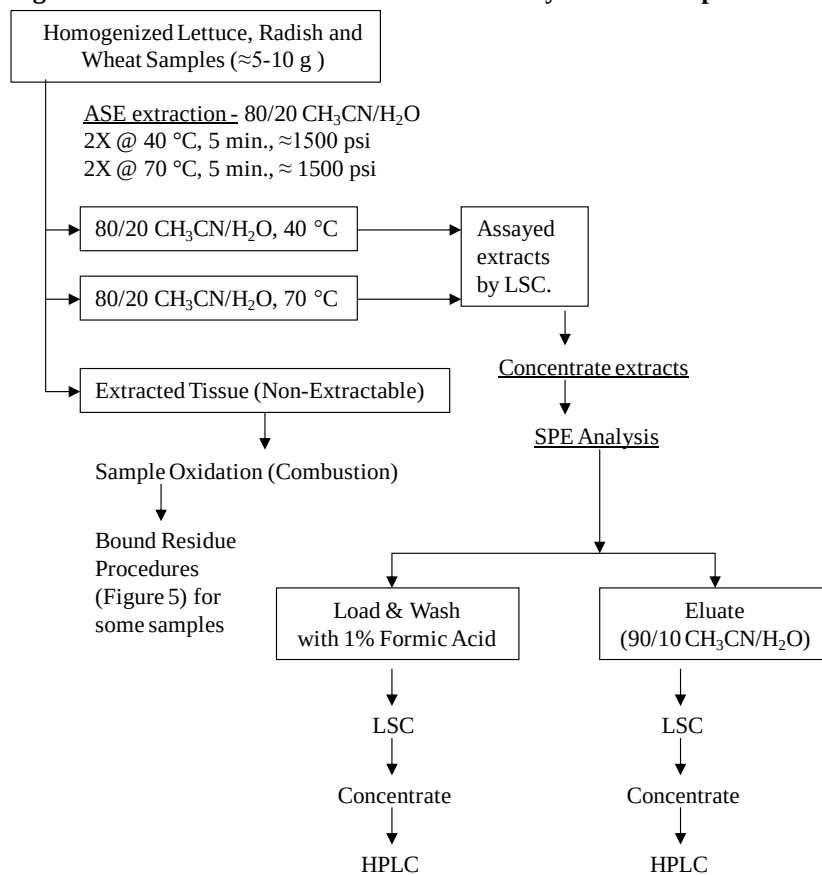
^a 180 DAT wheat samples were not necessary.^b Control was BBCH 49. Treated samples were not collected due to poor growth resulting in the absence of harvestable agricultural product.^c No samples were collected due to replanting this plot at 180 DAT so that better growth could be obtained.^d The only radish samples reaching the BBCH 49 mature growth stage were the 30 DAT controls. The 30 DAT PH and IZ treated radish samples reached BBCH 42 and 44, respectively and the 120 DAT PH and IZ treated radish samples reached BBCH 42 and 43.**Table 7.9.1-2 Organization of the plots**

The table below shows how crops, treatments and plant-back intervals were organized in the plots.

	Leafy Vegetable Crop	Root Crop	Cereal Crop	
Control Boxes	30-DAT plant-back	30-DAT plant-back	30-DAT plant-back	
¹⁴ C-Treated Boxes, Phenyl Label	30, 120, 180 & 365-DAT plant-back	30, 120, 180 & 365-DAT plant-back	30 & 365-DAT plant-back	120-DAT plant-back
¹⁴ C-Treated Boxes, Isoxazole Label	30, 120, 180 & 365-DAT plant-back	30, 120, 180 & 365-DAT plant-back	30 & 365-DAT plant-back	120-DAT plant-back

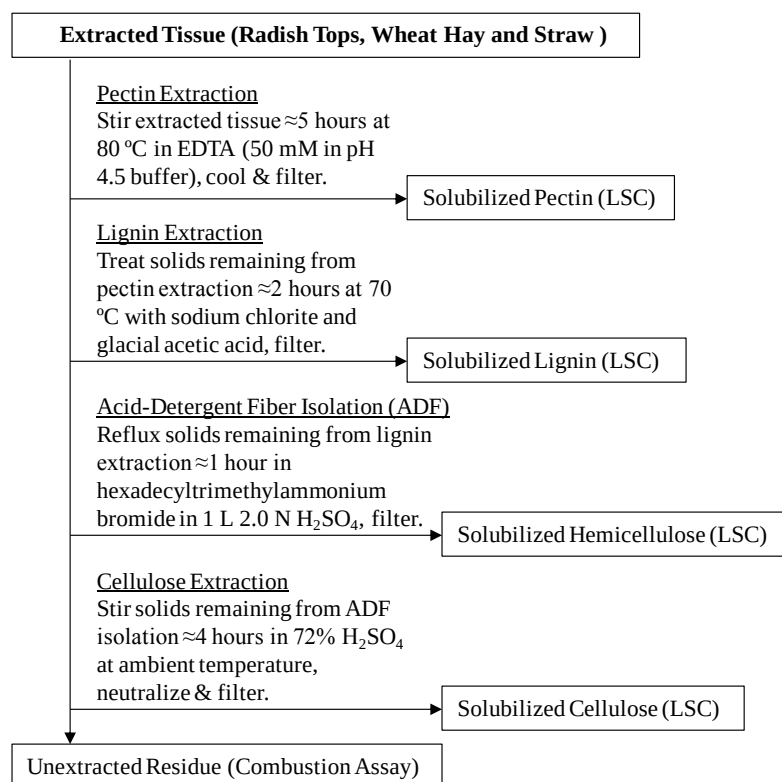
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Figure 7.9.1-1 Schematic flowchart for the analysis of the crop**Figure 7.9.1-2 Schematic flowchart for the analysis of bound residues in selected crop fractions**

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**Results****B.7.9.2 Magnitude of residue in the rotational crops lettuce, radish and wheat****Sampling**

The 30 DAT and 120 DAT lettuce and radish demonstrated a slow rate of development and abnormal growth (stunted) in comparison to control label. This phenomenon was observed in treatment with both labels (isoxazole and phenyl). However, the rate of development and abnormal growth was more severe in the phenyl labelled samples.

Distribution and quantity of TRR in various harvested plant parts

Table 7.9.2-1 presents the distribution of the total tissue residues within all of the crop harvests and the reasons for when samples were not collected.

