

REGISTRATION REPORT

Part B

Section 9

Ecotoxicology

Detailed summary of the risk assessment

Product code: SAP09I

Product name(s): VOLKETE MAX

Chemical active substance:

Spinosad, 480 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

(New product authorization according to article 33 and 34 of
Regulation (EC) 1107/2009)

Applicant: Albaugh TKI d.o.o.

Submission date: January 2025

MS Finalisation date: December 2025

Version history

When	What
January 2025	V0 – Original dRR version from applicant Albaugh TKI d.o.o. for submission to zRMS Poland in the frame of a new authorization according to Article 33/34 of Regulation (EC) No. 1107/2009.
October 2025	zRMS finalised dRR evaluation
November 2025	RR update with additional risk assessment for soil organisms
December 2025	zRMS finalised evaluation after commenting stage

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9 Ecotoxicology (KCP 10)

Review Comments:

This is the application for registration plant protection product – SAP09I (Volkete Max), according to Article 33 of Regulation 1107/2009 based on unprotected data for compositionally comparable formulation – SpinTor 480 SC (acc. Art. 34 of Reg. 1107/2009). Further detail are given in separate document titled: Compositions comparison in accordance with Article 34 – Volkete Max only for PL authority. SAP09I is a suspension concentrate type formulation (SC) containing 480 g/L spinosad and is used as an insecticide in numbers of crops.

This Part B document only reviews data and additional information that has not previously been considered within the EU review process.

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.

During the commenting process, the Applicant introduced modifications to the proposed use pattern of SAP09I (Volkete Max). Additional risk assessment was performed for mammals and soil organisms.

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 saf- ener/ synergist per ha	Conclusion						
					Method / Kind	Timing / Growth stage of crop & season	Max. number a) per use b) per crop/ season	Min. interval between applications (days)	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 arthropods	Soil organisms	Non-target plants
Zonal uses (field or outdoor uses)																				
1	PL	Head cabbage [BRSOL]	F	<i>Pieris brassicae</i> [PIERBR] <i>Artogeia rapae</i> [PIERRA] <i>Mamestra brassicae</i> [BARABR]	Spray	Young caterpillars / first damages BBCH 10- 49	a) 3 b) 3	7-10	a) 0.2 L/ha b) 0.6 L/ha	a) 96 b) 288	200-600	3	Dose rate 0.1-0.2 L/ha	A	A	R SPe3	R SPe8	R SPe3	N	A
2	PL	Broccoli [BRSOK] Cauliflower [BRSOB]	F	<i>Artogeia rapae</i> [PIERRA] <i>Mamestra brassicae</i> [BARABR]	Spray	Young caterpillars / first damages BBCH 10- 49	a) 3 b) 3	7-10	a) 0.2 L/ha b) 0.6 L/ha	a) 96 b) 288	200-600	3	Dose rate 0.1-0.2 L/ha	A	A	R SPe3	R SPe8	R SPe3	N	A
3	PL	Onion [ALLCE]	F	<i>Thrips tabaci</i> [THRITB]	Spray	Notice of the pest / first damages BBCH 10- 39	a) 3 b) 3	10	a) 0.2 L/ha b) 0.6 L/ha	a) 96 b) 288	200-600	7	Dose rate 0.15-0.2 L/ha Mammals: BBCH≥40 – non safe	A	A	R SPe3	R SPe8	R SPe3	N	A
4	PL	Leek [ALLCE]	F	<i>Thrips tabaci</i> [THRITB]	Spray	Young caterpillars / first damages BBCH 10- 39	a) 3 b) 3	10	a) 0.2 L/ha b) 0.6 L/ha	a) 96 b) 288	200-600	7	Dose rate 0.15-0.2 L/ha Mammals: BBCH≥40 – non safe	A	A	R SPe3	R SPe8	R SPe3	N	A

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
5	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spray	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10 21	a) 0.3 L/ha b) 0.6 L/ha	a) 144 b) 288	300-1500	14	Maximum number of treatments per growing season: 2 0.23 L/1000 m ² of LWA Mammals: BBCH<40 – non safe	A	A	R SPe3	R SPe8	R SPe3	N	A
Minor uses according to Article 51																				
6	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes orana</i> [CAPURE]	Spray	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10 21	a) 0.3 L/ha b) 0.6 L/ha	a) 144 b) 288	300-1500	14	Maximum number of treatments per growing season: 2 0.23 L/1000 m ² of LWA Mammals: BBCH<40 – non safe	A	A	R SPe3	R SPe8	R SPe3	N	A
7	PL	Garlic [ALLSA] Shallot [ALLAS]	F	<i>Thrips</i> sp. [THRISP]	Spray	Notice of the pest / first damages BBCH 10-39	a) 3 b) 3	10	a) 0.2 L/ha b) 0.6 L/ha	a) 96 b) 288	200-600	7	Dose rate 0.15-02 L/ha Mammals: BBCH≥40 – non safe	A	A	R SPe3	R SPe8	R SPe3	N	A

