FINAL REGISTRATION REPORT

Part B

Section 7

Metabolism and Residues

Detailed summary of the risk assessment

Product code: GLOB2013F

Product name(s): Observer

Chemical active substance:

Zoxamide, 450 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

Applicant: Globachem NV

Submission date: January 2024

Update: July 2024

MS Finalisation date: 19/12/2024

Version history

When	What
January 2024	Initial dossier submission by applicant for approval of new product
April 2024	Dossier sent for evaluation
July 2024	Applicant revision 01 to address zRMS initial comments
September 2024	zRMS finalised evaluation
December 2024	zRMS finalised evaluation after commenting period

Table of Contents

zRMS comments:
The text highlighted in grey was provided by the evaluator.

7 Metabolism and residue data (KCA section 6)

7.1 Summary and zRMS Conclusion

Comparison of intended and critical EU GAPs

Zoxamide

Type of GAP	Crop	Max number of applications	Method of application	Growth stage at last application	Max appl. rate per treatment (g a.s./ha)	PHI (days)
critical NEU GAP, RAR (RMS LV, 2017)	Potatoes	5	Foliar treatment-spraying	BBCH 20-80	180	7
Critical NEU GAP (EFSA Journal. 2023;21:e8427)	Potatoes	4	Foliar treatment-spraying	BBCH 31-93	180	7
Intended GAP	Potatoes	3	Downward spraying	BBCH 21-79	135	7
EFSA Journal 2017;15(9):4980	Table/wine grapes	5	3-d broadcast with mist blower	BBCH 15-79	180	28
Critical NEU GAP (EFSA Journal. 2023;21:e8427)	Table/wine grapes	4	Foliar treatment-spraying	BBCH 14-89	132	28
Intended critical GAP	Table/wine grapes	2	Air assisted	BBCH 53-83	166	28

EFSA Journal. 2023;21:e8427: The metabolism of zoxamide in plant was investigated in primary and rotational crops. According to the results of the metabolism and the hydrolysis studies, the residue definition for enforcement in primary, rotational crops, honey and processed commodities can be proposed as zoxamide (sum of constituent isomers) while for risk assessment can be proposed as sum of zoxamide and metabolite RH-141452, expressed as zoxamide. For processed commodities, a separate residue definition for risk assessment for metabolite RH-150721 was also proposed.

The proposed uses of zoxamide in the formulation GLOB2013F do not represent unacceptable acute and chronic risks for the consumer.

After the entry into force of the approved draft Commission Regulation PLAN/2024/307, there will be no change to the MRLs for potatoes and grapes. Therefore, the proposed uses will not pose chronic and acute risks for the consumer when applied according to intended GAPs.

7.1.1 Critical GAP(s) and overall conclusion

Selection of critical uses and justification

The critical GAPs with respect to consumer intake and risk assessment for the preparation GLOB2013F are presented in Table 7.1-1. They have been selected from the individual GAPs in the central zone for potato and grapes (table and wine grapes). A list of all intended uses within the central zone is given in Part B, Section 0.

Overall conclusion

The data available are considered sufficient for risk assessment. An exceedance of the current MRL of 0.02 mg/kg for zoxamide in potato and 5 mg/kg in table/wine grapes as laid down in Reg. (EU) 396/2005 is not expected.
The chronic and the short-term intakes of zoxamide residues are unlikely to present a public health concern. As far as consumer health protection is concerned, PL, zRMS agrees with the authorization of the intended use.

According to available data, no specific mitigation measures should apply.

Data gaps

- None in the framework of this application.

EU data gaps:
Zoxamide
Data gaps identified in peer review of the pesticide risk assessment of the active substance zoxamide for residue section, EFSA Journal 2017;15(9):4980:

- The nature of residues for zoxamide including the nature of RH-141455 and RH-141452 under standard hydrolysis conditions representative of pasteurisation, baking/brewing/cooking, sterilisation is required (relevant for all representative uses evaluated; submission date proposed by the applicant: unknown; see Section 3).
- A consistent reporting of the growth stage at application time according to BBCH scale for the potato plant in accordance with requested GAP (BBCH 20–80) shall be provided to allow the assessment of the field trials (relevant for representative uses in potato; submission date proposed by the applicant: unknown; see Section 3).
- Sufficient field trials in potatoes in NEU and SEU supporting the critical GAP for the full residue definition for RA are required (relevant for representative uses in potato; submission date proposed by the applicant: unknown; see Section 3).
- A poultry metabolism study is required (relevant for representative uses in potato; submission date proposed by the applicant: unknown; see Section 3).
- Log Po/w for RH-141288 and RH-127450 and an assessment regarding their potential fat-solubility according to FAO (2009) should be provided (relevant for representative uses in potato; submission date proposed by the applicant: unknown; see Section 3).
- Finalisation of the residue definitions and a dietary exposure assessments should be performed that need to account for potential changes in the isomer ratio of zoxamide and its metabolites that contain a chiral centre (relevant for all representative uses evaluated; submission date proposed by the applicant: unknown; see Section 3).

Issues that could not be finalised in residue section at EU level, EFSA Journal 2017;15(9):4980:

- The human health and environmental risk assessment consequent to potential changes in the isomer composition for zoxamide and metabolites RH-127450, RH-163353, (RH-150721 human health only)] could not be finalised (see Sections 2, 3, 4 and 5).3)

- The consumer risk assessment could not be finalised due to a number of data gaps that likely have an impact on the assessment of residue levels and due to the pending toxicological evaluation of metabolites which are included in the residue definition for risk assessment and monitoring (see Section 3

Table 7.1-1: Acceptability of critical GAPs (and respective fall-back GAPs, if applicable)

GAP rev. 4.0, date: 13/11/2023

PPP (product name/code):	GLOB2013F	Formulation type:	SC (suspension concentrate)
Active substance 1:	zoxamide	Conc. of as 1:	450 g/L
Applicant:	Globachem NV	Professional use:	☒
Zone(s):	Central	Non professional use:	☐
Verified by MS:	yes		
Field of use:	fungicide		

1	2	3	4	5	6	7	8	9	10	11	12	13	14	Conclusion
Use-No. (e)	**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 (f)	
					Method / Kind	Timing / Growth stage of crop & season	Max. number a) per use b) per crop/ season	Min. interval between applications (days)	kg or L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg 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	CZ, HU, IE, PL, RO, SK	Seed, ware and starch potato (SOLTU) (code:0211000)	F	*Phytophthora infestans* (PHYTIN)	Downward spraying	BBCH 21-79	a) 3 b) 3	7	a) 0.3 b) 0.9	a) 0.135 b) 0.405	150-300	7		A
2	CZ, HU, IE, PL, RO, SK	Table and wine grape (VITVI) (codes: 0151010 and 0151020)	F	*Plasmopara viticola* (PLASVI)	Air assisted	BBCH 13-52	a) 2 b) 2	8	a) 0.3 b) 0.6	a) 0.135 b) 0.270	100-1000	28	Maximum 2 apps per season. 0.23 L/10.000 m² LWA corresponding to 0.1035kg a.i./10.000 m² LWA. For early BBCH stages (13-52), the maximum rate allowed per ha soil	A
3	CZ, HU, IE, PL, RO, SK	Table and wine grape (VITVI) (codes: 0151010 and 0151020)	F	*Plasmopara viticola* (PLASVI)	Air assisted	BBCH 53-83	a) 2 b) 2	8	a) 0.368 b) 0.736	a) 0.166 b) 0.332	100-1000	28		A

													is set at 0.3L/ha soil corresponding to 13000 m^2 LWA. For later stages (BBCH53-83), the maximum rate allowed per ha soil is set at 0.368L/ha soil corresponding to 16000 m^2 LWA. **RO, SK, HU:** Range from 0.17 to 0.23 L/10.000 m^2 LWA	
4	CZ, HU, IE, PL, RO, SK	Seed, ware and starch potato (SOLTU) (code:0211000)	F	*Phytophthora infestans* (PHYTIN)	Downwards spraying	BBCH 21-79	a) 3 b) 3	7	a) 0.29 b) 0.87	a) 0.130 b) 0.390	150-300	7	Alternative GAP with a slightly lower dose rate in order to maintain a mitigation of maximum 10 m VFS only where necessary	A

* Use number(s) in accordance with the list of all intended GAPs in Part B, Section 0 should be given in column 1
** Use also code numbers according to Annex I of Regulation (EU) No 396/2005
*** 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 11 “Conclusion”

A	Exposure acceptable without risk mitigation measures, safe use
R	Further refinement and/or risk mitigation measures required
N	Exposure not acceptable, no safe use

7.1.2 Summary of the evaluation

Table 7.1-2: Toxicological reference values for the dietary risk assessment of zoxamide and ~~propamocarb-HCl~~ RH-150721

Reference value	Source	Year	Value	Study relied upon	Safety factor
zoxamide					
ADI	SANTE/10052/2018 Rev 2	2018	0.5 mg/kg bw per day	1-year dog	100
ARfD	SANTE/10052/2018 Rev 2	2018	not allocated – not necessary		
RH-150721, zoxamide metabolite in processed commodities					
ADI	EFSA	2023	0.04 mg/kg bw per day	90-day rats	1000
ARfD	EFSA	2023	0.22 mg/kg bw	14-day rats	300

7.1.2.1 Summary for zoxamide

Table 7.1-3: Summary for zoxamide

Use-No.*	Crop	Plant metabolism covered?	Sufficient residue trials?	PHI sufficiently supported?	Sample storage covered by stability data?	MRL compliance	Chronic risk for consumers identified?	Acute risk for consumers identified?
1,4	Potato	Yes	Yes (n=4 for zoxamide, RH-141452, RH-141455; all below LOQ)	Yes	Yes	Yes	No	No
2	Table and wine grape	Yes	Yes (n=10 for zoxamide; n=4 for RH-141452 – below LOQ)	Yes	Yes	Yes		No
3	Table and wine grape	Yes	Yes (n=10 for zoxamide; n=4 for RH-141452 – below LOQ)	Yes	Yes	Yes		No

* Use number(s) in accordance with the list of all intended GAPs in Part B, Section 0 should be given in column 1

The effects of processing on the nature of zoxamide residues have been investigated. Data on effects of processing on the amount of residue have been submitted.
These data were considered for risk assessment.

Residues in succeeding crops have been sufficiently investigated taking into account the specific circumstances of the cGAP uses being considered here. It is very unlikely that residues will be present in succeeding crops.

Considering dietary burden and based on the intended uses, no significant modification of the intake was calculated for livestock. Further investigation of residues as well as the modification of MRLs in commodities of animal origin is therefore not necessary.

7.1.2.2 Summary for GLOB2013F

Table 7.1-4: Information on GLOB2013F (KCA 6.8)

Crop	PHI for GLOB2013F proposed by applicant	PHI/ Withholding period* sufficiently supported for	PHI for GLOB2013F proposed by zRMS	zRMS Comments (if different PHI proposed)
		zoxamide		
Potato	7 days	Yes	7 days	-
Grape (table and wine)	28 days	Yes	28 days	-
Grape (table and wine)	28 days	Yes	28 days	-

NR: not relevant
* Purpose of withholding period to be specified
** F: PHI is defined by the application stage at last treatment (time elapsing between last treatment and harvest of the crop).

Waiting periods before planting succeeding crops

Not relevant

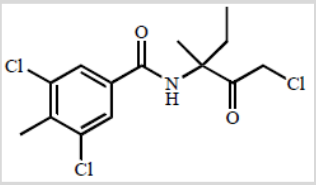
Assessment

Reference is made to the EU review of zoxamide where the critical uses were evaluated.

7.2 Zoxamide

General data on zoxamide are summarized in the table below.

Table 7.2-1: General information on Zoxamide

Active substance (ISO Common Name)	Zoxamide
IUPAC	*(RS)*-3,5-dichloro-*N*-(3-chloro-1-ethyl-1-methyl-2-oxopropyl)-*p*- toluamide.
Chemical structure	3,5-dichloro-*N*-(3-chloro-1-ethyl-1-methyl-2-oxo[rp[ul)-4-methylbenzamide 
Molecular formula	$C_{14}H_{16}NO_2Cl_3$
Molar mass	336.65 g/mol
Chemical group	benzamides
Mode of action (if available)	Arrests nuclear division and destroys the microtubule cytoskeleton of oomycete organisms.
Systemic	No
Company (ies)	Gowan Crop Protection Ltd
Rapporteur Member State (RMS)	Latvia
Approval status	Approved (01/07/2018) Reg. (EU) No 540/2011 as amended by Reg. (EU) 2018/84 and Reg. (EU) 2018/692
Restriction	~~Only use as a fungicide~~ None
Review Report	SANTE/10052/2018 Rev 2 (23 March 2018)
Current MRL regulation	Regulation (EC) No 2017/171
Peer review of MRLs according to Article 12 of Reg No 396/2005 EC performed	~~Pending~~ Yes, EFSA Journal. 2023;21:e8427
EFSA Journal : Conclusion on the peer review	EFSA Journal 2017; 15 (9):4980
EFSA Journal: conclusion on article 12	EFSA Journal 2023;21:e8427
Current MRL applications on intended uses	-

* Notifier in the EU process to whom the a.s. belong(s)
** If yes: EFSA, YYYY - see list of references

7.2.1 Stability of Residues (KCA 6.1)

7.2.1.1 Stability of residues during storage of samples

Available data

Three new stability studies have been submitted by the applicant in the framework of this application. Results are summarized in the Table below. The detailed assessment of this/these stud(y/ies) is/are presented in Appendix 2.

Table 7.2-2: Summary of stability data achieved at ≤ - 18°C

Matrix	Characteristics of the matrix	Acceptable Maximum Storage duration	Reference
Data relied on in EU			
Plant products			
Potato	High starch content	24 months	EFSA, 2017
Grapes	High acid content	18 months	EFSA, 2017
Wine	High acid content	24 months	EFSA, 2017
Honey		85 days	EFSA Journal 2023;21:e8427
New data			
Plant products			
Potato	High starch content	13 months	Gustloff, C. & Mohaupt, R., 2023, S21-07041
Grapes	High acid content	13 months	Gustloff, C. & Mohaupt, R., 2023, S21-07041
Wine	High acid content	13 months	Gustloff, C. & Mohaupt, R., 2023, S21-07043
Honey		6 months	Gustloff, C., S23-100693

Conclusion on stability of residues during storage

Storage stability studies of zoxamide assessed in this sections cover the requested use on potatoes and grapes for GLOB2013F.

zRMS comments:
Reference: EFSA, 2017
“Storage stability was sufficiently demonstrated for zoxamide and its metabolites in grapes (berries, juice, wine, raisins) and potato tubers.”
References: Latvia, 2016
“Storage stability of zoxamide (RH-7281) residues was shown to be acceptable for at least 24 months in potatoes and for at least 18 months in grapes. Stability of zoxamide was demonstrated over 24 months in grape juice, raisins and wine when stored frozen at ≤-18°C.
Residues of RH-141452 and RH-141455 were shown to be stable for at least 24 months in potato tubers when stored frozen at ≤-18°C.
Residues of RH-150721 were shown to be stable for at least 18 months in grape berries and at least 24

months in wine when stored frozen at ≤-18°C."

The storage periods from sampling to analysis are sufficiently covered by data.

7.2.1.2 Stability of residues in sample extracts (KCA 6.1)

Not relevant.

zRMS comments:
Procedural recoveries obtained during residue analysis demonstrate the stability of residues in sample extracts and fully support the residue data presented in the submission.

7.2.2 Nature of residues in plants, livestock and processed commodities

7.2.2.1 Nature of residue in primary crops (KCA 6.2.1)

Available data

One new metabolism study has been submitted by the applicant in the framework of this application. All metabolism studies are summarized in the table below. The detailed assessment of new study is presented in Appendix 2.
The new metabolism study submitted in the context of this dossier acts as supplementary information to the metabolism study as submitted and evaluated in the RAR of zoxamide to confirm results.

Table 7.2-3: Summary of plant metabolism studies

Crop Group	Crop	Label position	Application and sampling details					Reference
			Method, F or G (a)	Rate (kg a.s./ha)	No	Sampling (DAT)	Remarks	
EU data								
Fruits and fruiting vegetable	Grape	Uniformly isotopically labelled in the phenyl ring	Foliar application	1.867	3	1	Zoxamide was the only significant component of the residue (58.3 TRR)	EFSA, 2017
				0.5	3	28	Zoxamide was the only significant component of the residue (92.2% TRR)	
Root and tuber vegetables	potato	Uniformly isotopically labelled in the phenyl ring	Foliar application	0.9	3	14	No parent zoxamide was found on potato tubers. The main components of the residue in potato tubers were the metabolites RH-141452	EFSA, 2017

							and RH-141455.	
New data								
Fruits and fruiting vegetable	Grape	Uniformly isotopically labelled in the phenyl ring	Foliar application	0.180 g as/ha	2	28	Parent zoxamide was the only major residue detected (96.6% TRR on leaves and 94.6% TRR on fruits).	Maric, A., 2023, S22-01899

Summary of plant metabolism studies reported in the EU

The metabolism in primary crops was investigated after foliar spray application in fruit crops (grapes) and root crops (potato) using zoxamide ^{14}C-labelled in the phenyl ring. In fruits, zoxamide was the main component of the total radioactive residue (TRR)with 98% in grapes. The remaining TRR was extensively metabolised to a range of metabolites representing less than 10% TRR in this commodity. In potatoes in contrast to grapes where metabolism was investigated two major metabolites RH-141452 and RH-141455 were observed at 21% and 39% , respectively. Parent zoxamide was not found in potato tubers.
The metabolic pathways in the primary crop groups were adequately elucidated but a comparable pathway could not be confirmed yet. Because of outstanding toxicological issues on the metabolites RH-141452 and RH-141455, an overall residue definition (RD) for plants cannot be set.
For risk assessment (RA) it is therefore proposed to set provisional residue definitions for fruit crops as the sum of zoxamide and RH-141452 and for root crops the sum of metabolites RH-141452 and RH-141455.
The residue definition for monitoring for the root crops is proposed as zoxamide for fruit crops given that zoxamide is a good marker for these crop groups. For the root crops, the sum of metabolites RH-141455 and RH-141452 is proposed.

Summary of new plant metabolism studies

The metabolism of zoxamide in grape plants was investigated after two foliar applications at BBCH 81 and BBCH83 at a total application rate of 413 g a.s./ha. Fore metabolic analyses, grape leaves and fruits were harvested at BBCH 89, corresponding to 28 days after the last treatment.
The parent compound zoxamide was the only major residue detected in conventional extracts of grape leaves and fruits accounting for 96.6% of TRR and 94.6% of TRR respectively.
On the basis of the nature and amount of the residue in the extracts in this new study, it can be concluded that zoxamide does not undergo any relevant metabolic reaction in grape plants.

Conclusion on metabolism in primary crops

The metabolism of zoxamide in all primary crops was similar in the three crop groups whereas additional metabolites were identified in potatoes. The metabolism in rotational crops is similar to the metabolism observed in primary crops and the processing of zoxamide leads to the generation of several degradates. As the parent compound was found to be a sufficient marker in fruits, roots and pulses and oilseeds, rotational crops and processed commodities, the residue definition for enforcement is proposed as zoxamide only (sum of constituent isomers) (for primary, rotational crops, honey and processed commodities).
For risk assessment, parent and metabolite RH-141452 are toxicologically relevant and thus should be considered in the consumer exposure. Metabolite RH-141452 encountered in the rat metabolism and was

considered covered by the toxicological profile of the parent compound. Therefore, the residue for risk assessment in raw commodities from primary and rotational crops and for honey is proposed as the sum of zoxamide and RH-141452, expressed as zoxamide.
The only metabolite consistently found at levels above the LOQ in processed commodities was RH-150721 (e.g. grape juice, must, wine, raisins, and tomato juice, puree, canned). For metabolite RH-150721 an ADI lower than the parent was set; also, an ARfD was derived for this metabolite while for parent was considered not necessary. Therefore, for processed commodities, two separated residue definitions for risk assessment are proposed: sum of zoxamide and RH-141452, expressed as zoxamide; metabolite RH-150721.