The residues in crops for human consumption (lettuce, radish roots and wheat grain) ranged from 0.02 mg eq./kg-(wheat grain) to 0.074 mg eq./kg (radish roots) at 30 DAT. At 365 DAT the residues in these crops ranged from 0.001-0.006 mg eq./kg, and thus at a very low level. Residues in immature lettuce and radish roots and tops showed a significant decline from 30 to 180 DAT and then to 365 DAT. Residues in wheat grain for both labels increased from 30 to 120 DAT and then declined to 365 DAT. At 365 DAT residues of isoxazole and phenyl labelled isoxaben in wheat grain were 0.006 mg eq./kg and 0.003 mg eq./kg respectively.

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Residues in crops for livestock consumption (hay, forage, and straw) were higher than for human consumption and ranged from 0.012- 0.576-mg eq./kg. At 365 DAT these residues varied from 0.012-0.111 mg eq./kg. Residues in wheat forage showed a slight increase from 30 to 120 DAT and a significant decline from 120 DAT to 365 DAT. Residues in wheat hay and straw for the phenyl label declined from 30 to 120 DAT, but increased for the isoxazole label. At 365 DAT, residues in wheat hay and straw had declined significantly compared to those at 30 and 120 DAT and were at the most 0.054 mg eq./kg and 0.111 mg eq./kg, respectively. For 9 of the 13 corresponding sample types at 30 and 120 DAT, the isoxazole label samples contained higher residues than did the phenyl label samples.

While the difference in residue levels for most of the isoxazole and phenyl labels samples at 30 and 120 DAT were not large, residues for the isoxazole radish tops at 30 and 120 DAT and wheat hay and straw at 120 DAT were approximately twice that of the corresponding phenyl label samples. At 180 DAT, the phenyl label lettuce and radish samples contained higher residues than did the isoxazole label. At 365 DAT there were no significant differences between the crop residues for either label position. These results indicate that while the early isoxazole labelled samples sometimes possessed higher residues than did the phenyl label, it did not appear to be a consistent difference for every crop at every plant-back time.

Table 7.9.2-1. Total radioactive residues in plant samples collected for isoxaben confined rotational crop study

Phenyl label plant-back intervals

Sample ^a	30 DAT Treated		120 DAT Treated		180 DAT Treated		365 DAT Treated	
	<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>	
	dpm/g		dpm/g		dpm/g		dpm/g	
Lettuce-immature	3,706 ^c	0.060 ^c	- ^d	- ^d	1,716	0.028	228	0.004
Lettuce-mature	- ^c	- ^c	- ^d	- ^d	1,592	0.026	181	0.003
Radish tops	5,323 ^e	0.086 ^e	5,160 ^e	0.083 ^e	1,892	0.031	293	0.005
Radish roots	4,111 ^e	0.066 ^e	6,632 ^e	0.107 ^e	563	0.009	84	0.001
Wheat Forage	3,559	0.058	4,574	0.074	- ^f	- ^f	755	0.012
Wheat Hay	15,437	0.235	8,737	0.180	- ^f	- ^f	3,648	0.054
Wheat Straw	22,335	0.361	17,971	0.291	- ^f	- ^f	6,836	0.111
Wheat Grain	1,222	0.020	1,663	0.027	- ^f	- ^f	182	0.003

Isoxazole Label Plant-Back Intervals

Sample ^a	30 DAT Treated		120 DAT Treated		180 DAT Treated		365 DAT Treated	
	<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>		<i>mg eq./kg (ppm)^b</i>	
	dpm/g		dpm/g		dpm/g		dpm/g	
Lettuce-immature	5,436 ^c	0.041 ^c	- ^d	- ^d	1,734	0.013	243	0.002
Lettuce-mature	- ^c	- ^c	- ^d	- ^d	945	0.007	191	0.001
Radish tops	24,923 ^e	0.187 ^e	18,650 ^e	0.140 ^e	1,716	0.013	377	0.003
Radish roots	9,907 ^e	0.074 ^e	13,027 ^e	0.097 ^e	689	0.005	136	0.001

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Wheat Forage	7,691	0.058	12,552	0.094	- ^f	- ^f	1,824	0.014
Wheat Hay	30,337	0.209	22,970	0.294	- ^f	- ^f	6,460	0.048
Wheat Straw	51,359	0.384	76,941	0.576	- ^f	- ^f	12,758	0.095
Wheat Grain	3,094	0.023	3,974	0.030	- ^f	- ^f	742	0.006

^a Control samples (not shown) contained from 11 to 98 dpm/g and represented 0.001 to 0.002 mg eq./kg.^b In isoxaben equivalents.^c The 30 DAT treated immature lettuce samples experienced slow growth compared to controls. No samples were available for 30 DAT mature lettuces due to absence of harvestable agricultural product.^d The 120 DAT immature lettuce samples experienced slow growth and were harvested to allow an 180 DAT plant back. Consequently, there were no mature 120 DAT lettuce samples. The 120 DAT immature lettuce samples were not analyzed since 180 DAT immature lettuce samples were available.^e The 30 and 120 DAT plant-back samples for radish experienced slow growth and abnormal development.^f No 180 DAT plant-back samples were necessary for wheat.**Potential for the transfer of isoxaben residues from the soil to the rotational crops after tested rotational intervals**

Below follows a discussion based on TRR values presented in Table 7.9.2.1

Leafy vegetable (lettuce –immature and mature)

No samples were possible to take due to slow growth until 180 DAT. At this sample time point the TTR was quite high; from 0.007 to 0.028 mg eq. /kg for both labels, but at 365 DAT the residue in the leafy vegetable had declined to between 0.001 to 0.004 mg eq. /kg.

Root crop (radish tops and root)

The samples for 30 and 120 DAT experienced slow growth and abnormal development. The 120 DAT radish root samples showed quite high TRR of 0.094-0.107 mg eq./kg (isoxazole and phenyl labels respectively). The TRR residues then declined and were between 0.005-0.009 mg eq./kg in radish roots at 180 DAT. At 365 DAT the TRR declined to and 0.001 mg eq./kg for both labels.

Cereals (grain, straw, hay and forage)

TRR in grain for both labels increased from 30 to 120 DAT and then declined to 365 DAT. The residues in grain varied from 0.027-0.030 mg eq./kg at 120 DAT, but at 365 DAT the residue in grain was 0.006 mg eq./kg and 0.003 mg eq./kg for isoxazole and phenyl labelled isoxaben respectively.

TRR in wheat forage showed a slight increase from 30 to 120 DAT. At 120 DAT the TRR was 0.094-0.074 mg eq./kg for isoxazole and phenyl labels respectively while at 365 DAT the TRR was 0.014-0.012 mg eq./kg for isoxazole and phenyl labels respectively.