* 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

Table 9.1-2a: Critical use pattern of the formulated product SAP09I

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
Zonal uses (field or outdoor uses)														
1	PL	Head cabbage [BRSOL]	F	<i>Pieris brassicae</i> [PIERBR] <i>Artogeia rapae</i> [PIERRA] <i>Mamestra</i> <i>brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 10-49	a) 2-3 b) 2-3	10	a) 0.2 b) 0.6	a) 96 b) 288	200-600	3	Dose range 0.1-0.2 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 1a,b	Unacceptable risk for soil organisms
1a	PL	Head cabbage [BRSOL]	F	<i>Pieris brassicae</i> [PIERBR] <i>Artogeia rapae</i> [PIERRA] <i>Mamestra</i> <i>brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 20-49	a) 2 b) 2	10	a) 0.16 b) 0.32	a) 76.8 b) 153.6	200-600	3	Dose range 0.1-0.16 L/ha Intended Use just in case higher tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
1b	PL	Head cabbage [BRSOL]	F	<i>Pieris brassicae</i> [PIERBR] <i>Artogeia rapae</i> [PIERRA] <i>Mamestra</i> <i>brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 10-49	a) 2-3 b) 2-3	10	a) 0.1 b) 0.3	a) 48 b) 144	200-600	3	Intended Use just in case higher tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
2	PL	Broccoli [BRSOK] Cauliflower [BRSOB]	F	<i>Artogeia rapae</i> [PIERRA] <i>Mamestra brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 10-49	a) 2-3 b) 2-3	10	a) 0.2 b) 0.6	a) 96 b) 288	200-600	3	Dose range 0.1-0.2 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 2a	Unacceptable risk for soil organisms
2a	PL	Broccoli [BRSOK] Cauliflower [BRSOB]	F	<i>Artogeia rapae</i> [PIERRA] <i>Mamestra brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 20-49	a) 2 b) 2	10	a) 0.16 b) 0.32	a) 96 76.8 b) 288 153.6	200-600	3	Dose range 0.1-0.16 L/ha Intended Use just in case higher tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
2b	PL	Broccoli [BRSOK] Cauliflower [BRSOB]	F	<i>Artogeia rapae</i> [PIERRA] <i>Mamestra brassicae</i> [BARABR]	Spraying	Young caterpillars / first damages BBCH 20-49	a) 2-3 b) 2-3	10	a) 0.1 b) 0.3	a) 48 b) 144	200-600	3	Intended Use just in case higher tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
3	PL	Onion [ALLCE]	F	<i>Thrips tabaci</i> [THRITB]	Spraying	Notice of the pest / first damages BBCH 10-49	a) 1 b) 1	na	a) 0.15 b) 0.15	a) 72 b) 72	200-600	7	If higher tier assessment in Folsomia is not accepted please refer to Use No. 3a	Unacceptable risk for soil organisms

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
3a	PL	Onion [ALLCE]	F	<i>Thrips tabaci</i> [THRITB]	Spraying	Notice of the pest / first damages BBCH 40-49	a) 1 b) 1	na	a) 0.16 b) 0.16	a) 76.8 b) 76.8	200-600	7	Dose range 0.15-0.16 L/ha Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
4	PL	Leek [ALLPO]	F	<i>Thrips tabaci</i> [THRITB]	Spraying	Notice of the pest / first damages BBCH 10-49	a) 1 b) 1	na	a) 0.15 b) 0.15	a) 72 b) 72	200-600	7	If higher tier assessment in Folsomia is not accepted please refer to Use No. 4a	Unacceptable risk for soil organisms
4a	PL	Leek [ALLPO]	F	<i>Thrips tabaci</i> [THRITB]	Spraying	Notice of the pest / first damages BBCH 40-49	a) 1 b) 1	na	a) 0.16 b) 0.16	a) 76.8 b) 76.8	200-600	7	Dose range 0.15-0.16 L/ha Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
5	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10-21	a) 0.3 b) 0.6	a) 144 b) 288	300-1500	14	Dose range 0.18-0.3 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 5a,b,c	Unacceptable risk for soil organisms

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
5i	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	>BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.3 b) 0.6	a) 144 b) 288	300-1500	14	Dose range 0.18-0.3 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 5a,b,c	Unacceptable risk for soil organisms
5a	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spray	BBCH >70	a) 1 b) 1	NR	a) 0.3 b) 0.3	a) 144 b) 144	300-1500	14	Dose range 0.18-0.3 L/ha Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
5b	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	>BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.225 b) 0.450	a) 108 b) 216	300-1500	14	Dose range 0.18-0.225 Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
5c	PL	Apple [MABSD]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.225 b) 0.450	a) 108 b) 216	300-1500	14	Dose range 0.18-0.225 Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
Minor uses according to Article 51 (zonal uses)														
6	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10-21	a) 0.3 b) 0.6	a) 144 b) 288	300-1500	14	Dose range 0.18-0.3 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 6a,b,c	Unacceptable risk for soil organisms
6i	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes orana</i> [CAPURE]	Spraying	>BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.3 b) 0.6	a) 144 b) 288	300-1500	14	Dose range 0.18-0.3 L/ha If higher tier assessment in Folsomia is not accepted please refer to Use No. 6a,b,c	Unacceptable risk for soil organisms
6a	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes orana</i> [CAPURE]	Spray	BBCH >70	a) 1 b) 1	NR	a) 0.3 b) 0.3	a) 144 b) 144	300-1500	14	Dose range 0.18-0.3 L/ha Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
6b	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes orana</i> [CAPURE]	Spray	>BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.225 b) 0.450	a) 108 b) 216	300-1500	14	Dose range 0.18-0.225 Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Use- No.	Member state(s)	Crop and/or situation (crop destination / purpose of crop)	F, G or I	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	App. Method / Kind	App timing / Growth stage of crop & season	Max app n° a) per use b) per crop/ season	Min. interval between applications (days)	L product / ha a) max. rate per appl. b) max. total rate per crop/season	g or kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha (min / max)	PHI (days)	Remarks: Effective rates (L/ha)	zRMS Conclusions (Soil organisms)
6c	PL	Pear [PYUCO] Crab apple [MABSY] Quince [CYDOB]	F	<i>Adoxophyes</i> <i>orana</i> [CAPURE]	Spray	BBCH 40-59 (up to 1 treatment) >BBCH 70 (up to 2 treatments)	a) 2 b) 2	10	a) 0.225 b) 0.450	a) 108 b) 216	300-1500	14	Dose range 0.18-0.225 Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms
7	PL	Garlic [ALLSA] Shallot [ALLAS]	F	<i>Thrips</i> sp. [THRISP]	Spraying	Notice of the pest / first damages BBCH 10-49	a) 1 b) 1	na	a) 0.15 b) 0.15	a) 72 b) 72	200-600	7	If higher tier assessment in Folsomia is not accepted please refer to Use No. 7a	Unacceptable risk for soil organisms
7a	PL	Garlic [ALLSA] Shallot [ALLAS]	F	<i>Thrips</i> sp. [THRISP]	Spraying	Notice of the pest / first damages BBCH 40-49	a) 1 b) 1	na	a) 0.16 b) 0.16	a) 76.8 b) 76.8	200-600	7	Dose range 0.15-0.16 L/ha Intended Use just in case high-er tier assessment in Folsomia is not accepted	Acceptable risk for soil organisms

