FINAL REGISTRATION REPORT

Part B

Section 9

Ecotoxicology

Detailed summary of the risk assessment

Product code: SHA 4300 A

Product name(s): MIGHTY

Chemical active substance:

Mesotrione, 100 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

Applicant: Sharda Cropchem España S.L.

Submission date: December 2018

MS Finalisation date: 04/06/2024

Version history

When	What
December 2018	Submission to the Polish Ministry of Agriculture and Rural Development
February 2020	Submission to the evaluation
June 2020	zRMS finalised evaluation
August 2022	Applicant update
September 2023	zRMS finalised evaluation of additional data submitted by the Applicant
November 2023	Applicant update
April 2024	zRMS updated comments on effects on mammals following change in GAP
June 2024	Final version prepared by zRMS after the second commenting period

Table of Contents

9 Ecotoxicology (KCP 10)

Review Comments:

This application was submitted by Sharda Cropchem España S.L. for approval of MIGHTY (MESOTRIONE 10% SC), a Suspension Concentrate formulation containing 100 g/L of Mesotrione, for use on maize as an herbicide.

Since this document is based on the information provided by the applicant, all review comments, additions and corrections have been made using commenting boxes or highlighted in grey. Any incorrect data or text not evaluated by the zRMS has been crossed out.

9.1 Critical GAP and overall conclusions

Table 9.1-1: Table of critical GAPs

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Use-No. *	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, Fn, Fpn G, Gn, Gpn or I **	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	**Application**				**Application rate**			PHI (days)	Remarks: e.g. g safener/ synergist per ha	**Conclusion**						
					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			Birds	Mammals	Aquatic organisms	Bees	Non-target ...	Soil organisms	Non-target plants
Zonal uses (field or outdoor uses, certain types of protected crops)																				
1	CEU	**Maize**	F	Broadleaved- and grass weeds	Foliar Spray	BBCH 10-14	a) 1 b) 1	n.r.	a) 1.5 b) 1.5	a) 0.150 b) 0.150	200-600	NA	(*) Weeds at early stages	A	N#	C (R3, R4)	A	C	A	R
1	PL	**Maize**	F	Broadleaved and grass weeds	Foliar Spray	BBCH 10-14 (*)	a) 1 b) 1	N.A	a) 1.0* b) 1.0*	a) 100 b) 100	200-600	NA	(*) Weeds at early stages	A	R	C (R3, R4)	A	C	A	R

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

** 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

~~# The safe use is concluded only for application rate of 1.0 L product/ha (100 g a.s./ha). However, in the proposed use pattern of MIGHTY such a dose rate is not included.~~
~~As the risk to mammals via food and drinking water is still unacceptable following application of MIGHTY at 1.5 L/ha (corresponding to 150 g a.s./ha), the only dose included in the GAP, currently no safe use can be concluded for the product.~~

Explanation for column 15 – 21 “Conclusion”

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

Remarks table:

(1) Numeration necessary to allow references
(2) Use official codes/nomenclatures of EU
(3) For crops, the EU and Codex classifications (both) should be used; where relevant, the use situation should be described (*e.g.* fumigation of a structure)
(4) 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
(5) Scientific names and EPPO-Codes of target pests/diseases/ weeds or when relevant the common names of the pest groups (e.g. biting and sucking insects, soil born insects, foliar fungi, weeds) and the developmental stages of the pests and pest groups at the moment of application must be named
(6) Method, e.g. high volume spraying, low volume spraying, spreading, dusting, drench
Kind, e.g. overall, broadcast, aerial spraying, row, individual plant, between the plants - type of equipment used must be indicated
(7) Growth stage at first and last treatment (BBCH Monograph, Growth Stages of Plants, 1997, Blackwell, ISBN 3-8263-3152-4), including where relevant, information on season at time of application
(8) The maximum number of application possible under practical conditions of use must be provided
(9) Minimum interval (in days) between applications of the same product.
(10) For specific uses other specifications might be possible, e.g.: g/m³ in case of fumigation of empty rooms. See also EPPO-Guideline PP 1/239 Dose expression for plant protection products
(11) The dimension (g, kg) must be clearly specified. (Maximum) dose of a.s. per treatment (usually g, kg or L product / ha).
(12) If water volume range depends on application equipments (e.g. ULVA or LVA) it should be mentioned under “application: method/kind”.
(13) PHI - minimum pre-harvest interval
(14) Remarks may include: Extent of use/economic importance/restrictions

9.1.1 Overall conclusions

9.1.1.1 Effects on birds (KCP 10.1.1), Effects on terrestrial vertebrates other than birds (KCP 10.1.2), Effects on other terrestrial vertebrate wildlife (reptiles and amphibians) (KCP 10.1.3)

For birds a low acute risk via acute exposure was concluded at the screening step for the representative uses on maize. A low long-term risk was concluded at tier 1 for all the generic focal species via dietary exposure for the representative use. The risk for consumption of contaminated water was assessed as low.
For wild mammals a low acute risk via dietary exposure was concluded at the screening level for the representative use on maize. ~~Long-term risk to mammals was assessed as high for all scenarios at the tier 1, after refinement of the risk, the long-term risk for mammals are acceptable. No unacceptable risk for mammals was observed from drinking water after the toxicological endpoint refinement.~~
No safe use was concluded following application of MIGHTY at 1.5 L/ha (corresponding to 150 g a.s./ha) and further refinement is required. The high long-term and drinking water risk was identified.
No secondary poisoning assessment was triggered for Mesotrione and its metabolites (not required since LogKow<3).

The Applicant was requested to submit the additional data necessary to refined the risk to mammals and/or to consider a reduction of the maximum application rate.

Currently, the Applicant has access to following higher tier studies:
1. Von Blanckenhagen F., Grimm T., 2013: Generic field study on small mammals focal species and wood mouse (*Apodemus sylvaticus*) PT in maize fields in Germany. Report P12225. Applicant has access to this study and LoA is provided.
2. Katzschner I., Grimm T., 2019: Generic monitoring of European hares to determine proportion of time spent foraging in early maize in Central Europe. Report R1740045.
3. Allen L. (2019): Mesotrione – Foliage decline study on clover in Hungary, Germany, United Kingdom, Norther France and Belgium in 2019. Report CEMR-8397.
4. North L. (2016): Mesotrione – Foliage Decline study with A127379A on maize in Northern France and the United Kingdom in 2015. Report A12739A_11065.
The Letters of Access do not authorize Sharda CropChem España S.L. to inspect any of the studies or receive copies thereof, as a whole or in part, in original form or as a copy.
Those studies were evaluated and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Those studies were not re-evaluated.

The safe use is concluded only for application rate of 1.0 L product/ha (100 g a.s./ha). ~~However, in the proposed use pattern of MIGHTY such a dose rate is not included.~~
~~As~~ the risk to mammals via food (chronic) and drinking water is still unacceptable following application of MIGHTY at 1.5 L/ha (corresponding to 150 g a.s./ha), ~~the only dose included in the GAP, currently no safe use can be concluded for the product.~~

9.1.1.2 Effects on aquatic organisms (KCP 10.2)

Most PEC/RAC values for Mesotrione are below the trigger value of 1 at step 3, indicating that Mesotrione poses a low risk to aquatic organisms. However, some PEC/RACvalues taken from the assessment of *Lemna gibba* are above the trigger value of 1 (D3, D6, R1, R2 and R3 scenarios), indicating that Mesotrione poses a potential risk to higher plant. Based on the results of the risk assessment at step 4, the following conclusions regarding buffer zones and vegetative buffer strips may be

drawn for a maize use:

- D3 and D6 ditch scenarios: A 5m no spray buffer zone is required.
- R1 and R2 stream scenarios: A 20m no spray buffer zone and a 20m vegetative buffer strip are required.
- R3 stream scenario: A 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use for pH 5.1 and pH 6.5; only for pH 7.9 a 10m no spray buffer zone and a 10m vegetative buffer strip are required.
- For R4 stream scenario 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use.

The risk to aquatic organisms for metabolites AMBA, MNBA and SYN546974 was assessed as low at FOCUS step 1 for the representative use on maize.

In addition, no unacceptable risk for the formulated MIGHTY (MESOTRIONE 10% SC) is expected under GAP conditions.

~~***Spe3:** To protect aquatic organisms respect an unsprayed vegetated buffer zone of 20 m to surface water bodies.*~~

9.1.1.3 Effects on bees (KCP 10.3.1)

The evaluation of the risk for bees was performed in accordance with the recommendations of the "Guidance Document on Terrestrial Ecotoxicology", as provided by the Commission Services (SANCO/10329/2002 rev.2 (final), October 17, 2002).

A low risk to bees is expected after the application of MIGHTY (MESOTRIONE 10% SC) according to the proposed GAP.
Due to the lack of critical data for other groups of organisms (mammals and terrestrial plants), the Applicant was given the opportunity to submit those studies.
Currently, the chronic studies for bees are available, therefore the dRR was updated accordingly.

The data requirements in accordance with Commission Regulation (EU) No 284/2013 for the chronic toxicity to adult honeybees and honeybee larvae are fulfilled.

There is not harmonized approach for the chronic risk assessment for bees, therefore, Concerned Member States must decide on the requirements in this regard at national level.

9.1.1.4 Effects on arthropods other than bees (KCP 10.3.2)

No in-field ~~and off-field~~ risk to non-target arthropods is expected after the application of MIGHTY (MESOTRIONE 10% SC) according to the proposed GAP.
Based on the Tier II data for *Aphidius rhopalosiphi and Aleochara bilineata*, with effects below the triggers, the off-field risk is low. Nevertheless, for *Typhlodromus pyri* effects on reproduction were obtained below 38.3 g a.s./ha (lowest dose tested in extended laboratory study). Thus, no reproduction endpoint is available. In opinion of zRMS, in this case, based on the aged residue study, where only 4.82% and 9.30% of effects on mortality and reproduction respectively were found at 150 g a.s./ha in fresh residue (at 0DAA) the acceptable risk can be identified. Nevertheless, as this is not in accordance with ESCORT II, the decision whether the evaluation of the risk is sufficient should be taken on the national level.

9.1.1.5 Effects on non-target soil meso- and macrofauna (KCP 10.4), Effects on soil microbial activity (KCP 10.5)

A low acute and long-term risk was assessed for earthworms, other non-target soil meso- and macrofauna for the representative use on maize.

In addition, on the basis of the results of ~~carbon and~~ nitrogen transformation tests, it was concluded that MIGHTY (MESOTRIONE 10% SC) did not have any long-term adverse effect on the process of carbon and nitrogen transformation in aerobic surface soils

9.1.1.6 Effects on non-target terrestrial plants (KCP 10.6)

The risk assessment has been realized according to the seedling emergence ~~and vegetative vigour~~ data.

The calculated TER values are below the Annex VI trigger of 5 for seedling emergence ~~and vegetative~~ vigour when a distance of 1 m is considered. Therefore, no potential risk to non-target plants located outside the treated area after application of MIGHTY (MESOTRIONE 10% SC) according to the GAP table is expected when risk mitigation measures are considered.

No explanation was given as why for the herbicide only one formation study on terrestrial plants was performed. Moreover, based on data from LoEP the lowest endpoint for representative formulation is from vegetative vigour test. Nevertheless, the Applicant didn't performed this type of test, and only seedling emergence study for MIGHTY is available. There is also no explanation on what basis the Applicant assumed that a representative formulation covers the risk to MIGHTY. Since different plant species have been tested in the seedling emergence test, it is not possible to compare the toxicity of both formulations. Moreover, in seedling emergence study for MIGHTY the *Lactuca sativa* was not tested (the most sensitive species based on Callisto 100 SC data). Therefore, in opinion of zRMS, the vegetative vigour test for MIGHTY is required.

The Applicant was requested to submit the vegetative vigour test with MIGHTY.

~~***Spe3:*** *to protect non-target plants respect an unsprayed buffer zone of 30m to non-agricultural land OR an unsprayed buffer zone of 15m to non-agricultural land with the use of 50% drift reducing nozzles OR an unsprayed buffer zone of 10m to non-agricultural land with the use of 75% drift reducing nozzles OR an unsprayed buffer zone of 5m to non-agricultural land with the use of 90% drift reducing nozzles.~~

New study on vegetative vigour with MIGHTY is submitted by the applicant. After the risk assessment, no unacceptable risk on non-target plants is obtained with the following risk mitigation measures:

The Applicant' TERs calculations were performed for application rate of 1.0 L product/ha (100 g a.s./ha). However, in the proposed use pattern of MIGHTY such a dose rate is not included. Thus, additional calculations for application rate of 1.5 L product/ha, the only dose included in the GAP, were performed by zRMS.

Application rate 1.5 L/ha (150 g a.s./ha)
SPe3: *to protect non-target plants respect an unsprayed buffer zone of 5m to non-agricultural land OR the use of 75% drift reducing nozzles*

Application rate 1.0 L/ha (100 g a.s./ha)
Spe3: *to protect non-target plants respect an unsprayed buffer zone of 5m to non-agricultural land OR the use of 50% drift reducing nozzles.*

9.1.1.7 Effects on other terrestrial organisms (flora and fauna) (KCP 10.7)

Not relevant.

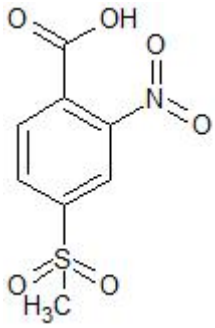
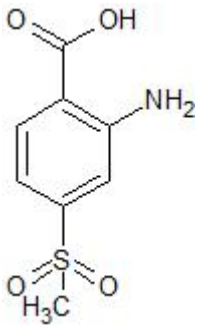
9.1.2 Grouping of intended uses for risk assessment

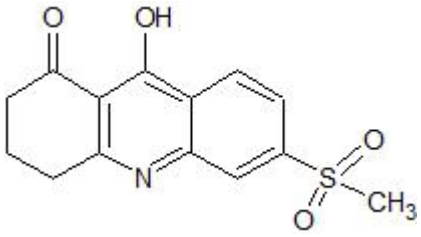
Not relevant because MIGHTY (MESOTRIONE 10% SC) is intended for only one crop (maize), one use (herbicide), for a single application, and with one application rate (1.5 L/ha).

9.1.3 Consideration of metabolites

A list of metabolites found in environmental compartments is provided below. The need for conducting a metabolite-specific risk assessment in the context of the evaluation of MIGHTY (MESOTRIONE 10% SC) is indicated in the table.

Table 9.1-2 Metabolites of Mesotrione

Metabolite	Chemical structure	Molar mass	Maximum occurrence in compartments	Risk assessment required?
MNBA	 4-(methylsulfonyl)-2-nitrobenzoic acid O=S(C)(=O)c1cc(c(cc1)C(=O)O)N(=O)=O	245	Soil: 57.2% Water/Sediment: 7.9%	Yes, for soil and water.
AMBA	 2-amino-4-(methylsulfonyl)benzoic acid O=S(C)(=O)c1cc(N)c(cc1)C(=O)O	215	Soil: 9.7% Water/Sediment: 24.6%	Yes, for water and soil.

Metabolite	Chemical structure	Molar mass	Maximum occurrence in compartments	Risk assessment required?
SYN546974	 9-hydroxy-6-(methylsulfonyl)-3,4-dihydroacridin- 1(2H)-one CS(=O)(=O)c1cc2nc3CCCC(=O)c3c(O)c2cc1	291	Soil: 1 x 10-10% Water/sediment: 33%	Yes, for water

9.2 Effects on birds (KCP 10.1.1)

9.2.1 Toxicity data

Avian toxicity studies have been carried out with Mesotrione. Full details of these studies are provided in the respective EU DAR.

Effects on birds of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione.
However, the provision of further data on the MIGHTY (MESOTRIONE 10% SC) is not considered essential, because birds are typically exposed to dry residues on their food items following the dilution and spraying of the formulated product. During these processes, much of the formulation constituents are likely to be lost by volatilisation. Since oral exposure is the main route exposure, toxicity data for the active substance are therefore used in preference to data from tests with the formulated material. On this basis, the risk to birds from the proposed uses of MIGHTY (MESOTRIONE 10% SC) will be assessed using data on Mesotrione.

The selection of studies and endpoints for the risk assessment is in line with the results of the EU review process.

Table 9.2-1: Endpoints and effect values relevant for the risk assessment for birds

Species	Substance	Exposure System	Results	Reference
Bobwhite quail (*Colinus virginianus*)	Active substance	Acute	LD_{50} >2000 mg a.s./kg bw (corrected to 3776 mg a.s./kg bw) NOEL = 2000 mg a.s./kg bw/d	EFSA Journal 2016;14(3):4419
Mallard duck (*Anas platyrhynchos*)	Active substance	Short-term	LC_{50} >5200 mg/kg diet NOEC = 5200 mg a.s./kg diet	EFSA Journal 2016;14(3):4419

Species	Substance	Exposure System	Results	Reference
Bobwhite quail (*Colinus virginianus*)	Active substance	Short-term	LC_{50} >5200 mg/kg diet NOEC = 5200 mg a.s./kg diet	EFSA Journal 2016;14(3):4419
Bobwhite quail (*Colinus virginianus*)	Active substance	Long-term	NOEC = 3000 mg a.s./kg diet	EFSA Journal 2016;14(3):4419
Mallard duck (*Anas platyrhynchos*)	Active substance	Long-term	NOEL 120 mg a.s./kg diet = 20.6 mg a.s./kg bw/d	EFSA Journal 2016;14(3):4419

9.2.1.1 Justification for new endpoints

The used endpoints were the EU agreed ones.

9.2.2 Risk assessment for spray applications

The risk assessment is based on the methods presented in the Guidance Document on Risk Assessment for Birds and Mammals on request from EFSA (EFSA Journal 2009; 7(12): 1438; hereafter referred to as EFSA/2009/1438).

9.2.2.1 First-tier assessment (screening/generic focal species)

The results of the acute and reproductive first-tier risk assessments are summarised in the following tables.

Table 9.2-2: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize				
Active substance/product	Mesotrione				
Application rate (g a.s./ha)	1 x 150				
Acute toxicity (mg a.s./kg bw)	> 2000				
TER criterion	10				
Crop scenario Growth stage	**Indicator/generic focal species**	**SV_{90}**	**MAF_{90}**	**DDD_{90} (mg/kg bw/d)**	**TER_a**
Maize	"Indicator species for screening"	158.8	1.0	23.82	>84.0

Reprod. toxicity (mg/kg bw/d)	20.6				
TER criterion	5				
Crop scenario Growth stage	**Indicator/generic focal species**	**SV_m**	**MAF_m × TWA**	**DDD_m (mg/kg bw/d)**	**TER_{lt}**
Maize BBCH 10-29	Medium herbivorous/granivorous bird “pigeon”	22.7	0.53	1.80	11.4
Maize BBCH 10-19	Small insectivorous bird “wagtail”	11.3	0.53	0.90	22.9
Maize BBCH 10-29	Small omnivorous bird “lark”	10.9	0.53	1.58	23.8
Maize Leaf development BBCH 10-19	Small insectivorous/worm feeding species “thrush”	5.7	0.53	0.45	45.5
Maize BBCH 10 – 29	Medium granivorous bird “gamebird”	3.0	0.53	0.24	86.4

SV: shortcut value; MAF: multiple application factor; TWA: time-weighted average factor; DDD: daily dietary dose; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

9.2.2.2 Drinking water exposure

When necessary, the assessment of the risk for birds due to uptake of contaminated drinking water is conducted for a small granivorous bird with a body weight of 15.3 g (*Carduelis cannabina*) and a drinking water uptake rate of 0.46 L/kg bw/d (*cf.* Appendix K of EFSA/2009/1438).

Leaf scenario

Since MIGHTY (MESOTRIONE 10% SC) is not intended to be applied on leafy vegetables forming heads or crop plants with comparable water collecting structures at principal growth stage 4 or later, the leaf scenario does not have to be considered.

Puddle scenario

Due to the characteristics of the exposure scenario in connection with the standard assumptions for water uptake by animals, no specific calculations of exposure and TER are necessary when the ratio of effective application rate (in g/ha) to relevant endpoint (in mg/kg bw/d) does not exceed 50 in the case of less sorptive substances (Koc < 500 L/kg) or 3000 in the case of more sorptive substances (Koc ≥ 500 L/kg).

With a K(f)oc of 14 (worst case value, EFSA Journal 2016;14(3):4419), Mesotrione belongs to the group of less sorptive substances. To achieve a concise risk assessment, the risk envelope approach is applied.

Effective application rate (g/ha)	=	150		
Acute toxicity (mg/kg bw)	=	> 2000	quotient =	< 0.08
Reprod. toxicity (mg/kg bw/d)	=	20.6	quotient =	7.28

Since the ratio of effective application rate (in g/ha) to relevant endpoint (in mg/kg bw/d) does not exceed the critical value of 50, a quantitative risk assessment (calculation of TER values) is not necessary.

9.2.2.3 Effects of secondary poisoning

The log P_{ow} of Mesotrione amounts to 0.11 (EFSA Journal 2016;14(3):4419) and thus does not exceed the trigger value of 3. A risk assessment for effects due to secondary poisoning is not required.

Risk assessment for earthworm-eating birds via secondary poisoning

Not required.

Risk assessment for fish-eating birds via secondary poisoning

Not required.

9.2.2.4 Biomagnification in terrestrial food chains

Not relevant.

9.2.3 Risk assessment for baits, pellets, granules, prills or treated seed

Not relevant.

9.2.4 Overall conclusions

Risk assessment for birds concludes in a low acute and long-term risk as well as for drinking water exposures (puddle and leaf scenarios). Therefore, no unacceptable acute and long-term risks are expected for birds.

Review Comments:

The acute and chronic risks of MIGHTY to birds were assessed from toxicity exposure ratios between toxicity endpoints, estimated from study with active ingredient and maximum residues occurring on food items. No acute toxicity test with the formulation was required.

All TER values exceed the relevant triggers indicating that MIGHTY does not pose an unacceptable risk to birds following applications according to recommended use pattern.

Evaluation of exposing to birds through the drinking water demonstrated the acceptable risk. Since the log P_{ow} value of Mesotrione and its relevant metabolites are all below the trigger of 3, the evaluation of the risk of secondary poisoning is not triggered.

9.3 Effects on terrestrial vertebrates other than birds (KCP 10.1.2)

9.3.1 Toxicity data

Mammalian toxicity studies have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on mammals of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Section 6 (Mammalian Toxicology) of this report.

However, mammals are typically exposed to dry residues on their food items following the dilution and spraying of the formulated product. During these processes, much of the formulation constituents are likely to be lost by volatilisation. Therefore, where oral exposure is the main route of exposure, toxicity data for the active substance are used in preference to data from tests with the formulated material. Exposure to MIGHTY (MESOTRIONE 10% SC) via dermal and inhalation routes is considered unlikely,

since at the time of application and for a short period thereafter, most wild mammals will leave the immediate vicinity of spray operations in response to the human disturbance.

The selection of studies and endpoints for the risk assessment is in line with from the results of the EU review process.

Table 9.3-1: Endpoints and effect values relevant for the risk assessment for mammals

Species	Substance	Exposure System	Results	Reference
Rat	Active substance	Acute	LD_{50} >5000 mg a.s./kg bw	EFSA Journal 2016;14(3):4419
Rat	Active substance	Long-term	NOEL = 2.5 mg a.s./kg feed = 0.3 mg a.s./kg bw/d	EFSA Journal 2016;14(3):4419
Rat	MNBA	Acute	LD_{50} >5000 mg MNBA/kg bw	EFSA Journal 2016;14(3):4419
Rat	AMBA	Acute	LD_{50} >5000 mg AMBA/kg bw	EFSA Journal 2016;14(3):4419

9.3.1.1 Justification for new endpoints

The used endpoints were the EU agreed ones. The acute risk assessment for metabolites MNBA and AMBA is covered by the acute risk assessment of the active substance Mesotrione.

9.3.2 Risk assessment for spray applications

The risk assessment is based on the methods presented in the Guidance Document on Risk Assessment for Mammals and Mammals on request from EFSA (EFSA Journal 2009; 7(12): 1438; hereafter referred to as EFSA/2009/1438).

9.3.2.1 First-tier assessment (screening/generic focal species)

The results of the acute and reproductive first-tier risk assessments are summarised in the following tables.

Table 9.3-2: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize				
Active substance/product	Mesotrione / Mesotrione 10% SC				
Application rate (g a.s./ha)	1 × 150				
Acute toxicity (mg/kg bw)	> 5000				
TER criterion	10				
Crop scenario Growth stage	**Indicator/generic focal species**	**SV_{90}**	**MAF_{90}**	**DDD_{90} (mg/kg bw/d)**	**TER_a**
Maize	"Indicator species for screening"	136.4	1.0	20.46	>244.4

Reprod. toxicity (mg/kg bw/d)	0.3				
TER criterion	5				
Crop scenario Growth stage	**Indicator/generic focal species**	**SV_m**	**MAF_m × TWA**	**DDD_m (mg/kg bw/d)**	**TER_{lt}**
Maize BBCH 10-29	Small herbivorous mammal "vole"	72.3	0.53	5.75	**0.05**
Maize BBCH 10-29	Small omnivorous mammal "mouse"	7.8	0.53	0.62	**0.48**
Maize BBCH 10-19	Small insectivorous mammal "shrew"	4.2	0.53	0.33	**0.90**

SV: shortcut value; MAF: multiple application factor; TWA: time-weighted average factor; DDD: daily dietary dose; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

9.3.2.2 Higher-tier risk assessment

Higher-tier risk assessment is required since the TERlt was below the trigger for mammals in maize.

In order to refine the risk assessment, the following parameters refined below were considered.

Toxicological endpoint

The TER calculation was based on the NOEL obtained in a multi-generations study in rats (specie chosen instead of mouse because more sensitive to mesotrione) based on reduced litter sizes. A NOAEL of 1.2 mg/kg bw/d corresponds to the reproduction and development endpoints of the first two generations F0 and F1 only. The continuous exposure of individuals for three generations can be considered unrepresentative of the exposure after one application of mesotrione.

Therefore, the NOAEL of 1.2 mg/kg bw/d considered as endpoint is more relevant for an exotoxicological assessment and can be used for a refined long-term risk assessment to mammals.

Moreover, in the course of national evaluation of mesotrione-containing product Elumis by the Comepetent Authority in Netherlands, the following findings were concluded:

"An additional study was provided and summarized by the Ctgb (section toxicology) In order to address the potential long-term effects caused by a short exposure to mesotrione residues, a 28-day study was carried out with male rats (the most sensitive organisms to tyrosinemia effects). The study mimicked the potential exposure to mesotrione residues by altering the exposure concentrations provided in the food from a maximum of 13.03 mg/kg bw/day to 0.025 mg/kg bw/day. This study indicated that shortterm exposure to rats will not cause tyrosinemia-linked effects at concentrations up to an initial maximum exposure of 100 mg/kg diet (13.03 mg/kg bw/day). For the purpose of the risk assessment, the average twa exposure value during the 28-day test period of 2.4 mg/kg bw/day has been used as the endpoint" (Toelatingsnummer 13192 N, 12.06 2009, http://www.ctb.agro.nl/ctb_files/13192_01.html).

Therefore, the NOEL of 2.4 mg/kg bw/d will be used as an option in the risk assessment

Review Comments:

The applicant's proposal to change the mammalian endpoint was not accepted. This issue was discussed at Pesticides Peer Review experts Meeting 136 in December 2015, where it was decided that the observed effects (e.g., litter size and pup survival) on the F2 generation should not disregard. Therefore the meeting agreed that the NOAEL of 0.3 mg/kg bw/day should be used in the risk assessment.

In zRMS opinion, the endpoint can be re-evaluated by using the benchmark dose approach. Further details can be found in the EFSA Journal 2017;15(1):4658.

Without additional data (BMD approach), it is not possible to change the mammalian endpoint.

DT_{50}

In the Tier I risk assessment, for the dissipation and degradation of residues from plant material a default DT_{50} value of 10 days was assumed. However, nine decline trials on cereals are available for CEU (please refer to the DAR of mesotrione, *Annex B.7, Residues data*) and the results of those studies are detailed below.

Reference	Crop	BBCH	Application rate (g a.s./ha)	Residues (mg/kg)	Time (days)	DT_{50} (days)
France (N) Barnes, J.P. 1997a Trial Ref. DP59806	Maize (Bangy)	16-18	150	4.58 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 63 80	1.58
France (N) Barnes, J.P. 1997b Trial Ref. DP59808	Maize (LG2243)	16-18	200	20 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 56 90	1.28
Germany Barnes, J.P. 1997c Trial Ref. DP59810	Maize (Diamant)	17	150	9.23 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 13 32 68	1.32
Germany Barnes, J.P. 1997c Trial Ref. DP59810	Maize (General)	17	150	10.31 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 32 73	1.40
Germany Barnes, J.P. 1997c Trial Ref. DP59810	Maize (Graf)	16-17	150	11.56 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 32 78	1.38
Germany Barnes, J.P. 1997c Trial Ref. DP59810	Maize (Anjou)	16-17	150	5.98 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 15 30 70	1.52
Germany Barnes, J.P. 1997d Trial Ref. DP59812	Maize (Janna)	16	150	23.2 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 44 86	1.25
Germany Barnes, J.P. 1997d Trial Ref. DP59812	Maize (Ilias)	17	200	10.9 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 35 110	1.39
Germany Barnes, J.P. 1997d Trial Ref. DP59812	Maize (Helix)	18	200	9.21 (immature) <0.01 (immature) <0.01 (forage) <0.01 (silage)	0 14 36 88	1.42
					Mean	**1.39**
				DT_{50}	**Geo mean**	**1.39**
					90^{th} percentile	**1.53**

The calculated mean DT_{50} and was 1.39, the geometric mean was 1.39 and the 90th percentile was 1.53. For the refinement of the long-term risk, the worst case 90th percentile DT_{50} value of 1.53 d was used. In addition, as indicated in table above, the residues in immature maize were shown to have declined to <0.01 mg a.s./kg (below the limit of quantification) within 14 days when application was made at proposed rates.

Review Comments:

The applicant's proposal to change the 10 day default DT_{50} value was not accepted. According EFSA Birds & Mammals guidance (2009) on measured residues and residue dynamics, the sampling points should be more closely spaced. Standard sampling time is on day 0, 1, 3-4, 7 and 14. Moreover, some recommendation indicate that sampled should have all taken place within three days of the application.

Therefore, the trials presented above were considered as insufficient data points to fit reliable kinetics.

The 10 day default value should be used in the risk assessment.

TWA

In the Tier I assessment, a default TWA = 0.53 was used (estimates time-weighted exposure over 21 days, assuming a default DT_{50} of 10 days). However, the estimated decline of the residues of Mesotrione on plants is lower than the default value of 10 days. Considering the 90th percentile DT_{50} of 1.53 d, the TWA factor was re-calculated considering the formula of the EFSA/2009/1438, and the resulting TWA was 0.11. This value was used for the refinement.

Review Comments:

The applicant's proposal to change the 10 day default DT_{50} value was not accepted. The default TWA value of 0.53 should be used in the risk assessment.

Identification of relevant focal mammal species in early maize (BBCH 10-14)

There are many reasons why the risk assessment for vole is considered to be covered through the assessment of other small mammalian species:

- High fecundity and population recuperation of the vole.
- Primary source of food outside crops fields for the vole.
- Necessity of population control measures since the vole is considered a crop pest when high population levels are reached.
- Other agricultural techniques being also means of population control

In addition, based in generic studies and literatura data, the Wood mouse (*Apodemus syvalticus*) and the European hare (*Lepus europaeus*) were identified as suitable focal species in maize fields. Same findings were reported in studies already evaluated by EFSA. In adittion, Mesotrione 10%SC is a herbicide used as a treatment against annual and perennial grass weeds and the symptoms appear a few days after application, these weeds assumed to be less attractive and palatable for herbivorous mammals, so it is unlikely that voles will prefer feeding in the treated areas (*Peer review of the pesticide risk assessment of the active substance fluazifop-P).*

Review Comments:

Agree that the focal species for maize at BBCH growth stages of 10-14 are wood mouse and brown hare.