TRR in wheat hay and straw for the phenyl label declined from 30 to 120 DAT, but increased for the isoxazole label. For wheat straw the TRR was 0.576 mg eq./kg at 120 DAT. At 365 DAT, residues in wheat hay and straw had declined significantly compared to those at 30 and 120 DAT and were at the most 0.054 mg eq./kg and 0.111 mg eq./kg.

Conclusion:

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No samples were possible to take from the leafy crop; immature as well as mature lettuce, until 180 DAT. At this time point TRR was quite high, that is 0.028 mg eq./kg., but declined to 0.001-0.004 mg eq./kg at 365 DAT. The root crop (radish) showed as the leafy crop phytotoxic symptoms until 180 DAT. At 120 DAT the TRR was above 0.1 mg eq./kg but decreased to 0.001 mg eq./kg at 365 DAT. The residues in grain varied from 0.027-0.030 mg eq./kg at 120 DAT, and then decreased to 0.006 mg eq./kg and 0.003 mg eq./kg at 365 DAT for isoxazole and phenyl labelled isoxaben respectively. No phytotoxicity was reported for cereals. A plant-back interval of at least 180 DAT is recommended for tested rotational crops after a pre-emergence soil application of isoxaben at 250 g. ai/ha.

B.7.9.3 Characterization and identification of residues in rotational lettuce, radish and wheat

Residue levels in radish roots at 180 DAT, isoxazole treated lettuce at 180 DAT and immature and mature lettuce, radish roots and tops and immature wheat grain at 365 DAT were below 0.010 mg eq./kg and were not subjected to characterization (Table 7.9.2-1). All other crop samples that possessed residues of 0.010 mg eq./kg or higher were subjected to characterization, except for the 120 DAT radish top and root samples. Slow growth and abnormal development of the 120 DAT radish top and root samples, a 5-month delay in receipt of these samples, plus the results of another intermediate plant-back interval (180 DAT) led to a decision by notifier to analyze these samples no further than extraction and SPE and to not include these data in this report.

Ten categories of metabolites plus parent isoxaben for the phenyl label and nine categories of metabolites plus parent isoxaben for the isoxazole label were characterized; see Table 7.9.3-1. For all samples that were analyzed, approximately 12 to almost 64% of the TRR, depending on the sample, was identified by co-chromatograph with reference compounds and for some samples, confirmed by MS. The category "Other" was associated from 8.4 to 33.4 % of the TRR, and consisted of 2-28 unidentified components. The largest metabolites in "Other" were found in isoxazole labelled wheat straw at 0.021 mg eq. /kg and 3.7% TRR. This declined to 0.005 mg eq. /kg and 5.7% TRR at 365 DAT.

Table 7.9.3-1 Isoxaben and metabolite levels of ¹⁴C-isoxaben in confined rotational crops**PHENYL LABEL**

DAT		Applied to HPLC ^a	X069203/ dimethoxy benzamide	dimethoxy benzoic acid	X11090 448/ 1-hydroxy -ethyl Isoxaben	glucose conjugates	glucose-malonyl conjugates & X1116287 9/ 2-hydroxy, 6-methoxy benzamide	X341970 / 1-hydroxy Isoxaben
Immature Lettuce								
30	%TRR	40.2%	13.7	- ^b	-	2.7	2.6	5.0
	mg eq./kg	0.024	0.008	-	-	0.002	0.002	0.003
180	%TRR	54.7%	34.7	-	-	1.9	2.8	2.7

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	<i>mg eq./kg</i>	0.015	0.010	-	-	0.001	0.001	0.001
Mature Lettuce								
180	%TRR	57.7%	33.6	-	-	2.0	4.5	4.2
	<i>mg eq./kg</i>	0.015	0.009	-	-	0.001	0.001	0.001
Radish Tops								
30	%TRR	64.5%	3.8	2.6	-	8.5	4.6	4.5
	<i>mg eq./kg</i>	0.055	0.003	0.002	-	0.007	0.004	0.004
180	%TRR	73.7%	1.5	2.3	-	20.6	12.4	4.4
	<i>mg eq./kg</i>	0.023	<0.001	0.001	-	0.006	0.004	0.001
Radish Roots								
30	%TRR	55.9%	3.4	1.6	-	1.3	2.7	3.2
	<i>mg eq./kg</i>	0.037	0.002	0.001	-	0.001	0.002	0.002
Wheat Forage								
30	%TRR	61.7%	18.0	0.7	-	8.3	15.7	4.3
	<i>mg eq./kg</i>	0.035	0.010	<0.001	-	0.005	0.009	0.002
120	%TRR	66.8%	11.4	-	-	10.3	18.3	3.0
	<i>mg eq./kg</i>	0.049	0.008	-	-	0.008	0.014	0.002
365	%TRR	87.9%	-	-	-	10.8	21.3	14.2
	<i>mg eq./kg</i>	0.011	-	-	-	0.001	0.003	0.002
Wheat Hay								
30	%TRR	60.3%	6.8	-	-	7.7	20.0	3.0
	<i>mg eq./kg</i>	0.142	0.016	-	-	0.018	0.047	0.007
120	%TRR	54.4%	10.3	-	-	8.2	9.5	4.2
	<i>mg eq./kg</i>	0.098	0.018	-	-	0.015	0.017	0.007
365	%TRR	73.6%	-	9.5	-	16.5	27.1	6.6
	<i>mg eq./kg</i>	0.040	-	0.005	-	0.009	0.015	0.004
Wheat Straw								
30	%TRR	53.6%	7.1	2.8	-	9.4	11.6	2.9
	<i>mg eq./kg</i>	0.194	0.026	0.010	-	0.034	0.042	0.011
120	%TRR	60.1%	9.4	2.6	-	7.4	11.5	4.0
	<i>mg eq./kg</i>	0.175	0.027	0.008	-	0.022	0.033	0.012
365	%TRR	64.3%	2.5	6.7	-	14.5	12.6	5.2
	<i>mg eq./kg</i>	0.071	0.003	0.007	-	0.016	0.014	0.006

^a Amount of extractable radioactivity after SPE and concentration that was injected on the HPLC column.