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 (<i>e.g.</i> 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 <u>and</u> 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
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9.1.1 Overall conclusions

The present dossier, risk assessment and conclusions are based on the data available for the active substance and the reference product since these are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Additionally, the risk assessment was performed with the lowest endpoints available (either coming from the active substance data or the reference product). Please see Part C of this dossier for the ecotoxicological bridging between the two formulations (SAP09I and the reference product).

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, dated January 2004, may be used for the purposes of current evaluation of SAP09I. Spintor 480 SC (GF-976/NAF-85) is a representative formulation. Consequently, the tests used to determine the endpoints provided in the document SANCO/1428/2001- rev. final dated 14 July 2006 were not reassessed.

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)

The risk assessment to birds and mammals was conducted according to the EFSA Guidance Document for the Risk Assessment for Birds and Mammals.

An acceptable risk was obtained for SAP09I at the screening phase for acute and long-term exposure. The risk assessment for drinking water is also acceptable as well as for the secondary poisoning scenarios. Overall, birds present an acceptable risk towards SAP09I when used according to the proposed application patterns.

~~An acceptable risk was obtained for SAP09I at the screening phase for acute exposure. After refinement of endpoint or deposition factor or residue per unit dose with more ecologically relevant figure, an acceptable risk is also observed for the long term exposure in all uses. The risk assessment for drinking water is also acceptable as well as for the secondary poisoning scenarios. Overall, mammals present an acceptable risk towards SAP09I when used according to the proposed application patterns.~~

Review Comments:

The performed risk assessment for mammals demonstrates acceptable risk for following uses of SAP09I:

- Leafy vegetables (use no. 1, 2) – BBCH 10-49, 3 x 96 g a.s./ha with min. interval of 10 days;
- Bulbs and onion like crops (use no. 3, 4, 7) – BBCH 10-39, 3 x 96 g a.s./ha with min. interval of 10 days;
- Bulbs and onion like crops - 1 x 76.8 g a.s./ha) and BBCH $\geq$ 40;
- Orchards (uses no. 5, 6) – BBCH $\geq$ 40, 1-2 x 144 g a.s./ha with min. interval of 21 days
- Orchards – BBCH $\geq$ 40, 2 x 108 g a.s./ha with min. interval of 10 days

An unacceptable chronic risk to mammals (voles) has been identified for the following uses:

- Leafy vegetables (use no. 1, 2) – BBCH 10-49, 3 x 96 g a.s./ha with min. interval of 7 days;
- Bulbs and onion like crops (use no. 3, 4, 7) – BBCH $\geq$ 40, 3 x 96 g a.s./ha with min. interval of 10 days;
- Orchards (uses no. 5, 6) – BBCH $<$ 40, 1-2 x 144 g a.s./ha with min. interval of 10 days
- Orchards (uses no. 5, 6) – BBCH $>$ 70, 2 x 144 g a.s./ha with min. interval of 10 days
- Orchards – BBCH $<$ 40, 2 x 108 g a.s./ha with min. interval of 10 days

Evaluation of exposing to mammals through the drinking water demonstrated the acceptable risk. The risk to earthworm- and fish-eating animals from secondary poisoning is acceptable.

9.1.1.2 Effects on aquatic organisms (KCP 10.2)

The risk assessment to aquatic organisms was conducted according to EFSA Journal 2013; 11(7):3290. An acceptable risk is obtained for SPA09I application according to the intended uses when the following mitigation measures are implemented:

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 10m with vegetated buffer of 5m to surface water bodies in applications to leafy vegetables (uses no. 1, 2) and bulb and onion like vegetables (uses no. 3, 4, 7).

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 40m or 20 m with 95% DRN to surface water bodies in early applications to orchards (uses no. 5, 6).

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 30m or 10 m with 90% DRN or 20 m with 50% DRN to surface water bodies in late applications to orchards (uses no. 5, 6).

9.1.1.3 Effects on bees (KCP 10.3.1)

The risk assessment for bees was conducted according to SANCO/10329/2002 rev 2 final and EFSA Journal 2013;11(7):3295. An unacceptable risk is found for bees ~~and bumble bees~~ exposure to SAP09I. Therefore, a restriction must be imposed in the application of this product according to the proposed application patterns:

SPe8: Dangerous to bees. To protect bees and other pollinating insects do not apply on flowering crops. Do not use where bees are actively foraging. Do not apply when flowering weeds are present. Remove weeds before flowering.