PD values

For woodmouse, the modification of diet is proposed following literature data (Peltz, 1989), as already accepted by the EFSA (*Conclusion on the peer review of the pesticide risk assessment of the active substance topramezone*. EFSA Journal 2014;12(2):3540). In May, wood mouse diet consists of: 0.16 plant leaves, 0.34 weed seeds, 0.1 large arthropods and 0.4 earthworms. RUD in earthworms was calculated according to the bioaccumulation equation and the soil PEC value.

Review Comments:

As no detail were presented about literature data (Peltz, 1989), the applicant's proposal to use PD values from EFSA Journal 2014;12(2):3540 for wood mouse was not accepted. Moreover, it should be highlighted that he risk assessment for topramezone was performed in accordance with SANCO 4145/2000. In zRMS opinion is more reasonable to use shortcut value of 7.8 (combination: 25% weeds 50% weed seeds 25% ground arthropods) from EFSA B&M guidance.

PT values

For woodmouse, PT of 0.98 (generic studies for the wood mouse) is proposed, following EFSA (*Conclusion on the peer review of the pesticide risk assessment of the active substance topramezone*. EFSA Journal 2014;12(2):3540).

For brown hare, a 90th %ile PT of 0.93 is proposed. This value is relevant for the consumers only in cereal crops in spring according to Prosser, 2010. It must be, however, emphasized that hare are highly mobile animals and it is very unlikely it may feed on treated maize plants thorough all breeding period.

Review Comments:

The applicant's proposal to use PT of 0.93 value was not accepted for maize BBCH 10-14. In zRMS opinion it is not reasonable to extrapolate PT for brown hare in newly-drilled cereals to early maize. In freshly drilled cereal fields, available green plant material is likely to be less than in germinated maize fields. Thus maize may be more attractive to mammals.

The default value of 1 from B&M guidance should be used in the risk assessment.

DF value

In maize the leave development growth stages may overlap with shoot elongation, therefore a consideration of deposition factor is justified. Moreover, as the mesotrione is a herbicide, the complete cover of crop by spray is not secured and both lower leaves and stems and weeds growing under crop plants are not completely covered with mesotrione solution. Here, the applicant proposes a deposition factor of 0.5 (BBCH 10-39) already agreed (*Conclusion on the peer review of the pesticonsideration of ide risk assessment of the active substance pyridate*. EFSA Journal 2014; 12 (8): 3801).

Review Comments:

The applicant's proposal to use DF of 0.5 value was not accepted for maize BBCH 10-14. The default value of 1 from B&M guidance should be used in the risk assessment.

~~*Higher tier risk assessment for woodmouse*~~

~~A FIR/bw corresponding to modified diet of woodmouse was calculated in accordance to the EFSA GD~~

~~**Table 9.3-3: Calculation of FIR for woodmouse**~~

Species	Body weight	Diet item	Daily energy expenditure, DEE [kJ/d]	Food energy, FE [kJ/d]	Moisture content, MC [%]	Assimilation efficiency, AE [%]	FIR	FIR/bw
Small omnivorous mammal "mouse"	21.70	Non-grass weeds & leafy crops	58.83	17.8	88.1	76	36.54	1.68
		earthworms	58.83	19.3	84.6	85	23.28	1.07
		Small seeds	58.83	21.7	9.9	84	3.58	0.17
		Ground arthropods	58.83	22.7	68.8	87	9.55	0.44

As it was mentioned above, RUD in earthworms was calculated according to the bioaccumulation equation and the soil PEC value.

Bioconcentration factor for the earthworm ($BCF_{earthworm}$) was calculated using the equation:

$$BCF_{earthworm} = \frac{0.84 + 0.012Kow}{f_{oc} x K_{oc}}$$

With:
Koc = Organic carbon adsorption coefficient
foc = Organic carbon content of soil (take 0.02 as a default value)

Estimate residues in earthworms were then calculated using the equation:
$PEC_{earthworm} = PEC_{soil} \times BCF_{earthworm}$

Table 9.3-4: Calculation of RUD surrogate value in earthworms

Kow	Foc	Koc	$BCF_{earthworm}$	PEC_{soil}	PEC_{worm}
1.29	0.02	109	0.3924	0.15	0.0589

Table 9.3-5: Refined reproductive risk assessment for woodmouse following application of mesotrione to maize

Species	FIR/bw	Rate, kg as/ha	RUD unit	RUD	PD	PT	TWA	MAF	AV	DF	ETE	∑ETE	NOEL	TER
Small omnivorous mammal "mouse"	1.68	0.15	Non-grass weeds & leafy crops	28.7	0.16	0.98	0.11	1	1	1	0.12	0.23	1.20	5.24
	1.07		earthworms	0.06	0.4	0.98	0.53	1	1	0.5	0.00			
	0.17		Small seeds	40.2	0.34	0.98	0.53	1	1	0.5	0.09		2.40*	10.48
	0.44		Ground arthropods	7.5	0.1	0.98	0.53	1	1	0.5	0.01			

* please refer to *Toxicological endpoint*

As the TER is above the trigger, it is concluded that no unacceptable risk to small herbivorous mammals is expected.

Higher tier risk assessment for brown hare

Table 9.3-6: Refined reproductive risk assessment for brown hare following application of mesotrione to maize

Species	bw (g)	FIR/bw	Rate, kg as/ha	RUD unit	RUD	PD	PT	TWA	MAF	AV	DF	ETE	NOEL	TER
Large herbivorous mammal brown hare	3230	0.334	0.15	Crop shoots	54.2	1	0.93	0.11	1	1	1	0.28	1.20	4.32
													2.40*	8.64

* please refer to *Toxicological endpoint*

As the TER is above the trigger, it is concluded that no unacceptable risk to large herbivorous mammals is expected.

Applicant update August 2022

Application rate
The Applicant wishes to consider a reduction of the maximum application rate from 150 g a.s./ha to 100 g a.s./ha for the refinement.

Focal species
As agreed by the zRMS, the relevant focal species for maize at BBCH growth stages 10-14 are wood mouse and brown hare.

PD
Wood mouse
For wood mouse, in the higher tier risk assessment, the default diet as given in EFSA/2009/1438 is considered: 25% weeds, 50% weed seeds and 25% ground arthropods.

Brown hare
For brown hare in the higher tier risk assessment it was considered that will feed exclusively on maize shoots (monocots).

Food intake rate
Wood mouse
According to EFSA/2009/1438, the FIR/bw for wood mouse considering the default diet is 0.27. Therefore, this value is used in the higher tier refinement.

Brown hare
The FIR/bw for hare was calculated by the applicant considering a body weight of 3800 g as indicated in the EFSA (2009) and considering that hare will feed exclusively on maize shoots (monocots). Calculation is presented below. The FIR/bw was calculated using the equation provided in Appendix G of the Guidance Document.

$$FIR = \left(\frac{DEE}{FE * \left(1 - \frac{MC}{100}\right) * \left(\frac{AE}{100}\right)} \right) \quad [\text{g fresh weight/d}]$$

In which

$$\log DEE = \log a + b \times \log bw$$

Table 9.3-7: Calculation of FIR/bw for hare

Food type	Daily energy expenditure (DEE)	bw (g)	Food energy (kJ/dry g)	Moisture content (%)	Assimilation efficiency (%)	FIR (g/day)	FIR/bw
Monocots (maize shoots)	2363.444	3800	17.6[a]	76.4[a]	47[b]	1210.66	0.32

[a] From table 3 of Appendix G in EFSA (2009)
[b] From table 4 of Appendix G in EFSA (2009)

Review Comments:

The Applicant' calculation of FIR/bw is correct. The body weight of 3800g is recommended in EFSA B&M guidance. The FIR/bw of 0.32 was accepted and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021, too.

PT

In order to refine the PT value, Applicant refers to data from two field monitoring studies.

Wood mouse

Von Blanckenhagen F., Grimm T., 2013: Generic field study on small mammals focal species and wood mouse (*Apodemus sylvaticus*) PT in maize fields in Germany. Report P12225. Applicant has access to this study and LoA is provided.

From this study, conducted in Germany in early stages of maize fields, a PT of 0.139 for the wood mouse was agreed at EU level. Therefore, the PT value of 0.139 is used in the higher tier risk assessment.

This study was evaluated at zonal level and the PT value of 0.139 was used (Callisto).

Review Comments:

This study was evaluated and used in the risk assessment at EU peer review and in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Study was not re-evaluated.

The PT of 0.139 is accepted by the zRMS.

Brown hare

Katzschner I., Grimm T., 2019: Generic monitoring of European hares to determine proportion of time spent foraging in early maize in Central Europe. Report R1740045. Applicant has access to this study and LoA is provided.

In this study, hares were monitorized in two Central Zone countries (Hungary and Germany) in maize field at early growth stages. From this study a mean PT of 0.36 and a 90th percentile PT of 0.62 were estimated from 21 radiotracked individuals that were observed to be crop consumers. For the higher tier risk assessment, the 90th percentile PT of 0.62 is used.

This study was evaluated at zonal level and the PT value of 0.62 was used (Callisto).

Review Comments:

This study was evaluated and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Study was not re-evaluated.

The PT of 0.62 is accepted by the zRMS.

DT_{50}

Weeds

In order to determine a DT_{50} value in weeds, Applicant refers to the residue decline study of Allen L. (2019): Mesotrione – Foliage decline study on clover in Hungary, Germany, United Kingdom, Norther France and Belgium in 2019. Report CEMR-8397. Applicant has access to this study and LoA is provided. The study was evaluated at zonal level (Callisto).

Although the study of Allen was conducted with a different formulation product that Mighty, it was conducted also with a solo formulation, a 480 SC formulation. Although the active substance content in the formulation used in the study of North was higher, the type of formulation is the same than for Mighty and therefore it is considered appropriate to use the data regarding the decline of mesotrione residues in treated clover from the study of Allen to determine a DT_{50} value in weeds that can be used also for the higher tier refinement of Mighty.

In this study, eleven residue decline trials were conducted on clover in Hungary, Germany, United Kingdom, Northern France and Belgium. Following the application, treated clover plant samples were collected at < 1hour after application (HAA), 8 HAA, 24 HAA, 32 HAA, 48 HAA, 3 days after application (DAA), 4 DAA and 7 DAA and untreated clover whole plant samples were collected < 1 hour before application (HBA). Samples were analysed for Mesotrione and results of analyses are given in the below table.

Time	**SRUK 18-001-037HR**	**SRUK 18-002-037HR**	**SRHU 18-053-037HR**	**SRHU 18-054-037HR**	**FRFR 18-010-037HR**	**SRFR 18-011-037HR**	**SRDE 18-001-037HR**	**SRDE 18-002-037HR**	**SRPL 18-014-037HR**	**SRPL 18-015-037HR**	**G006-18H**
0	6.10	3.63	11.97	11.69	11.51	8.75	4.46	9.11	6.15	6.50	8.58
8 HAA	6.03	4.20	11.41	8.99	8.78	9.98	5.66	2.71	4.34	4.58	8.48
24 HAA	4.58	3.39	11.02	8.76	9.86	8.73	4.59	2.59	2.28	4.72	8.17
32 HAA	2.69	4.09	9.02	8.80	6.72	4.77	3.98	3.29	2.06	6.37	5.65
48 HAA	2.73	2.61	7.14	6.89	5.47	4.66	4.21	2.54	1.78	5.78	5.54
72 HAA	1.95	2.17	6.06	5.51	2.00	4.77	0.22	2.61	3.82	1.8	3.26
96 HAA	0.58	2.76	0.14	0.12	0.58	4.37	0.08	0.43	1.67	1.66	3.43
168 HAA	0.27	0.19	0.11	0.07	0.09	0.70	0.09	0.05	1.23	0.27	1.70
0 DBA	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

DBA: days before application, HAA: hours after application

The kinetic evaluation of results of the study of Allen was evaluated at zonal level (Callisto) and a summary of the DT_{50} values derived is presented in table below.

Trial	**DT_{50} (d)**	**DT_{90} (hours)**
SRUK18-001-037HR	1.50	119
SRUK18-002-037HR	3.58	285
SRHU18-053-037HR	1.99	159
SRHU18-054-037HR	2.01	160
SRFR18-010-037HR	1.55	123
SRFR18-011-037HR	2.57	205
SRDE18-001-037HR	1.77	141
SRDE18-002-037HR*	1.05	83.9
SRPL18-014-037HR	2.40	192
SRPL18-015-037HR	2.63	210
G006-18H	2.65	211
Geometric mean	**2.19**	-

* Not used for geomean calculations

The geometic mean DT_{50} determined from the study was 2.19 days and this value was used for the refinement.

Review Comments:

This study was evaluated and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Study was not re-evaluated.
The DT_{50} of 2.19 days is accepted by the zRMS.

Maize
In order to determine a DT_{50} value in maize, Applicant refers to the residue decline study of North L. (2016): Mesotrione – Foliage Decline study with A127379A on maize in Northern France and the United Kingdom in 2015. Report A12739A_11065. Applicant has access to this study and LoA is provided. The study was evaluated at zonal level (Callisto).
Although the study of North was conducted with a different formulation product that Mighty, it was conducted also with a solo formulation, a 100 EC formulation. The active substance content in the formulation used in the study of North was the same than in Mighty and although the type of formulation is an EC, since the study was conducted to determine decline of mesotrione residues in maize plants (not to determine toxic effects on plants), it is considered appropriate to refer to the study of North to determine a DT_{50} value in maize that can be used also for Mighty.
In this study, five residue decline field trials on maize were conducted in Northern France and the United Kingdom. Plant samples were collected at < 1 hour after application (HAA), 4 HAA, 10 HAA, 24 HAA, 34 HAA, 48-51 HAA, 72-78 HAA and 96-99 HAA. Samples were analysed for mesotrione and its metabolite MNBA. The results of Analysis for samples for Mesotrione is given in the below table.

Application rate (g a.s./ha)	Crop part	Sampling	Residue mesotrione (mg/kg) Trial S15-02057-01 (UK)	Residue mesotrione (mg/kg) Trial S15-02057-03 (UK)	Residue mesotrione (mg/kg) Trial S15-02057-04 (UK)	Residue mesotrione (mg/kg) Trial S15-02057-06 (UK)	Residue mesotrione (mg/kg) Trial S15-02057-05 (FR)
1 x 150 g a.s./ha	Whole plant	< 1 HAA	7.09	13.96	4.24	3.09	14.99
		4 HAA	8.48	7.75	2.98	2.74	12.63
		10 HAA	4.11	6.25	3.33	2.05	8.61
		24 HAA	3.86	3.57	1.69	0.91	4.30
		34 HAA	2.79	2.95	0.50	0.80	2.19
		48-51 HAA	0.92	1.37	0.41	0.36	1.07
		72-78 HAA	0.16	0.63	0.14	<0.01	0.31
		96-99 HAA	0.12	0.11	0.06	0.10	0.13
		DT_{50}	0.803	0.512	0.663	0.531	0.636

The study, as well as the kinetic evaluation, were evaluated at zonal level (Callisto). The highest DT_{50} value determined from this study is 0.803 days. Therefore, a DT_{50} of 0.803 days has been considered by the Applicant for the higher tier refinement.

Review Comments:

This study was evaluated and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Study was not re-evaluated.
The DT_{50} of 0.803 days is accepted by the zRMS.

Ftwa
The ftwa factor was recalculated considering the formula of the EFSA/2009/1438.

Weeds

Considering the DT_{50} value of 2.19d, the resulting ftwa is 0.150 which is used in the higher tier risk assessment.

Maize
Considering the DT_{50} value of 0.803d, the resulting ftwa is 0.055 which is used in the higher tier risk assessment.

Refined TER calculations for wood mouse and hare are presented in the tables below.

Table 9.3-8: Refined reproductive risk assessment for woodmouse following application of mesotrione to maize – refined parameters (*) are further described and justified in the text

Intended use		Maize						
Active substance/product		Mesotrione						
Application rate (g/ha)		1 x 100*						
Reprod. Toxicity (mg/kg bw/d)		0.3						
TER criterion		5						
Crop scenario Growth stage	**Generic focal species**	**PD/diet type**	**FIR/bw**	**RUD**	**MAFm x f_{twa}**	**PT**	**DDD_m (mg/kg bw/d)**	**TER_{lt}**
BBCH 10-29	Small omnivorous mammal "mouse"	0.25 (weeds)	0.27	28.7	1 x 0.150*	0.139*	0.0040	
		0.25 (ground arthropods)		3.5[a]	1 x 0.53	0.139*	0.0017	
		0.50 (weed seeds)		40.2	1 x 0.53	0.139*	0.040	
						Total	0.0458	6.56

[a] According to Appendix A from EFSA (2009), RUD values for arthropods with interception are relevant to maize at BBCH 10-29

Table 9.3-9: Refined reproductive risk assessment for hare following application of mesotrione to maize – refined parameters (*) are further described and justified in the text

Intended use		Maize						
Active substance/product		Mesotrione						
Application rate (g/ha)		1 x 100*						
Reprod. Toxicity (mg/kg bw/d)		0.3						
TER criterion		5						
Crop scenario Growth stage	**Generic focal species**	**PD/diet type**	**FIR/bw**	**RUD**	**MAFm x f_{twa}**	**PT**	**DDD_m (mg/kg bw/d)**	**TER_{lt}**
BBCH 10-29	Large herbivorous mammal "hare"	1 / Maize shoots	0.32*	54.2	1 x 0.055*	0.62*	0.06	5.07

The TER values are above the trigger showing no risk and hence an acceptable risk for wood mouse and hare are expected.

9.3.2.3 Drinking water exposure

When necessary, the assessment of the risk for mammals due to uptake of contaminated drinking water is conducted for a small omnivorous mammal with a body weight of 21.7 g (*Apodemus sylvaticus*) and a drinking water uptake rate of 0.24 L/kg bw/d (*cf.* Appendix K of EFSA/2009/1438).

Puddle scenario

Due to the characteristics of the exposure scenario in connection with the standard assumptions for water uptake by animals, no specific calculations of exposure and TER are necessary when the ratio of effective application rate (in g/ha) to relevant endpoint (in mg/kg bw/d) does not exceed 50 in the case of less sorptive substances (Koc < 500 L/kg) or 3000 in the case of more sorptive substances (Koc ≥ 500 L/kg).

With a K(f)oc of 14 (worst case value, EFSA Journal 2016;14(3):4419), Mesotrione belongs to the group of less sorptive substances.

Effective application rate (g/ha)	=	150			
Acute toxicity (mg/kg bw)	=	5000	quotient	=	0.03
Reprod. toxicity (mg/kg bw/d)	=	0.3	quotient	=	**625**

Since the ratio of effective application rate to relevant endpoint exceeds 50 for reproductive assessment, quantitative long-term risk assessment is necessary.

Table 9.3-10: Assessment of the long-term risk for mammals due to exposure to Mesotrione via contaminated drinking water in puddles

Intended use	Maize				
Active substance	Mesotrione				
Application rate (g a.s./ha)	1 × 150				
Reprod. toxicity (mg/kg bw/d)	0.3				
TER criterion	5				
Soil-relevant applic. rate (g/ha)	**Koc (L/kg)**	**PEC_{puddle} (mg/L)**	**DW uptake (L/kg bw/d)**	**Daily dose (mg/kg bw/d)**	**TER_{lt}**
150	14	0.37	0.24	0.09	**3.4**

PEC_{puddle}: concentration in puddles; DW: drinking water; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

~~Considering the worst case Koc of 14 L/kg (pH >7), the TERlt is lower than the long term trigger of 5 indicating an unacceptable long term risk to mammals from drinking water. A refinement of the toxicological endpoint with a NOAEL of 1.2 mg/kg bw/d was performed. The risk assessment was presented below.~~

~~**Table 9.3-11: Assessment of the long-term risk for mammals due to exposure to Mesotrione via contaminated drinking water in puddles**~~

~~**Intended use**~~	~~Maize~~
~~**Active substance**~~	~~Mesotrione~~

Application rate (g a.s./ha)	1 × 150				
Reprod. toxicity (mg/kg bw/d) TER criterion	1.2 5				
Soil-relevant applic. rate (g/ha)	Koc (L/kg)	PEC_{puddle} (mg/L)	DW uptake (L/kg bw/d)	Daily dose (mg/kg bw/d)	TER_{lt}
150	14	0.37	0.24	0.09	13.7

PEC_{puddle}: concentration in puddles; DW: drinking water; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

The value of the TERlt is higher than the long-term trigger of 5 not indicating an unacceptable long-term risk to mammals from drinking water

Applicant wishes to consider a reduction of the maximum application rate from 150 g a.s./ha to 100 g a.s./ha for the refinement. Calculation is presented below.

Table 9.3-12: Assessment of the long-term risk for mammals due to exposure to Mesotrione via contaminated drinking water in puddles

Intended use **Active substance** **Application rate (g a.s./ha)**	Maize Mesotrione 1 × 100				
Reprod. toxicity (mg/kg bw/d) **TER criterion**	0.3 5				
Soil-relevant applic. rate (g/ha)	**Koc (L/kg)**	**PEC_{puddle} (mg/L)**	**DW uptake (L/kg bw/d)**	**Daily dose (mg/kg bw/d)**	**TER_{lt}**
100	14	02436	0.24	0.0585	5.125

PEC_{puddle}: concentration in puddles; DW: drinking water; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.
*Application rate considering an interception factor of 25%.

The value of the TERlt is higher than the long-term trigger of 5 not indicating an unacceptable long-term risk to mammals from drinking water

9.3.2.4 Effects of secondary poisoning

The log P_{ow} of Mesotrione amounts to 0.11 and thus does not exceed the trigger value of 3. A risk assessment for effects due to secondary poisoning is not required.

Risk assessment for earthworm-eating mammals via secondary poisoning

Not required.

Risk assessment for fish-eating mammals via secondary poisoning

Not required.

9.3.2.5 Biomagnification in terrestrial food chains

The log P_{ow} of Mesotrione amounts to 0.11 and thus does not exceed the trigger value of 3. A risk assessment for effects from biomagnification in terrestrial food chains is not necessary.

9.3.3 Risk assessment for baits, pellets, granules, prills or treated seed

Not relevant.

9.3.4 Overall conclusions

~~There is no unacceptable acute risk for mammals. A long-term unacceptable risk was observed for mammals in maize in the initial assessment. The election for the wood mouse (*Apodemus syvalticus*) and the european hare (*Lepus europaeus*) as suitable focal species in maize fields based on generic studies and literatura data, after the refinement of the risk by refinement of the foliar DT_{50} twa, PT, PD, DF and FIR/bw values, allowed to obtain a long-term acceptable risk for the wood mouse (*Apodemus syvalticus*) and, after the refinement of the toxicological endpoint, allowed to obtain a long-term acceptable risk for European hare (*Lepus europaeus*).~~

~~No unacceptable risk for mammals was observed from drinking water after the toxicological endpoint refinement.~~

~~Therefore, there is no acute and long-term risk for mammals after an application of MIGHTY (MESOTRIONE 10% SC) according to the proposed GAP.~~

Review Comments:

The acute and chronic risks of MIGHTY to mammals were assessed from toxicity exposure ratios between toxicity endpoints, estimated from study with active ingredient, and maximum residues occurring on food items.

No safe use was concluded following application of MIGHTY at 1.5 L/ha (corresponding to 150 g a.s./ha) and further refinement is required. The high long-term and drinking water risk was identified.

The Applicant was requested to submit the additional data necessary to refined the risk to mammals and/or to consider a reduction of the maximum application rate.

Currently, the Applicant has access to following higher tier studies:
1. Von Blanckenhagen F., Grimm T., 2013: Generic field study on small mammals focal species and wood mouse (*Apodemus sylvaticus*) PT in maize fields in Germany. Report P12225. Applicant has access to this study and LoA is provided.
2. Katzschner I., Grimm T., 2019: Generic monitoring of European hares to determine proportion of time spent foraging in early maize in Central Europe. Report R1740045.
3. Allen L. (2019): Mesotrione – Foliage decline study on clover in Hungary, Germany, United Kingdom, Norther France and Belgium in 2019. Report CEMR-8397.
4. North L. (2016): Mesotrione – Foliage Decline study with A127379A on maize in Northern France and the United Kingdom in 2015. Report A12739A_11065.
The Letters of Access do not authorize Sharda CropChem España S.L. to inspect any of the studies or receive copies thereof, as a whole or in part, in original form or as a copy.
Those studies were evaluated and used in the risk assessment in the CEU Core Assessment for Callisto (A12739A), dated 21/09/2021. Having regard to the clauses in LoA, in current evaluation the accepted values/approaches were taken directly from mentioned above zonal report, without any details. Those studies were not re-evaluated.

The safe use is concluded only for application rate of 1.0 L product/ha (100 g a.s./ha). ~~However, in the proposed use pattern of MIGHTY such a dose rate is not included.~~
~~As~~ the risk to mammals via food (chronic) and drinking water is still unacceptable following application of MIGHTY at 1.5 L/ha (corresponding to 150 g a.s./ha), ~~the only dose included in the GAP, currently no safe use can be concluded for the product.~~

9.4 Effects on other terrestrial vertebrate wildlife (reptiles and amphibians) (KCP 10.1.3)

No additional studies on other terrestrial vertebrates are required.

9.5 Effects on aquatic organisms (KCP 10.2)

9.5.1 Toxicity data

Studies on the toxicity to aquatic organisms have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on aquatic organisms of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

The selection of studies and endpoints for the risk assessment is in line with the results of the EU review process.

Table 9.5-1: Endpoints and effect values relevant for the risk assessment for aquatic organisms – Mesotrione and relevant metabolites

Species	Substance	Exposure System	End point	Toxicity (mg/L)	Reference
Rainbow trout (*Oncorhynchus mykiss*)	Mesotrione	Acute 96-hr (static)	Mortality, LC_{50}	>120 mg a.s/L^{nom}	EFSA Journal 2016;14(3):4419
Bluegill sunfish (*Lepomis macrochirus*)	Mesotrione	Acute 96-hr (static)	Mortality, LC_{50}	>120 mg a.s/L^{nom}	EFSA Journal 2016;14(3):4419
Fathead minnow (*Pimephales promelas*)	Mesotrione	Chronic 36-d (flow-through)	NOEC (physical symptoms) *	12.5 mg a.s/L ^{nom}	EFSA Journal 2016;14(3):4419
Common carp (*Cyprinus carpio*)	Preparation	Acute 96-hr (static)	Mortality, LC_{50}	71 mg/L ^{nom}	EFSA Journal 2016;14(3):4419
Rainbow trout (*Oncorhynchus mykiss*)	AMBA	Acut 96-hr (static)	Mortality, LC_{50}	150 mg a.s/L ^{nom}	EFSA Journal 2016;14(3):4419
Rainbow trout (*Oncorhynchus mykiss*)	MNBA	Acute 96-hr (static)	Mortality, LC_{50}	>120 mg a.s/L ^{nom}	EFSA Journal 2016;14(3):4419
Water flea (*Daphnia magna*)	Mesotrione	Acute 48-h (static)	EC_{50}	> 622** mg a.s./L ^{mm}	EFSA Journal 2016;14(3):4419
Water flea (*Daphnia magna*)	Preparation	Acute 48-h (static)	EC_{50}	> 49 mg a.s./L ^{nom}	EFSA Journal 2016;14(3):4419
Water flea (*Daphnia magna*)	Mesotrione	Chronic 21-d (semi-static)	NOEC (reproduction & length)	180 mg a.s/L ^{nom}	EFSA Journal 2016;14(3):4419
Water flea (*Daphnia magna*)	MNBA	Acute 48-h (static)	EC_{50}	130 mg a.s./L ^{nom}	EFSA Journal 2016;14(3):4419
Water flea (*Daphnia magna*)	AMBA	Acute 48-h (static)	EC_{50}	160 mg a.s./L (nom)	EFSA Journal 2016;14(3):4419
Green microalgae (*Pseudokirchneriella subcapitata*)	Mesotrione	Chronic 120-hr (static) 120-hr 120-hr 120-hr 72-hr 72-hr	E_bC_{50} E_rC_{50} $NOEC_b$ E_bC_{10} E_bC_{20}	3.5 mg a.s/L (1.9-6.1) 13 (nom) (3.2->48) 0.75 (nom) 0.692 (nom) ^{L} 0.958 (nom) ^{L}	EFSA Journal 2016;14(3):4419
Green microalgae (*Pseudokirchneriella subcapitata*)	Callisto 100 SC	Chronic 96-hr (static) 96-hr 96-hr 72/96-hr 72-hr 72-hr	E_bC_{50} E_rC_{50} $NOEC_{b,r}$ E_bC_{10} E_bC_{20}	72 (nom) > 100 (nom) 10 (nom) 4.6 (2005)(nom)^{L} NE (2005)(nom) ^{L}	EFSA Journal 2016;14(3):4419
Green microalgae (*Pseudokirchneriella subcapitata*)	MNBA	Chronic 72-hr (static)	E_bC_{50} E_rC_{50}	38 (nom) 42 (nom)	EFSA Journal 2016;14(3):4419

Species	Substance	Exposure System	End point	Toxicity (mg/L)	Reference
Green microalgae (*Pseudokirchneriella subcapitata*)	AMBA	Chronic 72-hr (static)	E_bC_{50} E_rC_{50} NOECb,r E_yC_{10} E_yC_{20}	9.4 (nom) (5.3-16) 14 (nom) (8.5-26) 7.7 (nom) 2.58 (nom) 4.04 (nom)	EFSA Journal 2016;14(3):4419
Gibbous duckweed (*Lemna gibba*)	Mesotrione	14-d chronic (semi-static)	E_rC_{50} E_bC_{50} $NOEC_b$ (for dry weight) E_yC_{10} (for dry weight) E_yC_{20} (for dry weight)	0.022 mg a.s/L (0.02-0.026) 0.0077 (nom) (0.0066-0.009) 0.002 (nom) 0.0014 (nom) 0.0022 (nom)	EFSA Journal 2016;14(3):4419
Gibbous duckweed (*Lemna gibba*)	Callisto 100 SC	7-d chronic (semi-static)	E_rC_{50} (for dry weight) E_yC_{50} (for dry weight) NOEC (for all) E_yC_{10} (for frond no.) E_yC_{20} (for frond no.)	0.117 (nom) 0.0269 (nom) 0.0114 (nom) 0.00461 (nom) 0.0108 (nom)	EFSA Journal 2016;14(3):4419
Gibbous duckweed (*Lemna gibba*)	MNBA	7-d chronic (semi-static)	E_rC_{50} (for both) E_yC_{50} (for both) NOEC (for frond no) E_yC_{10} (for all) E_yC_{20} (for all)	> 97 (mm) > 97 (mm) 3.3 (mm) > 97 (mm) > 97 (mm)	EFSA Journal 2016;14(3):4419
Gibbous duckweed (*Lemna gibba*)	AMBA	7-d chronic (semi-static)	E_rC_{50} (for both) E_yC_{50} (for both) NOEC (for both) E_yC_{10} (for frond no) E_yC_{20} (for all)	> 90 (mm) > 90 (mm) 90 (mm) 24 (mm) > 90 (mm)	EFSA Journal 2016;14(3):4419
Gibbous duckweed (*Lemna gibba*)	SYN546974	7-d chronic (semi-static)	E_rC_{50} (for both) E_yC_{50} (for frond no) NOEC (for frond no) E_yC_{10} (for frond no) E_yC_{20} (for frond no)	> 95 (mm) 93 (mm) 2.9 (mm) 9.9 (mm) 21 (mm)	EFSA Journal 2016;14(3):4419
Higher-tier studies (micro- or mesocosm studies)					
Not needed					

nom: based on nominal concentrations; mm: based on mean measured concentrations; im: based on initial measured concentrations

^{L} – the lowest value, estimation was carried out on all available timescales;

*Physical symptoms: loss of balance, less activity, spinal deformity, skin lesions and internal bleeding;

**In the LoEP of Review Report of mesotrione (January 2013, SANCO/1416/2001 - Final), the value of > 900 mg mesotrione/L was misreported for acute invertebrate toxicity.