The new metabolism study submitted in the context of this dossier (Maric, A., 2023) as supplementary information, confirms the conclusions drawn in the metabolism study submitted and evaluated in the RAR of zoxamide.

zRMS comments:
According to EFSA Journal. 2023;21:e8427: In the framework of the peer review, the metabolism in primary crops was investigated after foliar spray application in fruits (grape, tomato, cucumber), pulses and oilseeds (pea) and root crops (potato) using zoxamide ^{14}C-labelled in the phenyl ring.
In fruits, zoxamide was the main component of the total radioactive residue (TRR) in grape (98%). The remaining TRR was extensively metabolised to a range of metabolites representing less than 10% TRR in these commodities.
In potatoes, two major metabolites RH-141452 and RH-141455 were observed at 21% TRR(0.037 mg/kg) and 39% TRR (0.067 mg/kg), respectively. Parent zoxamide was not found in potato tubers.
The Applicant provided one new metabolism study in grape plants. Zoxamide was the main component of the total radioactive residue (TRR) in grape leaves (96.6%) and in grape fruits (95.6%).
The metabolism of zoxamide in grape plants and potatoes following foliar application is sufficiently addressed to support the proposed uses of GLOB2013F.

7.2.2.2 Nature of residue in rotational crops (KCA 6.6.1)

Available data

No new data submitted in the framework of this application.

Table 7.2-4: Summary of metabolism studies in rotational crops

Crop group	Crop	Label position	Application and sampling details					Reference
			Method, F or G *	Rate (kg a.s./ha)	Sowing intervals (DAT)	Harvest Intervals (DAT)	Remarks	
EU data								
Root and tuber vegetables	Radish, turnip	Uniformly isotopically labelled in the phenyl ring	F	4 x 0.5	30;137;210;365	137	The 210 and 365 mature mustard and radish samples lost due to freezer failure.	EFSA, 2017
Leafy vegetables	Mustard		F		30;145;210;365	145		

Cereals (small grain)	Sorghum		F		30;137;210;365	137		
Pulses and oilseeds	Soybean		F		30;137;210;365	137		

* Outdoor/field application (F) or glasshouse/protected/indoor application (G)

Summary of plant metabolism studies reported in the EU

In a rotational crop study, ^{14}C-labelled zoxamide was applied directly to bare soil at a rate of 4 × 0.50 kg/ha equivalent to 2.2N rate for potatoes. Zoxamide was not detected in any of the analysed plant parts, instead several metabolites were observed amongst which RH-141452 was identified mainly in the immature parts of the crops (12% TRR in immature mustard leaves, 15% TRR in immature radish tops, 23.5% TRR in immature soybean forage) and to a lower extend in mature crops (3% TRR in mustard leaf, 7% TRR in radish tops, not detected in soybean seeds). However, it remains unclear whether degradation of zoxamide occurred in soil with the preferential plant uptake of metabolites or whether the degradation of zoxamide is part of the metabolism in plants. In view of the representative uses, it can be concluded that significant individual residue compounds are unlikely to be present in rotational crops while for a more critical use pattern in terms of application rate the issue may have to be reconsidered for future uses.

Conclusion on metabolism in rotational crops

Rotation crop and primary crop metabolisms seems to be similar. Very little uptake of residues from soil. Parent zoxamide not detected in following crops. The crops metabolite RH-141452 was found at trace levels in following crops. No detectable residues of zoxamide or related metabolites are expected in rotational crops.

zRMS comments:
Metabolism in primary and rotational crops was found to be similar and a specific residue definition for rotational crops is not deemed necessary. In addition according to EFSA Journal. 2023;21:e8427: the geometric mean DT_{50} in soil of zoxamide is 5.5 days, while the DT_{50} of soil metabolites are less than 60 days (EFSA, 2017). Therefore, in principle further investigation on residues in rotational crops are not required.

7.2.2.3 Nature of residues in processed commodities (KCA 6.5.1)

Available data

One new hydrolysis study has been submitted by the applicant in the framework of this application. This study is summarized in the table below. The detailed results of this study are presented in Appendix 2.

Table 7.2-5: Nature of the residues in processed commodities

Conditions (Duration, Temperature, pH)	Identified compound(s) (%)	Reference
EU data		
Stability of zoxamide under standard hydrolytical conditions was not addressed, studies were not submitted (data gap). Radiolabelled vinification study showed that the major residue in wine is metabolite RH-150721		EFSA,2017
Vinification study	The concentration of parent zoxamide decreased slowly in wine during fermenation and storage.	**RAR (**Mamouni, A. 1998 – Report No. 34-98-151)

Conditions (Duration, Temperature, pH)	Identified compound(s) (%)	Reference
	RH-150721 was the only major degradation product. Ten other minor degradates were detected, including RH-139432 and RH-24549.	
Zoxamide		
Pasteurisation (20 minutes, 90°C, pH 4)	Zoxamide (35.3 %); RH-24549 (9.7 %); RH-150721 (46.7 %)	EFSA Journal 2023;21:e8427
Baking, boiling, brewing (60 minutes, 100°C, pH 5)	Zoxamide (1.2 %); RH-24549 (62.7 %); RH-150721 (11.0 %); RH-129151 (13.1 %)	
Sterilisation (20 minutes, 120°C, pH 6)	Zoxamide (0.8 %); RH-24549 (43.0 %); RH-129151 (20.8 %); RH-141288 (27.3 %)	
RH-141455		
Pasteurisation (20 minutes, 90°C, pH 4)	99.39 %	EFSA Journal 2023;21:e8427
Baking, boiling, brewing (60 minutes, 100°C, pH 5)	98.95 %	
Sterilisation (20 minutes, 120°C, pH 6)	99.75 %	
RH-141452		
Pasteurisation (20 minutes, 90°C, pH 4)	99.22 %	EFSA Journal 2023;21:e8427
Baking, boiling, brewing (60 minutes, 100°C, pH 5)	100.8 %	
Sterilisation (20 minutes, 120°C, pH 6)	99.29 %	
RH-129151		
Pasteurisation (20 minutes, 90°C, pH 4)	RH-129151 (3.4 %); RH-24549 (13.6 %); RH-150721 (70.8 %)	EFSA Journal 2023;21:e8427
Baking, boiling, brewing (60 minutes, 100°C, pH 5)	RH-129151 (26.8 %); RH-24549 (73.2 %)	
Sterilisation (20 minutes, 120°C, pH 6)	RH-129151 (53.9 %); RH-24549 (13.8 %); RH-141288 (26.7 %)	
New data		
Pasteurisation (20 minutes, 90°C, pH 4)	Major metabolite RH-150721 (35.9%) and three minor metabolites RH-139432 (0.6%), RH-24549 (1.5%), RH-129151 (0.6%) were detected	Maric, A., 2023, S21-07903
Baking, boiling, brewing (60 minutes, 100°C, pH 5)	Three major metabolites RH-150721(6.1%), RH-24549(69.9%), RH-129151 (17.5%) and two minor metabolites RH-139432 (1.2%), RH-141288 (4.4%) were detected	
Sterilisation (20 minutes, 120°C, pH 6)	Three major metabolites RH-	

Conditions (Duration, Temperature, pH)	Identified compound(s) (%)	Reference
	141288 (58%), RH-24549 (39.2%), RH-129151 (6%) and one minor metabolite RH-139432 (1.3%) were detected	

Conclusion on nature of residues in processed commodities

EFSA Journal 2023;21:e8427: In the framework of the peer review, a radiolabelled vinification study analysing for zoxamide, RH-150721 (sum of isomers) and RH-139432 was provided as a surrogate of the standard hydrolysis study. Data from this study give an indication that a significant degradation will occur to RH-150721 (sum of isomers). However, it was noted that the nature of residue under the standard hydrolysis conditions at processing was not addressed with this study for the whole range of full processes and a new hydrolysis study performed in accordance with the current recommendations was requested (EFSA, 2017).
New hydrolysis studies with zoxamide, RH-141455, RH-141452 and RH-129151 were reported in the framework of MRL Art. 12 application (Latvia, 2019) and MRL review (Latvia, 2023).
In the studies performed with parent, zoxamide hydrolysed to form RH-150721 (up to a mean maximum of 46.7% TRR upon pasteurisation), RH-141288 (up to a mean maximum of 27.3% TRR upon sterilisation), RH-24549 (up to a mean maximum of 62.7% TRR upon baking/boiling/brewing), RH-129151 (up to a mean maximum of 20.8% TRR upon sterilisation). Thus, zoxamide is not stable under processing conditions leading to the formation of several degradates upon processing.
Studies performed with RH-141455 and RH-141452 showed that these metabolites are stable under all processing conditions. Studies performed with RH-129151, showed that it degrades into RH-24549 up to 13.6% TRR, upon pasteurisation, up to 73.2% upon baking/boiling/brewing and up to 13.8% TRR upon sterilisation; into RH-150721 up to 70.8%TRR upon pasteurisation; and into RH-141288 up to 26.7% TRR upon sterilisation.

Based on the new study simulating hydrolytic conditions for industrial processing, submitted by the applicant in the context of this dossier, to fulfil this requirement, there was no significant change in the total radioactivity following processing under the three different conditions.
The results of the study demonstrated that significant hydrolysis occurred and rection products were formed under conditions representative for pasteurisation, baking/brewing/boiling and sterilisation.
[^{14}C]Zoxamide partly degraded under conditions representative for pasteurisation. One major metabolite and three minor metabolites were formed.
Under the conditions representative for baking/brewing/boiling [^{14}C]zoxamide degraded completely and three major metabolite and two minor metabolites were observed.
Under the conditions representative for sterilization [^{14}C]zoxamide degraded completely and three major metabolites and one minor metabolite were observed.

zRMS comments:
According to the EFSA Journal 2017;15(9):4980: A radiolabelled vinification study was provided as a surrogate analysing for zoxamide, RH-150721(sum of isomers) being the major one in the processed commodity, and RH-139432. Data from this study give an indication that a significant degradation will occur to RH-150721 (sum of isomers) for which toxicological data are needed. However, the nature of residue under the standard hydrolysis conditions at processing is not addressed with this study for the whole range of full processes and a new hydrolysis study performed in accordance with the current recommendations is requested (data gap).
To address the data gap highlighted during the peer review EU approval, the Applicant provided new study in the framework of this application, which demonstrate the nature of residues of zoxamide under conditions representative for pasteurisation, baking/brewing/boiling and sterilisation.

Under the processes representative for the processing of food commodities at higher temperatures the following conclusions can be made:
RH-150721 appears >10%AR only under conditions representative for pasteurisation
RH-129151 appears >10%AR only under conditions representative for baking/brewing/boiling
RH-24549 appears >10%AR under conditions representative for baking/brewing/boiling and sterilisation
RH-141288 appears >10%AR only under conditions representative for sterilisation.
Thus, zoxamide is not stable under processing conditions leading to the formation of several degradates upon processing. Similar conclusion were drawn from the studies assessed during the review of MRLs for zoxamide according to article 12 of Reg. (EC) No 396/2005, EFSA Journal. 2023;21:e8427.

7.2.2.4 Conclusion on the nature of residues in commodities of plant origin (KCA 6.7.1)

Table 7.2-6: Summary of the nature of residues in commodities of plant origin

Endpoints	
Plant groups covered	Root and tuber vegetables (Potatoes) Fruit crops (Grapes)
Rotational crops covered	Yes
Metabolism in rotational crops similar to metabolism in primary crops?	Yes
Processed commodities	a.s. is not stable under standard hydrolysis conditions radiolabelled vinification study showed that the major residue in wine is metabolite RH-150721, EFSA Journal 2017;15(9):4980 zoxamide is not stable under processing conditions leading to the formation of several degradates upon processing
Residue pattern in processed commodities similar to pattern in raw commodities?	~~Yes~~ No, EFSA Journal. 2023;21:e8427
Plant residue definition for monitoring	primary, rotational crops, honey and processed commodities: zoxamide Proposed: Grapes (fruit crops): zoxamide Potato (root crops): metabolites RH-141452 and RH-141455 (EFSA, 2017) Proposed in EFSA Journal. 2023;21:e8427: Zoxamide (sum of constituent isomers) Current: Zoxamide, Reg. (EU) 2017/171
Plant residue definition for risk assessment	primary, rotational crops, honey: sum of zoxamide and RH-141452, expressed as zoxamide processed commodities: RD-1: sum of zoxamide and RH-141452, expressed as zoxamide; RD-2: metabolite RH-150721.
Conversion factor from enforcement to RA	1 (EFSA Journal 2023;21:e8427) for table/wine grapes and potatoes

* If residue pattern in processed commodities is not similar to that in raw commodities

zRMS comments:
According to EFSA Journal. 2023;21:e8427:
"The metabolism of zoxamide in all primary crops was similar in the three crop groups whereas additional metabolites were identified in potatoes. The metabolism in rotational crops is similar to the metabolism observed in primary crops and the processing of zoxamide leads to the generation of several degradates. As the parent compound was found to be a sufficient marker in fruits, roots and pulses and oilseeds, rotational crops and processed commodities, the residue definition for enforcement is proposed as zoxamide only (sum of constituent isomers) (for primary, rotational crops, honey and processed commodities).
In available trials supporting the existing uses and the MRL application, residues of metabolite RH-141452 in fruits occurred at levels above the LOQ in grapes, cucumbers and onions, while metabolite RH-141455 was observed always below the LOQ in all plant commodities. Furthermore, an extensive data set of more than 20 overdosed trials on potatoes reported in the RAR and its addenda (Latvia, 2016, 2017) demonstrate that metabolites RH-141452 and RH-141455 are not expected to occur at levels above the LOQ in potatoes. Metabolite RH-150721 was never found at levels above the LOQ in the raw commodities.
For risk assessment, parent and metabolite RH-141452 are toxicologically relevant and thus should be considered in the consumer exposure. Metabolite RH-141452 encountered in the rat metabolism and was considered covered by the toxicological profile of the parent compound. Therefore, the residue for risk assessment in raw commodities from primary and rotational crops and for honey is proposed as the sum of zoxamide and RH-141452, expressed as zoxamide.
The only metabolite consistently found at levels above the LOQ in processed commodities was RH-150721 (e.g. grape juice, must, wine, raisins, and tomato juice, puree, canned). For metabolite RH-150721 an ADI lower than the parent was set; also, an ARfD was derived for this metabolite while for parent was considered not necessary. Therefore, for processed commodities, two separated residue definitions for risk assessment are proposed: sum of zoxamide andRH-141452, expressed as zoxamide; metabolite RH-150721."

7.2.2.5 Nature of residues in livestock (KCA 6.2.2-6.2.5)

Available data

No new data submitted in the framework of this application.

Table 7.2-7: Summary of animal metabolism studies

Group	Species	Label position	No of animal	Application details		Sample details		Reference
				Rate (mg/kg bw/d)	Duration (days)	Commodity	Time of sampling	
EU data								
Lactating ruminants	Goat	Uniformly isotopically labelled in the phenyl ring	1	2.82	7	Milk	twice daily	EFSA, 2017
						Urine and faeces	twice daily	

Summary of plant metabolism studies reported in the EU

Calculation of the livestock dietary burden considering residues in potato triggered poultry and ruminants metabolism studies. This calculation should be considered as provisional based on the outstanding data on the toxicity of these compounds (RH-141452 and RH-141455) and the nature of residues in processed

commodities. However, a poultry metabolism study was not presented (data gap) but triggered consequent to a provisional dietary intake calculations. Only a goat metabolism study with ^{14}C-labelled zoxamide is available. Zoxamide was not detected in the goat study and the metabolites RH-141452 and RH-141455 were observed as terminal compound of a minor metabolic pathway. It should be noted that from the potato metabolism study it is evident that animals will be mainly exposed to RH-141452 and RH-141455, and therefore the metabolic picture depicted with zoxamide cannot be quantitatively representative for the fate of RH-141452 and RH-1414555 in ruminants. The metabolites RH-141288 (sum of isomers) and RH-127450 (sum of isomers) were found in high concentrations in fat than in muscle leading to the assumption that zoxamide residues can be considered as fat soluble. In order to finally conclude on the fat-solubility of zoxamide residues, log P_{ow} for RH-141288 and RH-127450, and an assessment regarding their potential fat-solubility according to FAO (2009) should be provided (data gap). For ruminants, metabolism is addressed and can be considered consistent with rat. For the time being and considering the outstanding toxicological data on the metabolites RH-141452 and RH-141455 and the open question on the fat solubility of the residues, the derivation of the RD for ruminant matrices is not possible.

Conclusion on metabolism in livestock

Only goat metabolism study was submitted. Poultry metabolism study is required according to provisional dietary burden calculation.

zRMS comments:
According to EFSA Journal. 2023;21:e8427:
“Zoxamide is authorised for use on potatoes that might be fed to livestock. Livestock dietary burden calculations were therefore performed for different groups of livestock according to OECD guidance (OECD, 2013), which has now also been agreed upon at European level. Since the calculated dietary burdens for all groups of livestock were found to be below the trigger value of 0.004 mg/kg bw per day further investigation of residues as well as the setting of MRLs in commodities of animal origin is unnecessary. Although not needed for the current MRL review, the metabolism of zoxamide residues in livestock was investigated in lactating goats (Latvia, 2017) and assessed in the framework of the peer review (EFSA, 2017). In this study, zoxamide radiolabelled in the phenyl ring of the molecule was administered orally to a lactating goat once a day for 7 consecutive days. The test material was dosed at levels equivalent to a dietary concentration of 60.7 mg/kg (equivalent to 2.82 mg/kgbw per day). Zoxamide was not detected in the goat study and the metabolites RH-141452 and RH-141455 were observed as terminal compounds of a minor metabolic pathway (EFSA, 2017). It was also noted in the peer review that from the potato metabolism study it was evident that animals will be mainly exposed to RH-141452 and RH-141455, and therefore, the metabolic picture depicted with zoxamide cannot be quantitatively representative for the fate of RH-141452 and RH-141455in ruminants (EFSA, 2017).”

7.2.2.6 Conclusion on the nature of residues in commodities of animal origin (KCA 6.7.1)

Table 7.2-8: Summary on the nature of residues in commodities of animal origin

	Endpoints
Animals covered	Lactating goats
Time needed to reach a plateau concentration	4 days in milk
Animal residue definition for monitoring	Pending (data gap; EFSA, 2017, EFSA Journal 2023;21:e8427)

	Not triggered, EFSA Journal. 2023;21:e8427
Animal residue definition for risk assessment	Pending (data gap; EFSA, 2017, EFSA Journal 2023;21:e8427) Not triggered, EFSA Journal. 2023;21:e8427
Conversion factor	-
Metabolism in rat and ruminant similar	Yes
Fat soluble residue	Yes, log P_{ow} 3.76 for zoxamide (data gap for RH-141288 and RH-127450) (EFSA,2017)

7.2.3 Magnitude of residues in plants (KCA 6.3)

7.2.3.1 Summary of European data and new data supporting the intended uses

New studies on the magnitude of residue for grapes have been submitted by the applicant in the framework of this application. These studies are summarized in the Table below. The detailed assessment of these studies is presented in Appendix 2.
No new data are submitted in the framework of this application on potato.