^b A dash (-) indicates that no residues was detected

Table 7.9.3-1 continuation

PHENYL LABEL

DAT		2-hydroxy Isoxaben isomer A & 3-hydroxy Isoxaben	2-hydroxy Isoxaben isomer B	X1109129 6/oxoisoxa- ben	X285341/ Isoxaben	desmeth yl isoxa- ben	Other ^a	# ^b	Large st Other ^c
Immature Lettuce									
30	%TRR	- ^d	-	-	1.0	-	15.1	11	2.8
	<i>mg eq./kg</i>	-	-	-	0.001	-	0.009		0.002
180	%TRR	- ^d	1.3	-	0.8	-	10.5	6	4.4
	<i>mg eq./kg</i>	-	<0.001	-	<0.001	-	0.003		0.001
Mature Lettuce									
180	%TRR	-	-	-	-	-	13.3	4	7.1
	<i>mg eq./kg</i>	-	-	-	-	-	0.003		0.002
Radish Tops									
30	%TRR	-	-	2.1	14.1	1.7	22.6	13	4.9

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a	The category 'Other' includes radioactive peaks that did not co-chromatograph with any of the ten categories of metabolites or parent isoxaben.					
b	Maximum number of radioactive peaks in the chromatogram that did not co-chromatograph with any of the ten categories of metabolites or parent isoxaben.					
c	Largest of the 'Other' radioactive peaks for the analysis or analyses of a sample.					
d	A dash (-) indicates that no residues was detected.					

Table 7.9.3-1 continuation

ISOXAZOLE LABEL

		X1109044 8/ 1-					2-hydroxy Isoxaben isomer A	2- hydroxy Isoxaben isomer B
DAT		Applied to HPLC ^a	hydroxy- ethyl Isoxaben	glucose conjugate s	glucose- malonyl conjugates	X341970/ 1-hydroxy Isoxaben	& 3- hydroxy Isoxaben	
Immature Lettuce								
30	%TRR	44.4%	- ^b	8.1	4.9	12.7	0.9	2.2
	mg eq./kg	0.018	-	0.003	0.002	0.005	<0.001	0.001
180	%TRR	55.4%	-	8.4	6.1	27.2	-	-
	mg eq./kg	0.007	-	0.001	0.001	0.004	-	-
Radish Tops								
30	%TRR	41.1%	-	5.3	2.6	2.5	0.5	0.3
	mg eq./kg	0.077	-	0.010	0.005	0.005	0.001	0.001
180	%TRR	53.8%	-	15.1	3.6	4.2	-	-
	mg eq./kg	0.007	-	0.002	<0.001	0.001	-	-
Radish Roots								
30	%TRR	30.5%	-	2.0	1.5	1.0	0.5	0.4
	mg eq./kg	0.023	-	0.001	0.001	0.001	<0.001	<0.001
Wheat Forage								

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30	%TRR	57.4%	-	8.7	17.3	3.0	-	0.5
	<i>mg eq./kg</i>	<i>0.033</i>	-	<i>0.005</i>	<i>0.010</i>	<i>0.002</i>	-	<i><0.001</i>
120	%TRR	56.1%	-	8.9	14.9	3.7	-	-
	<i>mg eq./kg</i>	<i>0.053</i>	-	<i>0.008</i>	<i>0.014</i>	<i>0.003</i>	-	-
365	%TRR	74.6%	-	14.6	18.5	6.9	4.9	-
	<i>mg eq./kg</i>	<i>0.010</i>	-	<i>0.002</i>	<i>0.003</i>	<i>0.001</i>	<i>0.001</i>	-
Wheat Hay								
30	%TRR	69.2%	-	11.4	20.3	5.2	3.2	1.6
	<i>mg eq./kg</i>	<i>0.146</i>	-	<i>0.024</i>	<i>0.042</i>	<i>0.011</i>	<i>0.007</i>	<i>0.003</i>
120	%TRR	50.9%	-	6.6	13.3	3.3	2.1	0.8
	<i>mg eq./kg</i>	<i>0.150</i>	-	<i>0.020</i>	<i>0.039</i>	<i>0.010</i>	<i>0.006</i>	<i>0.002</i>
365	%TRR	74.9%	-	11.0	21.9	4.1	1.7	1.7
	<i>mg eq./kg</i>	<i>0.036</i>	-	<i>0.005</i>	<i>0.011</i>	<i>0.002</i>	<i>0.001</i>	<i>0.001</i>
Wheat Straw								
30	%TRR	56.9%	-	10.3	11.8	3.5	4.4	2.2
	<i>mg eq./kg</i>	<i>0.219</i>	-	<i>0.040</i>	<i>0.045</i>	<i>0.014</i>	<i>0.017</i>	<i>0.008</i>
120	%TRR	56.1%	-	10.4	12.5	2.9	2.6	1.7
	<i>mg eq./kg</i>	<i>0.323</i>	-	<i>0.060</i>	<i>0.072</i>	<i>0.017</i>	<i>0.015</i>	<i>0.010</i>
365	%TRR	65.4%	-	9.6	14.5	3.8	4.0	2.1
	<i>mg eq./kg</i>	<i>0.062</i>	-	<i>0.009</i>	<i>0.014</i>	<i>0.004</i>	<i>0.004</i>	<i>0.002</i>

^a Amount of extractable radioactivity after SPE and concentration that was injected on the HPLC column^b A dash (-) indicates that no residues was detected

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Table 7.9.3-1 continuation**ISOXAZOLE LABEL**

DAT		5-isoxazole amine	X11091296/ oxoisoxaben	X285341/ isoxaben	desmethyl isoxaben	Other ^a	# ^b	Largest Other ^c
Immature Lettuce								
30	%TRR	- ^d	-	1.8	-	13.9	9	2.7
	mg eq./kg	-	-	0.001	-	0.006		0.001
180	%TRR	-	-	-	-	13.7	3	6.1
	mg eq./kg	-	-	-	-	0.002		0.001
Radish Tops								
30	%TRR	1.2	0.5	2.5	0.6	25.1	26	2.6
	mg eq./kg	0.002	0.001	0.005	0.001	0.047		0.005
180	%TRR	-	-	-	-	30.9	11	7.5
	mg eq./kg	-	-	-	-	0.004		0.001
Radish Roots								
30	%TRR	1.3	0.7	3.6	0.7	18.7	28	2.8
	mg eq./kg	0.001	<0.001	0.003	0.001	0.014		0.002
Wheat Forage								
30	%TRR	0.9	-	4.9	-	22.1	11	6.0
	mg eq./kg	0.001	-	0.003	-	0.013		0.003
120	%TRR	-	-	2.3	-	26.3	12	5.2
	mg eq./kg	-	-	0.002	-	0.025		0.005
365	%TRR	-	-	-	-	29.7	13	7.1
	mg eq./kg	-	-	-	-	0.004		0.001
Wheat Hay								
30	%TRR	-	-	3.8	-	23.8	10	3.4
	mg eq./kg	-	-	0.008	-	0.050		0.007
120	%TRR	0.3	0.2	2.5	-	21.8	18	3.2
	mg eq./kg	0.001	0.001	0.007	-	0.064		0.009
365	%TRR	-	-	3.1	-	31.4	12	7.6
	mg eq./kg	-	-	0.002	-	0.015		0.004
Wheat Straw								
30	%TRR	0.4	0.4	2.1	-	21.8	13	4.3
	mg eq./kg	0.002	0.002	0.008	-	0.084		0.016
120	%TRR	0.5	-	1.9	-	23.7	20	3.7
	mg eq./kg	0.003	-	0.011	-	0.136		0.021
365	%TRR	-	-	1.9	-	29.6	17	5.7
	mg eq./kg	-	-	0.002	-	0.028		0.005

^a The category 'Other' includes radioactive peaks that did not co-chromatograph with any of the nine categories of metabolites or parent isoxaben.