Table 9.5-2: Endpoints and effect values relevant for the risk assessment for aquatic organisms – MIGHTY (MESOTRIONE 10% SC)

Species	Substance	Exposure System	End point	Toxicity	Reference
Oncorhynchus mykiss	Mesotrione 10% SC	96 h, s	LC_{50}	~~70.54 mg/L (nom) (equivalent to 7.05 mg a.s./L)~~ >50 mg/L (nom) (equivalent to 5 mg a.s./L)	KCP 10.2.1-01 Świerkot, A. 2016 W/112/16
Daphnia magna	Mesotrione 10% SC	48 h, s	EC_{50}	88.39 mg/L (equivalent to 8.84 mg a.s./L)	KCP 10.2.1-02 Brzozowska-Wojoczek, K. 2016 W/148/16
Pseudokirchneriella subcapitata	Mesotrione 10% SC	72 h	E_rC_{50} E_yC_{50}	E_rC_{50} = 80.18 mg/L (nom) (equivalent to 8.02 mg a.s./L) E_yC_{50} = 43.13 mg/L (nom) (equivalent to 4.31 mg a.s./L)	KCP 10.2.1-03 Brzozowska-Wojoczek, K. 2016 W/147/16
Lemna gibba	Mesotrione 10% SC	7 d,s	E_rC_{50} E_yC_{50}	E_rC_{50} = 0.383 mg/L (nom) (equivalent to 0.038 mg a.s./L) E_yC_{50} = 0.078 mg/L (nom) (equivalent to 0.008 mg a.s./L)	KCP 10.2.1-04 Brzozowska-Wojoczek, K. 2016 W/149/16
Higher-tier studies (micro- or mesocosm studies)					

s: static; ss: semi-static; f: flow-through; nom: based on nominal concentrations; mm: based on mean measured concentrations

9.5.1.1 Justification for new endpoints

The used endpoints are the EU agreed ones, except for formulation, corresponding to data proper to MIGHTY (MESOTRIONE 10% SC) formulation.

9.5.2 Risk assessment

The evaluation of the risk for aquatic and sediment-dwelling organisms was performed in accordance with the recommendations of the "Guidance document on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters in the context of Regulation (EC) No 1107/2009", as provided by the Commission Services (SANTE-2015-00080, 15 January 2015).
The relevant global maximum FOCUS Step 1, 2 and 3 PEC_{SW} for risk assessments covering the proposed use pattern and the resulting PEC/RAC ratios are presented in the table below.

In the following table, the ratios between predicted environmental concentrations in surface water bodies (PEC_{SW}, PEC_{SED}) and regulatory acceptable concentrations (RAC) for aquatic organisms are given per intended use for each FOCUS scenario and each organism group.

Table 9.5-3: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for MESOTRIONE 10%SC for each organism group based on Rautmann Spray Drift calculations for the use of MESOTRIONE 10%SC in maize

Group			**Fish acute**	**Inverteb. acute**	**Algae**	**Higher plant**
Test species			***Oncorhynchus mykiss***	***Daphnia magna***	***Pseudokirchneriella subcapitata***	***Lemna gibba***
Endpoint			LC_{50}	EC_{50}	E_rC_{50}	E_rC_{50}
(µg/L)			~~70540~~ 50000	88390	80180	383
AF			100	100	10	10
RAC (µg/L)			~~705.4~~ 500	883.9	8018	38.3
Distance (m)	**Drift(%)**	**PEC gl-max (µg/L)**				
1	2.77	14.45	~~0.02~~ 0.03	0.02	<0.01	0.38

AF: Assessment factor; PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC ratios above the relevant trigger of 1 are shown in bold

Table 9.5-4: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Mesotrione for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of MIGHTY (MESOTRIONE 10% SC) in maize

Group		**Fish acute**	**Fish prolonged**	**Inverteb. acute**	**Inverteb. prolonged**	**Algae**	**Higher plant**
Test species		***Oncorhynchus mykiss / Lepomis macrochirus***	***Lepomis macrochirus***	***Daphnia magna***	***Daphnia magna***	***Pseudokirchn. subcapitata***	***Lemna gibba***
Endpoint		LC_{50}	NOEC	EC_{50}	NOEC	~~E_bC_{50}~~ E_rC_{50}	E_bC_{50}/E_rC_{50}

Group		**Fish acute**		**Fish prolonged**	**Inverteb. acute**		**Inverteb. prolonged**	**Algae**		**Higher plant**	
(µg/L)		~~7050~~ 5000*	>120000	12500	8840*	>622000	180000	13000	~~3500~~ 8020*	7.7#	38*
AF		100		10	100		10	10		10	
RAC (µg/L)		~~70.5~~ 50	1200	1250	88.40	6220	18000	1300	~~350~~ 802	0.77	3.8
FOCUS Scenario	**PEC $_{gl\text{-}max}$ (µg/L)**										
Step 1											
	50.25**	~~0.713~~ **1.005**	0.42	0.040	0.568	0.008	0.003	0.039	~~0.144~~ 0.063	**65.260**	**13.224**
Step 2											
S-Europe	12.33**	0.247	-		-	-		-		**16.013**	**3.245**
N-Europe	15.22**	0.304	-		-	-		-		**19.766**	**4.005**
Step 3											
D3/ditch	0.787	-	-		-	-		-		**1.02**	0.207
D4/pond	0.084**	-	-		-	-		-		0.11	0.022
D4/stream	0.678**	-	-		-	-		-		0.88	0.178
D5/pond	0.047**	-	-		-	-		-		0.06	0.012
D5/stream	0.689**	-	-		-	-		-		0.89	0.181
D6/ditch	0.792**	-	-		-	-		-		**1.03**	0.208

Group		Fish acute		Fish prolonged	Inverteb. acute		Inverteb. prolonged	Algae	Higher plant	
R1/pond	0.114**	-		-	-		-	-	0.15	0.03
R1/stream	2.397**	-		-	-		-	-	**3.11**	0.631
R2/stream	3.277**	-		-	-		-	-	**4.26**	0.862
R3/stream	5.929**	-		-	-		-	-	**7.70**	**1.56**
R4/stream	~~0.546***~~ 6.261	-		-	-		-	-	~~0.71~~ **8.13**	0.144 **1.65**

AF: Assessment factor; PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC ratios above the relevant trigger of 1 are shown in bold
*endpoint in terms of a.s. from Sharda study ~~is used as worst case~~.
**Worst case for the three pH's simulated.
*** Worst case at pH 7.9 since the right pH for this scenario is 8.4 (please, refer to Table 8.9-6 from dRR B8).
# no E_rC_{50} available

For the intended uses, calculated PEC/RAC ratios did indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for higher plants as characterised by an EC_{50} for *Lemna gibba* of 7.7 µg/L in connection with an assessment factor of 10) in several FOCUS Steps 1-3 scenarios. Therefore, further PEC/RAC ratios were calculated based on FOCUS Step 4 PEC_{SW} considering reduced exposure of surface water bodies.

Table 9.5-5: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for Mesotrione based on FOCUS Step 4 calculations and toxicity data for higher plant with mitigation of spray drift and run-off for the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use		Maize			
Active substance		Mesotrione			
Application rate (g a.s./ha)		1 x 150			
Nozzle reduction	**No-spray buffer (m)**	None (step 3 value)	5	10	20
	Vegetated filter strip (m)	None (step 3 value)	None	10	20
None	D3 ditch ~~(pH 5.1) Linear~~ (all)	0.787	0.258	-	-
None	D6 ditch (pH 7.9) Log	0.787	0.258	-	-
None	D6 ditch (pH 7.9) Linear	0.787	0.258	-	-
None	D6 ditch (pH 5.1) Log	0.789	0.263**	-	-
None	R1 stream (pH 6.5) Linear*	1.850	-	0.837	0.438
None	R1 stream (pH 5.1) Log	2.396	-	1.085**	0.567**
None	R2 stream (pH 5.1) Log	~~1.813~~ 1.551	-	0.800	~~0.414~~ 0.412
None	R2 stream (pH 6.5) Log	2.479	-	1.446**	0.749**
None	R3 stream (pH 7.9)*** Linear	0.956	-	0.432	-
None	R3 stream (pH 6.5) Log	5.426	-	2.677**	1.400**
RAC (µg/L)					
0.77		**PEC/RAC ratio**			
None	D3 ditch (pH 5.1) Linear	**1.02**	0.34	-	-
None	D6 ditch (pH 7.9) Log	**1.02**	0.34	-	-
None	D6 ditch (pH 7.9) Linear	**1.02**	0.34	-	-
None	D6 ditch (pH 5.1) Log	**1.025**	0.34	-	-
None	R1 stream (pH 6.5) Linear*	**2.40**	-	**1.09**	0.57
None	R1 stream (pH 5.1) Log	**3.11**	-	**1.41**	0.74
None	R2 stream (pH 5.1) Log	~~2.35~~ **2.014**	-	**1.04**	0.54
None	R2 stream (pH 6.5) Log	**3.22**	-	**1.88**	0.97
None	R3 stream (pH 7.9) *** Linear/log	**1.24**	-	0.56	-
None	R3 stream (pH 6.5) Log	**7.05**	-	**3.48**	**1.82**

PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC ratios above the relevant trigger of 1 are shown in bold
*For R1 scenario PECsw at pH 6.5 instead of 7.9 has been chosen as worst case.
** worst case PECsw Step 4
***only for this pH PECsw is below RAC (linear and log)

Review Comments:

For R4 stream scenario 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use as the lowest PECsw value is 1.211 µg/L.

For scenarios D3 ditch, D6 ditch, R1 stream, R2 stream and R3 stream, calculated PEC/RAC ratios indicate an acceptable risk for the most sensitive group of aquatic organisms with mitigation of spray drift and run-off, therefore the risk is acceptable with the application of the following mitigation measure:

- D3 and D6 ditch scenarios: A 5m no spray buffer zone is required.
- R1 and R2 stream scenarios: A 20m no spray buffer zone and a 20m vegetative buffer strip are required.
- R3 stream scenario: A 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use for pH 5.1 and pH 6.5; only for pH 7.9 a 10m no spray buffer zone and a 10m vegetative buffer strip are required.
- For R4 stream scenario 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use.

Metabolites of Mesotrione

Table 9.5-6: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for AMBA for each organism group based on FOCUS Steps 1 and 2 calculations for the use of MIGHTY (MESOTRIONE 10% SC) in maize

Group			**Fish acute**	**Inverteb. acute**	**Algae**	**Higher plant**
Test species			***Oncorhynchus mykiss***	***Daphnia magna***	***Pseudokirchn. subcapitata***	***Lemna gibba***
Endpoint (µg/L)			LC_{50} 150000	EC_{50} 160000	E_rC_{50}/E_yC_{50} 9400	EC_{50} >90000
AF			100	100	10	10
RAC (µg/L)			1500	1600	940	9000
FOCUS Scenario	**Period / waterbody**	**PEC $_{gl\text{-}max}$ (µg/L)**				
Step 1						
		10.84*	0.007	0.007	0.012	0.001
Step 2						
SEU	Oct-Feb Mar - May	2.73*	0.002	0.002	0.003	<0.001
NEU	Oct-Feb	3.37*	0.002	0.002	0.004	<0.001

AF: Assessment factor; PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC ratios above the relevant trigger of 1 are shown in bold

*Worst case for pH's simulated.

Table 9.5-7: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for MNBA for each organism group based on FOCUS Steps 1 and 2 calculations for the use of MIGHTY (MESOTRIONE 10% SC) in maize

Group			**Fish acute**	**Inverteb. acute**	**Algae**	**Higher plant**
Test species			***Oncorhynchus mykiss***	***Daphnia magna***	***Pseudokirchn. subcapitata***	***Lemna gibba***
Endpoint (µg/L)			LC_{50} >120000	EC_{50} 130000	E_rC_{50}/E_bC_{50} 38000	EC_{50} >97000
AF			100	100	10	10
RAC (µg/L)			1200	1300	3800	9700
FOCUS Scenario	**Period / waterbody**	**PEC $_{gl-max}$ (µg/L)**				
Step 1						
		23.50	0.020	0.018	0.006	0.002
Step 2						
SEU	Mar - May	3.71*	0.003	0.003	0.001	<0.001
NEU	Oct – Feb Mar - May	4.46*	0.004	0.003	0.001	<0.001

AF: Assessment factor; PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC ratios above the relevant trigger of 1 are shown in bold
*Worst case for pH's simulated.

Table 9.5-8: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for SYN546974 for each organism group based on FOCUS Steps 1 and 2 calculations for the use of MIGHTY (MESOTRIONE 10% SC) in maize

Group			**Higher plant**
Test species			***Lemna gibba***
Endpoint (µg/L)			EC_{50} >93000
AF			10
RAC (µg/L)			9300
FOCUS Scenario	**Period / waterbody**	**PEC $_{gl-max}$ (µg/L)**	
Step 1			
		1.60	0.0002
SEU	Oct-Feb Mar – May Jun-Sep	0.39	<0.0001
NEU	Oct-Feb	0.46*	<0.0001

AF: Assessment factor; PEC: Predicted environmental concentration; RAC: Regulatory acceptable concentration; PEC/RAC

ratios above the relevant trigger of 1 are shown in bold
*Worst case for pH's simulated.

9.5.3 Overall conclusions

Most PEC/RAC values for Mesotrione are below the trigger value of 1 at step 3, indicating that Mesotrione poses a low risk to aquatic organisms. However, some PEC/RACvalues taken from the assessment of *Lemna gibba* are above the trigger value of 1 (D3, D6, R1, R2 and R3 scenarios), indicating that Mesotrione poses a potential risk to higher plant. Based on the results of the risk assessment at step 4, the following conclusions regarding buffer zones and vegetative buffer strips may be drawn for a maize use:

- D3 and D6 ditch scenarios: A 5m no spray buffer zone is required.
- R1 and R2 stream scenarios: A 20m no spray buffer zone and a 20m vegetative buffer strip are required.
- R3 stream scenario: A 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use for pH 5.1 and pH 6.5; only for pH 7.9 a 10m no spray buffer zone and a 10m vegetative buffer strip are required.
- For R4 stream scenario 20 m of no spray buffer and 20 m vegetated filter strip is not sufficed to confirmed safe use.

The risk to aquatic organisms for metabolites AMBA, MNBA and SYN546974 was assessed as low at FOCUS step 1 for the representative use on maize.

In addition, no unacceptable risk for the formulated MIGHTY (MESOTRIONE 10% SC) is expected under GAP conditions.

Spe3: *To protect aquatic organisms respect an unsprayed vegetated buffer zone of 20 m to surface water bodies.*

Review Comments:
The relevant predicted environmental concentrations in water (PEC_{sw}) for risk assessments covering the proposed use pattern are taken from Part B Section 8 (Environmental Fate). The risk assessment was based on the worst case PEC values and the results of laboratory toxicity testing.
MIGHTY pose no unacceptable risk to aquatic organisms according to the label with appropriate buffer zone with exceptions of scenario R3 and R4. The acceptability risk mitigation measures used in refined risk assessment for aquatic plants should be checked on national level.

9.6 Effects on bees (KCP 10.3.1)

9.6.1 Toxicity data

Studies on the toxicity to bees have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on bees of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

The selection of studies and endpoints for the risk assessment is in line with the results of the EU review process. Justifications are provided below.

Table 9.6-1: Endpoints and effect values relevant for the risk assessment for bees

Species	Substance	Exposure System	End point	Toxicity	Reference
Apis mellifera	Mesotrione	Acute	Oral toxicity (LD_{50})	> 11 µg a.s./bee	EFSA Journal 2016;14(3):4419
Apis mellifera	Mesotrione	Acute	Contact toxicity (LD_{50})	>100 µg a.s./bee	EFSA Journal 2016;14(3):4419
Apis mellifera	Callisto 100 SC (A12739A)	Acute	Oral toxicity (LD_{50})	79.7 µg a.s./bee (equivalent to 877.4 µg A12739A /bee)* (NOED = 25.0 µg a.s./bee)	EFSA Journal 2016;14(3):4419
Apis mellifera	Callisto 100 SC (A12739A)	Acute	Contact toxicity (LD_{50})	52.5 µg a.s./bee (equivalent to 578.2 µg A12739A /bee) (NOED = 32.4 µg a.s./bee)	EFSA Journal 2016;14(3):4419
Apis mellifera	Callisto 100 SC (A12739A)	Chronic	10 day-LD_{50}	19.2 µg a.s./bee/day** (LD_{20} = 11.8 µg a.s./bee/day) (LD_{10} = 9.2 µg a.s./bee/day) (NOED = 8.1 µg a.s./bee/day)	EFSA Journal 2016;14(3):4419
Apis mellifera	Callisto 100 SC (A12739A)	Bee brood development (7 day study; with dietary exposure on days 3, 4, 5 and 6)	NOEDlarvae	57.8 µg a.s./larva (LD_{50} = 118.5 µg a.s./larva) (LD_{20} = 39.185 µg a.s./larva) (LD_{10} = 18.836 µg a.s./larva)	EFSA Journal 2016;14(3):4419
Apis mellifera	Mesotrione 10% SC	Acute	Oral toxicity (LD_{50})	>200 µg/honeybee (equivalent to 20.4 µg a.s./bee)	KCP 10.3.1.1.1 Pawel Parma, 2015 B/132/15
Apis mellifera	Mesotrione 10% SC	Acute	Contact toxicity (LD_{50})	>200 µg/honeybee (equivalent to 20.4 µg a.s./bee)	KCP10.3.1.1.2 Paweł Parma, 2015 B/133/15
Apis mellifera	Mesotrione 10% SC	Chronic, 10d	LDD50 NOEDD	>57.21 µg a.s./bee/day ≥57.21 µg a.s./bee/day	KCP10.3.1.2 Gimeno, I., 2019, TRC17-006BA**

Species	Substance	Exposure System	End point	Toxicity	Reference
Apis mellifera	Mesotrione 10% SC	Larval, repeated exposure, 22 d	NOED ED50	1.8 µg as/larva 14.3 µg as/larva	KCP 10.3.1.3 Scheller, K., 2018 17 48 BLC 0088**
Higher-tier studies (tunnel test, field studies)					
Not needed					

*As uncertainties were noted around this endpoint (poor fitting of the oral 48-hour mortality data curve).
**Study summaries are included in the dossier (see appendix 2). However, they have not been considered furthers as they are not relevant to the currently approved risk assessment scheme (SANCO/10329/2002).

9.6.1.1 Justification for new endpoints

The used endpoints were the EU agreed ones. Additionally, new acute oral and contact studies were conducted with the formulation MIGHTY (MESOTRIONE 10% SC), and the endpoints were considered also for the risk assessment.

9.6.2 Risk assessment

The evaluation of the risk for bees was performed in accordance with the recommendations of the "Guidance Document on Terrestrial Ecotoxicology", as provided by the Commission Services (SANCO/10329/2002 rev.2 (final), October 17, 2002).

9.6.2.1 Hazard quotients for bees

Table 9.6-2: First-tier assessment of the risk for bees due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use **Active substance** **Application rate (g a.s./ha)**	Maize Mesotrione 1 x 150		
Test design	**LD_{50} (lab.) (µg/bee)**	**Single application rate (g a.s./ha)**	**Q_{HO}, Q_{HC} criterion: $Q_H \leq 50$**
Oral toxicity	>11	150	<14
Contact toxicity	>100		<1.50
Product **Application rate (g/ha)**	Mesotrione 10% SC 1 × 1564.5		
Test design	**LD_{50} (lab.) (µg/bee)**	**Single application rate (g/ha)**	**Q_{HO}, Q_{HC} criterion: $Q_H \leq 50$**
Oral toxicity	>200	1564.5	7.82
Contact toxicity	>200		7.82

Q_{HO}, Q_{HC}: Hazard quotients for oral and contact exposure. Q_H values shown in bold breach the relevant trigger.

9.6.2.2 Higher-tier risk assessment for bees (tunnel test, field studies)

Not relevant since a low risk for bees was concluded in the first-tier assessment above.

9.6.3 Effects on bumble bees

Not relevant.

9.6.4 Effects on solitary bees

Not relevant.

9.6.5 Overall conclusions

A low acute contact and oral risk was concluded on the basis of the first-tier assessment described above. Therefore, Mesotrione do not present a risk for bees.

Review Comments:

The evaluation of the risk for bees was performed in accordance with the recommendations of the "Guidance Document on Terrestrial Ecotoxicology", as provided by the Commission Services (SANCO/10329/2002 rev.2 (final), October 17, 2002).

The submitted risk assessment, based on laboratory studies, has been accepted. It can therefore be concluded that there will be negligible risk associated with the exposure of bees to MIGHTY.

The data requirements in accordance with Commission Regulation (EU) No 284/2013 for the chronic toxicity to adult honeybees and honeybee larvae are fulfilled.

There is not harmonized approach for the chronic risk assessment for bees, therefore, Concerned Member States must decide on the requirements in this regard at national level.

9.7 Effects on arthropods other than bees (KCP 10.3.2)

9.7.1 Toxicity data

Studies on the toxicity to non-target arthropods have been carried out with the EU representative formulation. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on non-target arthropods of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

The selection of studies and endpoints for the risk assessment is in line with the results of the EU review process. Justifications are provided below.

Table 9.7-1: Endpoints and effect values relevant for the risk assessment for non-target arthropods

Species	Substance	Exposure System	Results*	Reference
Typhlodromus pyri (protonymphus)	Callisto 100 SC (A12739A)	Laboratory test	LR_{50} = 93.11 g a.s./ha ER_{50} > 81 g a.s./ha	EFSA Journal 2016;14(3):4419
Aphidius rhopalosiphi (adults)	Callisto 100 SC (A12739A)	Laboratory test	LR_{50} = 43.56 g a.s./ha ER_{50} > 25.6 g a.s./ha	EFSA Journal 2016;14(3):4419
Typhlodromus pyri (protonymphus)	Callisto 100 SC (A12739A)	Extended Laboratory, aged residue, 14 d, Leaf discs	Corrected mortality at 7DAT: 11% at 9.4 g a.s./ha 25% at 18.8 g a.s./ha 12% at 37.5 g a.s./ha 12% at 75 g a.s./ha 19% at 150 g a.s./ha 42% at 300 g a.s./ha LR_{50}>300 g a.s./ha Effect of reproduction: 20.7% at 9.4 g a.s./ha 36.2% at 18.8 g a.s./ha 19% at 37.5 g a.s./ha 41.6% at 75 g a.s./ha 47.4% at 150 g a.s./ha 56.9% at 300 g a.s./ha ER_{50}>150 g a.s./ha	EFSA Journal 2016;14(3):4419
Aphidius rhopalosiphi (adults)	Callisto 100 SC (A12739A)	Extended Laboratory, aged residue, 48 h, Barley seedlings	Corrected mortality: 0% at 44.4 g a.s./ha 0% at 66.7 g a.s./ha 0% at 100 g a.s./ha 3.3% at 150 g a.s./ha 3.3% at 225 g a.s./ha LR_{50}>225 g a.s./ha Effect of reproduction: n.d. at 44.4 g a.s./ha n.d. at 66.7 g a.s./ha -9.6% at 100 g a.s./ha -3.5% at 150 g a.s./ha -8.0% at 225 g a.s./ha ER_{50}>225 g a.s./ha	EFSA Journal 2016;14(3):4419

Species	Substance	Exposure System	Results*	Reference
Aleochara bilineata	Callisto 100 SC (A12739A)	Extended Laboratory, soil, 67 d	Corrected mortality: 5% at 12.5 g a.s./ha 20% at 25.0 g a.s./ha 7% at 50.0 g a.s./ha 4% at 100 g a.s./ha 4% at 200 g a.s./ha LR_{50}>200 g a.s./ha Effect of reproduction: 6.5% at 12.5 g a.s./ha 24.5% at 25.0 g a.s./ha 11.7% at 50.0 g a.s./ha 7.4% at 100 g a.s./ha 10.1% at 200 g a.s./ha ER_{50}>200 g a.s./ha	EFSA Journal 2016;14(3):4419
Pardosa sp.	Callisto 100 SC (A12739A)	Extended Laboratory, soil, 21 d	Corrected mortality at 21DAT: 9% at 4.7 g a.s./ha 3% at 9.4 g a.s./ha 3% at 19.0 g a.s./ha 9% at 37.5 g a.s./ha 26% at 75.0 g a.s./ha 41% at 150 g a.s./ha LR_{50}>150 g a.s./ha Effect of reproduction: -2.2% at 4.7 g a.s./ha -2.2% at 9.4 g a.s./ha -2.2% at 19.0 g a.s./ha -2.2% at 37.5 g a.s./ha 6.7% at 75.0 g a.s./ha 13.3% at 150 g a.s./ha ER_{50}>150 g a.s./ha	EFSA Journal 2016;14(3):4419
Aphidius rhopalosiphi (adults)	Mesotrione 10% SC	Extended laboratory	LR_{50} > 1.5 kg f.p./ha (153 g a.s./ha) ER_{50} > 1.5 kg f.p./ha (153 g a.s./ha)	KCP 10.3.2.2-01 Paweł Parma, 2016 B/131/15
Typhlodromus pyri	Mesotrione 10% SC	Extended Laboratory, Bean leaves (2D)	LR_{50} > 2.1 kg f.p./ha (214.2 g a.s./ha) ER_{50} < 0.375 kg f.p./ha (<38.3 g a.s./ha)	KCP 10.3.2.2-02 Parma, P. 2017 B/130/15

Species	Substance	Exposure System	Results*	Reference
Typhlodromus pyri	Mesotrione 10% SC	Aged residue study, maize leaves	Corrected mortality at 150 g a.s./ha: 4.82% at 0DAA -1.16*% at 7DAA 1.16% at 14DAA Reduction of reproduction at 150 g a.s./ha: 9.30% at 0DAA -7.20*% at 7DAA 7.93% at 14DAA	KCP 10.3.2.2-03 Luna, F. 2017 TRC17-090BA
Aleochara bilineata	Mesotrione 10% SC	Extended study, natural soil (LUFA 2.1)	LR_{50} > 6 L f.p./ha (>624 g a.s./ha) ER_{50} > 6 L f.p./ha (>624 g a.s./ha)	KCP 10.3.2.2-04 Soler, E. 2017 TRC17-115BA
Field or semi-field tests				
None				

*Negative value indicates an increase compared to control

9.7.1.1 Justification for new endpoints

Studies were conducted with the formulation MIGHTY (MESOTRIONE 10% SC), and the endpoints were considered also for the risk assessment.

9.7.2 Risk assessment

The evaluation of the risk for non-target arthropods was performed in accordance with the recommendations of the “Guidance Document on Terrestrial Ecotoxicology”, as provided by the Commission Services (SANCO/10329/2002 rev.2 (final), October 17, 2002), and in consideration of the recommendations of the guidance document ESCORT 2.

9.7.2.1 Risk assessment for in-field exposure

Table 9.7-2: First- and higher-tier assessment of the in-field risk for non-target arthropods due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize		
Active substance/product	Mesotrione/Mesotrione 10% SC		
Application rate (g a.s./ha)	1 x 150		
MAF	1		
Test species Higher-tier	**Rate with ≤ 50 % effect* (g a.s./ha)**	**$PER_{in-field}$ (g a.s./ha)**	**$PER_{in-field}$ below rate with ≤ 50 % effect?**
Aphidius rhopalosiphi	>153	150	yes
Typhlodromus pyri	<38.3 (reproduction)	150	**no**
	214.2 (mortality)	150	**yes**
Aleochara bilineata	>624	150	yes
Test species Higher-tier	**Rate with ≤ 50 % effect (g a.s./ha) at 0 DALT**	**$PER_{in-field}$ (g a.s./ha)**	**$PER_{in-field}$ below rate with ≤ 50 % effect?**
Typhlodromus pyri	150	150	yes

MAF: Multiple application factor; PER: Predicted environmental rate; HQ: Hazard quotient; DALT: Days after last treatment.
Criteria values shown in bold breach the relevant trigger.

* If an LR_{50} or ER_{50} from a relevant extended laboratory test is available, it should be considered in place of the rate with ≤ 50 % effect.

An extended laboratory on *A. rhopalosiphi* with MIGHTY (MESOTRIONE 10% SC) was used in higher-tier assessment, when at the rate of 150 g a.s./ha had no adverse effect on morality and fecundity. Extended study on *Typhodromus pyri* with MIGHTY (MESOTRIONE 10% SC) was performed by the Applicant where the rate with > 50 % effect on reproduction was 38.3 g a.s./ha, below the maximum $PER_{in-field}$ according to the proposed GAP. An aged residue study on this species was performed by the Applicant showing no in-field risk at rate of 150 g a.s./ha at 0 days after treatment. Therefore, potential in-field recovery was proved by the Applicant.

Extended study on *Aleochara bilineata* with Mesotrione 10%SC was performed since in-field risk was obtained on *T.Pyri*. According to results, the rate with ≤ 50 % effect on mortality or reproduction was 624 g a.s./ha, above the maximum $PER_{in-field}$ of 150 g a.s./ha. Therefore, no unacceptable in-field risk was obtained for this species.

9.7.2.2 Risk assessment for off-field exposure

Table 9.7-3: First- and higher-tier assessment of the off-field risk for non-target arthropods due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize				
Active substance/product	Mesotrione / Mesotrione 10% SC				
Application rate (g a.s./ha)	1 x 150				
MAF	1				
vdf	10 (Tier 1) / 1 (Higher-tier)				
Test species Higher-tier	**LR_{50} and ER_{50} (extended lab.) (g a.s./ha)***	**Drift rate**	**$PER_{off-field}$ (g a.s./ha)**	**CF**	**corr. $PER_{off-field}$ below rate with ≤ 50 % effect?**

Aphidius rhopalosiphi	>150	2.77	4.155	5	yes
Typhlodromus pyri	<38.3 (reproduction)		0.4155	5	yes
	214.2 (mortality)		0.4155	5	yes
Aleochara bilineata	>624		0.4155	5	yes

MAF: Multiple application factor; vdf: Vegetation distribution factor; (corr.) PER: (corrected) Predicted environmental rate; CF: Correction factor; HQ: Hazard quotient. Criteria values shown in bold breach the relevant trigger.

* If an LR_{50} or ER_{50} from a relevant extended laboratory test is available, it should be considered in place of the rate with ≤ 50 % effect.

According to the risk assessment above, no unacceptable off-field risk is expected for species *A. rhopalosiphi* and *A. bilineata*. Off field risk is not expected on *T. pyri* too although effects on reproduction were obtained below 38.3 g a.s./ha. ~~Nevertheless, the Applicant considers that it is an overestimation since the maximum corrected $PER_{off\text{-}field}$ of 2.08 g a.s./ha is well below of value of ER_{50} of 38.3 g a.s./ha. In addition,~~ the Applicant wishes to refer to the new aged residue study with formulation submitted (please, refer to KCP 10.3.2.2-.3), where only 4.82% and 9.30% of effects on mortality and reproduction respectively were found at 150 g a.s./ha in fresh residue (at 0DAA).

In addition, considering the results from the laboratory test on *T. pyri* with the formulation of Callisto 100 SC (A12739A) from Monograph, an HQ well below the trigger of 2 was obtained (HQ of 0.04) according to the proposed GAP, therefore, an acceptable off-field risk was obtained.