Table 7.2-9: Summary of EU reported and new data supporting the intended uses of GLOB2013F and conformity to existing MRL

Commodity	Source	Residue zone (N-EU, S-EU, EU, outside EU)	Evaluation GAP Residue levels (mg/kg) E = according to enforcement residue definition RA = according to risk assessment residue definition	STMR (mg/kg)	HR (mg/kg)	Unrounded OECD calculator MRL (mg/kg)	Current EU MRL (mg/kg) *	MRL compliance
Potato	RAR Wais, A. (1999): KCA 6.3.1/05, KCA 6.3.1/10, Grolleau, G. (1999): KCA 6.3.1/07 Wais, A. (2000): KCA 6.3.1/14	N-EU	GAP on which MRL is based: 10 x 150 g as/ha, PHI 7d RH-7281 (zoxamide) ~~All~~ 4 x <LOQ 0.02mg/kg RH-141452 ~~All~~ 4 x l < LOQ 0.02 mg/kg RH-141455 ~~All~~ 4 x <LOQ 0.02 mg/kg	N/A				
	~~EFSA, 2017, RAR~~	~~S-EU~~	~~GAP on which MRL is based: 10 x 150 g as/ha, PHI 7d~~ ~~RH-7281 (zoxamide) All <LOQ 0.02mg/kg~~ ~~RH-141452 All < LOQ 0.02 mg/kg~~ ~~RH-141455 all <LOQ 0.02 mg/kg~~					

			Data from SEU are not adequate for central zone					
	Overall supporting data for cGAP	N-EU	4 x <LOQ 0.02mg/kg	0.02	0.02	0.02	0.02	Yes
	~~Overall supporting data for cGAP~~	~~S-EU~~	~~4 x <LOQ 0.02mg/kg~~	~~0.02~~	~~0.02~~	~~0.02~~	~~0.02~~	~~Yes~~
Grapes	EFSA, 2017, DAR	N-EU	GAP on which MRL/EU a.s. assessment is based: 6/10 x 125-150 g as/ha, PHI 28d zoxamide: 0.41, 0.45, 0.55, 0.59, 0.60, 0.72, 0.89, 0.93, 0.76, 0.78, 0.83, 1.02, 1.37, 1.51, 1.67, 2.65, 0.09, 0.17, 0.19, 0.19, 0.33, 0.35, 0.45, 0.48, 0.50, 0.56, 0.77, 0.77, 0.88, 1.31, 1.55, 0.05, 0.11, 0.39, 0.42, 0.43, 0.46, 0.47, 0.48, 0.47, 0.50, 0.51, 0.55, 0.67, 0.81 RH-141452: no data**	N/A				
	New trials	N-EU	Trials GAP: 2 x 0.18 kg as/ha, 1 x 0.36 kg as/ha, PHI 28d zoxamide: ~~0.81~~ 1.3, 3 x 1.2 RH-141452: ~~4~~ x < 0.01**	N/A				
	Overall supporting data for cGAP	N-EU	zoxamide: 0.41, 0.45, 0.55, 0.59, 0.60, 0.72, 0.89, 0.93, 0.76, 0.78, 0.83, 1.02, 1.37, 1.51, 1.67, 2.65, 0.09, 0.17, 0.19, 0.19, 0.33, 0.35, 0.45, 0.48, 0.50, 0.56, 0.77, 0.77, 0.88, 1.31, 1.55, 0.05, 0.11, 0.39, 0.42, 0.43, 0.46, 0.47, 0.48, 0.47, 0.50, 0.51, 0.55, 0.67, 0.81, ~~0.81,~~ 3 x 1.2, 1.3 RH-141452: ~~4~~ x < 0.01**	zoxamide: 0.590 RH-141452: < LOQ	zoxamide: 2.65 RH-141452: <LOQ	zoxamide: 2.679	5	Yes
	Overall supporting data for cGAP (selection from zoxamide RAR: 6 trials***, 4 new trials) Total: 10 trials	N-EU	zoxamide: 0.67, 0.51, 0.47, 0.55, 0.5, 0.81, ~~0.81~~, 3 x 1.2, 1.3 RH-141452: 4 x < 0.01**	zoxamide: 0.740 RH-141452: < LOQ	zoxamide: ~~1.2~~ 1.3 RH-141452: <LOQ	zoxamide: ~~2.376~~ 2.523	5	Yes

	~~EFSA, 2017, RAR~~	~~S EU~~	~~GAP on which MRL/EU a.s. assessment is based: 6/10 x 125-150 g as/ha, PHI 28d~~ ~~zoxamide: 0.24, 0.27, 0.28, 0.29, 0.33, 0.34, 0.36, 0.48, 0.53, 0.54, 0.59, 0.65, 0.81, 1.17, 1.56, 1.92, 0.30, 0.32, 0.36, 0.38, 0.46, 0.51, 0.30, 0.32, 0.48, 0.56, 0.64, 0.66, 0.82, 1.21, 1.37, 1.42, 0.21, 0.21, 0.33, 0.42, 0.46, 0.49, 0.54, 0.58, 0.63, 1.07, 2.84, 0.13, 0.21, 0.22, 0.24, 0.25, 0.29, 0.42, 0.43, 0.44, 0.52, 0.52, 0.56, 0.58, 0.61, 0.62, 0.87, 1.11, 1.53, 0.22, 0.46, 0.63, 1.86~~ ~~RH 141452: no data~~** Data from SEU are not adequate for central zone	~~N/A~~				
	~~New trials~~	~~S EU~~	~~Trials GAP: 2 x 0.18 kg as/ha, PHI 28d~~ ~~zoxamide: 0.18, 0.3, 0.02, 0.32~~ ~~RH 141452: 4 x < 0.01~~**	~~N/A~~				
	~~Overall supporting data for eGAP~~***	~~S EU~~	~~zoxamide : 0.24, 0.27, 0.28, 0.29, 0.33, 0.34, 0.36, 0.48, 0.53, 0.54, 0.59, 0.65, 0.81, 1.17, 1.56, 1.92, 0.30, 0.32, 0.36, 0.38, 0.46, 0.51, 0.30, 0.32, 0.48, 0.56, 0.64, 0.66, 0.82, 1.21, 1.37, 1.42, 0.21, 0.21, 0.33, 0.42, 0.46, 0.49, 0.54, 0.58, 0.63, 1.07, 2.84, 0.13, 0.21, 0.22, 0.24, 0.25, 0.29, 0.42, 0.43, 0.44, 0.52, 0.52, 0.56, 0.58, 0.61, 0.62, 0.87, 1.11, 1.53, 0.22, 0.46, 0.63, 1.86, 0.18, 0.3, 0.02, 0.32~~ ~~RH 141452: 4 x < 0.01~~**	~~zoxamide: 0.48~~ ~~RH 141452: < LOQ~~	~~zoxamide: 2.84~~ ~~RH 141452: <LOQ~~	~~zoxamide: 2.84~~	~~5~~	~~Yes~~

* Source of EU MRL: Reg. (EU) 2017/171

** EFSA Journal 2023;21:e8427: Table and wine grapes: The number of residue trials analysing for metabolites RH-141452 and supporting the northern and southern GAPs is not compliant with the data requirements for these crops. Nevertheless, considering that in the available trials, this metabolite was always measured at low levels, further trials are not required.

*** The applicant selects the following trials:
KCA 6.3.2/02, Grolleau, G. (1999)

Report Number	Field code	Formulation	Total No of applications	Second last application (g/ha)	Last application (g/ha)	Interval between two last applications (d)	Zoxamide residue (mg/kg)	PHI (d)	Remarks

R 60.1	FR01	RH-117281 2F	10	149.1	142.2	7	0.67	28	-
R 60.1	FR02	RH-117281 2F	10	145.3	143.9	7	0.51	28	-

KCA 6.3.2/07, Grolleau, G. (1999)

Report Number	Field code	Formulation	Total No of applications	Second last application (g/ha)	Last application (g/ha)	Interval between two last applications (d)	Zoxamide residue (mg/kg)	PHI (d)	Remarks
R 62.3	FR01	RH-117281 2F	10	145	149	7	0.47	28	-
R 62.3	FR02	RH-117281 2F	10	150	151	7	0.55	28	Highest residue occurring at PHI 35
R 62.3	FR03	RH-117281 2F	10	156	148	7	0.5	28	Highest residue occurring at PHI 35
R 62.3	FR04	RH-117281 2F	10	154	153	7	0.81	28	Highest residue occurring at PHI 35

In the selected trials, the dose rate per application is within the 25% rule (acceptable rates to support 166 g/ha: 124.5-207.5 g/ha) and therefore it is appropriate to support the required GAP. The number of applications exceeds the required number of applications in this submission, however this represents a worst-case, and additionally, the level of the residue is similar to the one obtained in the new trials performed by Globachem, demonstrating that the residue level is driven mainly by the last two applications. The previous additional applications are therefore not shown to influence the residue level and the trials are considered fit for purpose. Moreover, the trial dataset will not be used to set an MRL but only to demonstrate that the existing MRL will not be exceeded.

7.2.3.2 Effects on the residue level in pollen and bee products

In Regulation (EU) No. 283/2013 for active substances, the residue level in pollen and bee products for human consumption resulting from residues taken up by honeybees needs to be determined.

Two studies on residues in honey are provided in the context of the current submission.
In the first study (Knoll, M., 2024), formulation GLOB2013F (SC) was foliar applied three times to buckwheat, once at a target rate of 135 g zoxamide/ha at growth stage BBCH 51-63, and twice at a target rate of 176 g zoxamide/ha, once at BBCH 63 and once at BBCH 65. A summary of the residue data determined according to the proposed residue definition is given in the following table.
In the second study (Poráczki, K., 2023), formulation GLOB2013F (SC) was foliar applied twice to spring oilseed rape at a target rate of 180 g zoxamide/ha, once at BBCH 33 (stem elongation) and once at BBCH 59 (inflorence emergence).

Table 7.2-10: Summary of Zoxamide residue data for the proposed use in honey

EU region	sum of zoxamide and RH-141452, expressed as zoxamide (mg/kg)			
	Residues*	HR	STMR	MRL
N-EU, S-EU (Knoll, M., 2024)	3 x < 0.01*, 0.01	0.01	0.01	0.015
N-EU, S-EU (Poráczki, K., 2023)	4 x < 0.01*	0.01	0.01	0.01

* The applicant hereby presents the MRL calculation considering the parent only. In both studies, RH-141452 residues were below LOD of 0.003 mg/kg. Besides, in EFSA Journal 2023;21:e8427 it is stated: Residue trials were analysed for parent only, however, since there is a wide margin of safety and the contribution of honey to the overall exposure is minimal, additional trials analysing for metabolite RH-141452 are not required.

An MRL of 0.015 mg/kg is calculated based on residue data for honey and other apiculture products, which does not exceed the current default EU MRL of 0.05 mg/kg for zoxamide in honey and other apiculture products. Furthermore, in the recent Art. 12 review for zoxamide it is proposed to increase the EU MRL to 0.2 mg/kg for zoxamide in honey and other apiculture products (1040000) (EFSA Journal 2023;21:e8427).

zRMS comments:
Studies provided by the Applicant indicate that the applicable MRL for zoxamide in honey (0.05 mg/kg, Reg (EU) 2017/1711) is not expected to be exceeded following GLOB2013F applications consistent with the proposed GAP.
EFSA Journal 2023;21:e8427: “In the framework of the current MRL review, four semi-field (tunnel) trials were performed with honeybees with a use pattern of 3 × 180 g a.s./ha with a minimum interval of 7(+1) days during full flowering phase of *Phacelia tanacetifolia* (Latvia, 2023). This application pattern (3 applications during full flowering) can be considered a worst-case scenario of exposure from the application of zoxamide to the melliferous crops reported in this assessment (e.g. aubergines, cucurbits). Pollen analysis confirmed that the bees gathered nectar mainly from the treated Phacelia (Latvia, 2023). Based on the available residue data, residues up to 0.0784 mg/kg can be found in honey samples, when zoxamide is applied at a rate of 3 × 180 g a.s./ha with a minimum interval of 7(+1) days, resulting in an MRL proposal of 0.2 mg/kg. Residue trials were analysed for parent only, however, since there is a wide margin of safety and the contribution of honey to the overall exposure is minimal (see Section 4), additional trials analysing for metabolite RH-141452 are not required.”

7.2.3.3 Conclusion on the magnitude of residues in plants

Potato:

According to the available data, the intended use on potato is considered acceptable, for outdoor uses.
The data submitted show that no exceedance of the MRL will occur.
The use potato is considered acceptable.
The residue data for potatoes evaluated in the RAR of zoxamide and the GAP taken into account in the trials in terms of application time, frequency and total rate of applied zoxamide was considered to be more critical to the proposed GAP for GLOB2013F. At the proposed GAP in this dossier residues of zoxamide and metabolites RH-141452 , RH-141455, will be < 0.02 mg/kg and below the current EU MRL for zoxamide of 0.02 mg/kg.

zRMS comments:
Potato
The indicated results come from studies evaluated in RAR (RMS LV, 2017);
zoxamide: 4 x < 0.02 mg/kg
RH-141452: 4 x < 0.02 mg/kg
RH-141455: 4 x < 0.02 mg/kg
The GAP assessed in the RAR is more critical than that proposed for GLOB2013F:
Max 5 appl. in BBCH 20-80, interval between appl. 8 days, max appl. rate per treatment 180 g a.s./ha, PHI 7 days
As assessed by RMS (LV, 2017) in RAR:
"Total 4 residue trials in North zone and 5 residue trials in South zone are valid to support proposed GAP on potatoes. Residues of RH-141452 and RH-141455 were always below LOQ 0.02 mg/kg.
According to Commission Regulation (EU) No 283/2013 the number of studies to be performed may be reduced if residue trials show that residues in plant or plant products are lower than the LOQ. Minimum of four trials per zone shall be presented for major crop."
According to EFSA Journal 2017;15(9):4980:
"The available residue trials on potato were only partly supported by sufficient storage stability data. In order to judge the validity of the remaining trials, a consistent reporting of the application timing according to growth stages of mono- and dicotyledonous plants (BBCH) scale for the potato plant in accordance with the requested GAPs (BBCH 20–80) is necessary (data gap). Furthermore, sufficient field trials in potatoes in northern Europe (NEU) and southern Europe (SEU) supporting the critical GAP for the full RD for RA are required (data gap).
However, in Final Renewal report for the active substance zoxamide, SANTE/10052/2018 Rev 2., the use on potatoes assessed in the RAR was indicated as supported by available data.
According to EFSA Journal. 2023;21:e8427:
"Potatoes: The number of residue trials supporting the southern outdoor/indoor GAP is not compliant with the data requirements for this crop. However, it is noted that an extensive data set of more than 20 overdosed trials performed with 10–7 applications reported in the RAR and its addenda (Latvia, 2016, 2017) demonstrate that residues of parent and metabolites RH-141452 are not expected to occur at levels above the LOQ when zoxamide is applied according to the authorised uses. Therefore, the reduced number of residue trials is considered acceptable in this case because all results were below the LOQ and residues above the LOQ are not expected to occur. Further residue trials are therefore not required."

The data submitted show that no exceedance of the MRL (0.02 mg/kg, Reg. (EU) 2017/171) will occur.

Grapes:
According to the available data, the intended uses on grapes is considered acceptable, for outdoor uses.
The data submitted show that no exceedance of the MRL will occur.
The use grapes is considered acceptable.

zRMS comments:
Grapes
The GAP assessed in the RAR (RMS LV, 2017) is more critical than that proposed for GLOB2013F:

As assessed by RMS (LV, 2017) in RAR:
"The critical GAP for table and wine grapes is 5 applications at 180 g as/ha, minimum application interval of 8 days, PHI 28 days."
However, EFSA (EFSA Journal 2017;15(9):4980) concluded that:
"Field trials for grapes with mixed GAPs for NEU and SEU were presented. The merging of two different GAPs is not acceptable. Moreover, the trials were under dosed and the proportionality concept cannot be applied since the number of applications deviates from the critical GAP. For the SEU GAP on grapes, there are only two GAP-compliant residue trials. It is also noted that provisional RD for RA contains also the metabolite RH-141452 and results for this metabolite in grapes was not available. A sufficient number of residue trials for grapes conforming to the GAP analysing for the full RD were not available (data gap)."
The applicant selects the trials which can support GAP proposed for GLOB2013F. The indicated results come from studies evaluated in RAR (RMS LV, 2017);
zoxamide: 0.67, 0.51, 0.47, 0.55, 0.5, 0.81 mg/kg (n=6)
Residue results from new studies provided by the Applicant:
zoxamide: 3 x 1.2, 1 x 1.3 mg/kg (n=4)
RH-141452: 4 x < 0.003 mg/kg
Residues of RH-141452 were at levels below the LOD when zoxamide was applied according to the proposed GAP. Therefore, the reduced number of residue trials (n=4) is considered acceptable in this case. Further residue trials are therefore not required.

The data submitted show that no exceedance of the MRL (5 mg/kg, Reg. (EU) 2017/171) will occur.

7.2.4 Magnitude of residues in livestock

7.2.4.1 Dietary burden calculation

Zoxamide RAR: *Grape pomace is not fed to livestock. Potatoes may be fed to livestock, however residues of zoxamide and metabolites RH-141452 and RH-141455 in potatoes were <0.02 mg/kg in all trials and therefore will not result in significant residues occurring in the diets of livestock. The proposed residue definition in root crops is sum of RH-141452 and RH-141455, therefore the dietary burden calculations have been performed using the residue levels of both metabolites (sum of RH-141452 and RH-141455)*

Table 7.2-11: Input values for the dietary burden calculation (considering the uses under consideration)

Feed Commodity	Median dietary burden		Maximum dietary burden	
	Input value (mg/kg)	Comment	Input value (mg/kg)	Comment
Residue definition for root crops sum of RH-141452 and RH-141455				
Potato	0.04	STMR (0.04 mg/kg)	0.11	HR 0.11

Table 7.2-12: Results of the dietary burden calculation

Animal species	Median dietary burden (mg/kg bw/d)	Maximum dietary burden (mg/kg bw/d)	Highest contributing commodity	Max dietary burden (mg/kg DM)	Trigger exceeded (Y/N)
Risk assessment residue definition 1 (sum of RH-141452 and RH-141455)					
Beef cattle*	0.004			0.17	Y
Dairy cattle*	0.006			0.17	Y
Ram/ewe	0.006			0.17	Y
Lamb	0.006			0.11	Y
Breeding swine	0.006			0.28	N
Finishing swine*	0.008			0.28	N
Broiler poultry	0.004			0.06	Y
Layer poultry*	0.004			0.06	Y
Turkey	0.008			0.11	Y

* These categories correspond to those (formerly) assessed at EU level.

EFSA, 2017: *Calculation of the livestock dietary burden considering residues in potato triggered poultry and ruminants metabolism studies. This calculation should be considered as provisional based on the outstanding data on the toxicity of these compounds (RH-141452 and RH-141455) and the nature of residues in processed commodities. However, a poultry metabolism study was not presented (data gap) but triggered consequent to a provisional dietary intake calculation. Only a goat metabolism study with 14C–labelled zoxamide is available. Zoxamide was not detected in the goat study and the metabolites RH-141452 and RH-141455 were observed as terminal compounds of a minor metabolic pathway. It should be noted that from the potato metabolism study it is evident that animals will be mainly exposed to RH-141452 and RH-141455, and therefore, the metabolic picture depicted with zoxamide cannot be quantitatively representative for the fate of RH-141452 and RH-141455 in ruminants. The metabolites RH-141288 (sum of isomers) and RH-127450 (sum of isomers) were found in higher concentrations in fat than in muscle leading to the assumption that zoxamide residues can be considered as fat soluble. In order to finally conclude on the fat-solubility of zoxamide residues, log Po/w for RH-141288 and RH-127450, and an assessment regarding their potential fat-solubility according to FAO (2009) should be provided (data gap). For ruminants, metabolism is addressed and can be considered consistent with rat. For the time being and considering the outstanding toxicological data on the metabolites RH-141452 and RH-141455 and the open question on the fat solubility of the residues, the derivation of the RD for ruminant matrices is not possible.*

Open points relevant to active substance data to be addressed by Notifier at next renewal.

zRMS comments:
According to EFSA Journal. 2023;21:e8427:
"Zoxamide is authorised for use on potatoes that might be fed to livestock. Livestock dietary burden calculations were therefore performed for different groups of livestock according to OECD guidance (OECD, 2013), which has now also been agreed upon at European level. The input values for all relevant commodities are summarised in Appendix D. Since the calculated dietary burdens for all groups of livestock were found to be below the trigger value of 0.004 mg/kg bw per day further investigation of residues as well as the setting of MRLs in commodities of animal origin is unnecessary."

7.2.4.2 Livestock feeding studies (KCA 6.4.1-6.4.3)

No new data were submitted in the framework of this application.

7.2.5 Magnitude of residues in processed commodities (Industrial Processing and/or Household Preparation) (KCA 6.5.2-6.5.3)

7.2.5.1 Available data for all crops under consideration

Processing studies are not required for potato as residues of zoxamide (and the metabolites RH-141452, RH-141455) are < 0.02 mg/kg (LOQ).

New processing studies on grapes have been submitted by the applicant in the framework of this application. These studies are summarized in the table below. The detailed results are presented in Appendix 2.

Table 7.2-13: Overview of the available processing studies

Processed commodity	Number of studies	Median PF *	Median CF **	Comments	Reference
EU data					
Table grapes, raisins	4	1.97	-	-	EFSA Journal 2023;21:e8427
Wine grapes, limpid juice (pre-pasteurisation)	2	0.037	-	-	
Wine grapes, limpid juice (post-pasteurisation)	6	0.2	-	-	
Wine grapes, must	4	0.95	-	-	
Wine grapes, young wine	8	0.05	-	-	
Wine grapes, bottled wine	10	0.05	-	-	
New data					
Enforcement residue definition grapes (fruit crops) : Zoxamide					
Grapes, wet pomace	4	1.42	-	-	Kroniewicz, L. (2022), S21-07247
Grapes, pasteurized juice		0.013	-	-	
Grapes, raisins		0.39	-	-	
Metabolite RH-150721					
Grapes, pasteurized juice	4	(<LOQ – 0.02 mg/kg)	-	-	Gustloff, C. & Mohaupt, R. (2022), S22-08244

* The median processing factor is obtained by calculating the median of the individual processing factors of each processing study.

** The median conversion factor for enforcement to risk assessment is obtained by calculating the median of the individual conversion factors of each processing study.