^b Maximum number of radioactive peaks in the chromatogram that did not co-chromatograph with any of the nine categories of metabolites or parent isoxaben.

^c Largest of the 'Other' radioactive peaks for the analysis or analyses of a sample.

^d A dash (-) indicates that no residues was detected.

Table 7.9.3-2 presents sandy loam soil metabolites (DAR, 2006) for the different crop samples during the sampling period together with their respective DT90 values in sandy loam soil at aerobic conditions.

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Table 7.9.3-2 Residues of soil metabolites from isoxaben in different crop samples at different DAT.

Crop sample	X069203/ dimethoxy benzamide ¹		X341970/1-hydroxy isoxaben		X285341/ isoxaben	
	mg eq./kg	TRR	mg eq./kg	TRR	mg eq./kg	TRR
Immature Lettuce 30 DAT	0.008 phen.	13.7 phen.	0.003 phen. 0.005 iso	5 phen. 12.7 iso	0.001 phen. 0.001 iso	1.0 phen. 1.8 iso
Immature Lettuce 120 DAT	No data due to phyto-toxicity	No data due to phyto-toxicity	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -
Immature Lettuce 180 DAT	0.01 phen.	34.7 phen.	0.001 phen. 0.004 iso	2.7 phen. 27.2 iso	<0.001 phen. - ² Iso	0.8 phen. - iso
Immature Lettuce 365 DAT	No data ³	No data ³	No data ³	No data ³	No data ³	No data ³
Mature Lettuce 30 DAT	No data due to phyto-toxicity	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -
Mature Lettuce 120 DAT	No data due to phyto-toxicity	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity	No data due to phyto-toxicity -	No data due to phyto-toxicity -
Mature Lettuce 180 DAT	0.009 phen.	33.6 phen.	0.001 phen. No data iso	4.2 phen. No data iso	- No data iso	- No data iso
Mature Lettuce 365 DAT	No data ³	No data ³	No data ³	No data ³	No data ³	No data ³
Radish Tops 30 DAT	0.003 phen.	3.8 phen.	0.004 phen. 0.001 iso	4.5 phen. 1.0 iso	0.012 phen. 0.005 iso	14.1 phen. 2.5 iso
Radish Tops 120 DAT	No data due to phyto-toxicity	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -	No data due to phyto-toxicity -
Radish Tops 180 DAT	<0.001 phen.	1.5 phen.	0.001 phen. 0.001 iso	4.4 phen. 4.2 iso	- phen,iso	- phen,iso
Radish Tops 365 DAT	No data ³	No data ³	No data ³	No data ³	No data ³	No data ³
Radish Roots 30 DAT	0.002 phen.	3.4 phen.	0.002 phen.	3.2 phen.	0.014 phen	20.4 phen

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	X069203/ dimethoxy benzamide ¹		X341970/1-hydroxy isoxaben		X285341/ isoxaben	
Crop sample	mg eq./kg	TRR	mg eq./kg	TRR	mg eq./kg	TRR
			0.001 iso	1.0 iso	0.003	3.6 iso
Radish Roots 120 DAT	No data due to phyto- toxicity	No data due to phyto-	No data due to phyto-	No data due to phyto-	No data due to phyto-	No data due to phyto-
Radish Roots 180 DAT	No data ³	No data ³	No data ³	No data ³	No data ³	No data ³
Radish Roots 365 DAT	No data ³	No data ³	No data ³	No data ³	No data ³	No data ³
Wheat Forage 30 DAT	0.01 phen.	18.0 phen.	0.002 phen. 0.002 iso	4.3 phen. 3.0 iso	0.004 phen. 0.003 iso	6.3 phen. 4.9 iso
Wheat Forage 120 DAT	0.008 phen.	11.4 phen.	0.002 phen. 0.003 iso	3.0 phen. 3.7 iso	0.002 phen. 0.002 iso	2.8 phen. 2.3 iso
Wheat Forage 365 DAT	-	-	0.002 phen. 0.001 iso	14.2 phen 6.9 iso	- phen, iso	- phen,iso
Wheat Hay 30 DAT	0.016 phen.	6.8 phen.	0.007 phen. 0.011 iso	3.0 phen. 5.2 iso	0.008 phen. 0.008 iso	3.6 phen. 3.8 iso
Wheat Hay 120 DAT	0.018 phen.	10.3 phen.	0.007 phen 0.010 iso	4.2 phen 3.3 iso	0.003 phen. 0.007 iso	1.8 phen. 2.5 iso
Wheat Hay 365 DAT	-	-	0.004 phen 0.002 iso	6.6 phen. 4.1 iso	0.002 phen. 0.002 iso	4.2 phen. 3.1 iso
Wheat Straw 30 DAT	0.026 phen.	7.1 phen.	0.11 phen. 0.014 iso	2.9 phen. 3.5 iso	0.006 phen. 0.008 iso	1.6 phen. 2.1 iso
Wheat Straw 120 DAT	0.027 phen.	9.4 phen	0.012 phen. 0.017 iso	4.0 phen. 2.9 iso	0.006 phen. 0.011 iso	2.2 phen. 1.9 iso
Wheat Straw 365DAT	0.003 phen.	2.5 phen.	0.006 phen	5.2 phen	0.002 phen.	2.2 phen.

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	X069203/ dimethoxy benzamide ¹		X341970/1-hydroxy isoxaben		X285341/ isoxaben	
Crop sample	mg eq./kg	TRR	mg eq./kg	TRR	mg eq./kg	TRR
			0.004 iso	3.8 iso	0.002 iso	1.9 iso
DT 90* days	134		732		2103	

* DT 90 values are from DAR (2006) Annex B.8. A summary of laboratory studies on degradation of isoxaben and metabolites. The values presented are from aerobic conditions in a sandy loam soil.

Phen = phenyl label

Iso isoxaxol = label

¹ The metabolite is only present from phenyl labelled isoxaben.