~~Moreover, according to Monograph, *T. pyri* is not considered a relevant species in maize. Therefore, the Applicant consider no unacceptable risk for this species will be expected at proposed doses.~~

9.7.2.3 Additional higher-tier risk assessment

Not relevant.

9.7.2.4 Risk mitigation measures

No risk mitigation needed.

9.7.3 Overall conclusions

No in-field and off-field risk to non-target arthropods is expected after the application of MIGHTY (MESOTRIONE 10% SC) according to the proposed GAP.

Review Comments:

Based on the Tier II data for *Aphidius rhopalosiphi*, *Typhlodromus pyri*, *Aleochara bilineata* and aged residue for *Typhlodromus pyri*, the in-field risk to terrestrial arthropods other than bees following treatment with MIGHTY is low and acceptable for the intended worst-case use.

Based on the Tier II data for *Aphidius rhopalosiphi and Aleochara bilineata*, with effects below the triggers, the off-field risk is low. Nevertheless, for *Typhlodromus pyri* effects on reproduction were obtained below 38.3 g a.s./ha (lowest dose tested in extended laboratory study). Thus, no reproduction endpoint is available. In opinion of zRMS, in this case, based on the aged residue study, where only 4.82% and 9.30% of effects on mortality and reproduction respectively were found at 150 g a.s./ha in fresh residue (at 0DAA) the acceptable off-field risk can be identified. Nevertheless, as this is not in accordance with ESCORT II, the decision whether the evaluation of the risk is sufficient should be taken on the national level.

9.8 Effects on non-target soil meso- and macrofauna (KCP 10.4)

9.8.1 Toxicity data

Studies on the toxicity to earthworms and other non-target soil organisms (meso- and macrofauna) have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on earthworms and other non-target soil organisms (meso- and macrofauna) of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

Table 9.8-1: Endpoints and effect values relevant for the risk assessment for earthworms and other non-target soil organisms (meso- and macrofauna)

Species	Substance	Exposure System	Results	Reference
~~*Eisenia fetida*~~	~~Mesotrione~~	~~Acute, Mixed with soil, 10% peat~~	~~LC_{50} > 2000 mg a.s./kg d.w. soil~~	~~EFSA Journal 2016;14(3):4419~~
~~*Eisenia fetida*~~	~~MNBA~~	~~-~~	~~LC_{50} > 1000 mg a.s./kg d.w. soil~~	~~EFSA Journal 2016;14(3):4419~~
Eisenia fetida	Callisto 100 SC (A12739A)	Chronic, Mixed with soil, 10% peat	NOEC = 125 mg A12739A /kg d.w. soil (equivalent to 10.85 mg a.s./kg d.w. soil) EC_{10}, number of juveniles = 68.1 mg A12739A /kg d.w. soil (equivalent to 5.91 mg a.s./kg d.w. soil) EC_{20}, number of juveniles = 174.9 mg A12739A /kg d.w. soil (equivalent to 15.18 mg a.s./kg d.w. soil)	EFSA Journal 2016;14(3):4419
Eisenia fetida	AMBA	Chronic, mixed with soil using quartz sand. 5% peat	NOEC = 1050 mg /kg d.w. soil EC_{10} = 1050 mg /kg d.w. soil EC_{20} = 1050 mg /kg d.w. soil	EFSA Journal 2016;14(3):4419
Eisenia fetida	MNBA	Chronic, mixed with soil using quartz sand. 5% peat	NOEC = 1050 mg /kg d.w. soil EC_{10} > 1050 mg /kg d.w. soil EC_{20} > 1050 mg /kg d.w. soil	EFSA Journal 2016;14(3):4419

Species	Substance	Exposure System	Results	Reference
Folsomia candida	Callisto 100 SC (A12739A)	Mixed into substrate 28 d, chronic 5 % peat content	NOEC = 50.54 mg a.s. /kg d.w. soil (equivalent to 556 mg A12739A /kg d.w. soil) EC_{10} = 413 mg A12739A /kg d.w. soil EC_{20} = 620 mg A12739A /kg d.w. soil	EFSA Journal 2016;14(3):4419
Hypoaspis aculeifer	Callisto 100 SC (A12739A)	Mixed into substrate 14 d, chronic 5 % peat content	NOEC = 90.9 mg a.s. /kg d.w. soil (equivalent to 1000 mg A12739A /kg d.w. soil) EC_{10} > 1000 mg A12739A /kg d.w. soil EC_{20} > 1000 mg A12739A /kg d.w. soil	EFSA Journal 2016;14(3):4419
Eisenia fetida	Mesotrione 10% SC	Mixed into substrate 56 d, chronic 10 % peat content	**NOEC= 18 mg f.p./ kg d.w. soil** EC_{10} = 26.7 mg f.p./ kg d.w. soil	KCP 10.4.1.1 Weronika, Dec. 2016 G/243/15
Folsomia candida	Mesotrione 10% SC	Mixed into substrate 28 d, chronic 5 % peat content	**NOEC = 320 mg f.p./kg dw** EC_{10} = 367.8 mg f.p./ kg d.w. soil	KCP 10.4.2.1 Weronika Dec, 2016 G/244/15
Field studies				
None				
Litter bag test				
None				

* Corrected value derived by dividing the endpoint by a factor of 2 in accordance with the EPPO earthworm scheme 2002.

9.8.1.1 Justification for new endpoints

The used endpoints are the EU agreed ones, except for formulation, corresponding to data proper to MIGHTY (MESOTRIONE 10% SC) formulation.

9.8.2 Risk assessment

The evaluation of the risk for earthworms and other non-target soil organisms (meso- and macrofauna) was performed in accordance with the recommendations of the “Guidance Document on Terrestrial Ecotoxicology”, as provided by the Commission Services (SANCO/10329/2002 rev 2 (final), October 17, 2002).

9.8.2.1 First-tier risk assessment

The relevant PEC_{soil} for risk assessments covering the proposed use pattern are taken from Section 8 (Environmental Fate), Chapter 8.7.2, Table 8.7-3. According to the assessment of environmental-fate data, multi-annual accumulation in soil does not need to be considered for Mesotrione.

Table 9.8-2: First-tier assessment of the acute and chronic risk for earthworms and other non-target soil organisms (meso- and macrofauna) due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize		
~~**Acute effects on earthworms**~~			
~~**Product/active substance/metabolite**~~	~~**LC_{50} (mg/kg dw)**~~	~~**PEC_{soil} (mg/kg dw)**~~	~~**TER_a (criterion TER ≥ 10)**~~
~~Mesotrione~~	~~>2000~~	~~0.150~~	~~>13333.33~~
~~MNBA~~	~~>1000~~	~~0.062~~	~~>16129.03~~
Chronic effects on earthworms			
Product/active substance/metabolite	**NOEC (mg/kg dw)**	**PEC_{soil} (mg/kg dw)**	**TER_{lt} (criterion TER ≥ 5)**
Mesotrione	10.85	0.150	72.33
AMBA	1050	0.009	116666.67
MNBA	1050	0.062	16935.48
Mesotrione 10% SC	18	1.5645	11.51
Chronic effects on other soil macro- and mesofauna			
Product/active substance/metabolite	**NOEC or EC_{10} (mg/kg dw)**	**PEC_{soil} (mg/kg dw)**	**TER_{lt} (criterion TER ≥ 5)**
Folsomia candida			
Mesotrione	50.54	0.150	336.93
Mesotrione 10% SC	320	1.5645	204.54
Hypoaspis aculeifer			
Mesotrione	90.9	0.150	606.00

TER values shown in bold fall below the relevant trigger.

Chronic studies with MIGHTY (MESOTRIONE 10% SC) on earthworms and collembolan were submitted by the Applicant and no unacceptable risk was obtained after the risk assessment. In addition, according to data from Monograph no unacceptable chronic risk was obtained on *Hypoaspis aculeifer* for the technical. Moreover, the risk assessment for NTA was acceptable with endpoints for all tested species including the ground dwelling arthropod *Aleochara bilineata*. Therefore, the Applicant considers that an acceptable risk to *Hypoaspis aculeifer* for formulation MIGHTY (MESOTRIONE 10% SC) can be concluded on the basis that low risks to earthworms and other soil macro-organisms, and ground dwelling arthropod with formulation were concluded.

Therefore, it is expected that chronic toxicity on *Hypoaspis* will result from prolonged exposure and the formulation is not expected to remain intact in the environment.

9.8.2.2 Higher-tier risk assessment

Not relevant.

9.8.3 Overall conclusions

No unacceptable risk for meso- and macrofauna was reached at GAP application rates.

Review Comments:

The long-term risks of MIGHTY to soil meso- and macro-organisms were assessed from toxicity exposure ratios between toxicity endpoints and maximum PEC_{soil}. The relevant predicted environmental concentrations in soil (PEC_{soil}) for risk assessments covering the proposed use pattern are taken from Part B Section 8 (Environmental Fate).

Safe use of MIGHTY in maize were confirmed based on TER_{LT} calculations for active substances, their metabolites and for formulation.

9.9 Effects on soil microbial activity (KCP 10.5)

9.9.1 Toxicity data

Studies on effects soil microorganisms have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR.

Effects on soil microorganisms of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

Table 9.9-1: Endpoints and effect values relevant for the risk assessment for soil microorganisms

Endpoint	Substance	Exposure System	Results	Reference
N-mineralisation	Callisto 100 SC (A12739A)	28 d, aerobic soil type	7.8% effect at day 28 at 0.53 mg a.s./kg d.w. soil (equivalent to 5.84 mg A12739A/kg d.w. soil)	EFSA Journal 2016;14(3):4419
~~C-mineralisation~~	~~Callisto 100 SC (A12739A)~~	~~28 d, aerobic soil type~~	~~8.73% effect at day 28 at 2.18 mg a.s./kg d.w. soil (equivalent to 5.84 mg A12739A/kg d.w. soil)~~	~~RAR of Mesotrione~~
N-mineralisation	AMBA	28 d, aerobic soil type	-7.6% effect at day 28 at 1.13 mg /kg d.w. soil	EFSA Journal 2016;14(3):4419
~~C-mineralisation~~	~~AMBA~~	~~28 d, aerobic soil type~~	~~-4.8% effect at day 28 at 1.13 mg /kg d.w. soil~~	~~RAR of Mesotrione~~
N-mineralisation	MNBA	28 d, aerobic soil type	-4.8% effect at day 28 at 1.13 mg /kg d.w. soil	EFSA Journal 2016;14(3):4419

Endpoint	Substance	Exposure System	Results	Reference
~~C-mineralisation~~	~~MNBA~~	~~28 d, aerobic soil type~~	~~2.8% effect at day 28 at 1.13 mg /kg d.w. soil~~	~~RAR of Mesotrione~~
N-mineralisation	Mesotrione 10% SC	28 d, aerobic soil type	10.9% effect at 11 mg of the test item/kg of soil (equivalent to 1.0 mg a.s./kg of soil)	KCP 10.5-01 Weronika Dec, 08/2016, Report G/242/15
~~C-mineralisation~~	~~Mesotrione 10% SC~~	~~28 d, aerobic soil type~~	~~11.6% effect at 11 mg of the test item/kg of soil (equivalent to 1.0 mg a.s./kg of soil)~~	~~KCP 10.5-02 Weronika Dec, 08/2016, Report G/241/15~~

zRMS comments:

Since C-mineralization is no longer data requirement all studies were crossed out and thus not taken to consideration in risk assessment.

9.9.1.1 Justification for new endpoints

The used endpoints are the EU agreed ones, except for formulation, corresponding to data proper to MIGHTY (MESOTRIONE 10% SC) formulation.

9.9.2 Risk assessment

The evaluation of the risk for soil microorganisms was performed in accordance with the recommendations of the "Guidance Document on Terrestrial Ecotoxicology", as provided by the Commission Services (SANCO/10329/2002 rev 2 (final), October 17, 2002).

The relevant PEC_{soil} for risk assessments covering the proposed use pattern are taken from Section 8 (Environmental Fate), Chapter 8.7.2, Table 8.7-3 and were already used in the risk assessment for earthworms and other non-target soil organisms (meso- and macrofauna) (see 9.8).

Table 9.9-2: Assessment of the risk for effects on soil micro-organisms due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize		
N-mineralisation			
Product/active substance	**Max. conc. with effects ≤ 25 % (mg/kg dw)**	**PEC_{soil} (mg/kg dw)**	**Risk acceptable?**
Mesotrione	0.533 mg/kg soil (at 28 d)	0.150	yes
AMBA	1.13 mg/kg soil (at 28 d)	0.009	yes
MNBA	1.13 mg/kg soil (at 28 d)	0.062	yes
Mesotrione 10% SC	11 mg of the test item/kg of soil (at 28 d)	1.5645	yes
~~**C-mineralisation**~~			

Product/active substance	Max. conc. with effects ≤ 25 % (mg/kg dw)	PEC_{soil} (mg/kg dw)	Risk acceptable?
Mesotrione	2.18 mg/kg soil (at 28 d)	0.15	yes
AMBA	1.13 mg/kg soil (at 28 d)	0.009	yes
MNBA	1.13 mg/kg soil (at 28 d)	0.062	yes
Mesotrione 10% SC	11 mg of the test item/kg of soil (at 28 d)	1.5645	yes

9.9.3 Overall conclusions

On the basis of the results of carbon and nitrogen transformation tests, it was concluded that MIGHTY (MESOTRIONE 10% SC) did not have any long-term adverse effect on the process of carbon and nitrogen transformation in aerobic surface soils.

Review Comments:

MIGHTY had no significant effect on soil micro-organisms at 11 mg product/kg dry soil. This is approximately 7 times higher than the maximum PEC_{soil} of 1.5645 mg product/kg dry soil following the worst-case application to maize. This supports the conclusion that MIGHTY under field conditions, use of at the proposed rates poses no unacceptable risk to non-target soil micro-organisms.

9.10 Effects on non-target terrestrial plants (KCP 10.6)

9.10.1 Toxicity data

Studies on the toxicity to non-target terrestrial plants have been carried out with Mesotrione and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on non-target terrestrial plants of MIGHTY (MESOTRIONE 10% SC) were not evaluated as part of the EU assessment of Mesotrione. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

Table 9.10-1: Endpoints and effect values relevant for the risk assessment for non-target terrestrial plants

Species	Substance	Exposure System	Results	Reference
Allium cepa[1)] *Avena sativa*[2)] *Lolium perenne*[3)] *Brassica oleracea*[4)] *Brassica rapa*[5)] *Cucumis sativa*[6)] *Glycine max*[7)] *Lactuca sativa*[8)] *Linum usitatissimum*[9)] *Lycopersicon esculentum*[10)]	Callisto 100 SC (A12739A)	Seedling emergence	[1)]$ER_{50\ Biomass}$ = 33.2 g a.s./ha* [2)]$ER_{50\ Biomass}$ > 150 g a.s./ha [3)]$ER_{50\ Biomass}$ > 125 g a.s./ha* [4)]$ER_{50\ Biomass}$ = 19.8 g a.s./ha [5)]$ER_{50\ Biomass}$ = 20.6 g a.s./ha [6)]$ER_{50\ Biomass}$ > 150 g a.s./ha [7)]$ER_{50\ Biomass}$ > 150 g a.s./ha [8)]$ER_{50\ Biomass}$ = 13.8 g a.s./ha* [9)]$ER_{50\ Biomass}$ > 125 g a.s./ha* [10)]$ER_{50\ Biomass}$ = 19.7 g a.s./ha*	EFSA Journal 2016;14(3):4419
Allium cepa[1)] *Avena sativa*[2)] *Lolium perenne*[3)] *Brassica oleracea*[4)] *Brassica rapa*[5)] *Cucumis sativa*[6)] *Glycine max*[7)] *Lactuca sativa*[8)] *Linum usitatissimum*[9)] *Lycopersicon esculentum*[10)]	Callisto 100 SC (A12739A)	Vegetative vigour	[1)]$ER_{50\ Biomass}$ = 8.93 g a.s./ha [2)]$ER_{50\ Biomass}$ > 500 g a.s./ha [3)]$ER_{50\ Biomass}$ > 500 g a.s./ha [4)]$ER_{50\ Biomass}$ = 6.18 g a.s./ha [5)]$ER_{50\ Biomass}$ = 2.27 g a.s./ha [6)]$ER_{50\ Biomass}$ =1.53 g a.s./ha [7)]$ER_{50\ Biomass}$ = 6.70 g a.s./ha* [8)]$ER_{50\ Biomass}$ 0.883 g a.s./ha [9)]$ER_{50\ Biomass}$ = 264 g a.s./ha [10)]$ER_{50\ Biomass}$ = 1.50 g a.s./ha	EFSA Journal 2016;14(3):4419
Pisum sativum[1)], *Helianthus annuus*[2)], *Sinapis alba*[3)], *Solanum lycopersicon*[4)], *Zea mays*[5)], *Avena sativa*[6)]	Mesotrione 10% SC	Seedling emergence	[1)] ER_{50} plant weight = 41 g a.s./ha [2)] ER_{50} plant weight = 48.4 g a.s./ha [3)] ER_{50} plant weight = 13.8 g a.s./ha **[4)] ER_{50} plant weight = 10.9 g a.s./ha** [5)] ER_{50} plant weight = 140.4 g a.s./ha [6)] ER_{50} plant weight = 13.8 g a.s./ha	KCP 10.6.2-01 Weronika, D. 2017 G/246/15

Species	Substance	Exposure System	Results	Reference
Lactuca sativa[1)d], *Phaseolus vulgaris*[2)d], *Dacus carota*[3d)], *Beta vulgaris*[4)d], *Brassica campestris var. chinensis*[5)d], *Linum usitatissimum*[6)d] *Alluim cepa*[7)m] *Triticum aestivum*[8)m]	Mesotrione 10% SC	Vegetative vigour	[1)] ER_{50} dry shoot weight = 0.21 L f.p./ha [2)] ER_{50} dry shoot weight = 0.24 L f.p./ha [3)] ER_{50} dry shoot weight = 0.12 L f.p./ha [4)] ER_{50} shoot height = 0.22 L f.p./ha [5)] ER_{50} shoot height = 0.29 L f.p./ha **[6)] ER_{50} dry shoot weight = 0.09 L f.p./ha** [7)] ER_{50} shoot height = 0.22 L f.p./ha [8)] ER_{50} dry shoot weight = 0.24 L f.p./ha	KCP 10.6.2-02 Sadananda, T. S., 2019 BIO-ETX 034

m: monocotyledonous; d: dicotyledonous
*Modified to account for accuracy of spray concentrations

9.10.1.1 Justification for new endpoints

The used endpoints are the EU agreed ones, except for formulation, corresponding to data proper to MIGHTY (MESOTRIONE 10% SC) formulation.

9.10.2 Risk assessment

9.10.2.1 Tier-1 risk assessment (based screening data)

Not relevant.

9.10.2.2 Tier-2 risk assessment (based on dose-response data)

The risk assessment is based on the “Guidance Document on Terrestrial Ecotoxicology”, (SANCO/10329/2002 rev.2 final, 2002). It is restricted to off-field situations, as non-target plants are non-crop plants located outside the treated area.

Table 9.10-2: Assessment of the risk for non-target plants due to the use of MIGHTY (MESOTRIONE 10% SC) in maize

Intended use	Maize
Active substance/product	Mesotrione / Mesotrione 10% SC
Application rate (g/ha)	1 × 150* or 100 g a.s./ha ~~1 × 1 L fp./ha~~
MAF	1

Test species	**ER_{50} (g/ha)**	**Drift rate**	**$PER_{off\text{-}field}$ (g/ha)**	**TER criterion: TER ≥ 5**
~~*Lactuca sativa*~~ *Solanum lycopersicon* (tomato)	10.9 g a.s./ha (seedling emergence)	2.77	4.16* 2.77 g a.s./ha	**2.62* 3.94**
~~*Lactuca sativa* (lettuce)~~	~~0.883 (vegetative vigour)~~	~~2.77~~	~~4.16~~	~~**0.21**~~
~~*Linum usitatissimum* (flax)~~	~~0.09 L fp./ha (vegetative vigour)~~	~~2.77~~	~~0.03 L fp./ha~~	~~3.25~~
Linum usitatissimum (flax)	9 g a.s./ha (vegetative vigour)	2.77	4.16* 2.77 g a.s./ha	**2.16* 3.25**

MAF: Multiple application factor; PER: Predicted environmental rate; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

9.10.2.3 Higher-tier risk assessment

Not relevant.

9.10.2.4 Risk mitigation measures

In order to reduce the off-field exposure, risk mitigation measures can be implemented. These correspond to unsprayed in-field buffer strips of a given width and/or the usage of drift reducing nozzles. The results of the risk assessment using typical mitigation measures (no-spray buffer zones of 5 or 10 m; drift-reducing nozzles with reduction by 50 %, 75 %, or 90 %) are summarised in the following table.

Table 9.10-3: Risk assessment for non-target terrestrial plants due to the use of MIGHTY (MESOTRIONE 10% SC) in maize considering risk mitigation (in-field no-spray buffer zones, and drift-reducing nozzles)

Intended use **Active substance/product** **Application rate (g/ha)** **MAF**		Maize Mesotrione / Mesotrione 10% SC 1 × 150 1			
Buffer strip (m)	**Drift rate (%)**	**$PER_{off-field}$ (g/ha)**	**$PER_{off-field}$ 50 % drift red. (g/ha)**	**$PER_{off-field}$ 75 % drift red. (g/ha)**	**$PER_{off-field}$ 90 % drift red. (g/ha)**
1	2.77	4.16	2.08	1.04	0.42
5	0.57	0.86	0.43	0.21	0.09
10	0.29	0.44	0.22	0.11	-
15	0.20	0.30	0.15	-	-
30	0.10	0.15	-	-	-
~~**Toxicity value**~~ ~~ER_{50} = 0.883 g a.s./ha~~		~~**TER**~~ ~~**criterion: TER ≥ 5**~~			
~~1~~		~~**0.21**~~	~~**0.43**~~	~~**0.85**~~	~~**2.13**~~
~~5~~		~~**1.03**~~	~~**2.07**~~	~~**4.13**~~	~~10.33~~
~~10~~		~~**2.03**~~	~~**4.06**~~	~~8.12~~	~~-~~
~~15~~		~~**2.94**~~	~~5.89~~	~~-~~	~~-~~
~~30~~		~~5.89~~	~~-~~	~~-~~	~~-~~
Toxicity value ER_{50} = 9 g a.s./ha		TER criterion: TER ≥ 5			
1		**2.16**	**4.32**	8.65	-
5		10.11	-	-	-

Intended use **Product** **Application rate (L f.p./ha)** **MAF**		Maize Mesotrione 10% SC 1 × 1 L/ha 1			
Buffer strip (m)	**Drift rate (%)**	**$PER_{off-field}$ (L/ha)**	**$PER_{off-field}$ 50 % drift red. (L/ha)**	**$PER_{off-field}$ 75 % drift red. (L/ha)**	**$PER_{off-field}$ 90 % drift red. (L/ha)**
1	2.77	0.028	0.014	0.007	0.003
5	0.57	0.006	0.003	0.001	0.001
Toxicity value ER_{50} = 0.09 L f.p./ha		**TER** **criterion: TER ≥ 5**			
1		**3.25**	6.50	13.00	32.49
5		15.79	31.58	63.16	157.89

MAF: Multiple application factor; PER: Predicted environmental rates; TER: toxicity to exposure ratio. Criteria values shown in bold breach the relevant trigger.

9.10.3 Overall conclusions

The risk assessment has been realized according to the seedling emergence and vegetative vigour data.

The calculated TER values are below the Annex VI trigger of 5 for seedling emergence and vegetative vigour when a distance of 1 m is considered. Therefore, no potential risk to non-target plants located outside the treated area after application of MIGHTY (MESOTRIONE 10% SC) according to the GAP table is expected when risk mitigation measures are considered.

~~***Spe3:** to protect non-target plants respect an unsprayed buffer zone of 30m to non-agricultural land OR an unsprayed buffer zone of 15m to non-agricultural land with the use of 50% drift reducing nozzles OR an unsprayed buffer zone of 10m to non-agricultural land with the use of 75% drift reducing nozzles OR an unsprayed buffer zone of 5m to non-agricultural land with the use of 90% drift reducing nozzles.*~~

Application rate 1.5 L/ha (150 g a.s./ha)
Spe3: *to protect non-target plants respect an unsprayed buffer zone of 5m to non-agricultural land OR the use of 75% drift reducing nozzles*

Application rate 1.0 L/ha (100 g a.s./ha)
***Spe3**: to protect non-target plants respect an unsprayed buffer zone of 5m to non-agricultural land OR the use of 50% drift reducing nozzles*

Review Comments:

No explanation was given as why for the herbicide only one formation study on terrestrial plants was performed. Moreover, based on data from LoEP the lowest endpoint for representative formulation is from vegetative vigour test. Nevertheless, the Applicant didn't performed this type of test, and only seedling emergence study for MIGHTY is available. There is also no explanation on what basis the Applicant assumed that a representative formulation covers the risk to MIGHTY. Since different plant species have been tested in the seedling emergence test, it is not possible to compare the toxicity of both formulations. Moreover, in seedling emergence study for MIGHTY the *Lactuca sativa* was not tested (the most sensitive species based on Callisto 100 SC data). Therefore, in opinion of zRMS, the vegetative vigour test for MIGHTY is required.

The Applicant was requested to submit the vegetative vigour test with MIGHTY.

The new study was provided and used in the risk assessment.

The TERs calculations were performed for application rate of 1.0 L product/ha (100 g a.s./ha). However, in the proposed use pattern of MIGHTY such a dose rate is not included. Thus, additional calculations for application rate of 1.5 L product/ha, the only dose included in the GAP, were performed by zRMS.

It has been shown that application of 1.5 L product/ha with 5 m no spray buffer zone OR 75% of nozzles reduction are sufficient to prevent unacceptable risk to non-target terrestrial plants.

9.10.3.1 Effects on other terrestrial organisms (flora and fauna) (KCP 10.7)

9.11 Effects on other terrestrial organisms (flora and fauna) (KCP 10.7)

Not relevant.

9.12 Monitoring data (KCP 10.8)

Not relevant.

9.13 Classification and Labelling

	Mesotrione 10% SC
Common Name	Mesotrione 10% SC
Classification and proposed labelling	
With regard to ecotoxicological endpoints (according to the criteria in Reg. 1272/2008, as amended)	Hazard classes (s), categories: Aquatic Acute Category 1 Aquatic Chronic Category 2 Code(s) for hazard pictogram(s): GHS09 Signal word: Warning Hazard statement(s): H400, H411 (classification); H410 (label) Precautionary statement: P391, P501

Based on the information found in ECHA guidance for classification and labelling of substances and mixtures according to CLP criteria (version 4.1, June 2015) if substance/mixture is classified as: Aquatic Acute 1, H400 and Aquatic Chronic 2, H411, on the label the hazard statement H410 should be included. (please refer to part C).

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
KCP 10.2.1-01	xxxxxxx	2016	Mesotrione 10% SC Rainbow trout Acute toxicity test Study code: W/112/16 xxxxxxxxxxxxx GLP, unpublished	Y	Sharda Cropchem Limited
KCP 10.2.1-02	Brzozowska-Wojoczek, K.	2016	Mesotrione 10 % SC *Daphnia magna*, Acute Immobilization Test Study code: W/148/16 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.2.1-03	Brzozowska-Wojoczek, K.	2016	Mesotrione 10% SC *Pseudokirchneriella subcapitata* SAG 61.81 Growth inhibition test Study code: W/147/16 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.2.1-04	Brzozowska-Wojoczek, K.	2016	Mesotrione 10% SC *Lemna gibba* CPCC 310 Growth inhibition test Study code: W/149/16 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.1.1.1	Pawel Parma	2015	Mesotrione 10% SC; Honeybees (Apis mellifera L.), Acute Oral Toxicity Test Study code: B/132/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.1.1.2	Pawel Parma	2015	Mesotrione 10% SC; Honeybees (Apis mellifera L.), Acute Contact Toxicity Test Study code: B/133/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited

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 10.3.1.2	Gimeno, I.	2019	Mesotrione Technical – Chronic Toxicity to the Honey Bee, *Apis mellifera* L. under laboratory Conditions Study code: TRC17-006BA Trialcamp S.L.U. GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.1.3	Scheller, K.	2018	Mesotrione Technical - Repeated exposure of honey bee (*Apis mellifera* L.) larvae under laboratory conditions (in vitro) Study code: 17 48 BLC 0088 BioChem agrar GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.2.2-01	Pawel Parma	2016	An extended laboratory test for evaluating the effects of Mesotrione 10% SC on the parasitic wasp, *Aphidius rhopalosiphi* (De Stefani - Perez) Study code: B/131/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.2.2-02	Parma, P.	2017	An extended laboratory test for evaluating the effects of Mesotrione 10% SC on the predatory mite, *Typhlodromus pyri* (Sch.) Study code: B/130/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.2.2-03	Luna, F.	2017	Aged residue test with the formulation Mesotrione 10% SC on the predatory mite *Typhlodromus pyri* (Acari: Phytoseiidae) Study code: TRC17-090BA Trialcamp S.L.U. GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.3.2.2-04	Soler, E.	2017	Side-effects of the formulated product Mesotrione 10% SC on *Aleochara bilineata* (Coleoptera:Staphylinidae) under extended laboratory conditions Study code: TRC17-115BA Trialcamp S.L.U. GLP, unpublished	N	Sharda Cropchem Limited

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 10.4.1.1	Weronika Dec	2016	Mesotrione 10% SC Earthworm Reproduction Test (*Eisenia fetida*) Study code: B/243/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.4.2.1	Weronika Dec	2016	Mesotrione 10% SC Collembolan (*Folsomia candida*) Reproduction Test Study code: G/244/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.5-01	Weronika Dec	2016	Mesotrione 10% SC; Soil Microorganisms: Nitrogen Transformation Test Study code: G/242/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
~~KCP 10.5-02~~	~~Weronika Dec~~	~~2016~~	~~Mesotrione 10% SC; Soil Microorganisms: Carbon Transformation Test Study code: G/241/15 Institute of Industrial Organic Chemistry GLP, unpublished~~	~~N~~	~~Sharda Cropchem Limited~~
KCP 10.6.2-01	Weronika Dec	2017	Mesotrione 10% SC Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test Study code: G/246/15 Institute of Industrial Organic Chemistry GLP, unpublished	N	Sharda Cropchem Limited
KCP 10.6.2-02	Sadananda, T. S.	2019	Terrestrial plant test: Vegetative vigour test of Mesotrione 10% SC on plants Study code: BIO-ETX 034 Bioneeds India Private Limited GLP, unpublished	N	Sharda Cropchem Limited

Appendix 2 Detailed evaluation of the new studies

Review Comment:
In order to provide sufficient detail, where appropriate, the following study summaries have been adapted by the zRMS from the full study reports provided in the dossier. zRMS text is highlighted in grey. The comments on individual studies are provided in grey comment boxes.

A 2.1 KCP 10.1 Effects on birds and other terrestrial vertebrates

A 2.1.1 KCP 10.1.1 Effects on birds

A 2.1.1.1 KCP 10.1.1.1 Acute oral toxicity

No new study submitted.

A 2.1.1.2 KCP 10.1.1.2 Higher tier data on birds

No new study submitted.

A 2.1.2 KCP 10.1.2 Effects on terrestrial vertebrates other than birds

No new study submitted.

A 2.1.2.1 KCP 10.1.2.1 Acute oral toxicity to mammals

Please refer to section B6.

A 2.1.2.2 KCP 10.1.2.2 Higher tier data on mammals

No new study submitted.

A 2.1.3 KCP 10.1.3 Effects on other terrestrial vertebrate wildlife (reptiles and amphibians)

No new study submitted.