7.2.5.2 Conclusion on processing studies

The effect of industrial processing and/or household preparation was assessed on studies conducted on grapes and tomato (Latvia, 2019). Robust processing factors for zoxamide (fully supported by data) could be derived for all processed commodities: table grapes raisins, wine grapes (pre- and post-pasteurisation) juice, must, wine and tomato juice, puree and canned.

In the context of this dossier, new processing studies were performed and submitted by the applicant. In the first study (S21-07247), samples taken from 4 sites in Northern Europe were taken and processed. Residues of zoxamide and metabolite RH-1452 were below LOD (set at 30% of the LOQ), leading to the conclusion that following the use of the product at the suggested dose rates and timing no quantifiable residues are expected in processed commodities.
In the second study, the objective was to analyse samples of pasteurised juice originating from study S21-07247 for residues of RH-150721. Residues of metabolite RH-150721 was 0.02 mg/kg in samples originating from two sites and < LOQ for samples originating from the other two sites in Northern Europe. Based on these results, it can be concluded that no significant levels of residues of metabolite RH-150721 are expected in juice produced from raw commodities treated with the product at the suggested dose rates and timing.

zRMS comments:
According to the Reg. (EU) No 283/2013:
“If the level of residues is less than 0.1 mg/kg, processing studies shall be carried out if the contribution of the commodity under consideration to the theoretical maximum daily intake (TMDI) is ≥ 10 % of the ADI or if the estimated daily intake is ≥ 10 % of the ARfD for any European consumer group diet.”
Residues of zoxamide and the metabolites RH-141452, RH-141455 in potatoes are below LOQ. The contribution of the commodity under consideration (potato) to the TMDI is < 10 % of the ADI.
New processing studies have been submitted for grapes in the framework of this application. These studies investigate the magnitude of residues of zoxamide, RH-141452 and RH-141452 in grape processed fractions.
According to EFSA Journal. 2023;21:e8427:
“The effect of industrial processing and/or household preparation was assessed on studies conducted on grapes and tomato (Latvia, 2019). An overview of all available processing studies is available in Appendix B.2.2.3. Robust processing factors for zoxamide (fully supported by data) could be derived for all processed commodities: table grapes raisins, winegrapes (pre- and post-pasteurisation) juice, must, wine and tomato juice, puree and canned. Further processing studies are not required as they are not expected to affect the outcome of the risk assessment.”

7.2.6 Magnitude of residues in representative succeeding crops

~~Crops under evaluation are not expected to be grown in rotation.~~ Further investigation of residues in rotational crops is therefore not required.

zRMS comments:
According to EFSA Journal. 2023;21:e8427:
“Low residue levels were found in the rotational crop metabolism study using an exaggerated application rate (2.2 N). Residues in crops grown in rotation are not expected if zoxamide is to the GAPs reported in Appendix A.”
Residues in succeeding crops are not considered to be of concern.

7.2.6.1 Field rotational crop studies (KCA 6.6.2)

Available data

No new data submitted in the framework of this application.
As mentioned in the RAR of zoxamide:
No supervised field trials were conducted to investigate residues in succeeding crops. However , in the confined rotational crop metabolism study, the only crops to contain total radioactive residues greater than 0.1 mg/kg were immature radish (0.127 mg/kg) and soybean hay (0.189 mg/kg). Both crops were planted 30 days after bare soil was treated (4 applications at 18 day intervals) at a rate of 500 g/ha. Therefore residues in succeeding crops are not considered to be of concern.

7.2.7 Other / special studies (KCA6.10, 6.10.1)

The available data for the active substance sufficiently address aspects of the residue situation that might arise from the use of GLOB2013F Therefore, other special studies are not needed.

7.2.8 Estimation of exposure through diet and other means (KCA 6.9)

Toxicological reference values relevant for dietary risk assessment are reported in the summary of the evaluation (see 7.1.2).

7.2.8.1 Input values for the consumer risk assessment

Table 7.2-14: Input values for the consumer risk assessment (residue definition 1)

Commodity	Chronic risk assessment	
	Input value (mg/kg)	Comment
Risk assessment residue definition: sum of zoxamide and RH-141452, expressed as zoxamide		
211000 Potatoes	0.02	Proposed MRL (EFSA, 2023)
220010 Garlic	0.7	Proposed MRL (EFSA, 2023)
220020 Onions	0.7	Proposed MRL (EFSA, 2023)
220030 Shallots	0.7	Proposed MRL (EFSA, 2023)
220040 Spring onions/green onions and Welsh onions	0.02	Proposed MRL (EFSA, 2023)
231010 Tomatoes	2	Proposed MRL (EFSA, 2023)
231030 Aubergines/eggplants	0.5	Proposed MRL (EFSA, 2023)
1040000 Honey and other apiculture products	0.2	Proposed MRL (EFSA, 2023)
All other commodities	EU MRLs	Reg (EU) 2017/171

EFSA Journal 2023;21:e8427: Since metabolite RH-150721 was proposed for separate residue definition for processing commodities, consumer exposure calculations were performed for the potential exposure to

this metabolite in processed commodities. Considering the large margin of safety for the chronic exposure of raw commodities respect to the ADI of 0.5 mg/kg bw per day set for parent, exceedances of the ADI of 0.04 mg/kg bw per day set for metabolite RH-150721 are not expected. Therefore, only acute exposures were calculated for metabolite RH-150721. As a worst-case scenario, it was assumed a full conversion of the parent to the metabolite. The highest acute exposure was calculated for pumpkin (boiled), representing 49% of the ARfD.

Table 7.2-15: Input values for the consumer risk assessment (residue definition 2)

Commodity	Acute risk assessment	
	Input value (mg/kg)	Comment
Risk assessment residue definition 2: RH-150721		
Table grapes/raisins	19.4	EFSA Journal 2023;21:e8427
Wine grapes/wine	4.1	EFSA Journal 2023;21:e8427
Wine grapes/juice	0.79	EFSA Journal 2023;21:e8427
Potatoes/chips	0.01*	EFSA Journal 2023;21:e8427
Potatoes/dried (flakes)	0.04	EFSA Journal 2023;21:e8427
Potatoes/fried	0.02	EFSA Journal 2023;21:e8427
Onions/boiled	0.42	EFSA Journal 2023;21:e8427
Shallots/boiled	0.42	EFSA Journal 2023;21:e8427
Tomatoes/sauce/puree	0.19	EFSA Journal 2023;21:e8427
Tomatoes/juice	0.20	EFSA Journal 2023;21:e8427
Gherkins/pickled	0.10	EFSA Journal 2023;21:e8427
Courgettes/boiled	0.70	EFSA Journal 2023;21:e8427
Pumpkins/boiled	1.2	EFSA Journal 2023;21:e8427

7.2.8.2 Conclusion on consumer risk assessment

Extensive calculation sheets are presented in Appendix 3.

Table 7.2-16: Consumer risk assessment

	Risk assessment residue definition: sum of zoxamide and RH-141452, expressed as zoxamide	Risk assessment residue definition 2: RH-150721
TMDI (% ADI) according to EFSA PRIMo	8% (based on NL toddler)	-
IEDI (% ADI) according to EFSA PRIMo	No IEDI calculations were performed as the TMDI calculations using the MRLs were already acceptable. No refinement of the chronic risk assessment is required.	-
IESTI (% ARfD) according to EFSA PRIMo*	No ARfD value available	Pumpkins (boiled):

		49% of ARfD
NTMDI (% ADI) **	-	-
NEDI (% ADI)**	-	-
NESTI (% ARfD) **	-	-

* include raw and processed commodities if both values are required for PRIMo
** if national model is available

The proposed uses of zoxamide in the formulation GLOB2013F do not represent unacceptable chronic risks for the consumer.

zRMS comments:
The short-term and long-term intake of residues resulting from the use of zoxamide according to the proposed use on potato and grapes are unlikely to present a risk to consumer health.

7.3 Combined exposure and risk assessment

Not relevant. The product contains only one active substance.

7.4 References

EFSA (European Food Safety Authority), Bellisai, G., Bernasconi, G., Carrasco Cabrera, L., Castellan, I., del Aguila, M., Ferreira, L., Santonja, G. G., Greco, L., Jarrah, S., Leuschner, R., Perez, J. M., Miron, I., Nave, S., Pedersen, R., Reich, H., Ruocco, S., Santos, M., Scarlato, A. P., … Verani, A. (2023). Review of the existing maximum residue levels for zoxamide according to Article 12 of Regulation (EC) No 396/2005 and setting of an import tolerance for onions, garlic and shallots. EFSA Journal, 21(12), e8427. https://doi.org/10.2903/j.efsa.2023.8427

EFSA (European Food Safety Authority). (2017). Conclusion on the peer review of the pesticide risk assessment of the active substance zoxamide. EFSA Journal, 15(9), 25. https:// doi. org/ 10. 2903/j. efsa. 2017. 4980

EURLs (European Union Reference Laboratories for Pesticide Residues). (2023). Evaluation report prepared under Article 12 of Regulation (EC) No 396/2005. Analytical methods validated by the EURLs and overall capability of official laboratories to be considered for the review of the existing MRLs for zoxamide. February 2023. www.efsa.europa.eu

Latvia. (2016). Renewal assessment report on the active substance zoxamide prepared by the rapporteur Member State Latvia in the framework of regulation (EC) 1107/2009, July 2016. www.efsa.europa.eu

Latvia. (2017). Final addendum to the draft assessment report on the active substance zoxamide, compiled by EFSA, July 2017. www.efsa.europa.eu

Latvia. (2023). Evaluation report prepared under Article 12.2 of Regulation (EC) No 396/2005. Review of the existing MRLs for zoxamide, 05 April 2023. www.efsa.europa.eu

Appendix 1 Lists of data considered in support of the evaluation

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 6.1	Gustloff, C.	2023	Storage stability of Zoxamide and its metabolites RH-1452 and RH-1455 in Plant Matrices under Deep Frozen Conditions, Eurofins Agroscience Services Chem Gmbh, Report No.: S21-07041, GLP, Unpublished	N	Globachem NV
KCA 6.1	Gustloff, C.	2023	Storage Stability of Zoxamide and its metabolites RH-1452 and RH-150721 in Wine under Deep Frozen Conditions, S21-07043, Eurofins Agroscience Services Chem GmbH, GLP, Unpublished	N	Globachem NV
KCA 6.1	Gustloff, C.	2023	Storage Stability of Propamocarb-HCl, Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 in Honey under Deep Frozen Conditions, Eurofins Agroscience Services Chem Gmbh, Report No.: S23-100693, GLP, Unpublished	N	Globachem NV
KCA 6.2.1	Maric, A.	2023	Metabolism of [^{14}C]Zoxamide in Grape Plants, S22-01899, Eurofins Agroscience Services EcoChem GmbH, GLP, Unpublished	N	Globachem NV
KCA 6.3	Kroniewicz, L.	2022	Determination of residues of zoxamide after two applications of GLOB2013F in grape (outdoor) at 4 sites in Northern Europe 2021, on RAC samples and processed commodities, S21-07247, Eurofins Agroscience Services France SAS, GLP, Unpublished	N	Globachem NV
KCA 6.3	Kroniewicz, L.	2023	Determination of residues of zoxamide after two applications of GLOB2013F in grape (outdoor) at 4 sites in Southern Europe 2022, S22-01848, Eurofins Agroscience Service France SAS, GLP, Unpublished	N	Globachem NV
KCA 6.5.1	Maric, A.	2022	[^{14}C]Zoxamide Hydrolysis under Typical Conditions (pH, Temperature and Time) of Processing, S21-07903, Eurofins Agroscience Services EcoChem GmbH, GLP, Unpublished	N	Globachem NV

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 6.5.3	Gustloff, C.	2022	Determination of residues of RH-150721 after two applications of GLOB2013F in grape (outdoor) at 4 sites in Northern Europe 2021, on pasteurized juice samples, S22-08244, Eurofins Agroscience Services Chem GmbH, GLP, Unpublished	N	Globachem NV
KCA 6.10.1	Poráczki, K.	2023	Magnitude of residues of Zoxamide in Spring Oilseed Rape (Brassica napus L.) honey after two applications of GLOB2013F under semi-field conditions in Central and Southern Europe, Biochem Agrar Gmbh, Report No.: 23 48 BTR 0003, GLP, Unpublished	N	Globachem NV
KCA 6.10.1	Knoll, M.	2024	Determination of Residues of Zoxamide and Propamocarb-HCl in Honey after Five Applications in Total of GLOB2008F and GLOB2013F in Phacelia tanacetifolia at Four Sites in Central and Southern Europe in 2023, Eurofins Mitox Fopse Sarl, Report No.: S23-100687, GLP, Unpublished	N	Globachem NV

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
KCA 6.2.1/01	Reibach, PH, Spencer, WO	1998a	^{14}C-RH-117,281: Nature of the residue in Fruiting grape plants, Rohm and Haas Technical Report No. 34-98-49, October1, 1998 GLP, Unpublished	N	Gowan

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 6.2.1/05	Staffa, C. and Mondel, M.	2014	^{14}C-phenyl UL zoxamide: Plant metabolism in grape. RLP AgroScience GmbH, Breitenweg 71, 67435 Neustadt, Germany Report Number: S12-03952 Interim Report GLP, Not published	N	Gowan
KCA 6.3.1/05	Wais, A.	1999d	Determination of residues of RH-117281 and mancozeb in/on potatoes (RAC tubers) following treatment with RH-7281 2F and Dithane /RH-117,281 75 DG Blend from field trials in Germany; 1997 Report no. 652252, March 18, 1999 GLP, unpublished ER ref. no. R 64.4/R 64.5	N	Gowan
KCA 6.3.1/07	Wais, A.	1999f	Magnitude of the residue of RH-7281 and its metabolites RH-1452 and RH-1455 in Potato Raw Agricultural Commodity. Northern and Southern France, 1997 Report no. EA970131, April 6, 1999 GLP, unpublished ER ref. no. R 64.1	N	Gowan
KCA 6.3.1/08	Wais, A.	1999g	Determination of residues of RH-117281 and mancozeb in /on potatoes (RAC tubers) following treatment with RG-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Italy; 1997 Report no. 652285, March 25, 1999 GLP, unpublished ER ref. no. R65.3/R 65.4	N	Gowan

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 6.3.1/09	Wais, A.	1999h	Determination of residues of RH-117281 and mancozeb in/on potatoes (RAC tubers) following treatment with RH-7281 2F and Dithane/RH-117, 281 75 DG Blenc from field trials in Greece; 1997 Report no. 652307, March 17,1999 GLP, unpublished ER ref. no. R 64.2/64.3	N	Gowan
KCA 6.3.1/10	Wais, A.	1999i	Determination of residues of RH-117,281 and mancozeb in/on potato (RAC tubers) following treatment with Dithane/RH117,281 75 DG Blend (8:1) and Dithane/RH-117,281 75 WP Blend (8:1) from two field trials in Germany; 1998 Report no. 688904, April 13, 1999 GLP, unpublished,ER ref. no. R 68.1/R 68.2	N	Gowan
KCA 6.3.1/14	Wais, A.	2000	Determination of residues of RH-117,281 and its metabolites RH-141,452 and RH141,455 in/on potatoes (RAC tubers) following treatment with RH7281/mancozeb 75WG from a field trial (semi residue decline study) in the Netherlands; 1999 Report no. 734567, January 2000 GLP, unpublished ER ref. no. R 72.5	N	Gowan

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 6.3.1/17	Wais, A.	2000	Determination of residues of RH-117,281 and its metabolites RH-141,452 and RH141,455 in/on potatoes (RAC tubers and processing products) following treatment with RH7281/mancozeb 75WG from a field trial (semi residue decline study) in Italy; 1999 Report no. 734545, March 2000 GLP, unpublished ER ref. no. R 73.2	N	Gowan
KCA 6.3.2/01	Wais, A.	1999	Determination of residues of RH-117281 and mancozeb in/on vine (RAC grapes) following treatment with RH7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Germany; 1996 Report no. 553001/649765, April 16,1999 GLP, unpublished ER ref. no. R 69.4/R 69.5	N	Gowan
KCA 6.3.2/02	Grolleau, G.	1999b	Magnitude of the Residue of RH-7281 and Mancozeb in Grape Raw Agricultural Commodity and of RH-7281 in Wine and Processed Fractions – Northern and Southern France – 1996 Report no. EA960110, March 15,1999 GLP, unpublished ER ref. no. R 60.1	N	Gowan
KCA 6.3.2/03	Wais, A.	1999m	Determination of residues of RH-117281 and mancozeb in/on vine (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Italy, 1996 Report no. 553101/649787, April 16,1999 GLP, unpublished ER ref. no. R 70.1/ R 70.2	N	Gowan

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 6.3.2/04	Wais, A.	1999n	Determination of residues of RH-117281 and mancozeb in/on table grapes (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Italy, 1996 Report no. 553102/649798, April 16,1999 GLP, unpublished ER ref. no. R 71.3/ R 71.4	N	Gowan
KCA 6.3.2/05	Wais, A.	1999o	Determination of residues of RH-117281 and mancozeb in/on table grapes (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Spain, 1996 Report no. 553200/620875, April 16,1999 GLP, unpublished ER ref. no. R 70.5/ R 70.6	N	Gowan
KCA 6.3.2/06	Wais, A.	1999p	Determination of residues of RH-117281 and mancozeb in/on vine (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Germany, 1997 Report no. 652241, April 16,1999 GLP, unpublished ER ref. no. R 71.1/ R 71.2	N	Gowan
KCA 6.3.2/07	Grolleau, G.	1999c	Magnitude of the Residue of RH-7281 and Mancozeb in Grape Raw Agricultural Commodity and of RH-7281 in Wine – Northern and Southern France – 1997 Report no. EA 970130, March 15,1999 GLP, unpublished ER ref. no. R 62.3	N	Gowan

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 6.3.2/08	Wais, A.	1999q	Determination of residues of RH-117281 and mancozeb in/on vine (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Italy, 1997 Report no. 652274, April 14,1999 GLP, unpublished ER ref. no. R 68.5/ R 68.6	N	Gowan
KCA 6.3.2/09	Wais, A.	1999r	Determination of residues of RH-117281 and mancozeb in/on vine (RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Greece, 1997 Report no. 652296, April 14,1999 GLP, unpublished ER ref. no. R 69.2/ R 69.3	N	Gowan
KCA 6.3.2/10	Wais, A.	1999s	Determination of residues of RH-117281 and mancozeb in/on table grapes(RAC grapes) following treatment with RH-7281 2F and Dithane/RH-117,281 75 DG Blend from field trials in Italy, 1997 Report no. 660688, March 19,1999 GLP, unpublished ER ref. no. R 65.1/ R 65.2	N	Gowan
KCA 6.3.2/11	Wais, A.	1999t	Determination of residues of RH-117,281 and mancozeb in/on vine grapes (RAC grapes) following treatment with Dithane/RH-117,281 75 DG Blend (8:1), Dithane/RH-117,281 75 WP Blend (8:1) and RH-7281 2F Experimental fungicide from four field trials in Germany, 1998 Report no. 688893, April 13,1999 GLP, unpublished ER ref. no. R 67.2/ R 67.3	N	Gowan

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 6.3.2/12	Grolleau, G.	1999d	Magnitude of the Residue of RH-7281 and Mancozeb in Grape Raw Agricultural Commodity and of RH-7281 in Wine – Northern and Southern France – 1998 Report no. EA 980117, March 15,1999 GLP, unpublished ER ref. no. R 63.1	N	Gowan
KCA 6.3.2/13	Wais, A.	1999u	Determination of residues of RH-117,281 and mancozeb in/on vine grapes (RAC grapes) following treatment with Dithane/RH-117,281 75 DG Blend (8:1), Dithane/RH-117,281 75 WP Blend (8:1) from two field trials in Italy, 1998 Report no. 688961, April 12,1999 GLP, unpublished ER ref. no. R 66.2/ R 66.3	N	Gowan
KCA 6.3.2/14	Wais, A.	1999v	Determination of residues of RH-117,281 and mancozeb in/on vine grapes (RAC grapes) following treatment with Dithane/RH-117,281 75 DG Blend (8:1), from two field trials in Spain, 1998 Report no. 688915, April 14,1999 GLP, unpublished ER ref. no. R 67.4	N	Gowan
KCA 6.3.2/15	Wais, A.	1999w	Determination of residues of RH-117,281 and mancozeb in/on table grapes (RAC grapes) following treatment with Dithane/RH-117,281 75 DG Blend (8:1), from two field trials in Spain, 1998 Report no. 693674, April 12,1999 GLP, unpublished ER ref. no. R 66.1	N	Gowan

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 6.3.2/16	Wais, A.	1999x	Determination of residues of RH-117,281 and mancozeb in/on table grapes (RAC grapes) following treatment with Dithane/RH-117,281 75 DG Blend (8:1), from two field trials in Portugal, 1998 Report no. 688948, April 14,1999 GLP, unpublished ER ref. no. R 69.1	N	Gowan
KCA 6.3.2/19	Grolleau, G.	2000	Magnitude of the Residue of RH-7281/mancozeb 76.25WG in grapes raw agricultural commodity – Northern France – 1999 Report no. EA 990175, March 2000 GLP, unpublished ER ref. no. R 73.3	N	Gowan
KCA 6.3.2/20	Grolleau, G.	2000	Magnitude of the Residue of RH-7281/mancozeb 76.25WG in grapes raw agricultural commodity – Southern France – 1999 Report no. EA 990176, March 2000 GLP, unpublished ER ref. no. R 73.4	N	Gowan
KCA 6.5.1/01	Mamouni, A.	1998	Mamouni, A, 14C-RH-117281: Vinification Study, RCC Ltd., Rohm and Haas Technical Report No. 34-98-151 December 3, 1998 GLP, unpublished	N	Gowan

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 No Source GLP/non GLP/GEP/non GEP Published/Unpublished	Y/N	Owner

List of data relied on and 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 No Source GLP/non GLP/GEP/non GEP Published/Unpublished	Y/N	Owner

Appendix 2 Detailed evaluation of the additional studies relied upon

A 2.1 Zoxamide

A 2.1.1 Stability of residues

A 2.1.1.1 Stability of residues during storage of samples

A 2.1.1.1.1 Storage stability of residues in plant products

Comments of zRMS:	The study is accepted and adequate to confirm the stability of zoxamide and its metabolites RH-1452 and RH 1455 in homogenates of potato (tuber) and of zoxamide and its metabolite RH-1452 in grape (bunches, juice, raisin) upon storage at ≤ 18 °C for 13 months. The results, in accordance with OECD 506, should be presented as absolute values in mg/kg and not adjusted by recovery, as well as % of nominal spike value. The presented results are not given in mg/kg, but such data are included in the study report.