² A dash (-) indicates that no residues was detected

³ TRR below 0.01 mg eq./kg

Summary

The soil metabolites 2,6 dimethoxy benzamide with a DT90 of 134 days is transferred to the leafy vegetable (lettuce immature and mature) at least until 180 DAT. For the root crop (radish roots) the data show that the parent isoxaben with a DT90 of 2103 days, was transferred in amounts above 0.1 mg eq./kg until 30 DAT. At 120 DAT no data were presented due to phytotoxicity of the samples. For later sampling dates the TRR was below 0.01 mg eq./kg and no analyses were performed. For the cereal crop (wheat hay and straw) data presented show that 2,6 dimethoxy benzamide, 1-hydroxyisoxaben or the parent were present above 0.1 mg eq./kg until 120 DAT.

Conclusion

With a pre-emergence soil application of isoxaben to a sandy soil in amounts of 250 g ai/ha the soil metabolites 2,6 dimethoxybenzamide with DT90 of 134 days, 1-hydroxyisoxaben with DT90 of 732 days and the parent isoxaben with DT90 of 2103 days are transferred from soil to tested edible crops (leafy crop immature and mature lettuce), root crop (radish) or cereals (wheat) above 0.01 mg eq./kg until 180 DAT. At 30 and 120 DAT phytotoxicity was observed for lettuce and radish. A plant back interval of at least 180 days is therefore recommended for all three tested rotational crop groups.

Bound residues

All crop samples at each DAT contained a high level (from 15-44% TRR) of non-extractable residues as was also observed in the metabolism in primary crops (DAR, 2006). The non-extractable residues in grain was quite high, maximum 88% TRR and 0.024 mg eq./kg in this study (Table 7.9.3-2). This was also observed in the primary crop (barley grain) where the non-extractable residue was 33% TRR and 0.004 mg eq./kg for phenyl- and 36.8% TRR and 0.007 mg eq./kg for isoxalyl labelled isoxaben at a soil application of 125 g/ha ¹⁴C isoxaben (DAR 2006). Yet, the level of non-extractable residues in grains is below the trigger value 0.05 mg e.q/kg and it is not necessary to further characterize according to the guideline (EU guideline 7028/VI/95 rev.3).

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Samples from hay and straw however contained levels of non-extractable residues from 18.4-33.3% TRR and eight of them above 0.05 mg eq./kg. These samples were further analysed, see Table 7.9.3-3 below.

Table 7.9.3-2 Level of non-extractable residue of ¹⁴C-isoxaben in confined rotational wheat grain**PHENYL LABEL**

			80/20 CH ₃ CN/ H ₂ O 40 °C, #1	80/20 CH ₃ CN/ H ₂ O 40 °C, #2	80/20 CH ₃ CN/ H ₂ O 70 °C, #1	80/20 CH ₃ CN/ H ₂ O 70 °C, #2	Total Extract- able	Non- Extract- able	Total Recov- ered
Sample ID	Total ^a								
Wheat Grain									
30	%TRR	100.0 %	18.8%	6.8%	4.8%	1.7%	32.1%	69.1%	101.2 %
	mg eq./kg	0.020	0.004	0.001	0.001	<0.001	0.006	0.014	0.020
120	%TRR	100.0 %	7.2%	4.0%	3.9%	3.5%	18.6%	88.8%	107.3 %
	mg eq./kg	0.027	0.002	0.001	0.001	0.001	0.005	0.024	0.029
<u>ISOXAZOLE LABEL</u>									
Wheat Grain									
30	%TRR	100.0 %	18.1%	7.6%	4.5%	1.8%	32.0%	65%	97%
	mg eq./kg	0.023	0.004	0.002	0.001	<0.001	0.007	0.015	0.022
120	%TRR	100.0 %	11.6%	7.9%	7.4%	3.9%	30.9%	71.2%	102.0 %
	mg eq./kg	0.03	0.003	0.002	0.002	0.001	0.009	0.021	0.030

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Table 7.9.3-3 Fractionation of the bound residues in isoxaben treated radish tops and wheat hay and straw

Sample	Bound Residue ^a	EDTA Soluble (Pectin)	NaClO ₂ Soluble (Lignin)	ADF Soluble	H ₂ SO ₄ Soluble (Cellulose)	Remain- ing Pellet	Overall Recovery ^b
30 DAT Radish Tops, Isoxazole							
%TRR	41.3%	3.2%	10.3%	1.2%	- ^c	0.3%	36.4%
mg eq./kg	0.077	0.006	0.019	0.002	-	<0.001	
120 DAT Wheat Hay, Phenyl							
%TRR	30.6%	4.1%	8.9%	1.4%	-	0.2%	47.4%
mg eq./kg	0.055	0.007	0.016	0.003	-	<0.001	
120 DAT Wheat Hay, Isoxazole							
%TRR	20.5%	2.2%	7.2%	1.1%	0.1%	0.2%	52.4%
mg eq./kg	0.060	0.006	0.021	0.003	<0.001	0.001	
120 DAT Wheat Straw, Phenyl							
%TRR	33.3%	4.4%	10.7%	2.2%	0.4%	0.2%	54.1%
mg eq./kg	0.097	0.013	0.031	0.006	0.001	0.001	
120 DAT Wheat Straw, Isoxazole							
%TRR	29.5%	3.9%	8.1%	2.1%	0.3%	0.2%	49.4%
mg eq./kg	0.170	0.022	0.047	0.012	0.002	0.001	

^a % of TRR remaining in the extracted tissue.^b Procedural recovery for the 4-step bound residue process, ((Pectin + Lignin + ADF Soluble + Cellulose + Remaining Residue)/Bound Residue).^c A dash (-) indicates that nothing was detected in this fraction**Summary**

Non-extractable (bound residues) in radish tops, hay and straw from cereals, as also reported in DAR (2006), was associated primarily with the lignin and pectin fractions.