A 2.2 KCP 10.2 Effects on aquatic organisms

A 2.2.1 KCP 10.2.1 Acute toxicity to fish, aquatic invertebrates, or effects on aquatic algae and macrophytes

Comments of zRMS:	The study was conducted to OECD guideline 203 (1992) and according to the principles of GLP. Since there is the new version of OECD 203 adopted on 18 June 2019, zRMS evaluated the study according to the recent guideline. In the definitive test all validity criteria were met. The mortality of fish in the control was 0%. The dissolved oxygen concentration was in the range of 89-99% of air saturation value. Samples of each test item concentration and the control collected at exposure initiation and termination were chemically analyzed. One deviation to the current guideline were noted. The temperature was in the range 14.4-15.4°C instead recommended 10-14°C. However, this slight deviation did not affect the outcome of the study. The study is considered to be reliable and suitable for the risk assessment. All results refer to nominal concentrations. The test item concentrations up to 50.0 mg/L caused no effects, but at 100 mg/L all fish were dead. Therefore, in the zRMS opinion the LC_{50} should be stated as >50 mg formulation/L. This endpoint will be used in the risk assessment.

Reference:	KCP 10.2.1 - 01
Report	"Mesotrione 10% SC Rainbow trout Acute toxicity test". xxxxx 2016. Study code: W/112/16. xxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxx
Guideline(s):	Yes (OECD 203 (1992))
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Materials

Test item:

Description:	Mesotrione 10% SC
Production batch:	SWEPL-41203
A.i. content:	Mesotrione 10.2% w/w

Test system:

Species:	Rainbow trout (*Oncorhynchus mykiss*)
Strain:	-
Age:	approximately 2 months
Source:	Culture of the salmonidae fish in Zawoja', Poland (Hodowla Ryb Łososiowatych w Zawoi, Polska).
Acclimation period:	7 days
Diet:	During the adaptation the fish were fed with standard granulated fish food in the amount of 2% of their average body weight per day (standard fry food, Aller Aqua, Denmark). Feeding of the fish was stopped 24 h

before exposure initiation

Experimental conditions:

Temperature: 14.4 – 15.4 °C
Dissolved O_2: 96 – 99 %
Hardness: 63.6 mg/L $CaCO_3$.
pH of the control: 7.13-7.58
Light and photoperiod: 16h light and 8h dark.
Loading: 0.80 g/L.
Test procedure: -

Experimental period: 96h

Test design and treatment:

Static system (96 hours), one replicate of seven fish for each test item concentration and the control. A preliminary concentration range-finding test was conducted. In the preliminary test the following test item concentrations were used: 100, 10 and 1.0 mg/L plus the control. In the test item concentration of 100 mg/L after 3 h of exposure all fish were found dead. After 3, 6, 24, 48, 72 and 96 h of exposure in the test item concentrations of 10 and 1.0 mg/L and in the control neither mortality of fish nor symptoms of intoxication were observed. In the definitive test the rainbow trout were exposed to the test item concentrations of 100, 50, 25, 12.5 and 6.25 mg/L plus the control for 96 h in a static test design. The concentration of mesotrione was chemically determined using a validated liquid chromatographic method with DAD detection. Samples of each test item concentration and the control collected at exposure initiation and termination were chemically analyzed.

Results:

Table 8. Mortality - definitive test

Nominal test item concentration [mg/L]	Number of fish	Number of dead fish						Total mortality [%]
		3 h	6 h	24 h	48 h	72 h	96 h	
Control	7	0	0	0	0	0	0	0
6.25	7	0	0	0	0	0	0	0
12.5	7	0	0	0	0	0	0	0
25	7	0	0	0	0	0	0	0
50	7	0	0	0	0	0	0	0
100	7	7	7	7	7	7	7	100

Time of exposure: 12.08.2016 – 16.08.2016

Table 9. Intoxication symptoms - definitive test

Nominal test item concentration [mg/L]	Symptoms	Time of exposure					
		3 h	6 h	24 h	48 h	72 h	96 h
Control	UR	7 NN	7 NN	7 NN	7 NN	7 NN	7 NN
	SP	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	FO	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	U	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
6.25	UR	7 NN	7 NN	7 NN	7 NN	7 NN	7 NN
	SP	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	FO	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	U	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
12.5	UR	7 NN	7 NN	7 NN	7 NN	7 NN	7 NN
	SP	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	FO	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	U	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
25	UR	7 NN	7 NN	7 NN	7 NN	7 NN	7 NN
	SP	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	FO	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
	U	7 NR	7 NR	7 NR	7 NR	7 NR	7 NR
50	UR	7 NN	7 NN	7 NN	7 NN	7 NN	7 NN
	SP	3 U, 4 NR	3 U, 4 NR	3 U, 4 NR	3 U, 4 NR	3 U, 4 NR	3 U, 4 NR
	FO	4 F, 3 NR	5 F, 2 NR	5 F, 2 NR	5 F, 2 NR	5 F, 2 NR	6 F, 1 NR
	U	3 NT, 4 NR	3 NT, 4 NR	3 NT, 4 NR	3 NT, 4 NR	3 NT, 4 NR	5 NT, 2 NR
100	UR	-	-	-	-	-	-
	SP	-	-	-	-	-	-
	FO	-	-	-	-	-	-
	U	-	-	-	-	-	-

Symptoms:	**UR** – loss of equilibrium	**NN** – no, **T** – yes
	SP – swimming behavior	**NR** – normal, **U** – unbalanced
	FO – respiratory functions	**NR** – normal, **F** – faulty, problems
	U – pigmentation	**NR** – normal, **NT** – untypical

Table 10. Determined concentrations of mesotrione - definitive test

Nominal test item concentration [mg/L]	Nominal concentration of mesotrione in the test item [mg/L]	Mean concentration (n=3) of mesotrione determined in samples collected [mg/L]			
		at exposure initiation	% of nominal concentration	at exposure termination	% of nominal concentration
Control	0.000	< LoD	--	< LoD	--
6.25	0.625	0.570	91.20	0.567	90.72
12.5	1.25	1.231	98.48	1.092	87.36
25	2.5	2.452	98.08	2.378	95.12
50	5.0	5.034	100.68	5.033	100.66
100	10	9.547	95.47	9.989	99.89

LoQ = 0.005 mg/L
LoD = 0.001 mg/L

The endpoint values based on the mortality of rainbow trout after 96 hours of exposure to the nominal test item concentrations in a static design:

~~The LC_{50}/96 h is 70.54 mg/L~~.
The LOEC/96 h is 100 mg/L.
The NOEC/96 h is 50 mg/L.

The endpoint values determined based on the nominal concentrations of mesotrione in the test item and mortality of fish:

~~The LC_{50}/96 h is 7.05 mg/L.~~
The LOEC/96 h is 10 mg/L.
The NOEC/96 h is 5.0 mg/L.

Conclusion:

The endpoint values determined based on the nominal test item concentrations and mortality of fish:

The median concentration causing 50% mortality of rainbow trout after 96 hours of exposure LC_{50}/96 h value is 70.54 mg/L.
The Fisher's Exact Binomial Test with Bonferroni Correction performed with data for mortality at 96 h showed significant difference between the nominal test item concentration of 100 mg/L compared with the control. Therefore, the lowest test item concentration causing fish mortality the LOEC/96 h value is 100 mg/L. The test item concentration not causing effect on fish mortality the NOEC/96 h value is 50 mg/L.

The endpoint values determined based on the nominal concentration of mesotrione in the test item and mortality of fish:
The median concentration causing 50% mortality of rainbow trout after

96 hours of exposure LC_{50}/96 h value is 7.05 mg/L.
The Fisher's Exact Binomial Test with Bonferroni Correction performed with data for mortality at 96 h showed significant difference between the nominal mesotrione concentration of 10 mg/L compared with the control. Therefore, the lowest mesotrione causing fish mortality the LOEC/96 h value is 10 mg/L. The mesotrione concentration not causing effect on fish mortality the NOEC/96 h value is 5 mg/L

Comments of zRMS:	The study was conducted to OECD guideline 202 and according to the principles of GLP. In the definitive test the validity criteria were met according to OECD Guideline No. 202. The mortality of *Daphnia magna* in the control was 0%. The dissolved oxygen concentrations in the test vessels were within the range of 9.0-9.8 mg/L. One minor deviation to the guideline (described below) were noted. The study is considered to be reliable and suitable for the risk assessment. All results refer to nominal concentrations.

Reference: KCP 10.2.1 - 02

Report "Mesotrione 10 % SC *Daphnia magna*, Acute Immobilization Test". Brzozowska - Wojoczek, K. 2016. Study code: W/148/16. Institute of Industrial Organic Chemistry Branch Pszczyna

Guideline(s): Yes (OECD 202)

Deviations: Yes. During the study deviation from the study plan occurred. The study plan stated that concentration of microelement stock solution $Na_2MoO_4.2H_2O$ is 0.126 g/100 mL, the correct concentration of microelement stock solution $Na_2MoO_4.2H_2O$ is 0.123 g/100 mL.
The study plan stated the deadline for final report was October 2016. However, due to obligation acquire sponsor's acceptance of the report, the deadline was postponed.
The deviation did not have any impact on the study results.

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Materials and methods

Materials

Test item:

Description: Mesotrione 10% SC
Production batch: SWEPL-41203
A.i. content: Mesotrione 10.2% w/w

Test system:

Species: *Daphnia magna*
Strain: Straus
Age: neonatal daphnids (less than 24 h old.). not first brood progeny
Source: laboratory culture cultivated at the Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicology, Laboratory of Aquatic Toxicology
Acclimation period: -

Diet: mixture of two species *Pseudokirchneriella subcapitata* and *Desmodesmus subspicatus* (in 2:1 ratio) originating from separate cultures in the Laboratory of Aquatic Toxicology. Group B vitamins and micronutrients necessary for proper growth were supplied with the lyophilized suspension of *Spirulina* sp.

Experimental conditions:

Temperature: 19.3-20.0°C
pH of the control: 7.15 – 7.16
oxygen concentration: 9.0 – 9.8 mg/L
Light and photoperiod: 16h light and 8h dark, fluorescent light source.
No feeding; no aeration.

Experimental period: 48h

Test design and treatment:

The test with a reference substance, potassium dichromate, was performed with five concentrations ranging from 0.32 to 3.2 mg/L and a control were used. There were four replicates of each reference substance concentration and the control. Five young *Daphnia magna* were introduced into each replicate.
A preliminary range finding study was conducted with five groups of twenty daphnids per group with 0.1, 1.0, 10.0 and 100 mg/L of Mesotrione 10% SC and a control. In the first preliminary test, in the control and in the test item concentrations of 10, 1.0 and 0.1 mg/L no immobilization of *Daphnia magna* was observed during exposure.
The definitive test was performed using the following test item concentrations: 500, 250, 125, 62.5, 31.25, 15.63 mg/L plus the control were used. Four replicates of each test item concentration and the control with five Daphnia magna per replicate were used. *Daphnia magna* was observed for immobilization after 24 and 48 h of exposure. The *Daphnia magna* was considered immobile if they showed no ability to swim within 15 seconds after swirling the test vessel. At exposure termination, in the control and in the test item concentrations of 31.25, and 15.63 mg/L no immobilization of Daphnia magna was observed. In the test item concentrations of 500, 250, 125, and 62.5 mg/L the immobilization of Daphnia magna was 100, 100, 85, and 15%, respectively. The concentrations of mesotrione were chemically determined with a validated liquid chromatographic method with DAD. Samples of each test item concentration and the control collected at exposure initiation and at exposure termination were chemically analyzed.

Results:

At exposure initiation the determined concentration of mesotrione was in the range of 85.0 – 91.3% of the nominal concentration. The results confirm correct preparation of the test item concentrations. At exposure termination the determined concentration of mesotrione was in the range of 82.1 – 91.8% of the nominal concentration. Therefore, the concentrations of mesotrione were stable under test conditions. The endpoint values were determined on the basis of the nominal test item concentrations and the nominal concentration of mesotrione in the test item.

Table 8. Immobilization, definitive test.

Nominal test item concentration [mg/L]	Number of *Daphnia magna*	Number of immobilized *Daphnia magna*								Total immobilization of *Daphnia magna* [%]	
		24 h				48 h					
		Replicates									
		A	B	C	D	A	B	C	D	24 h	48 h
Control	20	0	0	0	0	0	0	0	0	0	0
15.63	20	0	0	0	0	0	0	0	0	0	0
31.25	20	0	0	0	0	0	0	0	0	0	0
62.5	20	0	0	0	0	1	1	1	0	0	15
125	20	2	2	3	3	4	5	4	4	50	85
250	20	5	5	5	5	5	5	5	5	100	100
500	20	5	5	5	5	5	5	5	5	100	100

Time of exposure: 30.08.2016 – 01.09.2016.

Table 10. Endpoint values based on nominal test item concentrations, definitive test.

Endpoint values [mg/L]	Time of exposure	
	24 h	48 h
EC_{50}	125.00 (n.d.)	88.39 (74.52 – 104.84)
EC_{20}	111.43 (n.d.)	67.04 (50.82 – 78.86)
EC_{10}	104.93 (n.d.)	58.02 (40.55 – 69.73)
LOEC	125	62.5
NOEC	62.5	31.25

Calculations according to [5], [SOP/W/68].
(-) - 95% confidence interval
n.d. – not determined

Table 11. Endpoint values based on the nominal concentrations of mesotrione in the test item, definitive test

Endpoint values [mg/L]	Time of exposure	
	24 h	48 h
EC_{50}	12.50 (n.d.)	8.84 (7.45 – 10.48)
EC_{20}	11.06 (n.d.)	6.70 (5.08 – 7.89)
EC_{10}	10.38 (n.d.)	5.80 (4.06 – 6.97)
LOEC	12.5	6.25
NOEC	6.25	3.13

Calculations according to [5], [SOP/W/68].
(-) - 95% confidence interval
n.d. – not determined

Conclusion

The EC_{50}/48 h = 88.39 mg/L (EC_{50}/48 h =8.84 mg a.s./L).
Based on the results generated in the study, the NOEC value equals 31.25 mg/L (3.13 mg a.s./L) (i.e. is the test item concentration used for the exposure) and the LOEC value is equal than 62.5 mg/L (6.25 mg a.s./L).

Comments of zRMS:	The study was conducted to OECD guideline 201 and according to the principles of GLP. The biomass in the control increased by a factor of 67.3 within the 72-hour test period. The coefficient of variation of the mean specific growth rate after the 72-hour test period (exposure initiation – exposure termination) in the control culture was 2.5%. The mean coefficient of variation for the section-by-section growth rate in the control culture was 31.2%. Thus, in the definitive test the validity criteria were met according to OECD Guideline No. 201. One minor deviation to the guideline (described below) were noted. The study is considered to be reliable and suitable for the risk assessment. All results refer to nominal concentrations.

Reference: KCP 10.2.1 - 03

Report "Mesotrione 10% SC *Pseudokirchneriella subcapitata* SAG 61.81 Growth inhibition test". Brzozowska - Wojoczek, K. 2016, Study code: W/147/16. Institute of Industrial Organic Chemistry Branch Pszczyna

Guideline(s): Yes (OECD 201)

Deviations: Yes. During the study one deviation from the study plan occurred. The study plan stated that concentration of macronutrient stock solution $MgSO_4.7H_2O$ is 1.47 g/100 mL, the correct concentration of macronutrient stock solution $MgSO_4.7H_2O$ should be 1.46 g/100 mL. The deviation did not have any impact on the study results.

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Materials and methods

Materials

Test item:

Description: Mesotrione 10% SC
Production batch: SWEPL-41203
A.i. content: Mesotrione 10.2% w/w

Test system:

Species: The unicellular freshwater green algae, *Pseudokirchneriella subcapitata (Reinsch)* Korshikov (syn. *Raphidocelis subcapitata, Selenastrum capricornutum* Prinz)
Strain: SAG 61.81
Age: -
Source: Cultivated at the Institute of Industrial Organic Chemistry. Obtained from the Culture Collection of Algae at Gottingen University, Germany
Acclimation period: Fresh medium and incubated at temperature between 21 – 24 ºC under constant illumination. The algal culture was renewed twice a week under sterile conditions.
Diet: AAP medium

Experimental conditions:

Temperature: 21.2 – 21.8 °C
pH values: 7.57– 8.55
Humidity: -
Air changes: -
Light and photoperiod: 7098-7380 lux

Experimental period: 72h

Test design and treatment:

72 hours of the exposure; three replicates of each test item concentration and six replicates of the control. Initial algal cell density: 1 x 104 cells/mL. The first preliminary test was performed using four test item concentrations: 100, 10, 1.0, 0.1 mg/L plus the control. The mean transmittance values were between 92.0 – 100.2% at exposure initiation and 83.3 – 99.7% at exposure termination when compared with the control. The second preliminary test was performed using one test item concentration 250 mg/L plus the control. The mean transmittance values was 81.6% at exposure initiation and 54.2% at exposure termination when compared with the control.
In the definitive test following test item concentrations were used 250, 125, 62.5, 31.25, 15.63 and 7.82 mg/L (with a separation factor of 2.0) plus control. Algal growth was monitored at 24, 48 and 72h. The APP medium was used as a culture medium for the test organism and a diluent/solvent of the test item.
The test item in the amount of 100.0 mg was weighed in a glass crystallizer and quantitatively transferred into a glass flask by multiple washing with the

AAP medium and filled up to 1000 mL. The control was the AAP medium. The reliability of the test system was confirmed using 3,5-Dichlorophenol as a positive control.

Statistics: Probit method calculations and analysis by Shapiro-Wilk's Test on NormalDistribution, Levene's Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure.

Results:

	The endpoint values (95% confidence limits)	
	Determined on the basis of the nominal test item concentrations:	Deterrmined on the basis of the nominal concentration of mesotrione in the test item:
ErC_{50}/72 hours (mg/L)	80.18 (75.53 – 85.36)	8.02 (7.55 – 8.54)
ErC_{20}/72 hours (mg/L)	56.91 (51.80 – 61.22)	5.69 (5.18 – 6.12)
ErC_{10}/72 hours (mg/L)	47.57 (41.92 – 52.21)	4.76 (4.19 – 5.22)
LOEC/72 hours (mg/L)	31.25	3.13
NOEC/72 hours (mg/L)	15.63	1.56
EyC_{50}/72 hours (mg/L)	43.13 (38.13 – 48.78)	4.31 (3.81 – 4.88)
EyC_{20}/72 hours (mg/L)	23.41 (18.60 – 27.45)	2.34 (1.86 – 2.75)
EyC_{10}/72 hours (mg/L)	17.00 (12.42 – 20.91)	1.70 (1.24 – 2.09)
LOEC/72 hours (mg/L)	31.25	3.13
NOEC/72 hours (mg/L)	15.63	1.56

Conclusion

The E_rC_{50}/72 hours of Mesotrione 10% SC is 80.18 mg/L (95% confidence interval: 75.53 – 85.36) = 8.02 mg/L (95% confidence interval: 7.55 – 8.54). The E_yC_{50}/72 hours of Mesotrione 10% SC is 43.13 mg/L (95% confidence interval: 38.13 – 48.78) = 4.31 mg/L (95% confidence interval: 3.81 – 4.88). The LOEC/72 hours and NOEC/72 hours were respectively 31.25 mg test item/L (3.13 mg a.s./L) and 15.63 mg test item/L (1.56 mg a.s./L).

Comments of zRMS:	The study was conducted to OECD guideline 221 and according to the principles of GLP. In the definitive test the validity criteria were met according to OECD Guideline No. 221. The doubling time of frond number in the control was 1.8 days. The factor of frond number in the control between 0 and 7 day was 12.6. The average specific growth rate in the control between day 0 and day 7 was 0.386 d^{-1}. The study is considered to be reliable and suitable for the risk assessment.

Reference: KCP 10.2.1 - 04

Report "Mesotrione 10% SC *Lemna gibba* CPCC 310 Growth inhibition test" Brzozowska - Wojoczek, K. 2016, Study code: W/149/16. Institute of Industrial Organic Chemistry Branch Pszczyna.

Guideline(s): Yes (OECD 221)

Deviations: No.

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study)

Materials and methods
Materials

Test item:

Description:	Mesotrione 10% SC
Production batch:	SWEPL-41203
A.i. content:	Mesotrione 10.2% w/w

Test system:

Species:	*Lemna gibba* L. cultivated at the Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicology, Laboratory of Aquatic Toxicology.
Strain:	CPCC 310
Age:	-
Source:	Canadian Phycological Culture Center (CPCC), Department of Biology, University of Waterloo, Canada.
Medium:	20X APP
Acclimation period: :	7 days

Experimental conditions:

Temperature:	24.3 – 25.0C°
pH values:	7.37 – 8.77
Mean light intensity:	8235 - 8278lux, illumination constant
Test vessels:	glass beakers containing 400 mL of each

	treatment.
Experimental period:	7 d

Test design:

Static system. 7 days of exposure. Three replicates of each test item concentration; six replicates of the control.
The preliminary test was performed under static test design. The test was performed using the following test item concentration: 10, 1.0, 0.1, 0.01 mg/L plus the control. In the definitive test, test item concentrations were: 32, 10, 3.1, 1.0, 0.31, 0.1, 0.03, 0.01, 0.003 mg/L plus the control. The 103.2 mg of the test item was weighed in a glass crystallizer and quantitatively transferred into a volumetric flask by multiple washing with 20X AAP medium. The flask was filled up to a total volume of 3225 mL with 20X AAP medium. The test item concentration of 32 mg/L was visually homogeneous. Each replicate was inoculated with a total of 9 fronds. The total number of fronds in each test vessel was counted twice during exposure and at exposure termination. The concentrations of mesotrione were chemically determined with a validated liquid chromatographic method with DAD detection. The sensitivity of the *Lemna gibba* culture is monitored with the reference material 3,5-dichlorophenol.

Statistics:

Probit method calculations (using linear max. likelihood regression) and analysis by Shapiro-Wilk's Test on Normal Distribution, Levene's Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure, Multiple Sequentially-rejective Welsh-t-test After Bonferroni-Holm.

Results:

Table 13. Concentration and stability of mesotrione, definitive test

Nominal test item concentration [mg/L]	Nominal concentration of mesotrione in the test item [mg/L]	Mean concentration (n=3) of mesotrione determined in samples collected [mg/L]			
		at exposure initiation	% of nominal concentration	at exposure termination	% of nominal concentration
Control	0.0000	< LoD	--	< LoD	--
0.003	0.0003	< LoD	--	< LoD	--
0.01	0.001	< LoD	--	< LoD	--
0.03	0.003	< LoQ	--	< LoQ	--
0.1	0.01	0.010	100.0	0.010	100.0
0.31	0.031	0.032	103.2	0.033	106.5
1.0	0.1	0.094	94.0	0.091	91.0
3.1	0.31	0.298	96.1	0.311	100.3
10	1.0	0.964	96.4	0.976	97.6
32	3.2	3.104	97.0	3.223	100.7

LoQ = 0.005 mg/L
LoD = 0.001 mg/L

	The endpoint values determined on the basis of the nominal test item concentrations. (95% confidence limits)	
	On the basis of the frond number:	On the basis of the dry weight:
ErC_{50}/7d (mg/l)	0.383 (0.345 – 0.426)	3.490 (2.979 – 4.123)
ErC_{20}/7d (mg/l)	0.043 (0.036 – 0.050)	0.069 (0.054 – 0.086)
ErC_{10}/7d (mg/l)	0.013 (0.011 – 0.017)	0.009 (0.006 – 0.012)
NOEC/7d (mg/l)	0.01	0.01
LOEC/7d (mg/l)	0.03	0.03
EyC_{50}/7d/ (mg/l)	0.078 (0.072 – 0.085)	0.109 (0.100 – 0.119)
EyC_{20}/7d/ (mg/l)	0.018 (0.016 – 0.021)	0.014 (0.012 – 0.016)
EyC_{10}/7d/ (mg/l)	0.009 (0.007 – 0.010)	0.005 (0.004 – 0.006)
NOEC/7d (mg/l)	0.003	0.03

LOEC/7d (mg/l)	0.01	0.1

	The endpoint values determined on the basis of the nominal concentration of mesotrione in the test item. (95% confidence limits)	
	On the basis of the frond number:	On the basis of the dry weight:
ErC_{50}/7d (mg/l)	0.038 (0.035 – 0.043)	0.349 (0.298 – 0.412)
ErC_{20}/7d (mg/l)	0.004 (0.004 – 0.005)	0.007 (0.005 – 0.009)
ErC_{10}/7d (mg/l)	0.001 (0.001 – 0.002)	0.001 (0.001 – 0.001)
NOEC/7d (mg/l)	0.001	0.001
LOEC/7d (mg/l)	0.003	0.003
EyC_{50}/7d/ (mg/l)	0.008 (0.007 – 0.009)	0.010 (0.010 – 0.012)
EyC_{20}/7d/ (mg/l)	0.002 (0.002 – 0.002)	0.001 (0.001 – 0.002)
EyC_{10}/7d/ (mg/l)	0.001 (0.001 – 0.001)	0.001 (0.000 – 0.001)
NOEC/7d (mg/l)	0.0003	0.003
LOEC/7d (mg/l)	0.001	0.01

A 2.2.2 KCP 10.2.2 Additional long-term and chronic toxicity studies on fish, aquatic invertebrates and sediment dwelling organisms

No new study submitted.

A 2.2.3 KCP 10.2.3 Further testing on aquatic organisms

No new study submitted.

A 2.3 KCP 10.3 Effects on arthropods

A 2.3.1 KCP 10.3.1 Effects on bees

A 2.3.1.1 KCP 10.3.1.1 Acute toxicity to bees

A 2.3.1.1.1 KCP 10.3.1.1.1 Acute oral toxicity to bees

Comments of zRMS:	The study was conducted to OECD guideline 213 and according to the principles of GLP. No deviations to the guideline were noted. The study is considered to be reliable and suitable for the risk assessment.

Reference:	KCP 10.3.1.1.1
Report	Mesotrione 10% SC; Honeybees (*Apis mellifera* L.), Acute Oral Toxicity Test, Paweł Parma, 2015, Study code B/132/15. Institute of Industrial Organic Chemistry Branch Pszczyna.
Guideline(s):	Yes (OECD 213)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Materials and methods

Mesotrione 10% SC (batch No. SWEPL - 41203) is a brown yellow liquid formulation containing the active substance Mesotrione (nominal concentration: 10.2% w/w). Five different doses (12.5, 25.0, 50.0, 100.0 and 200.0µg/honeybee) selected on the basis of the preliminary tests results were used on *Apis mellifera L.*. A group of 10 bees (3 replicates containing 10 bees each) was fed with 100µL of a 50% sucrose solution, containing the test item at the doses enumerated above, using a micropipette. During the experiment, the insects were caged in groups of 10.
After the application, the insects were observed for mortality and other signs of toxicity. These observations were made 4 hours after the beginning of the treatment and then every 24 hours after the beginning of the treatment. The acute oral toxicity test ended after the 48-hour exposure.

Results and discussions

Table 2.3.1.1.1.1-1: Acute oral toxicity on honeybees (*Apis mellifera L.*)

Dose (µg/bee)		Replicates	Toxicity symptoms*		
(µg test item/bee)	(µg a.i./bee)		4 h	24 h	48 h
0.0 (control)		I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
12.5	1.28	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
25.0	2.55	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
50.0	5.10	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
100.0	10.20	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10

200.0	20.40	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10

*bees with toxic symptoms (excitement, paralysis) / living bees

Findings:

- No deaths were observed during the study, at all tested doses
- No Sublethal toxicity effects were observed over the 48-hour exposure
- No abnormal behavioural effects were observed during the experiment with the reference item

The following validity criteria were met during the test:

- The average mortality for the total number of controls was 0.0% after 48h (criterion: it must not exceed 10%)
- The 24-hour LD_{50} of the reference item (dimethoate) was 0.11µ/bee (criterion: 0.10 – 0.35 µg a.i./bee)

Conclusion

Under the experimental conditions, the acute oral LD_{50} of the Mesotrione 10% SC is higher than 200.0 µg/honeybee, representing 20.40 µg a.i./honeybee.

A 2.3.1.1.2 KCP 10.3.1.1.2 Acute contact toxicity to bees

Comments of zRMS:	The study was conducted to OECD guideline 214 and according to the principles of GLP. No deviations to the guideline were noted. The study is considered to be reliable and suitable for the risk assessment.

Reference:	KCP 10.3.1.1.2
Report	Mesotrione 10% SC; Honeybees (*Apis mellifera* L.), Acute Contact Toxicity Test, Paweł Parma, 2015, Study code B/133/15. Institute of Industrial Organic Chemistry Branch Pszczyna.
Guideline(s):	Yes (OECD 214)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Materials and methods

Mesotrione 10% SC (batch No. SWEPL - 41203) is a brown yellow liquid formulation containing the active substance Mesotrione (nominal concentration: 10.2% w/w). Five different doses (12.5, 25.0, 50.0, 100.0 and 200.0µg/honeybee) selected on the basis of the preliminary tests results were used on *Apis mellifera L.*. The test item was applied to the dorsal part of thorax using a microapplicator, on a group of 10 bees (3 replicates containing 10 bees each). Each bee received 1µL of the solution containing an appropriate dose of the test item. During the experiment, the insects were caged in groups of 10.
After the application, the insects were observed for mortality and other signs of toxicity. These observations were made 4 hours after the beginning of the treatment and then every 24 hours after the beginning of the treatment. The acute contact toxicity test ended after the 48-hour observation.

Results and discussions

Table 2.3.1.1.2.1-1: Acute contact toxicity on honeybees (*Apis mellifera L.*)

Dose (µg/bee)		Replicates	Toxicity symptoms*		
(µg test item/bee)	(µg a.i./bee)		4 h	24 h	48 h
0.0 (control)		I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
12.5	1.28	0 / 10	0 / 10	0 / 10	0 / 10
		0 / 10	0 / 10	0 / 10	0 / 10
		0 / 10	0 / 10	0 / 10	0 / 10
25.0	2.55	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 9	0 / 9
		III	0 / 10	0 / 10	0 / 10
50.0	5.10	I	0 / 10	0 / 9	0 / 9
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
100.0	10.20	I	0 / 10	0 / 10	0 / 10
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 10	0 / 10
200.0	20.40	I	0 / 10	0 / 9	0 / 9
		II	0 / 10	0 / 10	0 / 10
		III	0 / 10	0 / 9	0 / 9

*bees with toxic symptoms (excitement, paralysis) / living bees

Findings:

- There were no dead bees in the control group at the end of the study
- The highest mortality rate at the end of the study was 6.67%, caused by test item at the doses of 200.0 µg/bee.
- No abnormal behavioural effects were observed during the test

The following validity criteria were met during the test:

- The average mortality for the total number of controls was 0.0% after 48h (criterion: it must not exceed 10%)
- The 24-hour LD_{50} of the reference item (dimethoate) was 0.23 µg/bee (criterion: 0.10 – 0.30 µg a.i./bee)

Conclusion

Under the experimental conditions, the acute contact LD_{50} of the Mesotrione 10% SC is higher than 200.0 µg/honeybee, representing 20.40 µg a.i./honeybee.

A 2.3.1.2 KCP 10.3.1.2. Chronic toxicity to bees

~~No new study submitted.~~

Comments of zRMS:	The study was conducted to OECD guideline and according to the principles of GLP. All validity criteria were fulfilled. The study is considered to be reliable.