Reference:	KCA 6.1
Report	Storage stability of Zoxamide and its metabolites RH-1452 and RH-1455 in Plant Matrices under Deep Frozen Conditions, Gustloff, C. & Mohaupt, R., 2023, Eurofins Agroscience Services Chem Gmbh, Report No.: S21-07041
Guideline(s):	Yes (OECD 506, 2007, SANTE/2020/12830, rev.1)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Materials and methods

The fortification level for storage samples was at ten times the limit of quantification (LOQ) of the method (0.1 mg/kg) with all analytes fortified separately on aliquots of homogenised control sample material. Storage samples were kept at ≤-18°C with no exceedance.
The day 0 testing was accompanied by analysis of a control sample while the testing after each storage interval was accompanied by analysis of a control sample and concurrent recovery samples. Storage samples allow assessment of storage stability, while concurrent recoveries demonstrate the performance of the analytical method.

For zoxamide, sample extraction and determination of analyte levels was performed according to the multi-residue QuEChERS. In brief, for zoxamide samples of potato (tuber) and grape (bunches, juice, raisin) are extracted with acetonitrile and if necessary, after addition of water. The ratio was 10 mL of extraction solvent per 10 g of potato tuber and grape (bunches, juice) or 5g of grape (raisin). Quantification was performed by use of LC-MS/MS.
For RH-1452 and RH-1455, sample extraction and determination of analyte levels was performed based on

Podhorniak, L.V. (2014). In brief, for RH-1452 and RH-1455 samples of potato (tuber) and grape (bunches, juice, raisin and wine) were extracted with glycine buffer. Liquid-liquid partition was performed twice with acetonitrile. Clean-up of the extract was performed with an ENVI-Carb SPE cartridge. Quantification was performed by use of LC-MS/MS.
The limit of quantification (LOQ) of the analytical method was 0.01 mg/kg for each analyte and each matrix with a limit of detection (LOD) set at 0.003 mg/kg (30% of the LOQ)

Results and discussions

Accuracy and Precision
For zoxamide the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was 98% for potato (tuber), 103% for grape (bunches), 103% for grape (juice) and 108% for grape (raisin).
For RH-1452 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was 96% for potato (tuber), 94% for grape (bunches), 102% for grape (juice) and 91% for grape (raisin).
For RH-1455 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was 93% for potato (tuber).
Relative standard deviations were ≤20% for all analytes and matrices.
For duplicate analysis of storage samples from the same time point the difference between the highest and lowest recovery did not exceed 20%. For triplicate analysis the relative standard deviation of the recoveries obtained for storage samples from the same time point did not exceed 20%.
These values demonstrate satisfying analytical performance for all analytes and matrices while analysing the storage samples.

Table A 1: Summary of recoveries of zoxamide and metabolites

Storage Period	Concurrent Recoveries	Storage Samples			
	Single Values (%)	Percentage of analyte found relative to the nominal fortification level (%)		Percentage recovered corrected for the (mean) concurrent recovery of the individual date of extraction [a]	Percentage recovered relative to the mean percentage recovered at Day 0[a]
		Single Values (%) [b]	Mean [a] in brackets: rel. std. deviation (%)		
Analyte: Zoxamide Matrix Type: Potato (tuber) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	98, 99, 99	98 (0.5)	-	-
6 months (181 days)	94	94, 91	92	98	94
13 months (393 days)	88	90, 95	93	105	94
Analyte: Zoxamide Test System: Grape (bunches) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	101, 106, 102	103 (2.4)	-	-
6 months (181 days)	94	99, 99	99	105	96
13 months (393 days)	115	115, 110	112	98	109
Analyte: Zoxamide Test System: Grape (juice) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	105, 103, 100	103 (2.5)	-	-

6 months (181 days)	99	99, 101	100	101	97
13 months (393 days)	100	101, 104	102	102	99
Analyte: Zoxamide Test System: Grape (raisin) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	110, 110, 103	108 (3.7)	-	-
6 months (181 days)	111	108, 104	106	96	98
13 months (393 days)	111	99, 97	98	88	91
Analyte: RH-1452 Test System: Potato (tuber) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	93, 95, 99	96 (3.3)	-	-
6 months (180 days)	89	80, 80	80	90	84
13 months (392 days)	91	88, 85	86	95	90
Analyte: RH-1452 Test System: Grape (bunches) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	96, 89, 96	94 (4.6)	-	-
6 months (182 days)	96	89, 91	90	93	96
13 months (392 days)	117	102, 99	101	86	107
Analyte: RH-1452 Test System: Grape (juice) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	102, 99, 106	102 (3.5)	-	-
6 months (183 days)	103	98, 88	93	91	91
13 months (393 days)	108	99, 103	101	93	99
Analyte: RH-1452 Test System: Grape (raisin) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	94, 88, 92	91 (3.2)	-	-
6 months (181 days)	92	88, 86	87	94	95
13 months (393 days)	98	102, 95	98	100	108
Analyte: RH-1455 Test System: Potato (tuber) Nominal Fortification Level: 0.1 mg/kg (10xLOQ)					
0 days	-	96, 93, 89	93 (3.7)	-	-
6 months (180 days)	84	79, 80	80	95	86
13 months (392 days)	96	89, 88	88	92	95

[a] calculated from unrounded values; [b] not corrected for concurrent recoveries

Conclusion

The study is deemed sufficient for assessing the stability of zoxamide and its metabolites RH-1452 and

RH-1455 in homogenates of potato (tuber) and of zoxamide and its metabolite RH-1452 in grape (bunches, juice, raisin) upon storage at ≤-18°C (target) in the dark over a storage period up to 13 months.

Comments of zRMS:	The study is accepted and adequate to confirm the stability of zoxamide and its metabolites RH-1452 and RH 150721 in grape under deep frozen conditions in the dark over a storage period up to 13 months. The results, in accordance with OECD 506, should be presented as absolute values in mg/kg and not adjusted by recovery, as well as % of nominal spike value. The presented results are not given in mg/kg, but such data are included in the study report.

Reference:	KCA 6.1
Report	Storage stability of Zoxamide and its metabolites RH-1452 and RH-150721 in Wine under Deep Frozen Conditions, Gustloff, C. & Mohaupt, R., 2023, Eurofins Agroscience Services Chem Gmbh, Report No.: S21-07043
Guideline(s):	Yes (OECD 506, 2007, SANTE/2020/12830, rev.1)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Materials and methods

The fortification level for storage samples was at ten times the limit of quantification (LOQ) of the method (0.1 mg/kg) with all analytes fortified separately on aliquots of homogenised control sample material. Storage samples were kept at ≤-18°C with no exceedance.
The day 0 testing was accompanied by analysis of a control sample while the testing after each storage interval was accompanied by analysis of a control sample and concurrent recovery samples. Storage samples allow assessment of storage stability, while concurrent recoveries demonstrate the performance of the analytical method.

For zoxamide, sample extraction and determination of analyte levels was performed according to the multi-residue QuEChERS. In brief, for zoxamide samples of grape (wine) are extracted with acetonitrile. The ratio was 10 mL of extraction solvent per 10 g of grape (wine). Quantification was performed by use of LC-MS/MS.
For RH-1452, sample extraction and determination of analyte levels was performed based on Podhorniak, L.V. (2014). In brief, for RH-1452 samples of grape (wine) were extracted with glycine buffer. Liquid-liquid partition was performed twice with acetonitrile. Clean-up of the extract was performed with an ENVI-Carb SPE cartridge. Quantification was performed by use of LC-MS/MS.
For RH-150721, sample extraction and determination of analyte levels was performed according to the analytical method validated in study no. S21-07042. In brief, for RH-150721 samples of grape (wine) were extracted with 1% potassium hydrogen carbonate. Clean-up of the extract was performed by SPE with an OASIS HLB cartridge. Quantification was performed by use of LC-MS/MS.
The limit of quantification (LOQ) of the analytical method was 0.01 mg/kg for each analyte and each matrix with a limit of detection (LOD) set at 0.003 mg/kg (30% of the LOQ)

Results and discussions

Accuracy and Precision
For zoxamide the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was

87% for grape (wine).
For RH-1452 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was 107% for grape (wine).
For RH-150721 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples) was 76% for grape (wine).
Relative standard deviations were ≤20% for all analytes and matrices.

For duplicate analysis of storage samples from the same time point the difference between the highest and lowest recovery did not exceed 20%. For triplicate analysis the relative standard deviation of the recoveries obtained for storage samples from the same time point did not exceed 20%.
These values demonstrate satisfying analytical performance for all analytes and matrices while analysing the storage samples.

Table A 2: Summary of recoveries of zoxamide and metabolites

Storage Period	Concurrent Recoveries	Storage Samples			
	Single Values (%)	Percentage of analyte found relative to the nominal fortification level (%)		Percentage recovered corrected for the (mean) procedural recovery of the individual date of extraction [a]	Percentage recovered relative to the mean percentage recovered at Day 0 [a]
		Single Values [b]	Mean [a] in brackets: rel. std. deviation (%)		
Analyte: Zoxamide					
0 days	-	92, 82, 87	87 (5.5)	-	-
6 months (181 days)	81	83, 83	83	102	95
13 months (393 days)	81	85, 83	84	103	96
Analyte: RH-1452					
0 days	-	108, 107, 106	107 (0.9)	-	-
6 months (183 days)	101	94, 80	87	86	81
13 months (393 days)	109	99, 95	97	89	91
Analyte: RH-150721					
0 days	-	77, 77, 73	76 (3.3)	-	-
6 months (182 days)	75	69, 70	70	92	92
13 months (391 days)	83	77, 75	76	91	100

[a] calculated from unrounded values; [b] not corrected for concurrent recoveries

Conclusion

The study is deemed sufficient for assessing the stability of zoxamide and its metabolites RH-1452 and RH-150721 in grape (wine) upon storage at ≤-18°C for 13 months.

A 2.1.1.1.2 Storage stability of residues in honey

A 2.1.1.1.2.1 Study 1

Comments of zRMS:	The study is accepted and adequate to confirm the stability of zoxamide and its metabolites RH-1452, RH-1455 and RH 150721 in honey under deep frozen conditions in the dark over a storage period up to 6 months. The results, in accordance with OECD 506, should be presented as absolute values in mg/kg and not adjusted by recovery, as well as % of nominal spike value. The presented results are not given in mg/kg, but such data are included in the study report.

Reference:	KCA 6.1
Report	Storage Stability of Propamocarb-HCl, Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 in Honey under Deep Frozen Conditions, Gustloff, C., 2024, Eurofins Agroscience Services Chem GmbH, Report No.: S23-100693
Guideline(s):	OECD 506
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Summary

Study Objective

The objective of the study was to obtain data about the storage stability of propamocarb-HCl, zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 in honey under deep frozen conditions in the dark over a storage period up to 6 months in accordance to OECD Guideline 506.

Study Setup

The fortification level for storage samples was at ten times the limit of quantification (LOQ) of the method (0.1 mg/kg) with all analytes fortified separately on aliquots of homogenised control sample material.
Storage samples were kept at ≤ -18 °C with no exceedance.
The day 0 testing was accompanied by analysis of a control sample while the testing after each storage interval was accompanied by analysis of a control sample and concurrent recovery samples.
Storage samples allow assessment of storage stability, while concurrent recoveries demonstrate the performance of the analytical method.

Analytical Method

For propamocarb-HCl and zoxamide, sample extraction and determination of analyte levels was performed according to the multi-residue QuEChERS [1] as validated for honey in S23-100692 [2].

In brief, samples of honey were extracted with acetonitrile after addition of water. The ratio was 10 mL of extraction solvent per 1.0 g of honey. Quantification was performed by use of LC-MS/MS.
For RH-1452 and RH-1455, sample extraction and determination of analyte levels was performed based on the method of Podhorniak, L. V. (2014) [3] as validated for honey in S23-100692 [2].
In brief, samples of honey were extracted with glycine buffer. Liquid-liquid partition was performed twice with ethyl acetate. Clean-up of the extract was performed with an ENVI-Carb SPE cartridge. Quantification was performed by use of LC-MS/MS.
For RH-150721, sample extraction and determination of analyte levels was performed based on the method of S21-07042 [4] as validated in S23-100692 [2].
In brief, samples of honey were extracted twice with methanol. Both extracts were combined, evaporated and resolved in final solvent. Quantification was performed by use of LC-MS/MS.
The limit of quantification (LOQ) of the analytical method was 0.01 mg/kg for each analyte with a limit of detection (LOD) set at 0.003 mg/kg (30 % of the LOQ).

Accuracy and Precision

For propamocarb-HCl the mean recovery for samples extracted without any storage (i.e. day 0 storage samples and concurrent recoveries) was 89 % for honey.

For zoxamide the mean recovery for samples extracted without any storage (i.e. day 0 storage samples and concurrent recoveries) was 98 % for honey.

For RH-1452 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples and concurrent recoveries) was 94 % for honey.

For RH-1455 the mean recovery for samples extracted without any storage (i.e. day 0 storage samples and concurrent recoveries) was 87 % for honey.

For RH-150721 the mean recovery for samples extracted without any storage (i.e., day 0 storage samples and concurrent recoveries) was 111 % for honey.

Relative standard deviations at each level were ≤ 20 % for all analytes.

For duplicate analysis of storage samples from the same time point the difference between the highest and lowest recovery did not exceed 20 %. For triplicate analysis the relative standard deviation of the recoveries obtained for storage samples from the same time point did not exceed 20 %.

These values demonstrate satisfying analytical performance for all analytes while analysing the storage samples.

Analysis of Storage Samples

The analyte levels detected in the storage samples allow the monitoring of the stability of the analytes upon storage. The values were as follows:

Storage Period	**Procedural Recoveries**		**Storage Samples**			
	Single Values (%)	Mean (%) [a]	Percentage of analyte found relative to the nominal fortification level (%)		Percentage recovered corrected for the (mean) procedural recovery of the individual date of extraction [a]	Percentage recovered relative to the mean percentage recovered at Day 0 [a]
			Single Values [b]	Mean [a] in brackets: rel. std. deviation (%)		
Analyte: Propamocarb-HCl; Test System: Honey; Nominal Fortification Level: 0.1 mg/kg (10xLOQ)						

0 days	-	-	83, 98, 100	93 (9.7)	-	-
6 months (193 days)	89, 74	82	124, 104	114	140	122
Analyte: Zoxamide; Test System: Honey; Nominal Fortification Level: 0.1 mg/kg (10xLOQ)						
0 days	-	-	100, 101, 105	102 (2.6)	-	-
6 months (181 days)	97, 89	93	86, 91	89	95	87
Analyte: RH-1452; Test System: Honey; Nominal Fortification Level: 0.1 mg/kg (10xLOQ)						
0 days	-	-	87, 91, 91	90 (2.6)	-	-
6 months (184 days)	99, 103	101	101, 92	97	96	108
Analyte: RH-1455; Test System: Honey; Nominal Fortification Level: 0.1 mg/kg (10xLOQ)						
0 days	-	-	79, 91, 91	87 (8.0)	-	-
6 months (184 days)	88, 88	88	81, 76	79	89	90
Analyte: RH-150721; Test System: Honey; Nominal Fortification Level: 0.1 mg/kg (10xLOQ)						
0 days	-	-	117, 113, 97	109 (9.7)	-	-
6 months (182 days)	116, 113	115	110, 102	106	93	97

[a] calculated from rounded values; [b] not corrected for concurrent recoveries

For all analytes the average amount of analyte recovered relative to the nominal fortification level at day 0 was ≥ 70 % at any testing interval.

Conclusion

The study is deemed sufficient for assessing the stability of propamocarb-HCl, zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 in honey upon storage at ≤ -18 °C for 6 months.

A 2.1.1.1.3 Storage stability of residues in animal products

No new studies were submitted.

A 2.1.2 Nature of residues in plants, livestock and processed commodities

A 2.1.2.1 Nature of residue in plants

A 2.1.2.1.1 Nature of residue in primary crops

Comments of zRMS:	The study is accepted.

Reference:	KCA 6.2.1
Report	Metabolism of [^{14}C] Zoxamide in Grape Plants, Maric, A., 2023, Eurofins Agroscience Services Chem GmbH, Report No.: S22-01899
Guideline(s):	Yes (OECD 501)

Deviations:	No
GLP:	Yes
Acceptability:	Supplementary

The metabolism of zoxamide in grape plants was investigated as a result of two foliar applications at BBCH 81 (beginning of ripening, berries begin to develop variety specific colour) and BBCH 83 (Berries developing colour). The test item [phenyl-U-^{14}C]Zoxamide was formulated as a suspension concentrate and applied at a nominal application rate of 2 * 180 g a.s./ha. The actual total application rate corresponded to 413 kg a.s./ha.

Grape leaves and fruits were harvested at BBCH 89 (Berries ripe for harvest0 corresponding to 28 days after the last treatment. The total radioactive residues (TRR) in all raw agricultural commodities (RACs) were determined by summing up the radioactivity in conventional extracts and remaining solids.

Materials and methods

Homogenised plant material from grape leaves and fruits was conventionally extracted with acetonitrile/water mixtures. The efficiency of conventional solvent extraction procedures, which is also the total extraction efficiency, was 99.2% of TRR for grape leaves and 99.5% of TRR for grape fruits. The post extraction solids for grape leaves and fruits accounted for 0.8% of TRR and 0.5% of TRR, respectively.

Residues in the extracts were analysed and quantified by HPLC. The parent compound zoxamide was identified by the methodology listed below:

- Comparison of metabolic profiles of grape leaves and fruits among themselves;
- Comparison of the HPLC retention times of reference items with the retention time of each component in the sample extracts;
- HPLC co-chromatography with stock solution of test item [phenyl-U-^{14}C]Zoxamide used as radiolabelled reference item in conventional extracts of grape leaves;
- Normal Phase Thin Layer co-chromatography (NP-TLC) of isolated parent fraction from grape fruits with non-labelled Zoxamide and conventional extracts from grape fruits.

Results and discussion

In conventional extracts of **grape leaves**, 96.6% of TRR (34.181 mg eq/kg) were identified in total. Parent compound Zoxamide was the only major residue detected and accounted for 96.6% of TRR (34.181 mg eq/kg). In addition, nine unknown metabolites have been found, representing 2.6% of TRR (0.923 mg eq/kg) in total, individually accounting for equal or less than 0.9% of TRR (0.312 mg eq/kg).

In conventional extracts of **grape fruits**, 94.6% of TRR (0.988 mg eq/kg) were identified in total. Parent compound Zoxamide was the only major residue detected and accounted for 94.6% of TRR (0.988 mg eq/kg). In addition, four unknown metabolites have been found, representing 4.4% of TRR (0.045 mg eq/kg) in total, individually accounting for equal or less than 2.0% TRR (0.021 mg eq/kg).