B.7.9.4 Metabolic pathway in rotational lettuce, radish and wheat

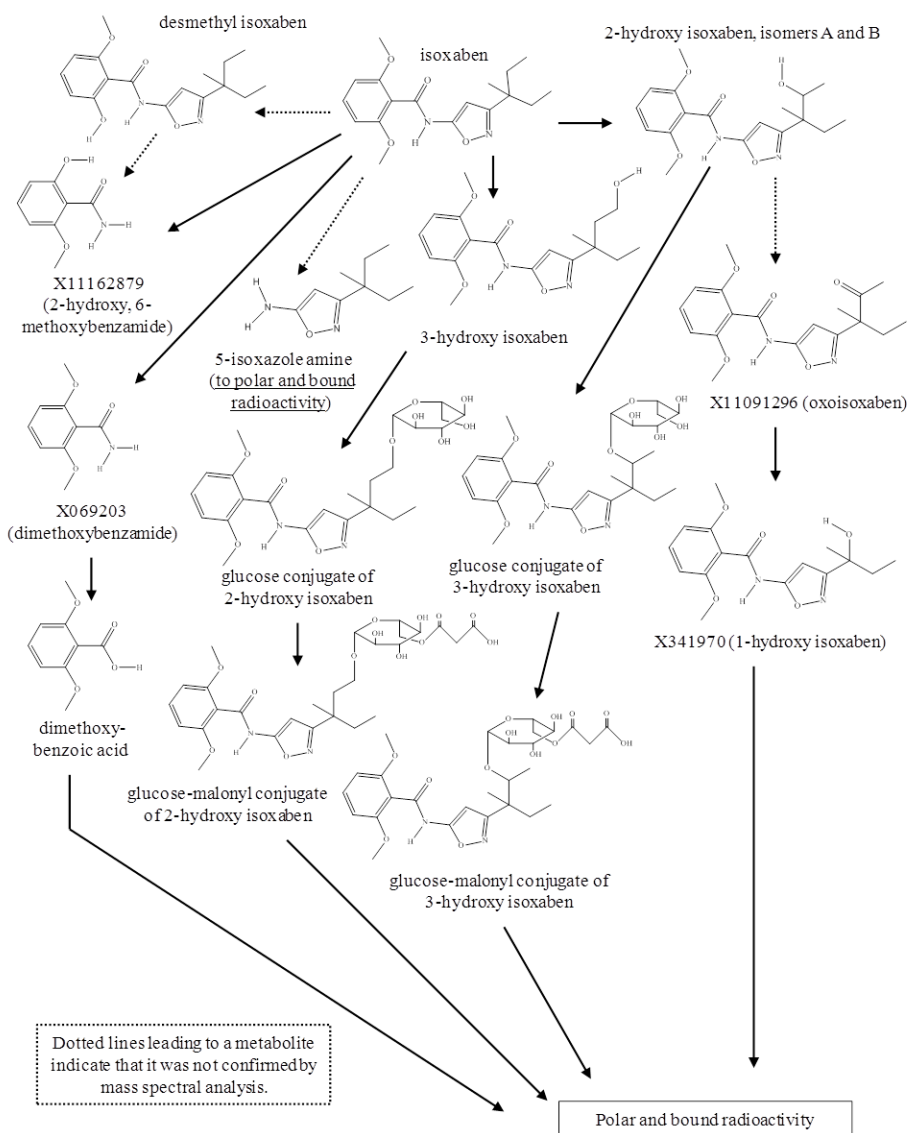
The proposed metabolic pathway is presented in Figure 7.9.4-1. In general, metabolism of isoxaben proceeded by hydroxylation at two different positions on the alkyl side chain of the isoxazole portion of the molecule to yield the 2-hydroxy isoxaben isomers A and B and 3-hydroxy isoxaben. One or both of these hydroxylated metabolites is subsequently conjugated with glucose and then glucose-malonyl. Demethylation of one of the methoxy groups on the phenyl portion of isoxaben to give desmethyl isoxaben is thought to be minimal or unlikely due to co-chromatography in only a few samples and the fact that it was not detected by mass spectral analysis (MS). Further degradation to X11162879(2-hydroxy, 6-methoxybenzamide), while possible, was not confirmed by MS. Cleavage of the amide bridge in isoxaben to produce X069203 (dimethoxybenzamide) and dimethoxybenzoic acid was demonstrated by MS and by co-chromatography, although whether this occurs in soil and is taken up by plants or whether metabolism is occurring in both plants or soil cannot be determined from available data. The 5-isoxazole amine is thought to be minimal or unlikely due to co-chromatography in only a few samples and the fact that it was not detected by MS. X11091296 (oxoisoxaben), which was suspected by co-chromatography in a few samples, is proposed as an intermediate during oxidation of 2-hydroxy isoxaben isomers A and B and then loss of a portion of the alkyl side chain to yield X341970 (1-hydroxy isoxaben). X341970 is known as the major soil metabolite and it is known to be a metabolite from a prior spring barley

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metabolism study although it could not be determined from this study whether it was produced from soil or plant metabolism or both. Ultimately metabolism is proposed to proceed to unidentified polar and bound radioactivity.

Fig 7.9.4-1 Proposed metabolic pathway for isoxaben in rotational crops



Comparison of residue profiles in primary and rotational crop

Metabolism in primary crops has previously been evaluated in wheat, barley and oil seed rape (DAR, 2006). The application rates in these studies ranged from 125 to 500 g/ha. Oilseed rape plants absorbed on average 1.5 times more isoxaben than wheat plants. Translocation was observed to be slower in oilseed rape compared to wheat. The same metabolic profile was observed in the two plant species with free and conjugated hydroxylated metabolites being produced at generally similar rates. Rape is sensitive to the herbicide, but wheat is not. Difference in absorption and translocation can partly explain the selectivity of the herbicide. Wheat plants

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absorbed less herbicide through their roots, and can quickly translocate it into their shoots, where its degradation is the most efficient. Rape plants which accumulate more isoxaben in their roots translocate it less easily into their leaves, and thus tend to keep it in organs that can be severely damaged.

Characterization of the residues in rotational in lettuce, radish and wheat from the three plant-back intervals (30, 120, 185, 365 DAT) after application of 250 g ai /ha of isoxaben, indicate that the residue profiles is similar as that seen in the primary crop metabolism studies of (DAR, 2006). This include residues of isoxaben the non-conjugated forms of the 1-hydroxy, 2- hydroxyl (both isomers) and the 3-hydroxy isoxaben metabolites and four glucose or glucose malonyl conjugates of the 2- and 3-hydroxy isoxaben metabolites, Tables 7.9.3-1. As in the winter cereal metabolism study, two amide bridge cleavage products, the dimethoxybenzamide and dimethoxybenzoic acid metabolites, were also observed in these samples, with the benzamide metabolite usually being the main cleavage product. For the tested rotational crops additional metabolites were identified, but all below 0.004 mg e.q./kg at 180 DAT in crops for human consumption

Summary

Under tested conditions the residue profile of isoxaben in rotational lettuce (leafy vegetable) and radish (root crop) and cereals was shown to be qualitatively the same as in the primary crops cereals and oil seed rape. As in the primary crop metabolism studies, most of the hay and straw samples also contained significant levels of non-extractable residues (18.4-33.3% of the TRR or 0.010-0.170 mg eq./kg).