9.1.1.4 Effects on arthropods other than bees (KCP 10.3.2)

The risk assessment for non-target arthropods was conducted according to SANCO/10329/2002 rev 2 final and ESCORT 2. No unacceptable risk is expected when SAP09I is applied according to the proposed application patterns when the following mitigation measures are in place:

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 5m or 1m with 75% DRN to non-agricultural land in applications to vegetables.

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 40m or 30m with 50% DRN or 15m with 90 % DRN to non-agricultural land in applications to orchards.

~~SPE3: To protect non target arthropods, respect an unsprayed buffer zone of 20m or 10m with 50% DRN or 5m with 75 % DRN to adjacent crops/non agricultural land in applications to vegetables.~~

~~SPE3: To protect non target arthropods, respect an unsprayed buffer zone of 20m or 10m with 50% DRN or 5m with 75 % DRN to adjacent crops/non agricultural land in applications to orchards.~~

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

The risk assessment for soil macro- and mesofauna was conducted according to SANCO/10329/2002 rev 2 final. ~~No unacceptable risk is expected for soil macro and mesofauna when exposed to SAP09I according to the proposed application patterns.~~

Review Comments:

The chronic TER values for earthworms and *H. aculeifer* exposed to spinosad and its relevant metabolites were greater than the Commission Regulation (EU) No. 546/2011 trigger of 5, indicating that the risk to earthworms and *H. aculeifer* is acceptable following use of SAP09I according to the proposed use pattern.

For *F. candida* the chronic TER value are lower than 5, therefore further assessment is required. The refined PEC_{SOIL} or additional higher tier data should be provided.

Currently, available data are considered as not sufficient and the unacceptable risk assessment for soil organisms is identified.

During the commenting process, the Applicant introduced modifications to the proposed scope of application of SAP09I (Volkete Max). Additional risk assessment was performed by the Applicant. Based on it, the acceptable risk was demonstrated for following uses:

- Use no. 1, 2 (cabbage, broccoli, cauliflower) application rate:

- 3 x 0.1 L/ha (3 x 48 g a.s./ha), interval: 10d, BBCH 10-49
- 2 x 0.16 L/ha (2 x 76.8 g a.s./ha), interval: 10d, BBCH 20-49

- Uses no. 3, 4, 7 (onion, leek, garlic) application rate:

- 1 x 0.15-0.16 L/ha (1 x 72-76.8g a.s./ha), BBCH 40-49

- Uses no. 5, 6 (pear, wild apple, quince) application rate:

- 1 x 0.3 L/ha (1x 144 g a.s./ha) BBCH >70
- 2 x 0.225 L/ha (2 x 108 g a.s./ha) BBCH >70, interval: 10d
- 2 x 0.225 L/ha (2 x 108 g a.s./ha) BBCH 10/40-59 (1st appl.) / BBCH>70 (2nd appl.), interval: 10d

The risk assessment for microorganisms was conducted according to SANCO/10329/2002 rev 2 final. An acceptable risk is proven for microorganisms when exposed to the active substance and its metabolites and consequently to the formulated product according to the proposed application patterns.

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

The risk assessment for non-target terrestrial plants was conducted according to SANCO/10329/2002 rev 2 final. Since spinosad is an insecticide, a Tier 1 risk assessment was conducted, and an acceptable risk is proven when SAP09I is applied at the proposed application rates.

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

No available data and not relevant as an acceptable is proven for all assessed organisms.

9.1.2 Grouping of intended uses for risk assessment

The following table documents the grouping of the intended uses to support application of the risk envelope approach (according to SANCO/11244/2011).

Table 9.1-3: Critical use pattern of SAP09I grouped according to application pattern

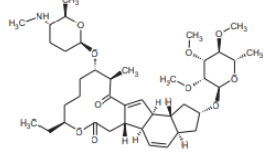
Grouping according to application pattern			
Group Critical use	Intended uses	relevant use parameters for grouping	relevant parameter or value for sorting
9.2 and 9.3 Effects on birds and vertebrates other than birds (screening assessment)			
Leafy vegetables Cabbage (use no. 1)	Cabbage, Broccoli, Cauliflower (use no. 1, 2)	Crop group according to EFSA/2009/1438	3 x 96 g a.s./ha, interval 7 days BBCH 10-49
Bulbs and onion like crops Onion (use no. 3)	Onion, Leek (use no. 3, 4, 7)		3 x 96 g a.s./ha, interval 10 days BBCH 10-49
Orchards (use no. 5, 6)	Apple, Pear, Wild apple, Quince (use no. 5, 6)		2 x 144 g a.s./ha, interval 10 days BBCH 10-59 & >70
9.4 Effects on aquatic organisms			
Leafy vegetables Cabbage (use no. 1)	Cabbage, Broccoli, Cauliflower (use no. 1, 2)	Grouped according to E-Fate.	3 x 96 g a.s./ha, interval 7 days BBCH 10-49
Bulbs and onion like crops Onion (use no. 3)	Onion, Leek (use no. 3, 4, 7)	Grouped according to E-Fate.	3 x 96 g a.s./ha, interval 10 days BBCH 10-49
Orchards (use no. 5, 6)	Apple, Pear, Wild apple, Quince (use no. 5, 6)	Grouped according to E-Fate.	1 x 144 g a.s./ha, BBCH 10-59 2 x 144 g a.s./ha, interval 10 days BBCH >70