The only residue found in grape leaves and fruits was the parent compound Zoxamide. No other major metabolites were detected in the conventional extracts of the crop commodities. Therefore, no metabolic pathway was described.

Table A 3: Total Radioactive Residues (TRRs) in grape leaves and grape fruits

RAC	Application	PHI* [days]*	Growth Stage [BBCH]	Fresh weight** [g]	TRR*** (= mg eq/kg)

Grape leaves	Two spray applications at a rate of 196 g a.s./ha 216 g a.s./ha (in total **Błąd! Nie można odnaleźć źródła odwołania.** g a.s./ha)	35 28	81 83	482.3	35.388
Grape fruits				810.7	1.044

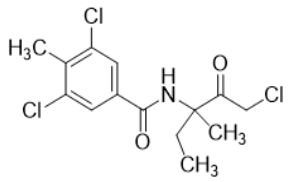
* PHI: pre-harvest interval (corresponds to days after last treatment (DAT) at harvest/sampling).
** Fresh weight after harvest.
** TRR values were determined by summing up the radioactivity measured in the extracts and in the remaining solids.

Table A 4: Distribution of radioactivity in the Extracts.

	grape leaves		grape fruits	
	% TRR	mg eq/kg	% TRR	mg eq/kg
	100.0	35.388	100.0	1.044
Conventional extraction	**99.2**	**35.107**	**99.5**	**1.038**
Analysed extracts	99.2	35.104	99.0	1.033
Losses (not analysed)*	<0.1	0.003	0.5	0.005
Total extracted	**99.2**	**35.107**	**99.5**	**1.038**
Unextractable (PES**)	0.8	0.280	0.5	0.006
Accountability	**100.0**	**35.388**	**100.0**	**1.044**

* not analysed subsamples e.g. distillates, SPE fractions
** post extraction solids

Table A 5: List of Reference Items and Identified Metabolites

Report/IUPAC name and remarks	Detected in RAC/amount	Structure identified by comparison with reference
Zoxamide (RS)-3,5-dichloro-N-(3-chloro-1-ethyl-1-methyl-2-oxopropyl)-p-toluamide	<u>Grape leaves</u> 96.6% of TRR; 34.181 mg eq/kg <u>Grape fruits</u> 94.6 % of TRR; 0.988 mg eq/kg	

Conclusions

On the basis of the nature and amount of the residue in the extracts in the summarised study above, it can be concluded that Zoxamide does not undergo any relevant metabolic reaction in grape plants.

A 2.1.2.1.2 Nature of residue in rotational crops

No new study submitted.

A 2.1.2.1.3 Nature of residues in processed commodities

Comments of zRMS:	The study is accepted. There was no significant change in the total radioactivity following processing under the three different conditions. The results of this study demonstrated that significant hydrolysis occurred and reaction products were formed under conditions representative for pasteurisation, baking/brewing/boiling and sterilisation. [^{14}C]Zoxamide partly degraded under conditions representative for pasteurisation. One major metabolite (RH-150721) and three minor metabolites RH-139432, RH-24549, RH-129151) were formed. Under the conditions representative for baking/brewing/boiling [^{14}C]Zoxamide degraded completely and three major metabolites (RH-150721, RH-24549, RH-129151) and two minor metabolites (RH-139432 RH-141288) were observed. Under the conditions representative for sterilisation [^{14}C]Zoxamide degraded completely and three major metabolites (RH-141288, RH-24549, RH-129151) and one minor metabolite (RH-139432) were observed.

Reference:	KCA 6.5.1
Report	[^{14}C]Zoxamide hydrolysis under Typical Conditions (pH, Temperature and Time) of Processing, Maric, A. (2022), Eurofins Agroscience Services EcoChem GmbH, Report No.: S21-07903
Guideline(s):	Yes (OECD 507)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

The objective of the study was to establish whether or not breakdown or reaction products arise from residues of the test item in raw agricultural commodities when subjected to processing. The parameter which is most likely to affect the nature of the residue during most processing operations is hydrolysis, because processes like heating would generally inactivate enzymes present in the substrate, leaving abiotic hydrolysis as the most important degradation mechanism.

Materials and methods

The following hydrolytic conditions, representative of processing procedures, were used:

Condition 1:	90°C for 20 min (pH 4), representative of pasteurization
Condition 2:	100°C for 60 min (pH 5), representative of baking, brewing, and boiling
Condition 3:	120°C for 20 min (pH 6), representative of sterilization (closed system under pressure)

Study was performed with radiolabeled [^{14}C]Zoxamide. An initial amount of 5.0 MBq/L per vessel, corresponding to 0.97 mg/L (at a specific activity of 5.18 MBq/mg) was applied.

Analysis of the samples was performed using LSC and HPLC. The detected transformation products were confirmed by TLC measurement and HPLC co-chromatography with non-radiolabeled reference items.

Results and discussion

The hydrolytic conditions and the respective results after processing are summarised in Table A6 below.

Table A 6: Hydrolytic conditions and results after processing

Item	Processing Conditions					
	pH 4, 90 °C, 20 min		pH 5, 100 °C, 60 min		pH 6, 120 °C, 20 min	
	%AR (mean)	mg/L (mean)	%AR (mean)	mg/L (mean)	%AR (mean)	mg/L (mean)
Test item	60.6	0.46	<LOD	<LOD	<LOD	<LOD
RH-150721	35.9	0.27	6.1	0.05	<LOD	<LOD
RH-139432	<LOD (0.6)	<LOD (<0.01)	1.2	0.01	1.3	0.01
RH-141288	<LOD	<LOD	4.4	0.03	58.0	0.42
RH-24549	1.5	0.01	69.9	0.52	39.2	0.29
RH-129151	<LOD (0.6)	<LOD (<0.01)	17.5	0.13	6.0	0.04
Total	99.2	0.76	99.1	0.74	104.5	0.76

LOD: limit of detection, <LOD = 0.79 % AR

The mean recoveries were between 99.1 % and 104.5 % AR, therefore there was no significant change in amount of radioactivity before and after processing.

Under the condition representative of pasteurization the test item was not fully stable and partly degraded. One major metabolite (RH-150721) was formed. In addition, three minor metabolites (RH-139432, RH-24549, RH-129151) were detected.

The test item was not stable under the condition representative of baking, brewing and boiling and degraded completely. Apart from three identified major metabolites (RH-150721, RH-24549, RH-129151) two minor metabolites were detected (RH-139432 RH-141288).

Under the condition representative of sterilization the test item was not stable and degraded completely. Apart from three identified major metabolites (RH-141288, RH-24549, RH-129151) one minor metabolite was detected (RH-139432).

Conclusions

Overall, there was no significant change in the total radioactivity following processing under the three different conditions.
The results of this study demonstrated that significant hydrolysis occurred and reaction products were formed under conditions representative for pasteurisation, baking/brewing/boiling and sterilisation.
[^{14}C]Zoxamide partly degraded under conditions representative for pasteurization. One major metabolite and three minor metabolites were formed.
Under the conditions representative for baking/brewing/boiling [^{14}C]Zoxamide degraded completely and three major metabolites and two minor metabolites were observed.
Under the conditions representative for sterilization [^{14}C]Zoxamide degraded completely and three major metabolites and one minor metabolite were observed.

A 2.1.2.2 Nature of residues in livestock

No new study submitted.

A 2.1.3 Magnitude of residues in plants

A 2.1.3.1 Grapes

Table A 7: Comparison of intended and critical EU GAPs

Type of GAP	Number of applications	Application rate per treatment max (g as/ha)	Interval between application	Growth stage at last application	PHI (days)
cGAP EU (RAR, Latvia 2017)	5	180	8d	BBCH 15 – 79	28
EFSA Journal. 2023;21:e8427	4	132	7d	BBCH 14-89	28
Intended cGAP (number*)	2	166	8-10d	BBCH 13 – 83	28

* Use number(s) in accordance with the list of all intended GAPs in Part B, Section 0

A 2.1.3.1.1 Study 1

Comments of zRMS:	The study is accepted. Four residue trials were conducted on grape during 2021 in NEU. Two applications of GLOB2013F were applied at 0.18 kg a.s./ha. For trials in Hungary, one additional plot received one application of GLOB2013F at 0.36 kg a.s/ha. Max. storage interval between sampling and analysis: RAC samples/zoxamide: 126 days RAC samples/RH-1452: 143 days The residue trials on grape are supported by sufficient storage stability data. Residues of zoxamide in RAC samples (bunches): 1.3 mg/kg, 3 x 1.2 mg/kg Residues of RH-1452 in RAC samples (bunches): 4 x < 0.003 mg/kg (not detectable).

Reference:	KCA 6.3
Report	Determination of residues of zoxamide after two applications of GLOB2013F in grape (outdoor) at 4 sites in Northern Europe 2021, on RAC samples and processed fractions, Kroniewicz, L., 2022, Eurofins Agroscience Services France SAS, Report No.: S21-07247
Guideline(s):	Yes (OECD 509, OECD 508, SANTE/2019/12752, SANTE/2020/12830)
Deviations:	Yes (No impact on the study)
GLP:	Yes
Acceptability:	Yes

The objective of the study was to determine residue levels of zoxamide and metabolite RH-1452 in the raw agricultural commodity grape and processed samples (pasteurized juice, wet pomace, raisins). Results of the processes samples are summarized in relevant point below.

Four residue trials were conducted on grapes during 2021, two in Hungary, and two in Germany. Two applications of GLOB2013F (450 g/L zoxamide) were applied at 0.18 kg ai/ha diluted with water immediately prior to application to a spray volume of 200-600 L/ha. For trials in Hungary , one additional plot received one application of GLOB2013F at 0.36 kg ai/ha.

Samples of grape from the untreated plots were taken by hand on the day of last application and 28 days after the final application.

For zoxamide, sample extraction and determination of residues were performed according to the multi-residue method QuEChERS that was previously validated at Eurofins Agroscience Services Chem GmbH in study S21-07039. For RH-1452, sample extraction and determination of residues were performed according to the analytical method that was previously validated at Eurofins Agroscience Services Chem GmbH in study S21-07040.

In brief, for zoxamide samples of grape (bunches, wet pomace, pasteurized juice, raisins) were extracted with acetonitrile and if necessary, after addition of water. Quantification was performed by use of LC-MS/MS detection. For RH-1452 grape (bunches, wet pomace, pasteurized juice, raisins) were extracted with glycine buffer. Liquid-liquid partition was performed twice with acetonitrile. Clean-up of the extract was performed with an ENVI-Carb SPE cartridge. Quantification was performed by use of LC-MS/MS.

The limit of quantification (LOQ) of the analytical method was 0.01 mg/kg for each analyte and each matrix with a limit of detection (LOD) set at 0.003 mg/kg (defined as the lowest calibration standard, which is 30 % of the LOQ).

The analytical method was validated during analysis. For zoxamide, fortifications were performed at the level of 0.01 mg/kg and 0.1 mg/kg for all matrices and additionally at 2.0 mg/kg for grape (bunches), at 3.0 mg/kg for grape (wet pomace) and at 1.0 mg/kg for grape (raisins) and were thus at or above the highest residue level found in a sample. For RH-1452, fortifications were performed at the level of 0.01 mg/kg and 0.1 mg/kg for all matrices and were thus at or above the highest residue level found in a sample.

No residues above 30 % of the LOQ were detected in the control (untreated) test portions used for recovery determinations. The accuracy and precision of the method during sample analysis was considered to be acceptable since the following criteria were fulfilled. For all combinations of analytes and matrices, single recoveries were in the range of 60 % - 130 %.

Table A 8: Summary of zoxamide and RH-1452 Residues in Bunches (RAC samples)

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Sample Type	Residue of Zoxamide (mg/kg)	Residue of RH-1452 (mg/kg)
Trial S21-07247-01 (Hungary)							
S1	28DAA2 (NCH)	U1	S21-07247-01-001A	1	Bunches	<0.003 n.d.	<0.003 n.d.
S1		2	S21-07247-01-002A	2	Bunches	1.3	<0.003 n.d.
Trial S21-07247-02 (Hungary)							
S1		U1	S21-07247-02-001A	3	Bunches	<0.003 n.d.	<0.003 n.d.

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Sample Type	Residue of Zoxamide (mg/kg)	Residue of RH-1452 (mg/kg)
S1	One day of application A2[#]	2	S21-07247-02-002A	4	Bunches	1.7	<0.003 n.d.
S2	7DAA2	2	S21-07247-02-003A	5	Bunches	1.6	<0.003 n.d.
S3	14DAA2	2	S21-07247-02-004A	6	Bunches	1.2	<0.003 n.d.
S4	21DAA2	2	S21-07247-02-005A	7	Bunches	1.3	<0.003 n.d.
S5	28DAA2 (NCH)	U1	S21-07247-02-006A	8	Bunches	<0.003 n.d.	<0.003 n.d.
S5	28DAA2 (NCH)	2	S21-07247-02-007A	9	Bunches	1.2	<0.003 n.d.
Trial S21-07247-03 (Germany)							
S1	28DAA2 (NCH)	U1	S21-07247-03-001A	10	Bunches	<0.003 n.d.	<0.003 n.d.
S1		2	S21-07247-03-002A	11	Bunches	1.2	<0.003 n.d.
Trial S21-07247-04 (Germany)							
S1	One day of application A2[#]	U1	S21-07247-04-001A	12	Bunches	<0.003 n.d.	<0.003 n.d.
S1		2	S21-07247-04-002A	13	Bunches	1.3	<0.003 n.d.
S2	6DAA2	2	S21-07247-04-003A	14	Bunches	1.2	<0.003 n.d.
S3	14DAA2	2	S21-07247-04-004A	15	Bunches	1.0	<0.003 n.d.
S4	21DAA2	2	S21-07247-04-005A	16	Bunches	1.1	<0.003 n.d.
S5	28DAA2 (NCH)	U1	S21-07247-04-006A	17	Bunches	<0.003 n.d.	<0.003 n.d.
S5	28DAA2 (NCH)	2	S21-07247-04-007A	18	Bunches	1.2	<0.003 n.d.

[#]the untreated sample may be collected just before the application; the treated sample must not be collected before the application

DAA = days after application; NCH = normal commercial harvest; 2 = treated; U1= untreated;

n.d. not detected (below LOD set at 30 % of the LOQ)

Table A 9: Summary of the study 1 trials

Trial No./ Location/ EU zone/ Year	Commodity / Variety	Date of 1.Sowing or planting 2.Flowering 3. Harvest	Application rate per treatment			Dates of treatment or no. of treatments and last date	Growth stage at last treatment BBCH	Portion analyzed	Residues (mg/kg)		PHI (days)	Details on trial
			kg a.s./ ha	Water (l/ha)	kg a.s./hl				Zoxamide	RH-1452		
	(a)	(b)				(c)					(d)	(e)
S21-07247 – 01 Hungary (Fejév) NEU 2021	Grape/ Cabernet Franc	1) 2003 2) Not available 3) 27 Oct 2021	0.186 0.180 0.334	620 600 556	0.030 0.030 0.060	21 Sep 2021 29 Sep 2021 29 Sep 2021	83	Bunch	1.3 0.81	n.d n.d	28	No residues found in untreated Processed samples: 1 application with double rate at PHI 28. Bunch results are from same samples taken from processing
S21-07247 – 02 Hungary (Veszprem) NEU 2021	Grape/ Olaszrizling	1) 2011 2) Not available 3) 28 Oct 2021	0.178 0.175	592 583	0.030 0.030	22 Sep 2021 30 Sep 2021	85	Bunch	1.7 1.6 1.2 1.3 1.2	n.d. n.d. n.d. n.d. n.d.	0 7 14 21 28	No residues found in untreated plots. Samples for processing were taken 1 day earlier.
S21-07247 – 03 Germany (Rhineland - Palatinate) NEU 2021	Grape/ Riesling	1) 11 Apr 2007 2) 17 Jun to 28 Jun 2021 3)27 Oct 2021	0.186 0.185	519 516	0.036 0.036	21 Sep 2021 29 Sep 2021	85	Bunch	1.2	n.d.	28	No residues found in untreated plots No untreated samples available for processing
S21-07247 – 04 Germany (Rhineland - Palatinate) NEU 2021	Grape/ Riesling	1) 12 Apr 2006 2) 20 Jun to 30 Jun 2021 3) 26 Oct 2021	0.183 0.181	510 505	0.036 0.036	20 Sep 2021 28 Sep 2021	83	Bunch	1.3 1.2 1.0 1.1 1.2	n.d. n.d. n.d. n.d. n.d.	0 7 14 21 28	No residues found in untreated plots

(*) Limit of quantification = 0.010 mg/kg; limit of detection = 0.003 mg/kg; n.d. = not detectable

A 2.1.3.1.2 Study 2

Comments of zRMS:	The study is accepted. However, the study was performed in SEU, so cannot support authorization for the central zone. Four residue trials were conducted on grape during 2022 in SEU. Two applications of GLOB2013F were applied at 0.18 kg a.s./ha. Max. storage interval between sampling and analysis: RAC samples/zoxamide: 126 days RAC samples/RH-1452: 143 days The residue trials on grape are supported by sufficient storage stability data. Residues of zoxamide in RAC samples (bunches): 0.18 mg/kg, 0.32 mg/kg, 0.02 mg/kg, 0.3 mg/kg Residues of RH-1452 in RAC samples (bunches): 1 x < 0.01 mg/kg, 3 x < 0.003 mg/kg

Reference:	KCA 6.3
Report	Determination of residues of zoxamide after two applications of GLOB2013F in grape (outdoor) at 4 sites in Southern Europe 2022, Kroniewicz, L. (2023), Eurofins Agroscience Service France SAS, Report No.: S22-01848
Guideline(s):	Yes (OECD 509,SANTE/2019/12752, SANTE/2020/12830)
Deviations:	Yes (No impact on the study)
GLP:	Yes
Acceptability:	Supplementary

The objective of the study was to determine residue levels of zoxamide and metabolite RH-1452 in the raw agricultural commodity grapes.

Four residue trials were conducted on grapes during 2022 in Southern Europe (France, Spain, Italy, Bulgaria). Two applications of GLOB2013F (450 g/L zoxamide) were applied at 180 g ai/ha, t 36 and 28 days before harvest, diluted with water immediately prior to application to a spray volume of 200-600 L/ha.

Samples of grapes from the untreated and treated plots were taken by hand 28 days 0r 29 days after the final application.

Crop samples were analyzed for residues of zoxamide using Eurofins Agroscience Services Chem GmbH Hamburg, Germany, S21-07039 (GLC-2110V). In brief, for zoxamide samples of grape (bunches) are extracted with acetonitrile after addition of water. Quantification was performed by use of LC-MS/MS. Crop samples were analysed for residues of For RH-1452 using method described by Podhorniak, L. V. (2014) and validated in Eurofins Agroscience Services Chem GmbH study no. S21-07040 (GLC-2111V). Samples of grape (bunches) were extracted with glycine buffer. Liquid-liquid partition was performed twice with ethyl acetate. Clean-up of the extract was performed with an ENVI-Carb SPE cartridge. Quantification was performed by use of LC-MS/MS.

The limit of quantification (LOQ) for both zoxamide and RH-1452 was 0.01 mg/kg with a limit of detection (LOD) of 0.003 mg/kg.

The analytical performance in terms of accuracy and repeatability was assessed for each analytical set by fortification of control (untreated) test portions of the respective matrix and subsequent determination of the concurrent recoveries upon applying the analytical method. The analytes were fortified and quantified separately. All values at fortification levels of 0.01 mg/kg and 0.1 mg/kg comply with the standard acceptance criteria of the guidance document SANTE/2020/12830, rev. 1).

Untreated and treated samples received a single assay. No residues above the limit of quantitation were found in any of the untreated samples. The residue levels of zoxamide and RH-1452 found in grapes are summarized in the table below.