Reference:	KCP 10.3.1.2
Report	"Mesotrione Technical - Chronic Toxicity to the Honey Bee, *Apis mellifera* L. under Laboratory Conditions", Ignacio Gimeno, 2019, Study code TRC17-006BA
Guideline(s):	Yes, OECD OECD test No. 245 Guideline for the Testing of Chemicals: Honey bee (*Apis mellifera* L.), Chronic Oral Toxicity Test – 10 Day Feeding (9 October 2017).
Deviations:	No.
GLP:	Yes
Acceptability:	Yes

Materials and methods

The aim of the study was to determine the effects of the test item Mesotrione Technical (Batch SCL-62581) on the honey bee *Apis mellifera* L. from chronic feeding exposure, the median Lethal Concentration (LC_{50}) and the median Lethal Dietary Dose (LDD_{50}) after 10 days of exposure, as well as the No Observed Effect Concentration and the No Observed Effect Dietary Dose (NOEC/NOEDD) were determined.

The test species was honey bee (*Apis mellifera* L.), young adult worker bees (not older than 48 hours) originating from commercial bee hives maintained by Trialcamp S.L.U..

The test was conducted as a dose-response test with an exposure phase duration of 10 days. Two control groups (a negative control and a solvent control), five test item groups and a toxic reference item group were used. Five different concentrations of Mesotrione Technical were applied to the bees in the test item groups, and one single concentration of the reference item was
applied to the bees in the toxic reference group. The analyzed content of Mesotrione was considered for calculation of the test item doses and dimethoate for the reference item dose. Control groups and treated groups were exposed concurrently to identical conditions.

1 control group, 5 test item groups with 157.56, 315.13, 630.25, 1260.50 and 2521.01 mg a.s./kg diet, equivalent to 18.75, 37.50, 75.00, 150.00 and 300.00 µg a.s./bee/day; 1 reference item group with 0.107 µg dimethoate/bee/day.Five replicates of 10 bees each were used for each group. Mortality and behavioural abnormalities were assessed daily over the 10 days test period.

The test conditions were: Air temperature: Min / Max: 32.1 / 33.0 °C, Relative air humidity: Min / Max: 43.8 % / 70.6 % RH, Exposure to light: Constant darkness except during feeding and assessments.
Endpoints were empirically estimated due to a lack of concentration – response.
Moreover no mortality resulted at the highest tested dose/concentration.

Results and discussions

Table 2.3.1.1.2-1: Cumulative mortality, overall mean consumption of feeding solution, dietary dose (DD), accumulated mean uptake, NOEC, NOEDD, LOEC, LOEDD, LC_{50} and LDD_{50}

Treatment	10-day cumulative mortaliy (corrected)[1]	Overall mean consumption of feeding solution	Dietary dose (based on actual measured consumption of feeding solution)	Mean accumulated uptake of test item during the test period

	[%]	[µL/bee/day]	[µg a.s./bee/day]	[µg a.s./bee]
Control				
C (0)	0.00	18.67	-	-
C_5(0)	0.00	20.02	-	-
Reference item: dimethoate [µg a.s./bee]				
R (0.107)	100.00	19.53	0.02	0.13
Test item: Mesotrione Technical [mg a.s./kg feeding solution]				
T1 (157.56)	2.00	20.81	3.90	39.02
T2 (315.13)	0.00	19.63	7.36	73.62
T3 (630.25)	2.00	18.96	14.22	142.19
T4 (1260.50)	2.00	19.63	29.44	294.38
T5 (2521.01)	0.00	19.07	57.21	572.10
NOEC	≥ 2521.01 mg a.s./kg feeding solution			
NOEDD	≥ 57.21 µg a.s./bee/day			
LC_{50} [95 % IC]	> 2521.01 mg a.s./kg feeding solution [not determined]			
LDD_{50} [95 % IC]	> 57.21 µg a.s./bee/day [not determined]			

Findings:

- The actual mean concentrations of mesotrione in all test item feeding solutions were in the range from 83.8 to 113.2 % of the nominal concentrations; therefore results are based on nominal.
- In control groups fed with pure 50 % (w/v) aqueous sucrose solution and 50 % (w/v) aqueous sucrose solution containing 5 % acetone, no mortality was observed at the final assessment after 10 days.
- The maximum mortality which was observed in the test item treatments concentration of 157.56, 630.25 and 1260.50 mg a.s./kg feeding solution was 2.00 % after 10 days.
- At the concentrations of 157.56, 315.13, 630.25, 1260.50 and 2521.01 mg a.s./kg feeding solution 2.00, 0.00, 2.00, 2.00 and 0.00 % mortality. Since no control(s) mortality occurred, no corrected mortality was calculated.
- Bees were recorded as affected, apathetic and moribund in the test item treatment group at all concentrations on different assessment days from day 1 until the end of the 10 day test period.
- The overall mean daily consumption of feeding solutions (i.e. the average consumption/bee over 10 days) in the test item concentrations of 157.56, 315.13, 630.25, 1260.50 and 2521.01 mg a.s./kg feeding solution was 20.81, 19.63, 18.96, 19.63 and 19.07 µL/bee/day, respectively. For the control and solvent control group 18.67 and 20.02 µL/bee/day, respectively. The values of food consumption were corrected for evaporation.
- After 10 days of continuous exposure, the mean accumulated uptake of mesotrione at the test item concentrations of 157.56, 315.13, 630.25, 1260.50 and 2521.01 mg a.s./kg feeding solution was 39.02, 73.62, 142.19, 294.38 and 572.10 µg a.s./bee, respectively. The corresponding average daily dose (DD) was therefore 3.90, 7.36, 14.22, 29.44 and 57.21 µg a.s./bee/day
- The NOEC for mortality after 10 days of continuous exposure was empirically estimated to be greater than or equal to 2521.01 mg a.s./kg feeding solution. The corresponding NOEDD, based on the actual consumption of the feeding solutions, was empirically estimated to be greater than or equal to 57.21 µg a.s./bee/day.
- After 10 days of continuous exposure, the LC50 was empirically estimated to be greater than 2521.01 mg a.s./kg feeding solution. The corresponding LDD50, based on the actual consumption of the feeding solutions, was empirically estimated to be greater than 57.21 µg a.s./bee/day.

The following validity criteria were met during the test:

- The mean mortality in the control was ≤ 15 % at the end of the test (actual 0.00 % for control and control solvent).
- The mean mortality in the reference item group was ≥50 % at the end of the test (actual 100.00 %)

Conclusion

The chronic toxicity of Mesotrione Technical to honey bees was tested under laboratory conditions over a period of 10 days.
The actual mean concentrations of Mesotrione in test item feeding solutions were in the range of 83.8 to 113.2 % of the nominal concentrations; therefore results are based on nominal.
The 10-day NOEC was empirically estimated to be greater than or equal to 2521.01 mg a.s./kg feeding solution.
The 10-day NOEDD was empirically estimated to be greater than or equal to 57.21 µg a.s./bee/day.
The 10-day LC50 was empirically estimated to be greater than 2521.01 mg a.s./kg feeding solution.
The 10-day LDD50 w was empirically estimated to be greater than 57.21 µg a.s./bee/day.
The test was deemed valid since all validity criteria were met.

A 2.3.1.3 KCP 10.3.1.3 Effects on honey bee development and other honey bee life stages

~~No new study submitted.~~

Comments of zRMS:	The study was conducted to OECD guideline and according to the principles of GLP. All validity criteria were fulfilled. The study is considered to be reliable.

Reference:	KCP 10.3.1.3
Report	"MesotrioneTechnical – Repeated exposure of honeyBee (*Apis mellifera* L.) larvae under laboratory conditions (in vitro)", Kathrin Scheller, 2018, Study code 17 48 BLC 0088
Guideline(s):	Guidance Document on Honey Bee Larval Toxicity Test following Repeated Exposure, Series on Testing and Assessment, No. 239, OECD (2016) with adaptations based on SCHMEHL et al. (2016)
Deviations:	No
GLP:	Yes
Acceptability:	Yes

Materials and methods

The objective of this study was to determine the effects of Mesotrione Technical (batch SCL-62581 on the honey bee larvae, Apis mellifera L., from repeated feeding exposure in an 22 day
in vitro test and to determine the No Observed Effect Dose/Concentration (NOED, NOEC), the Lowest Observed Effect Dose / Concentration (LOED, LOEC) and the corresponding Median Effect Dose/Concentration (ED_{50}, EC_{50}) and any ED_x/EC_x for day 22, where possible.

The test species was honey bee – Apis mellifera iberiensis Engel (Hymenoptera, Apoidea): first instar larvae; derived from three healthy and queen-right colonies; source: Beekeeper Joaquin Cordero, Paseo de Colón No. 19, 41370 Cazalla (Sevilla), Spain.

The test was conducted as a dose response test with a duration of 22 days from grafting on day 1
(D1) to the final assessment on day 22 (D22); from day 3 (D3) until day 6 (D6) of the test, test item (Mesotrione Technical) and reference item (dimethoate) were dissolved in the appropriate larval diet and provided to larvae once a day. The analysed Mesotrione content was used to calculate the test item doses and dimethoate content was used to calculate the reference item dose.

The experimental groups were: 1 untreated control group, 1 untreated control solvent group, 5 test item groups and 1 reference item group. The control groups and treated groups were exposed for the same period of time under identical conditions. Each treatment group consisted of 48 larvae from 3 different colonies (each colony representing a replicate); mortality assessments were performed on day 4 (D4), day 5 (D5), day 6 (D6), day 7 (D7), day 8 (D8), day 15 (D15) and day 22 (D22); the presence of uneaten food was qualitatively recorded on day 8 (D8).

The test concentrations were: 1 control group, 1 control solvent group (acetone 0.5 %), 5 test item groups with 317, 105, 35, 11 and 4 mg a.s./kg diet, equivalent to cumulative doses of 50.1, 16.5, 5.5, 1.8 and 0.6 µg a.s./larva; 1 reference item group with 48.00 mg dimethoate/kg diet, equivalent to a cumulative dose of 7.6 µg dimethoate/larva.

The test conditions were: Air temperature: Min / Max: 34.0 / 34.8 °C; Relative air humidity: D1 - D8: 90.1 – 99.9%; D8-D15: 80.1 – 84.4%; D15-D22: 61.4 – 65.9%. Exposure to light: constant darkness except during feeding and assessments.

Statistical calculations were made with Step-down Cochran-Armitage Test (one-sided greater, alpha = 0.05) for determination of NOED/NOEC. ED/ECx values were determined using the Probit analysis with linear maximum likelihood regression

Results and discussions

Table 2.3.1.1.2-1: Cumulative mortality, overall mean consumption of feeding solution, dietary dose (DD), accumulated mean uptake, NOEC, NOEDD, LOEC, LOEDD, LC_{50} and LDD_{50}

Endpoint	**µg a.s./larva**
22-Day NOED[1]	1.8
22-Day ED_{10} [95 % I.C.]	2.3 (1.4-3.8)
22-Day ED_{20} [95 % I.C.]	4.3 (2.9-6.4)
22-Day ED_{50} [95 % I.C.]	14.3 (10.2-20.2)
Endpoint	**mg a.s./kg diet**
22-Day NOEC[1]	11
22-Day EC_{10} [95 % I.C.]	14 (9-24)
22-Day EC_{20} [95 % I.C.]	27 (18-40)
22-Day EC_{50} [95 % I.C.]	91 (64-128)

[1]Step-down Cochran-Armitage Test Procedure
*Empirically estimated since no greater mortality than 50% occurred.

Findings:

- The actual mean concentrations of Acetamiprid Technical in test item application solutions were 80 and 108 % of the nominal concentrations; therefore results are based on nominal.
- In control and control solvent groups, cumulative larval mortality from day 4 (D4) until day 8 (D8) was 0.00 %. On day 22 (D22), the adult emergence rate in control and control solvent groups was 75.0 and 72.2 % of the initial grafted larvae, respectively. Cumulative mortality in the Reference Item treatment group was 63.9 % by D8. Therefore, the validity criteria were met.
- At day 8 (D8) of the test in the test item doses of 50.1, 16.5, 5.5, 1.8 and 0.6 µg a.s./larva, the cumulative mean mortality were; 0.0, 0.0, 0.0, 27.8 and 33.3% respectively. Larvae with presence of uneaten food were not observed.
- In the test item doses of 50.1, 16.5, 5.5, 1.8 and 0.6 µg a.s./larva, the cumulative mean mortalities at 22 days (D22) after grafting were 86.1, 63.9, 50.0, 33.3 and 25.0 % respectively. In consequence the mean emergence rates were 13.9, 36.1, 50.0, 66.7 and 75.0% respectively.

- Regarding EC_{50} / ED_{50} values on day 22, endpoints were calculated to be 91 mg a.s./kg diet and 14.3 µg a.s./larva respectively. The ED_{20} (successful adult emergence up to D22) was calculated to be 4.3 µg a.i./larva, which is equivalent to an EC_{20} of 27 mg a.i./kg food. The ED_{10} (successful adult emergence up to D22) was calculated to be 2.3 µg a.i./larva, which is equivalent to an EC_{10} of 14 mg a.i./kg food. The NOED determined to be 1.8 µg a.i./larva and the corresponding NOEC was determined to be 11 mg a.i./kg food.

The following validity criteria were met during the test:

- The cumulative larval mortality from day 4 (D4) to the day 8 (D8) was ≤ 15% across all replicates in control group and control solvent group.
- On day 22 (D22) the adult emergence rate was ≥ 70% across all replicates
- The cumulative larval mortality was ≥ 50 % across all replicates on day 8 (D8) and on day 22 (D22)

Conclusion

The repeated exposure of MesotrioneTechnical to honey bee larval was tested under laboratory conditions over a period of 22 days.
The ED_{50} (successful adult emergence up to D22) was calculated to be 14.3 µg a.i./larva, which is equivalent to an EC_{50} of 91 mg a.i./kg food.
The ED_{20} (D22) was determined to be 4.3 µg a.i./larva, which is equivalent to an EC_{20} of 27 mg a.i./kg food.
The ED_{10} (D22) was determined to be 2.3 µg a.i./larva, which is equivalent to an EC_{10} of 14 mg a.i./kg food.
The NOED was 1.8 µg a.i./larva and the corresponding NOEC was 11 mg a.i./kg food.
All validity criteria were met (cumulative mortality in control and reference treatment groups and adult emergence in the control).

A 2.3.1.4 KCP 10.3.1.4 Sub-lethal effects

No new study submitted.

A 2.3.1.5 KCP 10.3.1.5 Cage and tunnel tests

No new study submitted.

A 2.3.1.6 KCP 10.3.1.6 Field tests with honeybees

No new study submitted.

A 2.3.2 KCP 10.3.2 Effects on non-target arthropods other than bees

A 2.3.2.1 KCP 10.3.2.1 Standard laboratory testing for non-target arthropods

No data submitted.

A 2.3.2.2 KCP 10.3.2.2 Extended laboratory testing, aged residue with non-target arthropods

Comments of zRMS:	The study follows the guideline specified by Mead Briggs *et al.* and according to

	the principles of GLP. The following validity criteria were met: – after 48 hours mortality of the control group was 3.3% (criterion: a maximum of 10.0%), – after 48 hours mortality of the group treated with the reference item at the rate of 5.0 mL/ha was 62.1% (criterion: a minimum of 50%), – all wasps survived the 24-hour oviposition period (criterion: only wasps that survive oviposition can be examined for fecundity), – the mean number of mummies per female in the control group was 11.4 (criterion: a minimum of 5.0 mummies/female), – all wasps in the control group gave offspring (criterion: a maximum of 2 females giving no offspring). The study is considered to be valid.

Reference:	KCP 10.3.2.2-01
Report	An extended laboratory test for evaluating the effects of Mesotrione 10% SC on the parasitic wasp, *Aphidius rhopalosiphi* (De Stefani - Perez), Paweł Parma, 2016, Study code B/131/15. Institute of Industrial Organic Chemistry Branch Pszczyna.
Guideline(s):	Yes (ESCORT 1 (Barrett K.L. et al., 1994) and the ESCORT 2 (Candolfi M.P. et al., 2001) guidance documents and the guidelines developed by the IOBC, BART, and EPPO Joint Initiative (Mead-Briggs M.A. et al., 2000; Mead-Briggs M.A. et al., 2010))
Deviations:	Yes (the date of ending the study was a deviations from the Study Plan. It had no impact on the results).
GLP:	Yes
Acceptability:	Yes

Materials and methods

Materials

Test item:	
Description:	Mesotrione 10% SC
Production batch:	SWEPL - 41203
Active ingredients content:	Mesotrione 102 g/Kg
Vehicle and control:	-
Test system:	
Species:	*Aphidius rhopalosiphi*
Strain:	De Stefani-Perez
Age:	adult females (24 - 48 hours after emerging from mummies)
Source:	a laboratory culture at the Institute of Industrial Organic Chemistry, Branch Pszczyna; the culture was obtained from Katz Biotech AG (Baruth, Germany)
Acclimation period:	Not required
Diet:	fructose solution (supplementary food and attractant)
Experimental conditions:	
Test medium:	A test unit consisted of a transparent PMMA cylinder (isolator) with a diameter of 11 cm and a height of 20 cm put on a plastic pot with a diameter of 12 cm. When assessing mortality, the pots contained 7-day-old barley seedlings (8 seedlings per pot), whereas when assessing fecundity, the pots

	contained approximately 20 seedlings of 7-day-old barley infested with the bird cherry-oat aphid, *Rhopalosiphum padi* (> 100 aphids per pot). To provide good ventilation, the apex of each cylinder and two longitudinal openings on its two sides are covered with fine metal netting. There is a hole in the cylinder enabling introduction of the wasps to the test units. This port is filled with a cotton wool bung soaked with a 10% solution of fructose in water (w/v).
Temperature:	18-21°C
Humidity:	65-89%
Air changes:	-
Light and photoperiod:	16 hours light (mortality assessment and oviposition: 1017lx; fecundity assessment: 4740 lx) : 8 hours dark.

Study design and methods

Experimental period:	02.03.2016-16.03.2016
Test design and treatment:	5 test groups: – control (0.0 kg/ha) – Mesotrione 10% SC at the rate of 0.375 kg/ha (38.3 g a.i./ha) – Mesotrione 10% SC at the rate of 0.75 kg/ha (76.5 g a.i./ha) – Mesotrione 10% SC at the rate of 1.5 kg/ha (153.0 g a.i./ha) – Bi 58 Nowy 400 EC at the rate of 5.0 mL/ha (2.0 g a.i./ha) 6 replicates/group; 5 females/replicate The settling behaviour of the wasps in the test units was observed during the initial 3 hours (started 30 min after introduction of the was and took place every 30 min). The conditions of the wasps was observed 2, 24 and 48 hours after being introduced into the test units. After 48 h, 15 females from control group and 15 females from all groups treated with the test item were introduced into the fecundity units and after 24 h (oviposition) all surviving wasps were removed whereas the parasitized aphids were left for the next 12 days.
Statistics:	Shapiro-Wilk's test on normal distribution, Levene's test on variance homogeneity, one-way analysis of variances ANOVA, Chi2 2x2 table test with Bonferroni correction, probit analysis, Dunnett's Multiple t test procedure.

Results are summarized in the table below

Study group [application rate]		Parameter (endpoint)							
		Mortality				Fecundity			
Test item		Total [%]	Corrected*	LR_{50}		mean no. of mummies/ female	fecundity reduction Pr [%]	ER_{50}	
[kg/ha] [a]	[g a.i./ha] [c]			[kg/ha] [a]	[g a.i./ha] [c]			[kg/ha] [a]	[g a.i./ha] [c]
Control (0.0)		3.3	-	-		11.4	-	-	
0.375	38.3	6.7	3.5	> 1.5	> 153.0	12.3	(-8.2) [d]	> 1.5	> 153.0
0.75	76.5	13.3	10.3			13.5	(-18.1) [d]		
1.5	153	16.7	13.8			9.0	21.1		
$NOER_{mortality}$				≥ 1.5	≥ 153.0	$NOER_{fecundity}$		≥ 1.5	≥ 153.0
Reference item		-							
[mL/ha] [b]	[g a.i./ha] [c]								
5.0	2.0	62.1	not determined			not assessed			

[a]: [kg test item/ha]
[b]: [mL reference item/ha]
[c]: [g active ingredient/ha]
[d]: the negative value shows that the mean number of mummies was higher than in the control group
*: Abbott corrected mortality [11]

Conclusions:

The validity criterion concerning mortality was met, because mortality of the wasps in the control group was ≤ 10.0% after 48 hours of the exposure. Mortality of the control wasps and the ones exposed to Mesotrione 10% SC was equal to 3.3%.
The LR_{50} value (the application rate at which 50% mortality of wasps is observed) could not be determined on the basis of the obtained mortality results. The LR_{50} can be assumed to be > 1.5 kg Mesotrione 10% SC/ha (> 153.0 g a.i./ha).

The percentages of fecundity reduction (Pr) caused by Mesotrione 10% SC at the rates of 0.375, 0.75 and 1.5 kg/ha (38.25, 76.5 and 153.0 g a.i./ha) were (-8.2), (-18.1) and 21.1%,, respectively. The negative value shows that the mean number of mummies per female was higher in the group treated with the test item than in the control group. At the significance level of 0.05, there were no statistically significant differences in fecundity between the wasps exposed to the test item at the rates of 0.375, 0.75 and 1.5 kg/ha (38.25, 76.5 and 153.0 g a.i./ha) and the control group. On the basis of the obtained fecundity results, the ER_{50} and the $NOER_{fecundity}$ values could not be determined. It can be concluded that the ER_{50} appears to be higher than the maximum rate used in the fecundity assessment, i.e. > 1.5 kg/ha (153.0 g a.i./ha) and the $NOER_{fecundity}$ is higher than or equal to 1.5 kg /ha (153.0 g a.i./ha).

On the basis of the obtained results it can be concluded that Mesotrione 10% SC at all the rates of 0.375, 0.75 and 1.5 kg/ha (38.25, 76.5 and 153.0 g a.i./ha) has no adverse effect on mortality and fecundity of the wasps.

Comments of zRMS:	The study follows the guideline specified by Blümel *et al.* (2000) with modifications to create extended laboratory conditions and according to the principles of GLP. The following validity criteria were met: - mortality of the control group was 8.3% on day 7 of exposure (criterion: a

	maximum of 20%), - corrected mortality of the mites exposed to the reference item at the rate of 9.0 mL/ha was 89.1% on day 7 of exposure (criterion: a minimum of 50%), - the mean number of eggs per female in the control group was 5.8 (required: ≥ 4 eggs per female). The study is considered to be valid.

Reference:	KCP 10.3.2.2-03
Report	"An extended laboratory test for evaluating the effects of Mesotrione 10% SC on the predatory mite, *Typhlodromus pyri* (Sch.)", Parma, P. 2017.Study code: B/130/15. Institute of Industrial Organic Chemistry Branch Pszczyna
Guideline(s):	ESCORT 1 (Barrett K.L. et al., 1994) and the ESCORT 2 (Candolfi M.P. *et al.*, 2001) guidance documents and the guidelines developed by the IOBC, BART, and EPPO Joint Initiative (Blümel S. *et al.*, 2000)
Deviations:	No.
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:	name: Mesotrione 10% SC; content: 10.2% (w/w) of mesotrione as an active ingredient; batch number: SWEPL - 41203; manufacturing date: February 21, 2015; expiry date: February 20, 2017
Biological test system:	the predatory mite, *Typhlodromus pyri* (Sch.) (Acari: *Phytoseiidae*)
– age:	24-hour-old protonymphs
– source:	a laboratory culture at the Institute of Industrial Organic Chemistry, Branch Pszczyna [SOP/B/33]; the culture was obtained from the Research Institute of Pomology and Floriculture, Skierniewice, Poland
Experimental design:	6 study groups: – a control group (0.0 g/ha) – Mesotrione 10% SC at the rate of 0.375 kg/ha (38.3 g a.i./ha) – Mesotrione 10% SC at the rate of 0.75 kg/ha (76.5 g a.i./ha) – Mesotrione 10% SC at the rate of 1.5 kg/ha (153.0 g a.i./ha) – Mesotrione 10% SC at the rate of 3.0 kg/ha (306.0 g a.i./ha) – Bi 58 Nowy 400 EC at the rate of 9.0 mL/ha (3.6 g a.i./ha) number of replicates: 3; number of mites in each replicate: 20
Test conditions:	
– temperature:	24.0 - 26.0°C
– relative air humidity:	65 - 82%
– photoperiod:	16 h light (816 lux) : 8 h dark
Statistical analysis:	Shapiro-Wilk's test on normal distribution, Levene's test on variance homogeneity, probit analysis, step-down Cochran-Armitage test procedure, Williams Multiple Sequential t-test procedure,
Endpoints:	– mite mortality after 7 days of the treatment – LR_{50}, $NOER_{mortality}$ – reproduction reduction (Pr) after 14 days of the treatment – ER_{50}, $NOER_{reproduction}$

Results and discussions

Study group [application rate]		Parameter (endpoints)							
		Mortality				Reproduction			
Test item		Total (%)*	Corrected	LR_{50}		Mean number of eggs/female (Rr) (no.)	Reproduction reduction Pr (%)	ER_{50}	
(kg/ha) [a]	(g a.i./ha) [b]			(kg/ha) [a]	(g a.i./ha) [b]			(g/ha)[a]	(g a.i./ha) [b]
Control (0.0)		8.3	-	-		5.8	-	-	
0.375	38.3	10.0	1.8	2.1 (1.7-2.6)	214.2	1.3+	77.0	<0.375	<38.3
0.75	76.5	23.3+	16.4			1.3+	77.8		
1.5	153.0	43.3+	38.2			0.7+	87.4		
3.0	306.0	66.7+	63.6			Not assessed			
$NOER_{mortality}$				0.375	38.3	$NOER_{reproduction}$		<0.375	<38.3
Reference item		-							
(ml/ha)[a]	(g a.i./ha)[b]								
9.0	3.6	89.1	Not determined			Not assessed			

a: [kg test item/ha]
b: [g active ingredient/ha]
c: [mL reference item/ha]
*: mortality corrected using the formula of Abbott [1]
+: statistically significant difference

Conclusion

On the basis of the obtained results it can be concluded that Mesotrione 10% SC at the rates of 0.75, 1.5 and 3.0 kg/ha (76.5, 153.0 and 306.0 g a.i./ha) has an adverse effect on mortality of the mites and at rates of 0.375, 0.75 and 1.5 kg/ha (38.3, 76.5 and 153.0 g a.i./ha) has an adverse effect on reproduction.

Comments of zRMS:	The study follows the current guidelines and according to the principles of GLP. No deviations to the guideline were noted. The following validity criteria were met: - The maximum mortality (dead and escaped individuals) in the control was 17.0% on day 7 of exposure (criterion: a maximum of 20%), - Minimum mortality (corrected to control) in the toxic reference was 87.21 % at the studied ageing - More than 4 eggs per female in the control treatments were achieved (actual values ranged between 5.80 and 7.86 eggs per female) The study is considered to be valid.

Reference: KCP 10.3.2.2-04

Report "Aged residue test with the formulation Mesotrione 10% SC on the predatory mite *Typhlodromus pyri* (Acari: Phytoseiidae)". Francisco Luna. 2017. Study code: TRC17-090BA. Trialcamp S.L.U.

Guideline(s): IOBC WPRS/SROP (Blümel S. et al., 2000; Candolfi et al., 2000)

Deviations: Yes. 1. Reproduction in the exposure of 14 DAA was studied with 4 replicates instead of 5 in the test product treatment because one leaf fragment was damaged and the evaluation was not possible to be performed from day 11 after the exposure.
2. Final report signature has been delayed with respect to the indicated in the

study plan.
3. Mortality assessments were carried out at 1 and 7 days after each exposure. The Study Plan indicates, by mistake, “at 0, 7 and 14 days of each exposure”.

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Summary

The aim of the study was to evaluate the extent and persistence of effects on the survival and reproduction of the formulation Mesotrione 10% SC to *Typhlodromus pyri* Scheuten (Acari: Phytoseiidae), exposing the test organisms to treated maize leaves after different periods of ageing of the residues under outdoors conditions (with a roof closed only when it rains). The effects were evaluated with the rate of 150 g/ha of active substance (a.s.).
The study was carried out under GLP conditions and according to OECD Good Laboratory Practice Standards. The method used is based on the description of Blümel S. et al. (2000) with the exception of the substrate. The trial was codified as TRC17-090BA.

Material and methods

Test item: Mesotrione 10% SC
Nominal content: 100 g Mesotrione /L
Purity: 104 g Mesotrione /L
Batch No.: SCL-34891
Expiry date: March 11th, 2019

Biological test system: protonymphs of the parasitoid the predatory mite *T. pyri* Scheuten (Acari: Phytoseiidae)
– Age: Not older than 24 hours from moulting
– Source: From an in-house culture started with supplied eggs by Katz Biotech Ag. (Baruth – Germany)

Experimental design:

Code	Treatment	Application rate [g a.s./ha](1)	Application rate(1) [L product /ha]
C	Water (Control)	-	-
T1	Mesotrione 10% SC	150.0	1.4423
R (A1)*	Deltamethrin 2.5 % EC	12.20	0.500

(1): Rate of the formulated product (FP) according to the analytical certificate: Mesotrione 104 g/L. Purity of deltamethrin: 24.4 g/L.

Maize plants (Zea mays) of the variety AGROSTAR were used for trial purposes. Three plots with 10 potted plants per plot were used: One plot for water treated control, one plots for the test product and one plot for the toxic reference. The treated plot size was 10.5 m2 (15 m x 0.7 m) for the treatments and the plants were arranged in one crop row.
In order to select treated leaves with the same age for the different exposures, the youngest leaves were marked to avoid samplings of new (non-treated) or developing leaves (diluted residues) at the exposure periods.
A correct application of all leaves available was achieved thanks to the dispersed foliage of the plants at the time of the application.
Application was performed using a compressed air knapsack sprayer and one nozzle “Albuz Hollow Cone Lilac ATR-80” simulating an application in field (volume 600 L/ha) and applying the plants by 2-sides.

After application, plants were maintained outdoors to allow "natural" weathering of the test product residues.
The reference product was applied once just after the test product.

Statistics

Results of 7-d mortality and 7-14-d fecundity (eggs per female) were analysed with the Shapiro-Wilk test for normality of data distribution and with the Levene's test for homoscedasticity (Annex IV). The parametric T-test with Levene's test for equality of variances (α=0.05) were performed in order to study significant differences between the test product treatment and control according to the normality of data when mortality was higher or fecundity was lower than in the control treatment. No statistical analysis was performed with results in the test reference treatment.

Endpoints

- To study the mortality at 7 days after exposure (lethal effect) to residues on leaves aged for the following periods: 0, 7 and 14 days after application (DAA).
- To study the fecundity of the survivor females during 7 days following exposure to residues on leaves for the aforementioned ageing periods.

Results

Mortality
Based on mortalities being less than 20 % at the end of all exposure periods (actual maximum value was 17.0 % in the exposure of 0 DAA), reproductive performances above 4 eggs per female at the fecundity assessments 0, 7 and 14 DAA in the control (actual minimum value was 5.80 eggs per female in the exposure of 0 DAA) and a corrected mortality greater than 50 % in the toxic reference until the exposure of 14 DAA (87.21 % as minimum corrected mortality), the sensitivity of the test species and the suitability of the test system was confirmed and the study can be regarded to be valid.
The mortalities at the different bioassays are shown in Table. With fresh and dry residues (exposure of 0 DAA) and aged residues of 7 and 14 days, corrected mortality was less than 50 % with the tested rate of 150.0 g a.s./ha.
The mortality was not statistically significant higher than control (t-Test, α=0.05) at any studied period, and the corrected mortalities were always below 50 %; maximum corrected mortality compared to control was 4.82 % with fresh and dry residues.