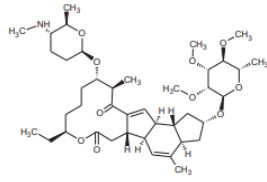
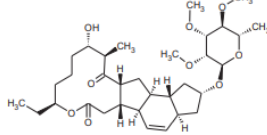
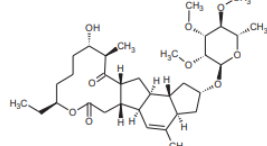
Grouping according to application pattern			
Group Critical use	Intended uses	relevant use parameters for grouping	relevant parameter or value for sorting
9.5 Effects on bees (screening assessment)			
Orchards (use no. 5, 6)	Cabbage, Broccoli, Cauliflower (use no. 1, 2) Onion, Leek (use no. 3, 4, 7) Apple, Pear, Wild apple, Quince (use no. 5, 6)	Crop group according to application pattern	1 x 144 g a.s./ha
9.6 Effects on non-target arthropods (screening assessment)			
Orchards (use no. 5, 6)	Cabbage, Broccoli, Cauliflower (use no. 1, 2) Onion, Leek (use no. 3, 4, 7) Apple, Pear, Wild apple, Quince (use no. 5, 6)	Crop group according to application pattern	2 x 144 g a.s./ha
9.7 and 9.8 Effects on soil macro- and mesofauna and microbial activity (screening assessment)			
Bulbs and onion like crops Onion (use no. 3)	Cabbage, Broccoli, Cauliflower (use no. 1, 2) Onion, Leek (use no. 3, 4, 7) Apple, Pear, Wild apple, Quince (use no. 5, 6)	Grouped according to E-Fate.	3 x 96 g a.s./ha, interval 10 days BBCH 10-49
9.9 Effects on non-target terrestrial plants (screening assessment)			
Leafy vegetables Cabbage (use no. 1)	Cabbage, Broccoli, Cauliflower (use no. 1, 2) Onion, Leek (use no. 3, 4, 7) Apple, Pear, Wild apple, Quince (use no. 5, 6)	Crop group according to application pattern	3 x 96 g a.s./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 SAP09I is indicated in the table.

Table 9.1-4 Metabolites of spinosad

Metabolite	Chemical structure	Molar mass	Maximum occurrence in compartments	Risk assessment required?
Spinosyn B		718	Soil: 67% Water: - Sediment: 9%	Yes, aquatic and soil organisms

Metabolite	Chemical structure	Molar mass	Maximum occurrence in compartments	Risk assessment required?
N-demethylated spinosyn D		732	Soil: 68% Water: - Sediment: 10%	Yes, aquatic and soil organisms
B-13,14-dihydropseudoglycone of spinosyn A		593	Soil: - Water: - Sediment: -	No
β-13,14-dihydropseudoglycone of spinosyn D		607	Soil: - Water: - Sediment: -	No

9.2 Effects on birds (KCP 10.1.1)

9.2.1 Toxicity data

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

Effects on birds of SAP09I were not evaluated as part of the EU assessment of spinosad.

However, the provision of further data on the formulation SAP09I is not considered essential, because the toxicity of the formulated product can be reliably predicted on the basis of the active substance data.

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
Mallard duck Bobwhite quail	spinosad	Acute	LD₅₀ > 2000 mg/kg bw	SANCO/1428/2001 – rev. final (2006)
Mallard duck Bobwhite quail	spinosad	Dietary	LC ₅₀ >5253 mg/kg feed (bobwhite quail) LC ₅₀ >5156 mg/kg feed (mallard duck)	SANCO/1428/2001 – rev. final (2006)
Mallard duck Bobwhite quail	spinosad	Reproductive toxicity	NOEC = 550 mg/kg feed (equivalent to 68.54 mg/kg bw/d for bobwhite quail¹ and 78.06 mg/kg bw/d for mallard duck ²) (2-generation rat, based on litter size, body weight and food consumption)	SANCO/1428/2001 – rev. final (2006)

1 Converted to daily dose accounting for a mean body weight of 210.17 g and feed consumption of 26.19 g feed/bird/d of the birds exposed at the 550 mg/kg diet treatment.

2 Converted to daily dose accounting for a mean body weight of 1149.5 g and feed consumption of 162.9 g feed/bird/d of the birds exposed at the 550 mg/kg diet treatment.

Review Comments:

The conversion of chronic endpoints from mg/kg feed to mg/kg bw/day is correct.

9.2.1.1 Justification for new endpoints

Not relevant.

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: Screening assessment of the acute and long-term/reproductive risk for birds due to the use of SAP09I in all intended uses

Intended use		all intended uses				
Active substance/product		spinosad				
Acute toxicity (mg/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	
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days	Small omnivorous bird	158.8	1.7 1.6	25.92 24.39	77.2 82.0	
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small omnivorous bird	158.8	1.5	22.87	87.5	
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days	Small insectivorous bird	46.8	1.3	8.76	228.3	

Reprod. toxicity (mg/kg bw/d)		68.54			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{tr}
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days	Small omnivorous bird	64.8	2.0 x 0.53	6.59	10.4
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small omnivorous bird	64.8	1.8 x 0.53	5.93	11.6
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days	Small insectivorous bird	18.2	1.5 x 0.53	2.08	33

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 Higher-tier risk assessment

Not needed as risk is acceptable at screening risk assessment.

9.2.2.3 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 SAP09I is 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 must be considered.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for leafy vegetables (use no. 1) covers the risk for birds from all intended uses (see 9.1.2).