Table A 10:Summary of zoxamide and RH-1452 Residues in grapes

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Sample Type	Residue of Zoxamide (mg/kg)	Residue of RH-1452 (mg/kg)
Trial S22-01848-01 (France)							
S1	28 (-0, +1) DAA2 (NCH)	U1	S22-01848-01-001A	11	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S1		2	S22-01848-01-002A	12	Grape (bunches)	0.18	<0.003 n.d.
Trial S22-01848-02 (Spain)							
S1	28 (-0, +1) DAA2 (NCH)	U1	S22-01848-02-001A	21	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S1		2	S22-01848-02-002A	22	Grape (bunches)	0.30	< 0.010 (0.008)
Trial S22-01848-03 (Italy)							
S1	On day of application A2	U1	S22-01848-03-001A	31	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S1		2	S22-01848-03-002A	32	Grape (bunches)	0.07	<0.003 n.d.
S2	7 (-1, +1) DAA2	2	S22-01848-03-003A	33	Grape (bunches)	0.03	<0.003 n.d.
S3	14 (-2, +2) DAA2	2	S22-01848-03-004A	34	Grape (bunches)	0.03	<0.003 n.d.
S4	21 (-2, +2) DAA2	2	S22-01848-03-005A	35	Grape (bunches)	0.03	<0.003 n.d.
S5	28 (-0, +1) DAA2 (NCH)	U1	S22-01848-03-006A	36	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S5		2	S22-01848-03-007A	37	Grape (bunches)	0.02	<0.003 n.d.
Trial S22-01848-04 (Bulgaria)							
S1	On day of application A2	U1	S22-01848-04-001A	41	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S1		2	S22-01848-04-002A	42	Grape (bunches)	1.1	<0.003 n.d.
S2	7 (-1, +1) DAA2	2	S22-01848-04-003A	43	Grape (bunches)	0.55	<0.003 n.d.
S3	14 (-2, +2) DAA2	2	S22-01848-04-004A	44	Grape (bunches)	0.38	<0.003 n.d.
S4	21 (-2, +2) DAA2	2	S22-01848-04-005A	45	Grape (bunches)	0.34	<0.003 n.d.

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Sample Type	Residue of Zoxamide (mg/kg)	Residue of RH-1452 (mg/kg)
S5	28 (-0, +1) DAA2 (NCH)	U1	S22-01848-04-006A	46	Grape (bunches)	<0.003 n.d.	<0.003 n.d.
S5		2	S22-01848-04-007A	47	Grape (bunches)	0.32	<0.003 n.d.

Table A 11: Summary of the study 2 trials

Trial No./ Location/ EU zone/ Year	Commodity / Variety	Date of 1.Sowing or planting 2.Flowering 3. Harvest	Application rate per treatment			Dates of treatment or no. of treatments and last date	Growth stage at last treatment BBCH	Portion analyzed	Residues (mg/kg)		PHI (days)	Details on trial
			kg a.s./ ha	Water (l/ha)	kg a.s./hl				Zoxamide	RH-1452		
	(a)	(b)				(c)					(d)	(e)
S22-01848-01 France Camaret-sur-Aigues (Vaucluse) SEU 2022	Grape/ Mourvedre	1) 15 Feb 2007 2) Not applicable 3) 07 Sep 2022	0.190 0.168	581 513	0.033 0.033	1 Aug 2022 9 Aug 2022	85	Bunch	0.18	n.d	29	No residues found in untreated samples
S22-01848 – 02 Spain Villanueva del Ariscal (Andalucia) SEU 2022	Grape/ Garrido Fino	1) 15 Sep 1998 2) Not applicable 3) 23 Aug 2022	0.183 0.188	611 626	0.030 0.030	18 Jul 2022 26 Jul 2022	81-83	Bunch	0.30	<0.010 (0.008)	28	No residues found in untreated samples
S22-01848 – 03 Italy Bagnarola di Budrioa (Emilia Romagna) SEU 2022	Grape/ Pignoletto	1) 2002 2) May 3) 14 Sep 2022	0.182 0.192	606 642	0.030 0.030	08 Aug 2022 16 Aug 2022	83	Bunch	0.07 0.03 0.03 0.03 0.02	n.d n.d n.d n.d n.d	0 8 13 23 29	No residues found in untreated samples
S22-01848 – 04 Bulgaria Lesichovo (Pazardzhik) SEU 2022	Grape/ Mavrud	1) Dec 2010 2) 02-13 Jun 2022 3) 27 Sep 2022	0.189 0.186	524 518	0.036 0.036	22 Sep 2022 30 Sep 2022	83	Bunch	1.1 0.55 0.38 0.34 0.32	n.d. n.d. n.d. n.d. n.d.	0 7 13 21 28	No residues found in untreated plots

(*) Limit of quantification = 0.010 mg/kg; limit of detection = 0.003 mg/kg; n.d. = not detectable

A 2.1.4 Magnitude of residues in livestock

No new studies were submitted.

A 2.1.5 Magnitude of residues in processed commodities (Industrial Processing and/or Household Preparation)

A 2.1.5.1 Distribution of the residue in peel/pulp

Comments of zRMS:	The study is accepted. Residue trials (n=4) were conducted on grapes in Hungary (n=2) and Germany (n=2). Two applications of GLOB2013F were applied at 0.18 kg ai/ha, PHI 28 days. Analytical methods were sufficiently validated in the course of the study. For zoxamide and RH-1452, the maximum storage interval of final sample extracts was 1 day for grape (bunches, raisins) and 0 days for grape (wet pomace, pasteurized grape juice).

Reference:	KCA 6.3
Report	Determination of residues of zoxamide after two applications of GLOB2013F in grape (outdoor) at 4 sites in Northern Europe 2021, on RAC samples and processed fractions, Kroniewicz, L. (2022), Eurofins Agroscience Service France SAS, Report No.: S21-07247
Guideline(s):	Yes (OECD 509, OECD 508, SANTE/2019/12752, SANTE/2020/12830)
Deviations:	Yes (No impact on the study)
GLP:	Yes
Acceptability:	Yes

The objective of the processing phase was to produce and sample processed fractions (wet pomace, pasteurized juice, raisins) from grape raw agricultural commodity.

Field specimens of grapes were transferred to the processing site and received in good conditions at ambient or chilled temperature.
After reception and before processing, the samples were stored in chilled condition (target +7°C).
The specimens sampled during processing were immediately stored deep frozen (target temperature -18°C) in a freezer at the processing test site and were available for a shipment to the analytical laboratory.

Materials and methods

Four residue trails generated specimens of RAC Grapes for the processing phase. Grapes specimens were sampled for each trial, with one untreated and one treated specimen.
The untreated specimen was processed prior to the treated one and kept separated at all times to avoid any contamination.

The following list details the equipment and material used during the study:
- Balances – Mettler Toledo
- Desiccator (for moisture analysis) – Ohaus

- Stopwatch – Fischer
- Thermometer – Testo
- Drying oven – France Etuves
- Hydraulic Press – Voran
- Autoclave - Techna

Results and discussions

The processing phase was conducted in accordance with good industrial procedures at a laboratory scale. Care was taken to avoid any contamination of the specimens and processed fractions. All specimens of processed fractions were stored deep frozen, until the beginning of the analytical phase.

Table A 12: Residue data from grape processing study with zoxamide

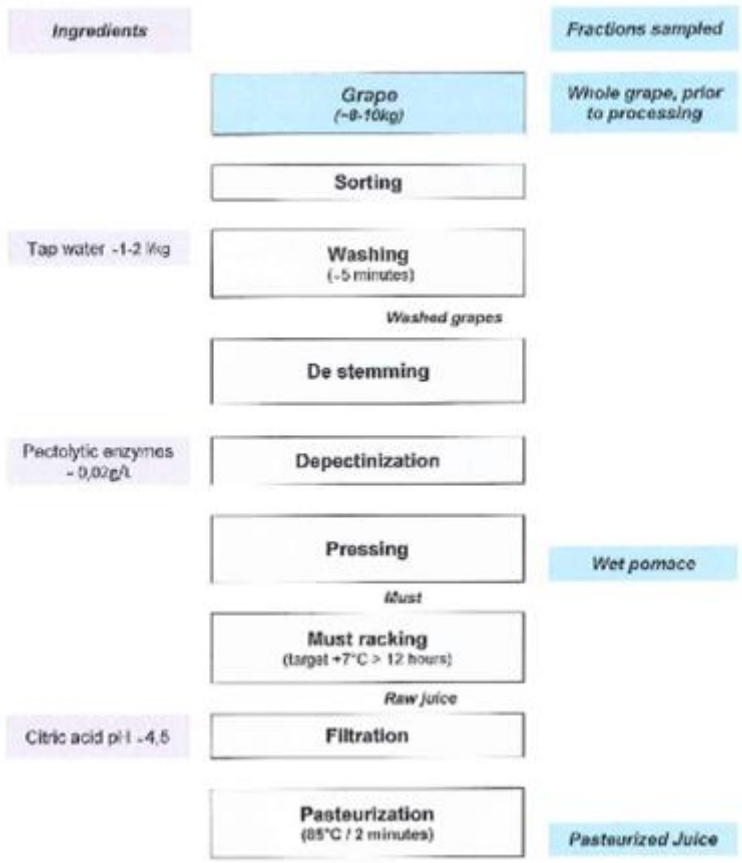
Process	zoxamide residue before processing (mg/kg)	zoxamide residue in processed commodity (mg/kg)	Transfer Factor
S21-07247-01			
Wet pomace	0.81	2.0	2.47
Pasteurized juice	0.81	< 0.010 (0.006)	0.007
Raisins	0.81	0.51	0.63
S21-07247-02			
Wet pomace	1.4	2.3	1.64
Pasteurized juice	1.4	0.021	0.015
Raisins	1.4	0.70	0.5
S21-07247-03			
Wet pomace	0.76	0.74	0.97
Pasteurized juice	0.76	< 0.010 (0.008)	0.011
Raisins	0.76	0.19	0.25
S21-07247-04			
Wet pomace	0.54	0.65	1.20
Pasteurized juice	0.54	< 0.010 (0.009)	0.017
Raisins	0.54	0.15	0.28

n.d = not detected (below LOD set at 30 % of the LOQ)

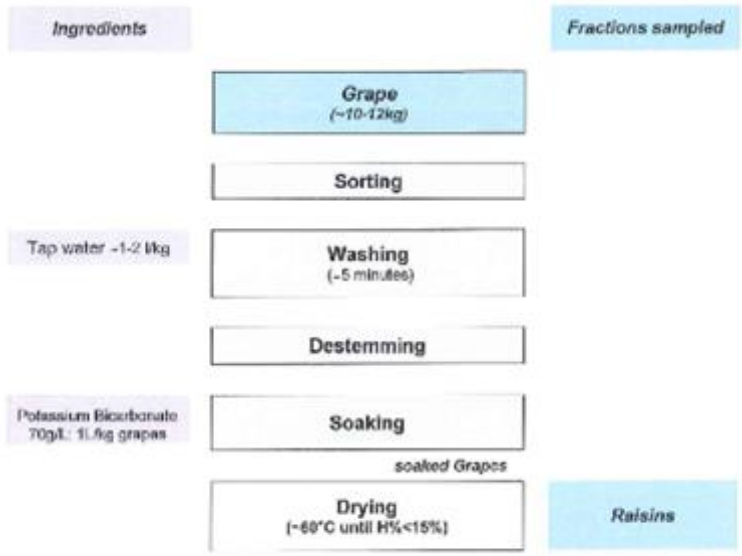
No residues for RH-1452 were found (<0.003 mg/kg) therefore no transfer factors can be calculated.

Figure A 1: Processing flowchart for grapes

Grape juice



Raisins



A 2.1.5.2 Processing studies on a core set of representative processes

Comments of zRMS:	The study is accepted. The samples of pasteurized juice originating from study S21-07247 were anaysed for residues of RH-150721. Analytical methods were sufficiently validated in the course of the study. Residues of RH-150721 (mg/kg): 2 x < 0.01, 2 x 0.02 The proof of stability of the analyte in samples upon storage in deep frozen conditions was not conducted as part of this study and was tested in the study no S21-07043 in grape (wine). Due to their similarity, the storage stability for grape (wine) are considered to be representative for grape (pasteurized juice). Days between Extraction and Analysis – 1.

Reference:	KCA 6.5.3
Report	Determination of residues of RH-150721 after two applications of GLOB2013F in grape (outdoor) at 4 sites in Northern Europe 2021, on pasteurized juice samples, Gustloff, C. & Mohaupt, R., 2022, Eurofins Agroscience Services Chem GmbH, Report No.: S22-08244
Guideline(s):	Yes (SANTE/2020/12830 rev.1, ENV/JM/MONO(2007)17)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

The objective of this study was to analyze samples of pasteurized juice originating from study S21-07247 for residues of RH-150721 in accordance to guidance document SANTE/2020/12830, rev. 1 for risk assessment and/or monitoring.

Materials and methods

Item	Activity, Result, Assessment
Analyte	RH-150721
Matrix	Grape (pasteurized juice)
Method Reference	Analytical method validated in EAS study no. S21-07042 [2]
LOQ	0.01 mg/kg
LOD	30 % of the LOQ (lowest calibration standard)
Principle of the Analytical Procedure	Extraction: 1 % potassium hydrogen carbonate (Ratio: 15 mL of extraction solvent per 5.0 g of matrix) Clean-up: OASIS HLB cartridge Sample concentration in final extract: 0.5 g sample per mL of extract Quantification: LC-MS/MS
Selectivity and Specificity	Demonstrated by one (1) mass transition for evaluation und one (1) mass transition monitored for confirmation Analyte levels or chromatographic interferences of unknown compounds in a reagent blank and control sample extracts were below of what would correspond to an analyte level of 30 % of the LOQ.
Matrix Effects on Analyte Detection	Insignificant (≤ 20 %).
Calibration	Matrix-matched calibration standards A minimum of eight (8) concentration levels Single determination Injection of standard solutions spread over the whole acquisition batch Concentration range: 1.5 ng/mL to 150 ng/mL Corresponding mass fraction range: 0.003 mg/kg to 0.3 mg/kg Coverage: 30 % of the LOQ to at least + 20 % of the highest analyte concentration level detected in a sample extract The validated range does not exceed two (2) orders of magnitude
Quantification	Linear regression with 1/x weighting Regression residuals randomly distributed Coefficients of determination (R^2) ≥ 0.99

<table>
<tr><td>Accuracy
and Precision</td><td>Three (3) fortifications at 0.01 mg/kg (LOQ)
Three (3) fortifications at 0.1 mg/kg (10x LOQ)
Mean recoveries comply with the standard acceptance criteria of the guidance document SANTE/2020/12830, rev. 1:
<table>
<tr><th>Concentration Level (mg/kg)</th><th>Range of Mean Recoveries (%)</th><th>Precision, Rel. Std. Dev. (%)</th></tr>
<tr><td>≤ 0.01</td><td>60 - 120</td><td>≤ 30</td></tr>
<tr><td>> 0.01 - ≤ 0.1</td><td>70 - 120</td><td>≤ 20</td></tr>
</table></td></tr>
<tr><td>Stability of Analyte in Standard Solutions</td><td>The stability of standard solutions upon storage at typically 1 °C to 10 °C for 7 days was demonstrated in S21-07042 [2] and would only need to be investigated if the reported intervals are exceeded.</td></tr>
<tr><td>Stability of Analyte in Sample Extracts</td><td>Recoveries within 70 % - 120 % in for 7 days when stored at typically 1 °C to 10 °C in the dark.</td></tr>
<tr><td>Conclusion</td><td></td></tr>
</table>

Results and discussions

The following residues were detected in the samples.

Table A 13: Residues of RH-150721 detected in the sample

Timing	Treatment	Sample Code Processing	Sample Type	Residue of RH-150721 (mg/kg)
28(-0,+1) DAA2 (NCH)	3	S21-07247-L2-007A	Grape (pasteurized juice)	0.02
28(-0,+1) DAA2 (NCH)	U1	S21-07247-L2-011A	Grape (pasteurized juice)	<0.003 n.d.
28(-0,+1) DAA2 (NCH)	2	S21-07247-L2-015A	Grape (pasteurized juice)	0.02
28(-0,+1) DAA2 (NCH)	2	S21-07247-L2-023A	Grape (pasteurized juice)	< 0.010 (0.008)
28(-0,+1) DAA2 (NCH)	U1	S21-07247-L2-027A	Grape (pasteurized juice)	<0.003 n.d.
28(-0,+1) DAA2 (NCH)	3	S21-07247-L2-031A	Grape (pasteurized juice)	< 0.010 (0.005)

DALA = days after last application; NCH = normal commercial harvest; 2 = treated; U1 = untreated;
n.d. = not detected (below LOD set at 30 % of the LOQ))
Residues are not corrected for concurrent recoveries

Conclusion

The analytical method was applied successfully when analysing the samples of the study according to the guidance document(s) SANTE/2020/12830, rev. 1 for risk assessment and/or monitoring.

A 2.1.6 Magnitude of residues in representative succeeding crops

No new studies were submitted.

A 2.1.7 Other/Special Studies

A 2.1.7.1 Effect on the residue level in pollen and bee products

A 2.1.7.1.1 Study 1

Comments of zRMS:	The study is accepted. Field trials (n=4) were conducted in Spain (n=2) and Germany (n=2). Two applications of GLOB2013F (first in BBCH 33 and the second at BBCH 59) were applied at 0.18 kg ai/ha on spring oilseed rape. Analytical methods were sufficiently validated in the course of the study. This procedure meets the quality criteria of the guideline SANTE/2020/12830 rev. 2. No residues of Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 above the LOD of 0.003 mg/kg were found in any untreated or treated honey sample.

Reference:	KCA 6.10.1
Report	Magnitude of residues of Zoxamide and its metabolites in Spring Oilseed Rape (Brassica napus L.) honey after two applications of GLOB2013F under semi-field conditions in Central and Southern Europe, Poráczki, K., 2023, BioChem agrar GmbH, Report No.: 23 48 BTR 0003
Guideline(s):	
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Summary

The objective of this study was to determine the inadvertent residues of Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 in Spring Oilseed Rape (*Brassica napus* L.) honey after two applications of GLOB2013F under semi-field conditions in Central and Southern Europe in order to allow dietary risk assessment and to establish scientifically-based Maximum Residue Levels (MRLs).

During the growing season in 2023 four separate field trials were conducted at various places in Southern Spain and Eastern Germany. Trials 23BTR0003_T1 and 23BTR0003_T2 (subsequently named T1 and T2) were performed in the area of Eastern Germany (Borsdorf and Grimma; Saxony; Germany), respectively. Trials 23BTR0003_T3 and 23BTR0003_T4 (subsequently named T3 and T4) were performed in the area of Southern Spain (Utrera and Adriano; Seville; Spain).

Two plots (tunnel) with one beehive each, were set up per trial. Each tunnel covered an area of 216 m² (192.5 m² effective crop size). GLOB2013F was applied twice at a rate equivalent to 180 g a.i./ha of Zoxamide (400 mL GLOB2013F/ha). The first application was carried out in BBCH growth stage 33 (Stem

elongation) and the second application at 59 (Inflorescence emergence) of Spring Oilseed Rape with a spray volume of 400 L/ha.

Two assessments of the colony condition were performed per plot, once before beehive setup in the tunnels and once directly after sampling. The beehives were placed in the tunnels when the Spring Oilseed Rape was in full-flowering and remained there until the honey showed a water content < 20%. Precisely 8 to 19 days after beehive setup, honey combs were removed from the colonies and gathered in the laboratory using a honey extractor.

The purpose of the analytical phase of the study was the determination of residues of Zoxamide and its metabolites in honey of *Brassica napus* L. Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 were determined in honey specimens. The determination was conducted by multiple methods at Eurofins Agroscience Services Chem GmbH, Hamburg, Germany using liquid chromatography (LC-MS/MS). The analytical methods were validated according to guideline SANTE 2020/12830 rev. 2.

The residue levels in honey were determined after two applications of GLOB2013F at a rate of 180 g a.i./ha in four independent trials, performed in Southern Spain and Eastern Germany during the 2023 growing season, respectively.

No residues of Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 above the LOD of 0.003 mg/kg were found in any untreated or treated honey sample.

The LOQ for all analytes was set to 0.01 mg/kg. The LOD was defined to be 30 % of the LOQ, corresponding to 0.003 mg/kg for all analytes. Detailed results of the analysed residues are presented in the following tables.