B.7.9.5 Residues definition in tested rotational crops in comparison to tested primary crop

Metabolism has been examined for rotational crops in three crop groups; leafy vegetable (lettuce), root crops (radish) and cereals (wheat). The route of metabolism appears to be the same as the same type of metabolites were found in these crops, though sometimes at different levels. The same types of metabolites were also found in the primary crops cereal and oil seed rape..

The residue profile of isoxaben in rotational lettuce (leafy vegetable) and radish (root crop) and cereals was shown to be qualitatively the same as in primary crops cereals and oil seed rape. It is therefore concluded that metabolism in primary and rotational crops is similar and a separate residue definition in rotational crops is not necessary provided that the GAP is within the tested conditions in this CRC study

B.7.9.6 Residue definition in livestock and animal products**Animal dietary burden**

The maximum intake of residues from rotational crops in dairy cattle, beef cattle and poultry was calculated for the parent isoxaben at 365 DAT, and presented in the table below.

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Table 7.9.6-1 Maximum feed intake of residues from tested rotational crops in different livestock species*Worst case scenario guideline EU guideline 7031/VI/95 rev.4*

Crop group	% of total diet	Intake of dry matter (kg/day)	Intake of fresh food (kg/day)	Residue level for phenyl label in fresh produce (mg/kg)	Residue intake (mg/kg)	Residue level for isoxazol label in fresh produce (mg/kg)	Residue intake (mg/kg)
Dairy cattle							
Wheat hay	80	16	18.8 (16:85x100)	0.054	1.02	0.048	0.90
Wheat straw	20	4	4.7 (4:86x100)	0.111	0.52	0.095	0.45
Total intake per day mg/cattle/day					1.54		1.35
Total intake mg/kg total diet (dry matter)					0.077		0.068
Total intake per day (mg/kg bw/day)					0.003		0.002
Beef cattle							
Wheat hay	50	7.5	8.8 (7.5:0.85)	0.054	0.48	0.048	0.42
Wheat straw	50	7.5	8.7 (7.5:0.86)	0.111	0.97	0.095	0.83
Total intake per day mg/beef cattle /day					1.45		1.25
Total intake mg/kg total diet (dry matter)					0.097		0.083
Total intake per day (mg/kg bw/day)					0.004		0.004
Chicken							
Root and tubers	20	0.24	2.4 (0.24:10x100)	0.001	0.002	0.001	0.002
Wheat grain	70	0.84	0.98(0.84:86x100)	0.003	0.003	0.006	0.006
Total intake per day mg/poultry /day					0.005		0.008
Total intake mg/kg total diet (dry matter)					0.005		0.008
Total intake per day (mg/kg bw/day)					0.003		0.003

* TRRs presented for 365 DAT from Table 7.9.2-1, are used in the calculations

The calculations in Table 7.9.6-1 above show that worst case intake of residues from tested rotational crops in the total diet for dairy cattle is 0.077 mg/kg for phenyl label and 0.068 mg/kg for isoxazol labelled isoxaben. For beef cattle the respectively values are 0.097 and 0.083 mg/kg. These values are 10 times-below trigger value 0.1 kg/mg (of total diet as received) for a feeding study (EU guideline 7031/VI/95 rev.4). For poultry the total intake of residues from isoxaben varied from 0.005(phenyl label) and 0.003mg/kg (isoxazol label) and thus below the trigger value. Thus, under tested condition with pre-emergence soil application of isoxaben at 250 g a.i/ha (2 X critical GAP) intakes of residues from rotational crops used as feed are of no concern.

RMS comments and conclusion

The study was performed according to OECD guideline 502, and the principal parts according to GLP. The study was well performed and reported.

As in the primary crop metabolism studies, most of the hay and straw samples contained significant levels of non-extractable residues (18.4-33.3% of the TRR or 0.010-0.170 mg/kg). The samples which contained residues

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Addendum to DAR following submission of confirmatory data

above 0.05 mg e.q./kg (all hay and straw samples) were further analysed. Equally as reported in the DAR (2006) these residues were under tested conditions shown to be associated primarily with the lignin and pectin fractions lignin, pectin and hemicelluloses and thus strongly bound and of low concern.

Under tested conditions the residue profile of isoxaben in rotational lettuce (vegetable) and radish (root crop) and cereals was shown to be qualitatively the same as in primary crop cereals and oil seed rape (vegetable). It is therefore concluded that metabolism in primary and rotational crops is similar and a separate residue definition in rotational crops is not necessary provided that the GAP is within the tested conditions in this CRC study.

Residues in cereal feed were at each plant back interval at a higher level than the residues in commodities for human consumption. Calculation of the maximum dietary burden for livestock at 365 DAT shows that residues from tested rotational crops in the total diet for dairy cattle and beef cattle is about 10 times below the trigger value 0.1 mg/kg for feeding study (EU guideline 7031/VI/95 rev.4) The qualitative nature as well as the level of the terminal residue in the RAC at the time of harvest or when utilized for livestock feed is of no concern if isoxaben is used within the tested condition (2X recommended GAP of 125 g ai/ha) at 365 DAT.

With a pre-emergence soil application of isoxaben to a sandy loam soil in amounts of 250 g ai/ha the soil metabolites 2,6 dimethoxybenzamide with DT90 of 134 days, 1-hydroxyisoxaben with DT90 of 732 days and the parent isoxaben with DT90 of 2103 days are transferred from soil to tested edible crops (leafy crop immature and mature lettuce), root crop (radish) or cereals (wheat) above 0.01 mg eq./kg until 180 DAT. At 30 and 120 DAT phytotoxicity was observed for lettuce and radish. A plant back interval of at least 180 days is therefore recommended for the three tested rotational crop commodities (cereal, leafy vegetable and root crops).

B.7.9.7 References relied on

Annex point / reference number	Author(s)	Year	Title Source (where different from company) Company, Report No GLP or GEP status (where relevant) Published or not	Data Protection Claimed* Y/N	Owner
IIA 8.2.8	Gaper L.K, Y. Adelfinskaya	2013	A confined Rotational Crop Study with ¹⁴ C-Isoxaben, Phenyl and Isoxazole Labels, Study ID 110534	Y	Dow Agro-Science
IIA 6.6.1		2006	Volume 3, Annex B.7. Residue data DAR (2006)	N	
IIA 6.6.1.		2006	Volume 3, Annex B.8. Environmental fate and behavior DAR (2006)	N	

*: Data protection claimed by the notifier, no assessment on acceptability at this stage.