Table 9.2-3: Assessment of the acute risk for birds due to exposure to spinosad via contaminated drinking water in leaf whorls in cabbage (use no. 1) – risk envelope approach

Intended use		leafy vegetables (use no. 1) – risk envelope approach				
Active substance		spinosad				
Application rate (g/ha)		1 x 96				
Acute toxicity (mg/kg bw)		2000				
TER criterion		10				
(Single) applic. rate (g/ha)	Water applic. rate (L/ha)	C_{spray-sol.} (g/L)	PEC_{leaf-whorl} = C_{spray-sol.}/5 (mg/L)	DW uptake (L/kg bw/d)	Daily dose (mg/kg bw/d)	TER_a
96	200	0.48	0.096	0.46	0.04416	45289.9

C_{spray-sol.}: concentration in spray solution; PEC_{leaf-whorl}: concentration in pools in leaf whorls; DW: drinking water; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

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 ($K_{oc} < 500$ L/kg) or 3000 in the case of more sorptive substances ($K_{oc} \geq 500$ L/kg).

With a $K(f)_{oc}$ of 8281.6 (geomean), spinosyn A belongs to the group of more sorptive substances. Spinosyn D $K(f)_{oc}$ is not available and it is assumed that it has sorption characteristics equal to spinosyn A.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 5) covers the risk for birds from all intended uses (see 9.1.2).

Effective application rate (g/ha)	=	144		
Acute toxicity (mg/kg bw)	=	2000	quotient =	0.072 < 3000
Reprod. toxicity (mg/kg bw/d)	=	68.54	quotient =	2.101 < 3000

Review Comments:

In the risk assessment for birds via drinking water the MAF value should be calculated based on DT_{50} in soil. Nevertheless, the Applicant not included in the evaluation any MAF value. Thus, zRMS corrected assessment accordingly.

The effective application rate should be calculated using following equations:

$$AR_{eff} = \text{Application rate (g/ha)} \times MAF_{mean}$$

where

$MAF_m = (1 - e^{-nki}) / (1 - e^{-ki})$ with $k = \ln(2)/DT_{50}$ (rate constant), n = number of applications and i = application interval [d]

Correct MAF_m for spinosad for use in orchards (2 x 0.144 kg a.s./ha with 10 days interval) is 1.895.

Effective application rate (g/ha)	=	272.91		
Acute toxicity (mg/kg bw)	=	2000	quotient =	0.14
Reprod. toxicity (mg/kg bw/d)	=	68.54	quotient =	3.98

*mean MAF(Spinosyn D DT_{50soil} = 62.6 d)

Correct MAF_m for spinosad for use in leafy vegetables (3 x 0.096 kg a.s./ha with 7 days interval) is 2.782.

Effective application rate (g/ha)	=	267		
Acute toxicity (mg/kg bw)	=	2000	quotient =	0.13
Reprod. toxicity (mg/kg bw/d)	=	68.54	quotient =	3.90

*mean MAF(Spinosyn D DT_{50soil} = 62.6 d)

9.2.2.4 Effects of secondary poisoning

The max log P_{ow} of spinosyn A and D amounts to 5.16 and 5.21, respectively, and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

Risk assessment for earthworm-eating birds via secondary poisoning

According to EFSA/2009/1438, the risk for vermivorous birds is assessed for a bird of 100 g body weight with a daily food consumption of 104.6 g. Bioaccumulation in earthworms is estimated based on predicted concentrations in soil.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for bulb and onion like vegetables (use no. 3) covers the risk for birds from all other intended uses (see 9.1.2).

Table 9.2-4: Assessment of the risk for earthworm-eating birds due to exposure to spinosad via bioaccumulation in earthworms (secondary poisoning) for the intended use in bulb and onion like vegetables (use no. 3) – risk envelope approach

Parameter	Spinosad	Comments
PEC _{soil} (twa = 21 d) (mg/kg soil)	0.237	Tier I PECs initial (worst case) for onion, leek, garlic (uses no. 3, 4, 7) – risk envelope approach
log P _{ow} / P _{ow}	5.185 / 153109	Mean of spinosyn A (5.16) and D (5.21)
Koc	8281.6	Geomean (n = 5)
foc	0.02	Default
BCF _{worm}	11.0932	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	2.6291	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	2.7606	$DDD = PEC_{worm} \times 1.05$
NOEL (mg/kg bw/d)	68.54	
TER _{lt}	24.83	>5, no further refinement needed

TER values shown in bold fall below the relevant trigger.

Risk assessment for fish-eating birds via secondary poisoning

According to EFSA/2009/1438, the risk for piscivorous birds is assessed for a bird of 1000 g body weight with a daily food consumption of 159 g. Bioaccumulation in fish is estimated based on predicted concentrations in surface water as a limit value for admissible concentrations of spinosad in water.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 6) covers the risk for birds from all intended uses (see 9.1.2).

Table 9.2-5: Assessment of the risk for fish-eating birds due to exposure to spinosad via bioaccumulation in fish (secondary poisoning) for the intended use in orchards (use no. 6) – risk envelope approach

Parameter	Spinosad	Comments
PEC _{sw} (mg/L)	0.0092 0.00956	Step 1 PEC _{sw} (worst case) for orchards late application (uses no. 6) onion (uses no. 3, 4, 7) - risk envelope approach
BCF _{fish}	114.5	Mean of spinosyn A (114 L/kg) and D (115 L/kg)
BMF	-	biomagnification factor (relevant for BCF ≥ 2000)
PEC _{fish}	1.0534 1.095	$PEC_{fish} = PEC_{water} \times BCF_{fish}$
Daily dietary dose (mg/kg bw/d)	0.1675 0.174	$DDD = PEC_{fish} \times 0.159$
NOEL (mg/kg bw/d)	68.54	
TER _{lt}	409.19 393.9	>5, no further refinement needed

TER values shown in bold fall below the relevant trigger.