			Exposure					
Code	Treatment	Rate	0 DAA(1)		14 DAA		28 DAA	
		[g a.s./ha]	% M	[%] Cm	% M	[%]Cm	% M	[%]Cm(2)
C	Control (water)	-	17.00	-	14.00	-	14.00	-
T1	Mesotrione 10% SC	150.00	21.00	4.82	13.00	-1.16	15.00	1.16
R	Deltamethrin 2.5 %, EC	12.20	100.00	100.00	95.00	94.19	89.00	87.21

(1): DAA = Days after application; M [%] = Mortality [%]; Cm [%] = Corrected mortality [%]
(2): Negative value indicates a decrease relative to the control

Fecundity
The reduction of number of eggs/female was below the ESCORT 2 trigger value of 50 % in the bioassays performed of 0, 7 and 14 DAA at the tested rate of 150.0 g a.s./ha.
Reproduction amounted to be 9.30 % reduction relative to control by fresh and dry residue but without significant differences (T-test, α=0.05) compared to the control.

			Exposure					
Code	Treatment	Rate	0 DAA(1)		14 DAA		28 DAA	
		[kg a.s./ha]	e/f(2)	[%] R	e/f	[%] R	e/f	[%] R(3)
C	Control (water)	-	5.80	-	6.71	-	7.86	-

T1	Mesotrione 10% SC	150.0	5.26	9.30	7.19	-7.20	7.24	7.93

(1): DAA = Days after application; "e/f"= eggs per female (mean); [%] R = Reduction [%]
(3): Negative value indicates an increase relative to the control

Test validity criteria
According to the guideline (Blümel et al. 2000) and the study plan, the test should be considered to be valid if:

- Mortality in the control treatment did not exceed 20 %
- Mortality in the reference treatment was in a range from 50 % to 100 %, at least until the exposure of 14 DAA
- More than 4 eggs per female in the control treatments were achieved

Conclusion
Based on the results of the present study it can be concluded that residues of the test product Mesotrione 10% SC applied to the rate of 150.0 g a.s./ha (equivalent to 1.4423 L of formulated product/ha according to the analysed content) causes mortality less than 50 % compared to the control and has less than 50 % reduction on the reproduction of Typhlodromus pyri from 0 days after the application; fresh and dry Residues.

Comments of zRMS:	The study follows the guideline specified by Grimm et al. (2000) and according to the principles of GLP. The following validity criteria were met: - Number of beetles emerging from the fly pupae should be above 400 in control (average of 831 adults was obtained from the 1500 fly pupae introduced per replicate). - A minimum reduction of 50 % reproductive capacity relative to the control should be achieved in the reference item treatment (obtained: 87.1%) The study is considered to be valid.

Reference:	KCP 10.3.2.2-04
Report	"Side-effects of the formulated product Mesotrione 10% SC on *Aleochara bilineata* (Coleoptera:Staphylinidae) under extended laboratory conditions". Eugenia Soler, 2017, Study code: TRC17-115BA. TRIALCAMP S.L.U.
Guideline(s):	Yes: A test for evaluating the chronic effect of plant protection products on the rove beetle *Aleochara bilineata* Gyll. (Coleoptera: Staphylinidae) under laboratory and extended laboratory conditions"; Grimm et al., 2000 and ESCORT workshops for higher tier testing (Barrett et al., 1994 and Candolfi et al., 2000)
Deviations:	Yes. 1. Introduction of the Delia antiqua at +7DAA (days after the application) was done at +8DAA due to a delay on the shipment. 2. Delay on the final report. This deviation did not have any negative impact on the outcome of the study.
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Materials

Test item:

Description:	Mesotrione 10% SC
Production batch:	SCL - 34891
Active ingredients content:	Mesotrione 10% w/v (nominal) Mesotrione 10.4% w/v (analytical)
Vehicle and control:	De-ionized water

Test system:

Species:	*Aleochara bilineata*
Age:	At the application date adult beetles 2 -5 days old.
Source:	The batch of larvae used in this study came from the supplier "De Groene Vlieg", reliable commercial supplier, which has permanent rearing and select A. bilineata according to the requirements for ecotoxicological assays.

Experimental conditions:

Test substrate:	LUFA 2.1 (86.2 ± 0.7% sand, 10.7 ± 1.2% silt, 3.1 ± 0.9% clay; 0.68 ± 0.07% organic carbon)
Test units:	The test units used to assess beetle mortality and fecundity consisted of glass containers (135x135x62 mm), with a base surface area of 182.25 cm^2 filled with a sandy soil (LUFA 2.1) layer 4 cm deep (760.80 g of wet LUFA 2.1). Each test unit was covered with a lid where an opening of approximately 100x100 mm was practiced to provide ventilation. The opening was covered with a fine mesh nylon netting (approximately 0.5x0.5 mm).
Temperature:	19.9 – 21.3 °C
Humidity:	77.0 – 88.6 %
Light and photoperiod:	1019.70 to 1246.30 lux.

Experimental period:
14/06/2017 – 28/08/2017

Summary:

A laboratory study was carried out to evaluate the effects of residues of freshly applied formulated product Mesotione 10%, SC on the rove beetle Aleochara bilineata (Coleoptera: Staphylinidae).

The study was carried out under GLP conditions and according to OECD Good Laboratory Practice Standards. The study was conducted in accordance with the recommendations of the SETAC/ESCORT 2 workshops (Candolfi et al., 2000). The method is based on guidelines proposed by Grimm et al., 2000.

The test substance Mesotrione 10%, batch "SCL-34891", was tested in a range of 5 different rates in a geometric series, with a spacing factor of 2 (0.3750 to 6.0000 L FP/ha). A reference product treatment, Dimethoate 40%, was tested at the rate of 1.1 L FP/ha. A water control group was also concurrently tested.

Application was performed with a Laboratory Track Sprayer (Schachtner). The Schachtner was calibrated to deliver a target of 4 ± 0.4 mg spray solution/cm² corresponding to 400 L/ha.

Tests exposing Aleochara bilineata under laboratory conditions to the treated soil were started immediately after the test substance was mixed with the substrate. Effect on reproductive capacity was used as the toxic endpoint.

A group of ten adult females and ten adult males were placed in each of the test units. The test units were held for the duration of the study in an environment controlled room at a temperature of 20±2ºC, a 16:8 light/dark photoperiod and a light intensity < 2000 lux.

Pupae of the onion fly Delia antiqua were added to the test substrate during the three weeks following the beginning of exposure as hosts for the A. bileneata larvae. The adult beetles were removed from the substrate after four weeks. The fly pupae were placed into the hatching units and the numbers of second generation adult beetles were recorded.

Under the conditions of the present study it can be concluded that:

The validity criteria were achieved since the average number of beetles emerged from the fly pupae was above 400 in the control treatment, and the reduction in the reproductive capacity relative to the control was greater than 50% with the maximum tested rate of the reference product.

The test substance Mesotione 10%, SC was tested with rates between 0.3750 and 6.0000 L FP/ha and the reference substance (Dimethoate 40% EC) was tested at 1.1 L FP/ha.

Adults' survival of the rove beetle Aleochara bilineata was not significantly affected (1.39% corrected mortality relative to the control; Bonferroni Chi2 test) by the maximum tested rate of 6.0000 L FP/ha, equivalent to 624 g Mesotrione/L (according to the certificate of analysis).

Offspring production of the rove beetle Aleochara bilineata was not significantly affected (19.7% of reduction relative to the control; Williams Multiple Sequential t-test) by the maximum tested rate 6.0000 L FP/ha; equivalent to 624g Mesotrione/L (according to the certificate of analysis).

Thus, the NOEC for both survival and reproductive output was 6.0000 L FP/ha (equivalent to 624 g Mesotrione/L).

Treatment		Rate [L FP/ha]	Mortality [%]	Corrected Mortality [%]	Offspring (Mean of adults emerged)	Reduction (relative to the control) [%]
C	Deionised water	0	10.00	--	83	--
T1	Mesotrione 10%, SC	0.3750	12.50	2.78	84	-1.6
T2		0.7500	10.00	0.00	76	8.8
T3		1.5000	10.00	0.00	70	15.7
T4		3.0000	13.75	4.17	78	5.8
T5		6.0000	11.25	1.39	67	19.7
R	Dimethoate 40 % EC	1.1	100.00	100.00	11	87.1

Conclusion
The validity criteria were achieved since the average number of beetles emerged from the fly pupae was above 400 in the control treatment, and the reduction in the reproductive capacity relative to the control was greater than 50% with the maximum tested rate of the reference product.
The test substance Mesotione 10%, SC was tested with rates between 0.3750 and 6.0000 L FP/ha and the reference substance (Dimethoate 40% EC) was tested at 1.1 L FP/ha.
Adults' survival of the rove beetle Aleochara bilineata was not significantly affected (1.39% corrected mortality relative to the control; Bonferroni Chi2 test) by the maximum tested rate of 6.0000 L FP/ha, equivalent to 624 g Mesotrione/L (according to the certificate of analysis).
Offspring production of the rove beetle Aleochara bilineata was not significantly affected (19.7% of reduction relative to the control; Williams Multiple Sequential t-test) by the maximum tested rate 6.0000 L FP/ha; equivalent to 624g Mesotrione/L (according to the certificate of analysis).
Thus, the NOEC for both survival and reproductive output was 6.0000 L FP/ha (equivalent to 624 g Mesotrione/L).

A 2.3.2.3 KCP 10.3.2.3 Semi-field studies with non-target arthropod

Not required.

A 2.3.2.4 KCP 10.3.2.4 Field studies with non-target arthropods

Not required.

A 2.4 KCP 10.4 Effects on non-target soil meso- and macrofauna

A 2.4.1 KCP 10.4.1 Earthworms

A 2.4.1.1 KCP 10.4.1.1 Earthworms - sub-lethal effects

Comments of zRMS:	The study was conducted to OECD guideline 222 and according to the principles of GLP. In the definitive test all validity criteria were met. One minor deviation to the guideline (described below) were noted. The study is considered to be reliable and suitable for the risk assessment.

Reference: KCP 10.4.1.1

Report "Mesotrione 10% SC Earthworm Reproduction Test (*Eisenia fetida*)". Weronika Dec. 2016, Study code: G/243/15. Institute of Industrial Organic Chemistry Branch Pszczyna.

Guideline(s):	Yes OECD Guideline 222
Deviations:	Yes. Three deviations from the study plan occurred: 1. The temperature in the test room was between 17.5 – 22°C. According to the Study Plan, it should have ranged from 18 to 22°C. It was a short-term deviation (for two hours) which did not affect the result of the experiment. 2. The SUP-30W was used in the experiment instead of the SUP-100G laboratory dryer. 3. The study was finished in December 2016 and not in September 2016.
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	-

Materials and methods

Materials

Test item:	
Description:	Mesotrione 10% SC
Production batch:	SWEPL-41203
A.i. content:	Mesotrione 10.2% w/v
Vehicle and control:	Artificial soil
Test system:	
Species:	*Eisenia fetida*
Strain:	-
Age:	adult (about 6 months old)
Body weight:	262– 324mg
Source:	Standard laboratory culture cultivated at the Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicology, Laboratory of Soil Toxicology.
Acclimation period:	24h
Diet:	During the experiment, the earthworms were fed on air-dried finely ground cow manure. At the beginning of the experiment, it was mixed with the soil substrate (5 g food/ 500 g dry soil). The food prepared in this way was provided once a week during the four-week period (3 - 4 g food/container). After 4 weeks (when the adult earthworms were removed from the soil), the juvenile worms were fed only once (5 g food/container).
Experimental conditions:	
Test medium:	The test was performed in soil (10% sphagnum peat, 20% kaolin clay, 70% air-dried quartz sand) in plastic containers (500 g dry soil/container).
Temperature:	17.5-22.0°C
Humidity:	29.90 – 31.90% (52.82 – 56.35% of the maximum water holding capacity); soil moisture content at the end of the experiment: 28.30 – 30.50% (49.99 – 53.88% of the maximum water holding capacity);
pH:	5.53 – 5.61 (beginning); 5.57 – 5.68 (end)
Air changes:	-

Light and photoperiod:	480 – 535 (16 hours light and 8 hours dark)

Study design and methods

Experimental period:	20/07/16 – 17/08/2016
Test design and treatment:	The experiment lasted 8 weeks. Number of replicates: 4 replicates/concentration + 8 replicates/control. Number of earthworms: 10 earthworms/replicate. The test item in the form of a water solution was mixed with the artificial soil. The concentrations of the test item were a control, 10, 18, 32, 56, 100, 180, 320, 560 and 1000 mg/kg dry soil. After 4 weeks, all adult worms were removed from the test containers and observed. All changes in their behaviour and morphology were recorded. The number of earthworms and their body weights were also determined. The impact of the test item on reproduction was evaluated after an additional 4-week period on the basis of the number of juveniles hatched from cocoons during the experiment. The obtained results served as a basis for the determination of the NOEC and the LOEC A test with a reference substance, carbendazim was conducted between 10.05.2016 – 07.07.2016. Five concentrations of the reference substance were used: 1, 1.5, 2.25, 3.37, and 5 mg/kg dry soil.
Statistics:	EC_{10}, EC_{20}, EC_{50}, LC_{50} – the probit method NOEC (reproduction) – the Shapiro-Wilk's Test on Normal Distribution, the Barlett's Test Procedure on Variance Homogeneity, the Williams Multiple Sequential t-test Procedure NOEC (survival) - Fisher's Exact Binomial Test with Bonferroni Correction LOEC : value suggested by the ToxRat Professional 2.10 statistical computer software.

Results and discussions

Mortality of *Eisenia fetida*	After 4 weeks of the exposure, mortality of the of the earthworms exposed to the test item at the concentrations ranging from 10 to 1000 mg/kg dry weight of artificial soil was not observed. Mortality of the control group was not recorded.
Observations of the earthworms	After 4 weeks of the experiment, the treated earthworms did not exhibit any changes in appearance and behaviour.
Body weight *Eisenia fetida*	After the application of the test item at the concentrations ranging from 10 to 1000 mg/kg dry weight of artificial soil, the body weight increase was between 24.5 – 43.2%. As for the control group, it was equal to 47.5%.
Reproduction of *Eisenia fetida*	After 8 weeks of the exposure, it was concluded that Mesotrione 10% SC had a significant effects on reproduction

of the earthworms at the concentrations ranging from 32 to 1000 mg/kg dry weight of artificial soil.

Results of the reproduction test are in the table below:

Concentration [mg/kg dry weight of artificial soil]	Replicate	Number of juveniles [no.]	Mean ±SD	Comparison to the control [%]	CV* [%]
0 (control)	1 2 3 4 5 6 7 8	133 112 145 122 141 135 145 119	**131.5 ± 12.5**	100	9.5
10	1 2 3 4	107 144 119 124	**123.5 ± 15.4**	93.9	12.5
18	1 2 3 4	148 132 132 136	**137.0 ± 7.6**	104.2	5.5
32	1 2 3 4	112 104 118 125	**114.8* ± 8.9**	87.3	7.8
56	1 2 3 4	103 91 135 111	**110.0* ± 18.6**	83.7	16.9
100	1 2 3 4	87 104 115 103	**102.3* ± 11.5**	77.8	11.3
180	1 2 3 4	89 93 74 87	**85.8* ± 8.2**	65.2	9.6
320	1 2 3 4	106 79 82 93	**90.0* ± 12.2**	68.4	13.6
560	1 2 3 4	69 66 105 90	**82.5* ± 18.4***	62.7	22.3
1000	1 2 3 4	58 59 63 66	**61.5* ± 3.7***	46.8	6.0

* - statistically significant differences (significance was Alpha = 0.05, one-sided smaller)

In order to determine significance of differences between the control and the treated groups, the Shapiro-Wilk's Test on Normal Distribution, Barlett's Test Procedure on Variance Homogeneity, and Williams Multiple Sequential t-test Procedure were used.

* - the coefficient of variation

Endpoints values are in the table below:

Parameter	Value [mg test item/ kg dry soil]	Value [mg active substance/ kg dry soil]
EC_{10}	26.7 (< 10 – 49.9)	2.5 (< 0.9 – 4.7)
EC_{20}	90.9 (48.3 – 137.1)	8.5 (4.5 – 12.8)
EC_{50}	948.9 (604.4 – > 1000)	88.8 (56.6 – > 93.6)
LOEC	32	3.0
NOEC	18	1.7
LC_{50}	>1000	> 93.6

Conclusion

The results are considered valid because the following criteria were satisfied in the controls:each replicate produced 131.5 juveniles (mean) at the end of the experiment - (criterion: ≥ 30 juveniles by the end of the experiment), the coefficient of variation of reproduction was 9.5% (criterion: ≤ 30%) and adult mortality over the initial 4 weeks of the experiment was 0.0% (criterion: ≤ 10%).
The highest concentration at which the test item is observed to have no statistically significant effects on reproduction (NOEC) is equal to 18 mg/kg dry weight of artificial soil (1.7 mg of a.s./kg dry weight of artificial soil). The lowest concentration at which the test item is observed to have a statistically significant effect on reproduction (LOEC) is equal to 32 mg/kg dry weight of artificial soil (3.0 mg of a.s./kg dry weight of artificial soil).

A 2.4.1.2 KCP 10.4.1.2 Earthworms - field studies

No new study submitted.

A 2.4.2 KCP 10.4.2 Effects on non-target soil meso- and macrofauna (other than earthworms)

A 2.4.2.1 KCP 10.4.2.1 Species level testing

Comments of zRMS:	The study was conducted to OECD guideline 232 and according to the principles of GLP. No deviations to the guideline were noted. In the definitive test all the validity criteria were met as: - mean adult mortality was 6.3% (criterion: ≤ 20%), - the mean number of juveniles per vessel was 236.3 at the end of the test (criterion: ≥ 100 juveniles at the end of the test), - the coefficient of variation calculated for the number of juveniles was 5.7% (criterion: ≤ 30%), The study is reliable and suitable for the risk assessment.

Reference:	KCP 10.4.2.1
Report	Mesotrione 10% SC Collembolan (*Folsomia candida*) Reproduction Test, Weronika Dec, 2016, Study code G/244/15. Institute of Industrial Organic Chemistry Branch Pszczyna.
Guideline(s):	Yes (OECD 232)
Deviations:	Yes (from the Study Plan) • The temperature in the test room was between 17 – 22°C. According to the Study Plan, it should have ranged from 18 to 22°C. It was a short-term deviation (for one hour) which did not affect the result of the experiment. • The study was finished in July 2016 and not in June 2016.).
GLP:	Yes
Acceptability:	Yes

Materials and methods

Materials

Test item:	
Description:	Mesotrione 10% SC
Production batch:	SWEPL-41203
Active ingredients content:	mesotrione - 10.2% w/v
Vehicle and control:	Artificial soil
Test system:	
Species:	*Folsomia candida*
Strain:	-
Age:	11 – 12 days old
Body weight:	-
Source:	Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicology, Laboratory of Soil Toxicology, Poland.
Acclimation period:	-
Diet:	Granulated dried baker's yeast once or twice a week.
Experimental conditions:	
Test medium:	The test was performed in Soil (5% sphagnum peat, 20% kaolin clay, and 75% sand).
Temperature:	17 – 22 °C
Humidity:	Soil moisture at the beginning of the experiment: 9.9 – 10.6%; soil moisture at the end of the experiment: 9.6 – 10.3%
pH:	pH at the beginning of the test: 6.10 – 6.46; pH at the end of the test: 6.20 – 6.38
Air changes:	-
Light and photoperiod:	light-dark cycle: 12 h light and 12 h dark; light intensity: 518 – 655 lux

Study design and methods

Experimental period:	02.03.2016 – 30.03.2016

Test design and treatment:	Eight concentrations of the test item were used. These included: 18, 32, 56, 100, 180, 320, 560, and 1000 mg/kg dry weight of artificial soil. The test item in the form of a water solution was mixed with the artificial soil. Each concentration was prepared in four replicates. At the same time, an untreated control group (8 replicates) was conducted. The experiment lasted 28 days. After that, the collembolans were extracted from the soil.. A collembolan was recorded as dead if not present in the extraction. The number of collembolans was counted using a magnifier. The number of adult and juvenile collembolans was determined separately. In order to determine the sensitivity of the test organisms to chemical substances and to verify that the response of the test organisms would not change over time, as test of a reference substance was used boric acid
Statistics:	EC_{10}, EC_{20}, EC_{50} – a probit method NOEC– Shapiro-Wilk's Test on Normal Distribution, Levene's Test on Variance Homogeneity, and Williams Multiple Sequential t-test Procedure (significance of differences)

Results and discussions

After the application of the test item at the concentrations ranging from 18 to 1000 mg/kg dry weight of soil, survival of the collembolans was between 77.5% and 95.0%. It did not depend on concentration of the test item. In case of the control group, survival was equal to 93.8%.
The mean number of juveniles after the application of the test item at the concentrations ranging from 18 to 1000 mg/kg dry weight of soil was between 159–235.8 per replicate. The mean number of juveniles in the control group was equal to 236.3 per replicate.
On the basis of the obtained results the mortality of the adult collembolans after 28 days of the experiment was ranged from 5.0 to 22.5%.

Table 5. Survival of adult collembolans *(Folsomia candida)* after 28 days of the experiment.

Concentration [mg/kg dry soil]	Replicate	Number of tested collembolans	Number of living collembolans after 28 days [no.]	Total	
				No.	%
0 (control)	1	10	10	75	93.8
	2	10	8		
	3	10	10		
	4	10	9		
	5	10	10		
	6	10	10		
	7	10	8		
	8	10	10		
18	1	10	10	38	95.0
	2	10	10		
	3	10	10		
	4	10	8		
32	1	10	10	38	95.0
	2	10	10		
	3	10	10		
	4	10	8		
56	1	10	10	38	95.0
	2	10	10		
	3	10	10		
	4	10	8		
100	1	10	10	36	90.0
	2	10	8		
	3	10	10		
	4	10	8		
180	1	10	10	36	90.0
	2	10	10		
	3	10	8		
	4	10	8		
320	1	10	10	35	87.5
	2	10	9		
	3	10	8		
	4	10	8		
560	1	10	10	33	82.5
	2	10	6		
	3	10	8		
	4	10	9		
1000	1	10	8	31*	77.5
	2	10	7		
	3	10	9		
	4	10	7		

*- statistically significant differences (significance was Alpha = 0.05, one-sided smaller)

In order to determine significance of differences between the control and the treated groups, the Shapiro-Wilk's Test on Normal Distribution, the Levene's Test on Variance Homogeneity, and the Williams Multiple Sequential t-test Procedure were used.

The endpoint values showing the impact of the test item on the survival of adult collembolans are presented in the table given below.

Endpoint	Value [mg/kg of dry soil]	Value [mg a.s./kg of dry soil]
LC_{10}	**473.4** (284.3 – 917.2)	**44.3** (26.6 – 85.8)
LC_{20}	**> 1000** (640.8 – > 1000)	**> 93.6** (60.0 – > 93.6)
LC_{50}	**> 1000**	**> 93.6**
NOEC	**560**	**52.4**
LOEC	**1000**	**93.6**

Table 7. Number of juvenile collembolans *(Folsomia candida)* after 28 days of the experiment.

Concentration [mg/kg dry soil]	Replicate	Number of juveniles	Mean ±SD	Comparison to the control [%]	CV [%]
0 (control)	1 2 3 4 5 6 7 8	222 223 264 236 245 229 235 236	236.3 ± 13.5	**100.0**	5.7
18	1 2 3 4	213 256 238 236	235.8 ± 17.6	**99.8**	7.5
32	1 2 3 4	231 211 244 235	230.3 ± 13.9	**97.5**	6.1
56	1 2 3 4	238 242 236 207	230.8 ± 16.0	**97.7**	6.9
100	1 2 3 4	232 213 222 241	227.0 ± 12.1	**96.1**	5.3
180	1 2 3 4	236 216 245 222	229.8 ± 13.2	**97.2**	5.7
320	1 2 3 4	206 226 240 231	225.8 ± 14.4	**95.6**	6.4
560	1 2 3 4	209 165 185 191	187.5* ± 18.1	**79.4**	9.7
1000	1 2 3 4	184 154 138 160	159.0* ± 19.1	**67.3**	12.0

*CV – coefficient of variation

* - statistically significant differences (significance was Alpha = 0.05, one-sided smaller)

In order to determine significance of differences between the control and the treated groups, the Shapiro-Wilk's Test on Normal Distribution, the Levene's Test on Variance Homogeneity, and the Williams Multiple Sequential t-test Procedure were used.

The endpoint values showing the impact of the test item on reproduction of *Folsomia candida* are presented in the table given below.

Endpoint	Value [mg/kg of dry soil]	Value [mg a.s./kg of dry soil]
EC_{10}	**367.8** (239.2 – 461.6)	**34.4** (22.4 – 43.2)
EC_{20}	**615.8** (500.6 – 722.0)	**57.6** (46.8 – 67.6)
EC_{50}	**> 1000**	**> 93.6**
NOEC	**320**	**29.9**
LOEC	**560**	**52.4**

Conclusion

The concentration of Mesotrione 10% SC causing a 10% reduction in the number of juveniles produced within the exposure period (EC_{10}) is 367.8 mg/kg dry soil.
The concentration of Mesotrione 10% SC causing a 20% reduction in the number of juveniles produced within the exposure period (EC_{20}) is 615.8 mg/kg dry soil.
The concentration of Mesotrione 10% SC causing a 50% reduction in the number of juveniles produced within the exposure period (EC_{50}) is above 1000.0 mg/kg dry soil.
The lowest concentration at which the test item is observed to have statistically significant effects on collembolan reproduction (LOEC) is 560 mg/kg dry soil.
The highest concentration at which the test item is observed to have no statistically significant effects on collembolan reproduction (NOEC) is 320 mg/kg dry soil.

A 2.4.2.2 KCP 10.4.2.2 Higher tier testing

No new study submitted.

A 2.5 KCP 10.5 Effects on soil nitrogen transformation

A 2.5.1 KCP 10.5.1 Soil Microorganisms: Nitrogen Transformation Test

Comments of zRMS:	The study was conducted to OECD guideline 216 and according to the principles of GLP. In the definitive test the validity criteria were met as the coefficients of variation (CV) in the control group were 12.7, 4.5, 2.5 and 1.6%, after 0, 7, 14, and 28 days of incubation. One minor deviation from the guideline (temporary temperature was above 22°C) occurred, which did not affect the result of the experiment. The study is reliable and suitable for the risk assessment.

Reference:	KCP 10.5-01
Report	Mesotrione 10% SC; Soil Microorganisms: Nitrogen Transformation Test, Weronika Dec, 2016, Study code: G/242/15. Institute of Industrial Organic Chemistry Branch Pszczyna.
Guideline(s):	Yes, OECD Guideline No. 216 (2000) / EU Method C.21.
Deviations:	Yes • The temperature in the test room was between 19 – 22.5°C. According to the study plan, it should have ranged from 18 to 22°C. It was a short-term deviation (approximately 12 hours) which did not affect the result of the experiment • The active substance content of the application rate (1.5L/ha = 1635 g of the test item/ha) should be 153 g a.s. of test item/ha, not 166.8 g a.s. of test item/ha. This is an editorial mistake. It did not affect the results.
GLP:	Yes
Acceptability:	Yes

Materials and methods

Mesotrione 10% SC (batch No. SWEPL - 41203) is a cream color liquid formulation containing the active substance Mesotrione (nominal concentration: 10.2% w/v). The test material was introduced into soil (manually cleared of large objects and then sieved to particle size equal to 2mm) at concentrations of 2.2 mg/kg of soil (0.2 mg a.s./kg of soil) – PEC and 11.0 mg/kg of soil (1.0 mg a.s./kg of soil) – 5 x PEC. The control soil and soil treated with test item were divided in three replicates. The soil incubation time was 28 days. On days 0, 7, 14 and 28 of incubation, soil samples were collected to determine the quantities of nitrates. The method involves a measurement of the nitrate ion concentration in a soil extract obtained by using 0.1M KCl. The pH/ION 7320 digital meter and the NO 800 nitrate electrode were used.
In order to determine significance of differences in nitrate transformation between control soil and soil treated with the test item, at the concentrations mentioned above, some statistical analysis was performed using ToxRat 2.10. computer software [17], [SOP/G/80], namely:

- Shapiro-Wilk's test on normal distribution
- Levene's Test on Variance Homogeneity
- Williams Multiple sequential t-test Procedure.

Results and discussions

Table 2.5.2-1: Mean nitrate ion concentration – deviations from the control [%]

Day of incubation	**PEC** 2.2 mg of the test item/kg of soil (0.2 mg a.s./kg of soil)	**5 x PEC** 11.2 mg of the test item/kg of soil (1.0 mg a.s./kg of soil)
0	2.3	-4.8
7	19.2^{+}	16.0^{+}
14	10.5^{+}	14.4^{+}
28	7.7^{+}	10.9^{+}

$^{+}$ Significant difference to control ($p \leq 0.05$)
"-" higher concentration of nitrate as compared to the control

Findings:

- On day 0 of incubation no statistically significant differences in the nitrate formation rate between the control soil and the soil treated with the test item at the concentrations mentioned above were noticed
- After 7, 14 and 28 days of incubation, statistically significant differences in the nitrate formation rate between the control soil and the soil treated with the test item at the concentrations mentioned above were noticed
- The difference in the nitrate formation rate between the control soil and the one treated with the test item at the concentrations corresponding to the PEC did not exceed 25% on any day of the analysis.
- As for the concentration of nitrates, the percent deviation of the soil treated with the test item at the concentrations: 2.2 mg of the test item/kg of soil (0.2 mg a.s./kg of soil) and 11.0 mg of the test item/ kg of soil (1.0 mg a.s./kg of soil) from the control soil did not exceed 25% on any day of the analysis.

On the basis of the obtained results, it was stated that the validity criterion was met. The coefficients of variation (CV) in the control group were 12.7, 4.5, 2.5 and 1.6% after 0, 7, 14 and 28 days of incubation (validity criterion: the variation between replicate control samples should be less than ± 15%).

Conclusion

On the basis of the results, it was concluded that Mesotrione 10% SC at the concentrations corresponding to the PEC: of 2.2 mg of the test item/kg of soil (0.2 mg a.s./kg of soil) and 5 x PEC: 11.0 mg of the test

item/kg of soil (1.0 mg a.s./kg of soil) did not have any long-term adverse effects on the process of nitrogen formation in aerobic surface soils.

A 2.5.2 KCP 10.5.2 Soil Microorganisms: Carbon Transformation test

Comments of zRMS:	The study Carbon Transformation Test is no longer a data requirement. Therefore, the study was not evaluated.