Residues of Zoxamide in honey samples

Field phase	Trial	Treatment group	Sampling day	Sample ID	DALA[1]	Zoxamide [mg/kg]
I	T1	Control	12.07.2023	23BTR0003_01-C-A	15	< LOD
		Test item	12.07.2023	23BTR0003_02-T-A	15	< LOD
	T2	Control	12.07.2023	23BTR0003_03-C-A	15	< LOD
		Test item	12.07.2023	23BTR0003_04-T-A	15	< LOD
II	T3	Control	27.06.2023	23BTR0003_05-C-A	25	< LOD
		Test item	27.06.2023	23BTR0003_06-T-A	25	< LOD
	T4	Control	27.06.2023	23BTR0003_07-C-A	14	< LOD
		Test item	27.06.2023	23BTR0003_08-T-A	14	< LOD

[1]DALA: Days after last application

Residues of RH-1452 and RH-1455 in honey samples

Field phase	Trial	Treatment group	Sampling day	Sample ID	DALA[1]	RH-1452 [mg/kg]	RH-1455 [mg/kg]
I	T1	Control	12.07.2023	23BTR0003_01-C-A	15	< LOD	< LOD
		Test item	12.07.2023	23BTR0003_02-T-A	15	< LOD	< LOD
	T2	Control	12.07.2023	23BTR0003_03-C-A	15	< LOD	< LOD
		Test item	12.07.2023	23BTR0003_04-T-A	15	< LOD	< LOD
II	T3	Control	27.06.2023	23BTR0003_05-C-A	25	< LOD	< LOD
		Test item	27.06.2023	23BTR0003_06-T-A	25	< LOD	< LOD
	T4	Control	27.06.2023	23BTR0003_07-C-A	14	< LOD	< LOD
		Test item	27.06.2023	23BTR0003_08-T-A	14	< LOD	< LOD

[1]DALA: Days after last application

Residues of RH-150721 in honey samples

Field phase	Trial	Treatment group	Sampling day	Sample ID	DALA[1]	RH-150721 [mg/kg]
I	T1	Control	12.07.2023	23BTR0003_01-C-A	15	< LOD
		Test item	12.07.2023	23BTR0003_02-T-A	15	< LOD
	T2	Control	12.07.2023	23BTR0003_03-C-A	15	< LOD
		Test item	12.07.2023	23BTR0003_04-T-A	15	< LOD
II	T3	Control	27.06.2023	23BTR0003_05-C-A	25	< LOD
		Test item	27.06.2023	23BTR0003_06-T-A	25	< LOD
	T4	Control	27.06.2023	23BTR0003_07-C-A	14	< LOD
		Test item	27.06.2023	23BTR0003_08-T-A	14	< LOD

[1]DALA: Days after last application

Overall conclusions

The residue levels in honey in four independent trials, performed in Southern Spain and Eastern Germany during the 2023 growing season were determined after two applications of GLOB2013F at a rate of 180 g a.i./ha under semi-field conditions. The first application was conducted during stem elongation of the crop in BBCH growth stage 33 (visible extended internodes) and the second application during inflorescence emergence at BBCH growth stage 59 (first petals visible, flower buds still closed).

No residues of Zoxamide and its metabolites RH-1452, RH-1455 and RH-150721 above the LOD of 0.003 mg/kg were found in any untreated or treated honey sample.

A 2.1.7.1.2 Study 2

Comments of zRMS:	The study is accepted. Analytical methods were sufficiently validated in the course of the study. This procedure meets the quality criteria of the guideline SANTE/2020/12830 rev. 2. Residues of zoxamide in the treated honey specimen were detected at levels of <LOD, <0.010 (0.008), <0.010 (0.006) and 0.01 mg/kg, respectively. No residues of RH-1452, RH-1455 and RH-150721 were detected in any of the treated samples.

Reference:	KCA 6.10.1
Report	Determination of Residues of Zoxamide and Propamocarb-HCl in Honey after Five Applications in Total of GLOB2008F and GLOB2013F in *Phacelia tanacetifolia* at Four Sites in Central and Southern Europe in 2023, Knoll, M., 2024, Eurofins Mitox Fopse Sarl, Report No.: S23-100687
Guideline(s):	OECD 509
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Summary

The objective of this study was the determination of residues of propamocarb-HCl and zoxamide in honey from *Phacelia tanacetifolia* collected by honeybees after five applications of GLOB2008F and three applications of GLOB2013F under semi-field conditions at four sites in Central and Southern Europe.

Residue trials were conducted in *Phacelia tanacetifolia*, with two field sites in Germany (S23-100687-01 and -02), one site in Spain (S23-100687-03) and one site in Southern France (S23-100687-04). Each trial site consisted of a control plot and a treated plot. In each trial, GLOB2008F (nominal content of 722 g/L propamocarb-HCl) was applied once at a target rate of 2800 mL product/ha (2021.6 g propamocarb-HCl/ha) and four times at a target rate of 1400 ml product/ha (1010.8 g propamocarb-HCl/ha). In each trial, GLOB2013F was applied once at a target rate of 300 ml product/ha (135 g zoxamide/ha) and twice at a target rate of 391 ml product/ha (176 g zoxamide/ha). The product was diluted in water immediately prior to application to a total spray volume of 300 L/ha. The control plot remained untreated. In trial -04 application 3 and 4 were accidentally interchanged. The application amounts of test items of A4 were used on the days A3 should be performed and the application amount of A3 was used on the day the 4th application should have been performed. The 5th and last application in this trial was performed like given in the study plan.

The first application of GLOB2008F was conducted before BBCH 50 in trial -01, -03 and -04 and at BBCH 50-51 in trial -02. The second application of GLOB2008F 5-10DAA1 in trial -01, -02 and -03 and 5-13DAA1 in trial -04. The third application was performed 5-10DAA2 and GLOB2008F and GLOB2013F were used. Application four was conducted in the morning before noon at BBCH 63 with GLOB2008F and GLOB2013F. Application five was conducted 5-7DAA4 4in the morning before noon with GOLB2008F und GLOB2013F. Honey sampling S1 was conducted 3DAA5 in trial -01, 4DAA5 after application in trial -02, 4DAA5 in trial -03 and 3DAA5 after application in trial -04.

Empty combs were inserted to the bee hives before application 4 and honey was collected from these combs. All honey samples from the untreated and treated plots were taken by hand using a spoon or syringe.

Honey was sampled when mature at the time of comb closure or water content < 20 % in the T tunnel. The honey samples were frozen shortly after sampling and were stored in frozen conditions (≤ -18°C) at the test sites. All samples were transported frozen to Eurofins Agroscience Services EcoChem GmbH where they were analysed for residues of propamocarb-HCl and zoxamide.

No residues of zoxamide, propamocarb-HCl, RH-1452, RH-1455 and RH-150721 were detected at or above the limit of detection (0.003 mg/kg) in any of the untreated specimen.

Residues of zoxamide in the treated honey specimen were detected for trials -01 to -04 at levels of <LOD, <0.010 (0.008), <0.010 (0.006) and 0.01 mg/kg, respectively.

Residues of propamocarb-HCl in the treated honey specimen were detected for trials -01 to -04 at levels of 0.02, 1.8, 0.44 and 3.4 mg/kg, respectively.

No residues of RH-1452, RH-1455 and RH-150721 were detected in any of the treated samples.

The following residues of zoxamide and Propamocarb-HCl were detected in the samples:

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Commodity	Residue of Zoxamide (mg/kg)	Residue of Propamocarb-HCl (mg/kg)
Trial S23-100687-01 (Germany)							

S1	At maturity of honey	C	L23-100687-01-C-S1-HO-A	11	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-01-T-S1-HO-A	12	Honey	<0.003 n.d.	0.02
Trial S23-100687-02 (Germany)							
S1	At maturity of honey	C	L23-100687-02-C-S1-HO-A	21	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-02-T-S1-HO-A	22	Honey	< 0.010 (0.008)	1.8
Trial S23-100687-03 (Spain)							
S1	At maturity of honey	C	L23-100687-03-C-S1-HO-A	31	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-03-T-S1-HO-A	32	Honey	< 0.010 (0.006)	0.44
Trial S23-100687-04 (Southern France)							
S1	At maturity of honey	C	L23-100687-04-C-S1-HO-A	41	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-04-T-S1-HO-A	42	Honey	0.01	3.4

T = treated; C = untreated; n.d. = not detected (below LOD set at 30 % of the LOQ)

The following residues of RH-1452 and RH-1455 were detected in the samples:

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Commodity	Residue of RH-1452 (mg/kg)	Residue of RH-1455 (mg/kg)
Trial S23-100687-01 (Germany)							
S1	At maturity of honey	C	L23-100687-01-C-S1-HO-A	11	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-01-T-S1-HO-A	12	Honey	<0.003 n.d.	<0.003 n.d.
Trial S23-100687-02 (Germany)							
S1	At maturity of honey	C	L23-100687-02-C-S1-HO-A	21	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-02-T-S1-HO-A	22	Honey	<0.003 n.d.	<0.003 n.d.
Trial S23-100687-03 (Spain)							
S1	At maturity of honey	C	L23-100687-03-C-S1-HO-A	31	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-03-T-S1-HO-A	32	Honey	<0.003 n.d.	<0.003 n.d.
Trial S23-100687-04 (Southern France)							
S1	At maturity of honey	C	L23-100687-04-C-S1-HO-A	41	Honey	<0.003 n.d.	<0.003 n.d.
		T	L23-100687-04-T-S1-HO-A	42	Honey	<0.003 n.d.	<0.003 n.d.

T = treated; C = untreated; n.d. = not detected (below LOD set at 30 % of the LOQ)

The following residues of RH-150721 were detected in the samples:

Sampling Code	Timing	Treatment	Sample Code	EAS Chem Internal code	Commodity	Residue of RH-150721 (mg/kg)
Trial S23-100687-01 (Germany)						
S1		C	L23-100687-01-C-S1-HO-A	11	Honey	<0.003 n.d.

	At maturity of honey	T	L23-100687-01-T-S1-HO-A	12	Honey	<0.003 n.d.
Trial S23-100687-02 (Germany)						
S1	At maturity of honey	C	L23-100687-02-C-S1-HO-A	21	Honey	<0.003 n.d.
		T	L23-100687-02-T-S1-HO-A	22	Honey	<0.003 n.d.
Trial S23-100687-03 (Spain)						
S1	At maturity of honey	C	L23-100687-03-C-S1-HO-A	31	Honey	<0.003 n.d.
		T	L23-100687-03-T-S1-HO-A	32	Honey	<0.003 n.d.
Trial S23-100687-04 (Southern France)						
S1	At maturity of honey	C	L23-100687-04-C-S1-HO-A	41	Honey	<0.003 n.d.
		T	L23-100687-04-T-S1-HO-A	42	Honey	<0.003 n.d.

T = treated; C = untreated; n.d. = not detected (below LOD set at 30 % of the LOQ)

Table A 11: Summary of the study 2 trials

Trial No./ Location/ EU zone/ Year	Crop /variety	Application rate per treatment (g a.s./ ha)		Growth stage at last treatment (BBCH)	Matrix	Residues (mg/kg)				
		zoxamide	propamocarb-HCl			Zoxamide	RH-1452	RH-1455	RH-150721	Propamocarb -HCl
S23-100687-01 / 76344 Leopoldshafen, Baden-Württemberg, Germany / NEU / 2023	*Phacelia tanacetifolia* /Stala	0 0 138.0 175.2 174.6	2055.2 1027.4 1033.0 1006.3 1002.6	15 32 51 - 52 63 67	Honey	<0.003	<0.003	<0.003	<0.003	0.02
S23-100687-02 / 74080 Böckingen, Baden-Württemberg, Germany / NEU / 2023	*Phacelia tanacetifolia* /Priora	0 0 133.7 175.4 171.0	2097.8 1015.9 1000.7 1007.6 982.0	50 - 51 55 - 57 61 - 62 63 65	Honey	< 0.01 (0.008)	<0.003	<0.003	<0.003	1.8
S23-100687-03 / 03400, Villena, Allicante, Spain / SEU / 2023	*Phacelia tanacetifolia* /Stala	0 0 133.2 180.6 172.3	2075.8 1047.0 997.4 1037.4 989.3	45 - 49 51 - 55 55 - 60 63 65	Honey	< 0.01 (0.006)	<0.003	<0.003	<0.003	0.44
S23-100687-03 / 47170, Sainte Maure de Peyriac, Lot et Garonne, France / SEU / 2023	*Phacelia tanacetifolia* /Natra	0 0 174 134 174.5	2033.7 960.8 1000.0 1000.0 1002.2	18 60 63 63 65	Honey	0.01	<0.003	<0.003	<0.003	3.4

Appendix 3 Pesticide Residue Intake Model (PRIMo)

A 3.1 TMDI calculations

European Food Safety Authority
EFSA PRIMo revision 3.1; 2021/01/06

Zoxamide (F)

LOQs (mg/kg) range from:	0.01	to:	0.05
Toxicological reference values			
ADI (mg/kg bw/day):	0.5	ARfD (mg/kg bw):	not necessary
Source of ADI:	EFSA	Source of ARfD:	EFSA
Year of evaluation:	2023	Year of evaluation:	2023

Input values | Details - chronic risk assessment | Supplementary results - chronic risk assessment | Details - acute risk assessment/children | Details - acute risk assessment/adults

Comments:

Normal mode

Chronic risk assessment: JMPR methodology (IEDI/TMDI)

No of diets exceeding the ADI : ---

	Calculated exposure (% of ADI)	MS Diet	Expsoure (µg/kg bw per day)	Highest contributor to MS diet (in % of ADI)	Commodity / group of commodities	2nd contributor to MS diet (in % of ADI)	Commodity / group of commodities	3rd contributor to MS diet (in % of ADI)	Commodity / group of commodities	Exposure resulting from MRLs set at the LOQ (in % of ADI)	Exposure resulting from commodities not under assessment (in % of ADI)
TMDI/NEDI/IEDI calculation (based on average food consumption)	8%	NL toddler	41.65	4%	Spinaches	2%	Table grapes	1%	Escaroles/broad-leaved endives	0.4%	
	6%	IT adult	28.39	2%	Lettuces	0.9%	Other lettuce and other salad plants	0.6%	Spinaches	0.0%	
	5%	GEMS/Food G06	26.74	1%	Tomatoes	1%	Table grapes	0.6%	Lettuces	0.1%	
	5%	ES adult	26.02	3%	Lettuces	0.5%	Chards/beet leaves	0.4%	Spinaches	0.1%	
	5%	DE child	25.29	1%	Table grapes	1%	Spinaches	0.5%	Lettuces	0.2%	
	5%	GEMS/Food G10	24.50	2%	Lettuces	0.5%	Tomatoes	0.4%	Wine grapes	0.1%	
	5%	GEMS/Food G07	23.50	1%	Wine grapes	1%	Lettuces	0.4%	Tomatoes	0.1%	
	5%	IT toddler	22.74	2%	Lettuces	0.7%	Other lettuce and other salad plants	0.6%	Tomatoes	0.1%	
	4%	GEMS/Food G08	22.22	1%	Lettuces	1%	Wine grapes	0.5%	Tomatoes	0.1%	
	4%	FR adult	21.53	2%	Wine grapes	0.9%	Other lettuce and other salad plants	0.3%	Spinaches	0.1%	
	4%	ES child	21.33	3%	Lettuces	0.5%	Spinaches	0.5%	Chards/beet leaves	0.1%	
	4%	NL child	21.14	2%	Spinaches	1%	Table grapes	0.6%	Escaroles/broad-leaved endives	0.2%	
	4%	GEMS/Food G11	20.13	1%	Wine grapes	0.6%	Spinaches	0.5%	Lamb's lettuce/corn salads	0.1%	
	4%	PT general	19.76	2%	Wine grapes	0.6%	Lettuces	0.4%	Tomatoes	0.0%	
	4%	SE general	19.17	2%	Lettuces	0.4%	Spinaches	0.3%	Tomatoes	0.1%	
	4%	IE adult	19.05	1%	Wine grapes	0.8%	Spinaches	0.5%	Lettuces	0.1%	
	3%	GEMS/Food G15	16.92	1%	Wine grapes	0.7%	Lettuces	0.5%	Tomatoes	0.1%	
	3%	NL general	16.74	0.9%	Spinaches	0.6%	Escaroles/broad-leaved endives	0.6%	Wine grapes	0.1%	
	3%	RO general	15.90	2%	Wine grapes	0.8%	Tomatoes	0.2%	Watermelons	0.1%	
	3%	FR child 3 15 yr	15.09	0.7%	Other lettuce and other salad plants	0.6%	Spinaches	0.4%	Wine grapes	0.1%	
	3%	DE women 14-50 yr	14.76	0.8%	Wine grapes	0.7%	Lettuces	0.3%	Table grapes	0.1%	
	3%	DE general	13.37	0.8%	Wine grapes	0.6%	Lettuces	0.3%	Spinaches	0.1%	
	2%	UK vegetarian	11.98	0.8%	Lettuces	0.8%	Wine grapes	0.2%	Tomatoes	0.0%	
	2%	FR infant	11.47	2%	Spinaches	0.2%	Chards/beet leaves	0.2%	Courgettes	0.1%	
	2%	UK adult	11.19	1%	Wine grapes	0.7%	Lettuces	0.2%	Tomatoes	0.0%	
	2%	DK child	10.96	0.8%	Lettuces	0.7%	Cucumbers	0.2%	Tomatoes	0.1%	
	2%	DK adult	10.95	1.0%	Wine grapes	0.5%	Lettuces	0.2%	Tomatoes	0.0%	
	2%	FR toddler 2 3 yr	10.42	0.9%	Spinaches	0.2%	Wine grapes	0.2%	Tomatoes	0.1%	
	2%	FI adult	9.02	0.8%	Lettuces	0.3%	Wine grapes	0.2%	Tomatoes	0.1%	
	2%	FI 3 yr	8.50	0.4%	Cucumbers	0.4%	Spinaches	0.2%	Table grapes	0.0%	
	2%	FI 6 yr	8.37	0.5%	Lettuces	0.3%	Spinaches	0.3%	Cucumbers	0.0%	
	1%	UK toddler	5.16	0.2%	Table grapes	0.2%	Tomatoes	0.2%	Spinaches	0.1%	
	1.0%	PL general	4.99	0.4%	Tomatoes	0.3%	Table grapes	0.1%	Lettuces	0.0%	
	0.9%	LT adult	4.33	0.4%	Lettuces	0.2%	Tomatoes	0.2%	Cucumbers	0.0%	
	0.5%	UK infant	2.28	0.1%	Tomatoes	0.1%	Milk: Cattle	0.1%	Spinaches	0.1%	
	0.2%	IE child	0.99	0.1%	Table grapes	0.0%	Lettuces	0.0%	Spinaches	0.0%	

Conclusion:
The estimated long-term dietary intake (TMDI/NEDI/IEDI) was below the ADI.
The long-term intake of residues of Zoxamide (F) is unlikely to present a public health concern.
DISCLAIMER: Dietary data from the UK were included in PRIMO when the UK was a member of the European Union.

A 3.2 IEDI calculations

Not required as TMDI does not exceed ADI.

A 3.3 IESTI calculations - Raw commodities

No ARfD value available.

A 3.4 IESTI calculations - Processed commodities

RH-150721

Acute risk assessment /children	Acute risk assessment / adults / general population
Details - acute risk assessment /children	Details - acute risk assessment/adults

The acute risk assessment is based on the ARfD. DISCLAIMER: Dietary data from the UK were included in PRIMO when the UK was a member of the European Union.

The calculation is based on the large portion of the most critical consumer group.

Processed commodities

Results for children
No of processed commodities for which ARfD/ADI is exceeded (IESTI): ---

IESTI

Highest % of ARfD/ADI	Processed commodities	MRL / input for RA (mg/kg)	Exposure (µg/kg bw)
49%	Pumpkins / boiled	2 / 1.21	107
16%	Wine grapes / juice	5 / 0.79	34
11%	Courgettes / boiled	2 / 0.69	25
3%	Shallots / boiled	0.7 / 0.42	6.8
2%	Tomatoes / juice	2 / 0.2	3.7
1%	Gherkins / pickled	2 / 0.1	2.4
0.8%	Potatoes / fried	0.02 / 0.02	1.8
0.8%	Tomatoes / sauce/puree	2 / 0.19	1.8
0.3%	Potatoes / dried (flakes)	0.02 / 0.04	0.56

Expand/collapse list

Results for adults
No of processed commodities for which ARfD/ADI is exceeded (IESTI): ---

IESTI

Highest % of ARfD/ADI	Processed commodities	MRL / input for RA (mg/kg)	Exposure (µg/kg bw)
30%	Pumpkins / boiled	2 / 1.21	67
18%	Wine grapes / wine	5 / 4.12	39
11%	Table grapes / raisins	5 / 19.38	24
7%	Wine grapes / juice	5 / 0.79	16
7%	Courgettes / boiled	2 / 0.69	16
2%	Onions / boiled	0.7 / 0.42	3.9
1%	Shallots / boiled	0.7 / 0.42	2.6
0.7%	Tomatoes / sauce/puree	2 / 0.19	1.5
0.04%	Potatoes / chips	0.02 / 0.01	0.08
0.02%	Potatoes / dried (flakes)	0.02 / 0.04	0.05

Conclusion:
The estimated short-term intake (IESTI) exceeded the toxicological reference value for 1 commodities.

For processed commodities, no exceedance of the ARfD/ADI was identified.