9.2.2.5 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

The risk assessment to birds was conducted according to the EFSA Guidance Document for the Risk Assessment for Birds and Mammals.

An acceptable risk was obtained for SAP09I at the screening phase for acute and long-term exposure. The risk assessment for drinking water is also acceptable as well as for the secondary poisoning scenarios. Overall, birds present an acceptable risk towards SAP09I when used according to the proposed application patterns.

Review Comments:

The acute and chronic risks of SAP09I to birds were assessed from toxicity exposure ratios between toxicity endpoints, estimated from studies with active ingredient, and maximum residues occurring on food items.

All TER values exceed the relevant triggers indicating that SAP09I 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. The risk to earthworm- and fish-eating animals from secondary poisoning is low.

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 spinosad. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on mammals of SAP09I were not evaluated as part of the EU assessment of spinosad.

However, the provision of further data on the formulation SAP09I is not considered essential, because the toxicity of the formulated product can be reliably predicted on the basis of the active substance data.

The selection of studies and endpoints for the risk assessment is in line with 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	spinosad	Acute	LD ₅₀ > 2000 mg/kg bw	SANCO/1428/2001 – rev. final (2006)

Species	Substance	Exposure System	Results	Reference
Rat	spinosad	Reproductive toxicity	NOEC = 100 mg/kg feed (equivalent to 10 mg/kg bw; 2-generation rat, based on litter size, bodyweight and food consumption)	SANCO/1428/2001 – rev. final (2006)

9.3.1.1 Justification for new endpoints

Not relevant.

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: Screening assessment of the acute and long-term/reproductive risk for mammals due to the use of SAP09I in all intended uses

Intended use	all intended uses				
Active substance/product	spinosad				
Acute toxicity (mg/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
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days	Small herbivorous mammal	136.4	1.6	20.95	95.5
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small herbivorous mammal	118.4	1.5	17.05	117.3
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days	Small herbivorous mammal	136.4	1.3	25.53	78.3

Reprod. toxicity (mg/kg bw/d)		10			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{lt}
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days	Small herbivorous mammal	72.3	2.0 x 0.53	7.36	1.40
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small herbivorous mammal	48.3	1.8 x 0.53	4.42	2.30
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days	Small herbivorous mammal	72.3	1.5 x 0.53	8.28	1.20

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.

Table 9.3-3: First-tier assessment of the long-term/reproductive risk for mammals due to the use of SAP09I in all intended uses

Intended use		all intended uses			
Active substance/product		spinosad			
Reprod. toxicity (mg/kg bw/d)		10			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{lt}
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days BBCH 10-49 2 x 96 g as/ha Min. interval 10 days	Small insectivorous mammal “shrew” BBCH 10 - 19	4.2	2.0 x 0.53	0.43	23.30
	Small insectivorous mammal “shrew” BBCH ≥20	1.9	2.0 x 0.53	0.19	52.60
	Small herbivorous mammal “vole” BBCH 40-49	72.3	2.0 x 0.53	7.36	1.40
	Small herbivorous mammal “vole” BBCH 40-49	72.3	1.5 x 0.53	5.52	1.81
	Large herbivorous mammal “lagomorph” All season	14.3	2.0 x 0.53	1.46	6.80
	Small omnivorous mammal “mouse” BBCH 10-49	7.8	2.0 x 0.53	0.79	12.70
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days 1x76.8 g as/ha	Small insectivorous mammal “shrew” BBCH 10 - 19	4.2	1.8 x 0.53	0.38	26.30
	Small insectivorous mammal “shrew” BBCH ≥20	1.9	1.8 x 0.53	0.17	58.80
	Small herbivorous mammal “vole” BBCH ≥40	43.4	1.8 x 0.53	3.97	2.50
	Small herbivorous mammal “vole” BBCH ≥40	43.4	1.0 x 0.53	1.77	5.65

Intended use	all intended uses				
Active substance/product	spinosad				
Reprod. toxicity (mg/kg bw/d)	10				
TER criterion	5				
Crop scenario Growth stage	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{it}
	Small omnivorous mammal “mouse” BBCH 10-39	7.8	1.8 x 0.53	0.71	14.10
	Small omnivorous mammal “mouse” BBCH ≥40	4.7	1.8 x 0.53	0.43	23.30
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days BBCH 10-59 & >70 1x144 g as/ha 2 x 108 g a.s./ha, 10d interval	Small herbivorous mammal “vole” BBCH 10- 19	57.8	1.5 x 0.53	6.62	1.50
	Small herbivorous mammal “vole” BBCH 10-19	57.8	1.5 x 0.53	4.96	2.01
	Small herbivorous mammal “vole” BBCH 20-40	43.4	1.5 x 0.53	4.97	2.00
	Small herbivorous mammal “vole” BBCH 20-40	43.4	1.5 x 0.53	3.73	2.68
	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.5 x 0.53	2.48	4.00
	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.0 x 0.53	1.67	6.04
	Frugivorous mammal “dormouse” BBCH 71-79	22.7	1.5 x 0.53	2.6	3.80
	Frugivorous mammal “dormouse” BBCH 71-79	22.7	1.0 x 0.53	1.73	5.77
	Large herbivorous mammal “lagomorph” BBCH 10- 19	11.5	1.5 x 0.53	1.32	7.60
	Large herbivorous mammal “lagomorph” BBCH 20-40	8.6	1.5 x 0.53	0.98	10.20
	Large herbivorous mammal “lagomorph” BBCH ≥40	4.3	1.5 x 0.53	0.49	20.40
	Small omnivorous mammal “mouse” BBCH 10- 19	6.2	1.5 x 0.53	0.71	14.10
	Small omnivorous mammal “mouse” BBCH 20-40	4.7	1.5 x 0.53	0.54	18.50
	Small omnivorous mammal “mouse” BBCH ≥40	2.3	1.5 x 0.53	0.26	38.50
Orchards Appl. rate: 1 x 144 g as/ha BBCH ≥ 40	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.0 x 0.53	1.66	6.02
Orchards Appl. rate: 2 x 108 g as/ha Min. interval: 10 days BBCH ≥ 40	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.5 x 0.53	1.86	5.36