~~Reference:~~ ~~KCP 10.5-02~~

~~Report~~ ~~Mesotrione 10% SC; Soil Microorganisms: Carbon Transformation Test, Weronika Dec, 2016, Study code: G/241/15. Institute of Industrial Organic Chemistry Branch Pszczyna.~~

~~Guideline(s):~~ ~~Yes, OECD Guideline No. 217 (2000) / EU Method C.22.~~

~~Deviations:~~ ~~Yes~~

- ~~The temperature in the test room was between 19 – 22.5°C. According to the study plan, it should have ranged from 19 to 22°C. It was a short-term deviation (approximately 12 hours) which did not affect the result of the experiment~~
- ~~The active substance content of the application rate (1.5L/ha = 1635 g of the test item/ha) should be 153 g a.s. of test item/ha, not 166.8 g a.s. of test item/ha. This is an editorial mistake. Did not affect the result.~~

~~GLP:~~ ~~Yes~~

~~Acceptability:~~ ~~Yes~~

~~Materials and methods~~

~~Mesotrione 10% SC (batch No. SWEPL – 41203) is a cream color liquid formulation containing the active substance Mesotrione (nominal concentration: 10.2% w/v). The test material was introduced into soil (manually cleared of large objects and then sieved to particle size equal to 2mm) at concentrations of 2.2 mg/kg of soil (0.2 mg a.s./kg of soil) – PEC and 11.0 mg/kg of soil (1.0 mg a.s./kg of soil) – 5 x PEC. The control soil and soil treated with test item were incubated in three replicates. The soil incubation time was 28 days. On 0, the 7th, 14th and 28th day of incubation, the respiration rates in the treated and control soil samples were determined. The *Substance-Induced Respiration* (*SIR*) method was used to determine the intensity of soil respiration [SOP/G/33], involving measurements of the pressure difference in a closed system.~~

~~In order to determine significance of differences in soil respiration between control soil and soil treated with the test item, at the concentrations mentioned above, some statistical analysis was performed using ToxRat 2.10. computer software [17], [SOP/G/80], namely:~~

- ~~Shapiro-Wilk's test on normal distribution~~
- ~~Levene's Test on Variance Homogeneity~~
- ~~Williams Multiple sequential t-test Procedure.~~

~~Results and discussions~~

~~Table 2.5.1-1: Oxygen consumption – deviations from the control [%]~~

~~Day~~	~~**PEC**~~ ~~2.2 mg of the test item/kg of soil~~ ~~(0.2 mg a.s./kg of soil)~~	~~**5 x PEC**~~ ~~11.2 mg of the test item/kg of soil~~ ~~(1.0 mg a.s./kg of soil)~~

0	1.1	15.0
7	1.2	9.1
14	3.9	9.9
28	4.7	11.6

" " higher concentration of oxygen (O_2) consumption as compared to the control

Findings:

- After 28 days of incubation, statistically significant differences in respiration intensity between the control soil and the soil treated with the test item at the concentrations corresponding to the PEC (2.2 mg of the test item/kg of soil) were observed
- On day 0 and after 7, 14 and 28 days of incubation, statistically significant differences in respiration intensity between the control soil and the soil treated with the test item at the concentrations corresponding to the 5 x PEC (11.0 mg of the test item /kg of soil were observed
- The percentage deviations between the control soil and the soil treated with the test item at the concentrations corresponding to the PEC and 5 x PEC did not exceed 25% on any day of analysis.

On the basis of the obtained results, it may be stated that the validity criterion was met. The coefficient of variation in the control group was as follows: 0.9, 2.5, 2.4 and 2.3% on 0, the 7th, 14th and 28th day of soil incubation, respectively. The criterion of validity: the variation between replicate samples in the control should be less than ± 15%.

Conclusion

Taking into account the obtained results, it was concluded that Mesotrione 10% SC at the concentrations corresponding to the PEC: 2.2 mg of the test item/kg of soil (0.2 mg a.s./kg of soil) and 5 x PEC: 11.0 mg of the test item/kg of soil (1.0 mg a.s./kg of soil) can be perceived as having no long-term influence on carbon transformation in soil

A 2.6 KCP 10.6 Effects on terrestrial non-target higher plants

A 2.6.1 KCP 10.6.1 Summary of screening data

A 2.6.2 KCP 10.6.2 Testing on non-target plants

Comments of zRMS:	The study was conducted to OECD guideline 208 and according to the principles of GLP. One minor deviation from the guideline (temporary humidity was below 70 ± 25%) occurred, which did not affect the result of the experiment. All validity criteria were met according as: - the seedling emergence in the control (validity criterion: at least 70%) was as follows: 100% – pea, 95% – sunflower, 100% – white mustard, 100% – tomato, 100% – corn, 100% – oats.

	- the mean survival of the emerged control seedlings was 100% (validity criterion: at least 90%). - the control seedlings did not exhibit any visible phytotoxic effects. - environmental conditions for all plants of the same species were identical. The study is reliable and suitable for the risk assessment.

Reference: KCP 10.6.2 - 01

Report "Mesotrione 10% SC Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test" Weronika Dec. 2017, Study code: G/246/15. Institute of Industrial Organic Chemistry Branch Pszczyna

Guideline(s): OECD Guideline 208

Deviations: Yes. Four deviations from the Study Plan were recorded.
1. The humidity in the test room was between 44.9 – 64.2%. According to the Study Plan, it should be 70 ± 25%. It was a short-term deviation (about one hour) which did not affect the result of the experiment.
2. The study was finished in January 2017 and not in August 2016.
3. The RTF II meter was used instead of TZ 18 td thermohygrograph.
4. The SUP-100G with the digital temperature controller was used in the experiment instead of the SUP-100G without the digital temperature controller laboratory dryer.

The deviations did not affect the results of the study..

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Materials and methods

Materials

Test item:
Description: Mesotrione 10% SC
Production batch: SWEPL-41203
Active ingredients content: mesotrione - 10.2% w/v
Vehicle and control: Distilled water
Test system:
Species: pea (*Pisum sativum*), sunflower (*Helianthus annuus*), white mustard (*Sinapis alba*), tomato (*Solanum lycopersicon*), corn (*Zea mays*), and oats (*Avena sativa*).

Experimental conditions:

Test species	Light intensity [lx]	Photoperiod day/night [h]	Humidity [%]	Temperature [°C]	CO_2 concentration [ppm]
Pea *(Pisum sativum)*	5030 – 5200	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386
Sunflower *(Hellanthus annus)*	4900 – 5100	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386
White mustard *(Sinapis alba)*	4900 – 5120	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386
Tomato *(Solanum lycopersicon)*	5080 – 5280	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386
Corn *(Zea mays)*	4800 – 5030	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386
Oats *(Avena sativa)*	5010 - 5220	16:08	44.9 – 64.2	22.1 – 27.1	330 - 386

Study design and methods

Experimental period: 08/07/2016 – 26/0/2016

Test design and treatment: number of concentrations: 7 application rates + a control
number of replicates: 4 replicates of each application rate and the control (pea, sunflower, white mustard, tomato, oats); 10 replicates of each application rate and the control (corn)
number of seeds: 5 seeds/replicate (pea, sunflower, white mustard, tomato, oats); 2 seeds/replicate (corn)
test termination: 14 days after the emergence of 50% of the control seedlings

Agricultural soil (Sandy loam soil) was used in the study collected from the place belonging to the Institute of Industrial Organic Chemistry, Branch Pszczyna (49˚ 59', 780 N; 18˚55', 190 E) where no plant protection products or organic and inorganic fertilizers had been used. Test item application rates were selected on the basis of the maximum recommended rate, i.e. 1500 mL/ha (i.e. 140.4 g a.s./ha). Seven rates of the test item were used. These were 2.06, 6.17, 18.52, 55.56, 166.67, 500.0 and 1500.0 mL/ha (i.e. 0.2, 0.6, 1.7, 5.2, 15.6, 46.8 and 140.4 g a.s./ha). One untreated control group was used for each species. The test system was the same for all test species. The test item was sprayed onto the soil using a suitable spraying chamber. The experiment was conducted in a special room where suitable environmental conditions were provided. The experiment finished 14 days after the emergence of 50% of the control seedlings. During the experiment, the plants were observed for emergence (every day and then every 2 – 3 days) and visual phytotoxicity (7 and 14 days after the emergence of 50% of the control seedlings). At the end of the experiment, the plants were counted, cut down, measured, dried to a constant weight at 60°C, and weighed.

Statistics: ER_{10}, ER_{25}, ER_{50} – probit analyses
NOER – Shapiro-Wilk's Test on Normal Distribution, Levene's Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure, Fisher's Exact Binomial Test with Bonferroni Correction, Welch-t test for Inhomogeneous Variances with Bonferroni-Holm Adjustment.

Results

Results are displayed on the tables below:

Phytotoxic symptoms on pea (pisum stivum)

After the application of the test item at the rates ranging from 166.67 to 1500 mL/ha, the mean plant damage was from 18.8 to 72.5% at the end of the test. At lower application rates plant damage was not observed (Table below). Phytotoxic symptoms, i.e. stunted growth, witling, chlorosis and dead plants were noticed.

Table 10. Pea *(Pisum sativum)* – plant damage.

Application rate [mL/ha]	Replicate	Plant damage					
		Day 7			Day 14		
		Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*	Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*
control	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
2.06	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
6.17	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
18.52	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
55.56	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
166.67	1	5	5.0	chl	20	18.8	chl, sg, w
	2	5			15		
	3	10			20		
	4	0			20		
500	1	15	11.3	chl, sg	50	55.0	chl, sg, w, d
	2	10			60		
	3	15			50		
	4	5			60		
1500	1	40	42.5	chl. sg	70	72.5	chl, sg, w, d
	2	45			70		
	3	45			80		
	4	40			70		

* **nc** – no changes, **sg** – stunted growth, **w** – witling, **chl** – chlorosis, **d** – dead plant

Phytotoxic symptoms of sunflower (*Helianthus annuus)*

After the application of the test item at the rates ranging from 166.67 to 1500 mL/ha, the mean plant damage was from 31.3 to 57.5% at the end of the test. At lower application rates plant damage was not observed (Table below). Phytotoxic symptoms, i.e. stunted growth, witling, chlorosis, and dead plants were noticed.

Table 15. Sunflower *(Helianthus annuus)* – plant damage.

Application rate [mL/ha]	Replicate	Plant damage					
		Day 7			Day 14		
		Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*	Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*
control	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
2.06	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
6.17	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
18.52	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
55.56	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
166.67	1	5	6.3	chl	30	31.3	sg, chl, w, d
	2	5			30		
	3	5			25		
	4	10			40		
500	1	20	20.0	sg, chl	30	35.0	sg, chl, w
	2	20			35		
	3	20			35		
	4	20			40		
1500	1	25	25.0	sg, chl	50	57.5	sg, chl, w, d
	2	25			70		
	3	25			50		
	4	25			60		

* **nc** – no changes, **sg** – stunted growth, **w** – witling, **chl** – chlorosis, **d** – dead plants

Phytotoxic symptoms of white mustard (*Sinapsis alba*)

After the application of the test item at the rates ranging from 55.56 to 1500 mL/ha, the mean plant damage was between 26.3 – 70.0% at the end of the test. At lower application rates plant damage was not observed (Table below). Phytotoxic symptoms, i.e. stunted growth, witling, chlorosis and dead plants were noticed.

Table 20. White mustard *(Sinapis alba)* – plant damage.

Application rate [mL/ha]	Replicate	Plant damage					
		Day 7			Day 14		
		Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*	Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*
control	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
2.06	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
6.17	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
18.52	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
55.56	1	10	12.5	sg, chl	20	26.3	sg, chl, w
	2	15			25		
	3	10			30		
	4	15			30		
166.67	1	15	15.0	sg, chl	30	35.0	sg, chl, w
	2	20			30		
	3	10			40		
	4	15			40		
500	1	20	15.0	sg, chl	45	50.0	sg, chl, w, d
	2	10			45		
	3	15			55		
	4	15			55		
1500	1	45	42.5	sg, chl	70	70.0	sg, chl, w, d
	2	45			80		
	3	35			60		
	4	45			70		

* **nc** – no changes, **sg** – stunted growth, **w** – witling, **chl** – chlorosis, **d** – dead plants

Phytotoxic symptoms of Tomato (*Solanum lycopersicon*)

After the application of the test item at the rates ranging from 2.06 to 1500 mL/ha, the mean plant damage was between 0.0 – 100% at the end of the test. At lower application rates plant damage was not observed (Table below). Phytotoxic symptoms, i.e. stunted growth, witling, chlorosis, and dead plants were noticed.

Table 25. Tomato *(Solanum lycopersicon)* – plant damage.

Application rate [mL/ha]	Replicate	Plant damage					
		Day 7			Day 14		
		Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*	Mean effects/ replicate [%]	Mean effects/ application rate [%]	Plant damage*
control	1	0	0.0	nc	0	0.0	nc
	2	0			0		
	3	0			0		
	4	0			0		
2.06	1	0	0.0	nc	0	2.5	d
	2	0			0		
	3	0			0		
	4	0			10		
6.17	1	0	0.0	nc	10	2.5	d
	2	0			0		
	3	0			0		
	4	0			0		
18.52	1	0	0.0	nc	0	2.5	d
	2	0			10		
	3	0			0		
	4	0			0		
55.56	1	0	0.0	nc	10	10.0	w
	2	0			10		
	3	0			10		
	4	0			10		
166.67	1	5	10.0	sg, chl, w	10	15	sg, w, chl, d
	2	15			20		
	3	5			10		
	4	15			20		
500	1	45	48.8	sg, chl, w	80	80	sg, w, chl, d
	2	50			80		
	3	50			80		
	4	50			80		
1500	1	80	75.0	sg, chl, w	100	100.0	d
	2	60			100		
	3	80			100		
	4	80			100		

* **nc** – no change, **sg** – stunted growth, **w** – witling, **chl** – chlorosis, **d** – dead plants

Phytotoxic symptoms of corn (*Zea mays*)

After the application of the test item at the rates ranging from 2.06 to 1500 mL/ha, no plant damage was observed at the end of the test.

Phytotoxic symptoms of Oats (*Avena sativa*)

After the application of the test item at the rates ranging from 2.06 to 1500 mL/ha, no plant damage was observed at the end of the test

The EC50 and NOER values determined on the basis of plant number, shoot length and dry shoot weight measurements expressed as ml of formulation/ ha for all test species are given below.

	Pea	Sunflower	White mustard	Tomato	Corn	Oats
	Pisum sativum	*Helianthus annuus*	*Sinapis alba*	*Solanum lycopersicon*	*Zea mays*	*Avena sativa*
Plant number at the end of the experiment						
ER_{10}	> 1500	238.3 (40.3 – > 1500)	31.2 (< 2.06 – 197.3)	10.0	> 1500	> 1500
ER_{25}	> 1500	> 1500 (577.9 – > 1500)	209.2	42.5	> 1500	> 1500
ER_{50}	> 1500	> 1500	> 1500 (261.3 – > 1500)	211.3	> 1500	> 1500
NOER	≥1500	≥1500	500	166.67	> 1500	≥1500
Post-emergence survival						
ER_{10}	> 1500	1153.7	472.5 (184.9 – 682.6)	33.1	> 1500	> 1500
ER_{25}	> 1500	> 1500	709.4 (403.6 – 974.8)	89.6	> 1500	> 1500
ER_{50}	> 1500	> 1500	1114.3 (798.2 – >1500)	271.1	> 1500	> 1500
NOER	≥1500	≥1500	500	166.67	> 1500	> 1500

	Pea	Sunflower	White mustard	Tomato	Corn	Oats
	Pisum sativum	*Helianthus annuus*	*Sinapis alba*	*Solanum lycopersicon*	*Zea mays*	*Avena sativa*
Shoot length (plants without roots)						
ER_{10}	80.5 (43.3 – 119.7)	146.5 (106.7 – 186.6)	12.9 (< 2.06 – 31.0)	30.5 (< 2.06 – 77.4)	> 1500	> 1500
ER_{25}	190.7 (130.5 – 249.2)	410.8 (347.0 – 473.3)	47.5 (15.7 – 87.7)	73.3 (2.4 – 148.8)	> 1500	> 1500
ER_{50}	496.9 (396.5 – 630.8)	1291.1 (1121.1 – > 1500)	202.7 (114.2 – 380.5)	194.3 (75.6 – 784.4)	> 1500	> 1500
NOER	55.56	55.56	18.52	55.56	≥1500	≥1500
Plant dry weight (plants without roots)						
ER_{10}	33.0 (4.1 – 77.7)	16.3 (2.1 – 42.1)	7.1 (< 2.06 – 21.0)	28.0 (12.8 – 42.7)	> 1500	> 1500
ER_{25}	112.5 (35.3 – 202.7)	83.9 (28.7 – 156.0)	29.9 (6.3 – 64.4)	55.0 (33.7 – 74.2)	> 1500	> 1500
ER_{50}	439.4 (251.2 – 894.7)	517.0 (289.1 – 1194.8)	147.8 (69.7 – 339.9)	116.6 (88.5 – 153.9)	> 1500	> 1500
NOER	55.56	55.56	18.52	18.52	≥1500	≥1500

The EC_{50} and NOER values determined on the basis of plant number, shoot length and dry shoot weight measurements expressed as g of active substance / ha for all test species are given below.

	Pea	Sunflower	White mustard	Tomato	Onion	Oats
	Pisum sativum	*Helianthus annuus*	*Sinapis alba*	*Solanum lycopersicon*	*Allium cepa*	*Avena sativa*
Plant number at the end of the experiment						
ER_{50}	> 140.4	> 140.4	> 140.4 (24.5 – > 140.4)	19.8	> 140.4	> 140.4
NOER	≥ 140.4	≥ 140.4	46.8	15.6	> 140.4	≥ 140.4
Post-emergence survival at the end of the experiment						
ER_{50}	> 140.4	> 140.4	104.3 (74.7 – >140.4)	25.4	> 140.4	> 140.4
NOER	≥ 140.4	≥140.4	46.8	15.6	> 140.4	> 140.4
Shoot length (plants without roots)						
ER_{50}	46.5 (37.1 – 59.0)	120.8 (104.9 – >1140.4)	19.0 (10.7 – 35.6)	18.2 (7.1 – 73.4)	> 140.4	> 140.4
NOER	5.2	5.2	1.7	5.2	≥ 140.4	≥ 140.4
Plant dry weight (plants without roots)						
ER_{50}	41.1 (23.5 – 83.7)	48.4 (27.1 – 111.8)	13.8 (6.5 – 31.8)	10.9 (8.3 – 14.4)	> 140.4	> 140.4
NOER	5.2	5.2	1.7	1.7	≥ 140.4	≥ 140.4

Conclusion

1. The test item i.e. Mesotrione 10% SC had a varied impact on the growth and seedling emergence of the test plant species. The impact depended on the concentration and species.
2. After the application of the test item at the rates ranging from 2.06 to 1500 mL/ha all test plant species: pea (Pisum sativum), sunflower (Helianthus annuus), white mustard (Sinapis alba), tomato (Solanum lycopersicon), corn (zea mays), and oats (Avena sativa) emerged. After the application of the test item at rates ranging from 2.06 to 1500 mL/ha, all test species emergence was not delayed in comparison to the control.
3. The death of pea and white mustard plants was noticed at application rates of 500 and 1500 mL/ha. The death of sunflower was noticed at application rates of 166.67 and 1500 mL/ha. The death of tomato plant was noticed almost at the all application rates, only at the application rate of 55.56 mL/ha the death of tomato was not recorded. The death of corn and oats was not observed at rates ranging from 2.06 to 1500 mL/ha.
4. On the basis of NOER, ER_{10}, ER_{25} and ER_{50} values determined from the shoot length and shoot dry weight, it was proved that the test item inhibited the process of growth of pea, sunflower, white mustard and tomato.
5. Phytotoxic symptoms were observed. They were stunted growth, witling, chlorosis and dead plants for pea, sunflower, white mustard and tomato. For corn and oats no phytotoxic symptoms were recorded.
6. The lowest ER_{50} values determined on the basis of the plant number, post-emergence survival, shoot length and shoot dry weight at the end of the experiment were equal to 211.3, 271.1, 194.3 and 116.6 mL/ha (i.e. 19.8, 25.4, 18.2 and 10.9 g a.s./ha) respectively and they were calculated for tomato.
7. The following order of the test plant sensitivity was noticed:

tomato > white mustard > pea, sunflower > corn, oats.

Comments of zRMS:	The study was conducted to OECD 227 guideline and according to the principles of GLP. All validity criteria were fulfilled. The study is considered to be reliable and suitable for the risk assessment.

Reference:	KCP 10.6.2-02
Report	"Terrestrial Plant Test: Vegetative Vigour Test of Mesotrione 10% SC on Plants" Dr. T. S. Sadamanda, 2019, Study number: BIO-ETX 034. Bioneeds India Privated Limited
Guideline(s):	OECD Guideline No. 227 (2006)
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:	Mesotrione 10% SC; Batch Number SCL-96311; active substance: mesotrione 10.4% (w/v)
Test species:	Lettuce (*Lactuca sativa*), Dwarf bean, French bean, Garden bean (*Phaseolus vulgaris*), Carrot (*Daucus carota*), Sugar beet (*Beta vulgaris*), Chinese cabbage (*Brassica campestris var. chinensis*), Flax (*Linum usitatissimum*), Onion (*Allium cepa*), Wheat (*Triticum aestivum*)
Soil:	Clay loam soil containing 1.5% organic carbon
Study design:	number of rates: 7 application rates + control; number of replicates: 4 pots/application rate and 5 seeds/plot. test termination: 21 days after the spraying.
Application rates:	Water control, 0.02, 0.05, 0.09, 0.19, 0.38, 0.75 and 1.5 L test item/ha Volume of deionised water used to prepare the highest rate: 200 L water/ha
Test conditions:	temperature: 22.9-29.8ºC, humidity: 70.0 – 84.0%, light – dark cycles (16h:8h), light intensity: 6599 – 8241 lux, carbon dioxide concentration: 344 – 398 ppm.
Statistical analysis:	The EC_{10}, EC_{25}, EC_{50} and NOEC values were determined by using a Probit analysis and one-way ANOVA followed by Dunnett´s test using SPSS Software version 22, respectively.
Endpoints:	EC_{10}, EC_{25}, EC_{50} and NOER

Results and Conclusions

The test item, i.e. Mesotrione 10% SC applied at rates ranging from 0.02 to 1.5 L test item/ha had a varied impact on vegetative vigour of all the plant species tested. The impact depended on the rate of the test item and species used.
There was mortality observed for all the plant species tested at rates ranging from 0.0125 to 0.2 kg test item/ha. The phytotoxic symptoms for all plant species tested were observed at all the rates of the test item used.
The endpoint values showing the impact of the test item on vegetative vigour of the plant species tested are presented in table given below:

Endpoint value		Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
Plant number									
ER_{10}	L/ha	>1.50	>1.50	>1.50	n.d.	n.d.	>1.50	n.d.	>1.50
ER_{20}	L/ha	>1.50	>1.50	>1.50	n.d.	n.d.	>1.50	n.d.	>1.50
ER_{50}	L/ha	>1.50	>1.50	>1.50	n.d.	n.d.	>1.50	n.d.	>1.50
NOER	L/ha	≥1.50	≥1.50	≥1.50	0.75	0.75	≥1.50	0.75	≥1.50
Fresh shoot weight									
ER_{10}	L/ha	0.04	0.05	0.02	0.08	0.06	0.03	0.03	0.04
ER_{20}	L/ha	0.09	0.11	0.06	0.15	0.14	0.06	0.09	0.10
ER_{50}	L/ha	0.24	0.27	0.20	0.34	0.38	0.14	0.28	0.28
NOER	L/ha	0.02	0.02	0.02	0.05	0.05	>0.02	0.02	0.05
Dry shoot weight									
ER_{10}	L/ha	0.03	0.05	0.01	0.07	0.07	0.02	0.04	0.04
ER_{20}	L/ha	0.08	0.11	0.03	0.13	0.15	0.04	0.09	0.10
ER_{50}	L/ha	0.21	0.24	0.12	0.27	0.35	0.09	0.23	0.24
NOER	L/ha	<0.02	0.02	0.02	0.05	0.05	<0.02	0.05	0.05
Shoot height									
ER_{10}	L/ha	0.05	0.07	0.012	0.05	0.05	0.01	0.03	0.05
ER_{20}	L/ha	0.12	0.15	0.093	0.10	0.11	0.03	0.07	0.12
ER_{50}	L/ha	0.36	0.35	0.28	0.22	0.29	0.11	0.22	0.33
NOER	L/ha	0.05	0.05	0.02	0.02	0.02	0.02	0.02	0.02

VALIDITY OF THE TEST

The following criteria have satisfied in the control, hence the test results are considered to be valid:

- The seedling emergence for all the plants tested was 100% (validity criterion: at least 70%).
- The control plants did not exhibit any visible phytotoxic effects. Plants exhibited normal variation in growth and morphology for that particular species.
- The mean survival of the emerged control seedlings was 100% in case of all experimental species (validity criterion: at least 90%),
- Environmental conditions for a particular species were identical and growing media contained the same amount of soil matrix, support media, or substrate from the same source.

TABLE 11. PERCENT INHIBITION OF PLANT NUMBER DURING THE MAIN STUDY

Group	Application rate (L test item/ha)	Percent inhibition of plant number							
		Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
G1	0.0	-	-	-	-	-	-	-	-
G2	0.02	0	0	0	0	0	0	0	0
G3	0.05	0	0	0	0	0	0	0	0
G4	0.09	0	0	0	0	0	0	0	0
G5	0.19	0	0	0	0	0	0	0	0
G6	0.38	0	0	0	0	0	0	0	0
G7	0.75	0	0	0	0	0	0	0	0
G8	1.50	0	0	0	100*	100*	0	100*	0
#G9	1200 mL glyphosate/ acres	95*	90*	0	95*	0	0	95*	90*

-: Not applicable; #: Positive control Broadband herbicide (Roundup ultra mix), Glyphosate 41% SL; *: Statistical significance at $P < 0.05$

TABLE 12. PERCENT INHIBITION OF FRESH SHOOT WEIGHT OF PLANTS DURING THE MAIN STUDY

Group	Application rate (L test item/ha)	Percent inhibition of fresh shoot weight							
		Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
G1	0.0	-	-	-	-	-	-	-	-
G2	0.02	6.88	2.31	8.81	0.41	1.75	10.79*	2.68	3.99
G3	0.05	11.91*	14.30*	32.70*	7.99	8.39	30.85*	22.35*	14.54
G4	0.09	23.74*	19.30*	33.96*	16.50*	19.91*	47.68*	32.35*	26.58*
G5	0.19	41.49*	29.33*	46.54*	25.87*	25.51*	67.36*	38.87*	36.66*
G6	0.38	55.89*	51.23*	57.23*	58.90*	45.43*	87.09*	48.99*	47.08*
G7	0.75	79.10*	79.51*	71.19*	71.47*	72.77*	91.72*	72.69*	72.20*
G8	1.50	92.45*	94.06*	90.60*	100*	100*	99.02*	100*	90.08*
#G9	1200 mL glyphosate/ acres	93.66*	97.34*	86.06*	95.06*	96.92*	98.71*	90.66*	91.95*

-: Not applicable; #: Positive control Broadband herbicide (Roundup ultra mix), Glyphosate 41% SL; *: Statistical significance at $P < 0.05$

TABLE 13. PERCENT INHIBITION OF DRY SHOOT WEIGHT OF PLANTS DURING THE MAIN STUDY

Group	Application rate (L test item/ha)	Percent inhibition of dry shoot weight							
		Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
G1	0.0	-	-	-	-	-	-	-	-
G2	0.02	11.18*	1.84	18.00	1.04	1.71	12.05*	6.11	2.01
G3	0.05	10.62*	15.25*	32.00*	5.95	5.21	31.09*	6.89	19.43
G4	0.09	24.68*	22.99*	51.03*	20.33*	16.33*	51.27*	36.89*	28.13*
G5	0.19	45.71*	31.31*	60.00*	29.73*	20.37*	70.55*	43.07*	37.98*
G6	0.38	54.28*	56.30*	59.00*	61.23*	49.71*	91.26*	60.95*	55.68*
G7	0.75	82.65*	83.82*	79.00*	84.39*	81.00*	95.19*	80.13*	77.94*
G8	1.50	93.68*	95.73*	93.00*	100*	100*	99.57*	100.0*	93.92*
#G9	1200 mL glyphosate/ acres	96.10*	97.87*	92.43*	97.91*	98.43*	99.52*	92.77*	93.56*

-: Not applicable; #: Positive control Broadband herbicide (Roundup ultra mix), Glyphosate 41% SL; *: Statistical significance at $P < 0.05$*

TABLE 14. PERCENT INHIBITION OF SHOOT HEIGHT OF PLANTS DURING THE MAIN STUDY

Group	Application rate (L test item/ha)	Percent inhibition of shoot height							
		Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
G1	0.0	-	-	-	-	-	-	-	-
G2	0.02	5.50	0.29	2.44	2.27	4.59	11.24	7.34	2.85
G3	0.05	8.43*	9.69	21.19*	10.14*	9.61*	37.87*	18.06*	8.58*
G4	0.09	23.20*	23.93*	32.19*	30.81*	18.74*	52.12*	34.57*	28.36*
G5	0.19	29.74*	36.14*	47.69*	39.01*	31.61*	67.96*	44.92*	41.61*
G6	0.38	33.76*	43.01*	56.45*	57.48*	56.12*	76.67*	55.27*	49.23*
G7	0.75	63.70*	61.23*	61.66*	89.65*	80.54*	85.73*	81.90*	59.56*
G8	1.50	91.50*	91.20*	86.16*	100*	100*	91.99*	100*	88.26*
#G9	1200 mL glyphosate/ acres	96.43*	93.93*	91.28*	92.07*	95.63*	94.97*	93.13*	93.87*

-: Not applicable; #: Positive control Broadband herbicide (Roundup ultra mix), Glyphosate 41% SL; *: Statistical significance at $P < 0.05$

TABLE 6. SUMMARY OF PLANT OBSERVATION DUIRNG THE MAIN STUDY

Group	Application rate (L test item/ha)	Dicotyledonous						Monocotyledonous	
		Lettuce (*Lactuca sativa*)	Dwarf bean (*Phaseolus vulgaris*)	Carrot (*Daucus carota*)	Sugar beet (*Beta vulgaris*)	Chinese cabbage (*Brassica campestris*)	Flax (*Linum usitatissimum*)	Onion (*Allium cepa*)	Wheat (*Triticum aestivum*)
G1	0	20(1)	20(1)	20(1)	20(1)	20(1)	20(1)	20(1)	20(1)
G2	0.02	18 (1), 2(2)	5(2,3),15(5)	18(1),2(2)	15(1),5(2)	18(2,3),2(10)	17(2),3(1)	11(1),9(2)	16(1),4(2)
G3	0.05	9(2),6(3),5(4)	5(2,3),15(5)	12(2),6(3), 2(10)	12(1),8(2,5)	20(2,3,4)	12(3,4,2), 8(10)	12(1),5(2,3), 3(5)	14(2), 6(5)
G4	0.09	9(2,3), 11(5,4)	9(2,3), 11(5,4)	16(4,2,3),4(10)	10(1),10(2,5)	17(2,3,4), 3(3)	14(3,4,2), 6(10)	15(1),5(3,2,5)	16(2), 4(5)
G5	0.19	15(5),5(2,3)	7(2,3,4),13(5)	20(2,5)	11(1), 9(2,5)	20(4,5)	3(2,3), 17(4,10)	12(1),5(2, 3),3(5)	12(2,3), 8(5)
G6	0.38	20(2,4)	8(3,2,10,4),12(5)	20(2,5)	20(4,5)	20(4,5)	12(2,3), 8(10,4)	8(2,3), 12(5)	13(2,3),7(5)
G7	0.75	20(2,4)	15(2,3,4,10),5(5)	20(2,5)	20(4,5)	20(10,5)	3(2,3), 11(10,4), 6(5)	16(2,3,10), 4(5)	18(5), 2(3,10)
G8	1.50	20(2,5)	20(2,5)	20(5)	20(*5)	20(*5)	20(2,5)	20(*5)	20(5,3,10)
#G9	1200 mL glyphosate/ acres	1(5), 19(5*)	2(5), 18(5*)	20(5)	1(5), 19(5*)	20(5)	20(5)	1(5), 19(*)	1(5), 19(*)

1: Normal; 2: Chlorosis; 3: Death of plant tissues; 4: Defoliation; 5: Dried leaves or stem; 10: In the process of drying; *: Complete plant dried; 5*: Completely dried leave and stem, plant observed complete mortality; Value inside the bracket and value outside the bracket signifies the clinical symptom code and number of plants exhibiting that particular symptom; #: Positive control Broadband herbicide (Roundup ultra mix), Glyphosate 41% SL

A 2.6.3 KCP 10.6.3 Extended laboratory studies on non-target plants

No new study submitted.

A 2.7 KCP 10.7 Effects on other terrestrial organisms (flora and fauna)

No new study submitted.

A 2.8 KCP 10.8 Monitoring data

No new study submitted.