Intended use	all intended uses				
Active substance/product	spinosad				
Reprod. toxicity (mg/kg bw/d)	10				
TER criterion	5				
Crop scenario Growth stage	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{tt}
Orchards Appl. rate: 2 x 144 g as/ha Min. interval: 21 days BBCH ≥ 40	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.2 x 0.53	1.99	5.03

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.

After first-tier risk assessment, further assessment is still needed for the vole and dormouse species in all intended uses.

9.3.2.2 Higher-tier risk assessment

The first-Tier risk assessment after the application of SAP09I indicates a long-term risk for small herbivorous mammal and frugivorous mammal. Thus, a refined risk assessment for long-term exposure of mammals is presented below following independent approaches:

- refinement of reproductive endpoint (NOEC vs BMD) or
- refinement of DF/IF and RUD.

Refinement of reproductive endpoint (NOEC vs BMD)

The NOEC approach has some flaws as the definition of such values is subject of the assessor's views, cannot be compared between studies, often does not represent the quality of the data available and, specially, is highly dependent on the dose gradient chosen. Therefore, nowadays the Benchmark Dose Approach (BMD) has shown to be more precise and accurate when defining values for risk assessment.

The BMD takes into account the dose-response relationship while estimating a BMDL (the lower confidence limit of the benchmark dose) which can be further used in the risk assessment.

Review Comments:

zRMS has concerns about the applicability of the benchmark dose (BMD) approach to spinosad. The current list of endpoints was established in 2006 based on an assessment of active substance data contained mainly in the DAR (2001). The information included in the old DAR regarding studies on mammals are very limited, making a robust analysis of the results almost impossible. Spinosad is currently undergoing re-evaluation, during whose course all available data will be verified for validity using currently accepted guidelines. Therefore, in zRMS opinion, the NOEL = 10 mg a.s./kg bw/d for mammals should not be revised. Thus, further analysis of the BMDL was not performed by the zRMS.

According to the endpoint description present in the spinosad review report SANCO/1428/2001 – rev. final (2006) (NOEC = 100 mg/kg feed; 2-generation rat, based on litter size, bodyweight and food consumption), the NOEC is defined based on three main parameters: litter size, bodyweight and food consumption. When looking at the study summaries available in spinosad DAR (2001) section 6, there are

two generational studies performed in rat and one performed in rabbit. The only parameter that is consistently affected in all three studies is the body weight. Thus, this will be used to derive the BMDL.

Species	Route	NOAEL mg/kg bw/day	LOAEL mg/kg bw/day	Critical effects	Notifier Reference
Reproduction studies					
rat	oral	parental: 10	parental: 100	- mortality, dystocia, vaginal bleeding, changes in body and organ weight, histological changes in several organs	Breslin <i>et al.</i> , 1994
		developmental: 10	developmental: 100	- decreased gestation survival, litter size, pup weight, and neonatal survival	
		reproductive: 10	reproductive: 100	- dystocia, vaginal bleeding, decreased litter size	
Teratogenicity studies					
rat	oral	maternal: 50	maternal: 200	- decreased body weights	Liberacki <i>et al.</i> , 1993
		developmental: >=200	developmental: -	- no effects	
		teratogenicity: >=200	teratogenicity: -	- no effects	
rabbit	oral	maternal: 10	maternal: 50	- decreased body weight gains, feed consumption, and fecal output, abortions	Liberacki <i>et al.</i> , 1994
		developmental: >=50	developmental: -	- no effects	
		teratogenicity: >=50	teratogenicity: -	- no effects	

The first study mentioned in the table above (Breslin *et al.*, 1994) assessed the effect of the active substance at the concentrations of 0, 3, 10 and 100 mg/kg bw/d in rats. The bodyweight of two generations of parents was affected at maximum 7 and 4% in P1 and P2, respectively, while the litter had an effect of around 10% in all assessed generations (F1a, F1b and F2).

The second study mentioned in the table above (Liberacki *et al.*, 1993) assessed the effect of the active substance at the concentrations of 0, 10, 50 and 200 mg/kg bw/d in rats. The bodyweight of pregnant females was transiently affected during the gestation (namely between days 6-12), but no statistically significant differences can be found at the end of the gestation. No changes were observed in the body weight of foetuses.

The third study mentioned in the table above (Liberacki *et al.*, 1994) assessed the effect of the active substance at the concentrations of 0, 2.5, 10 and 50 mg/kg bw/d in rabbits. The bodyweight of pregnant females was transiently affected during the gestation (namely between days 7-10), but no statistically significant differences can be found at the end of the gestation. No changes were observed in the body weight of foetuses.

Looking at all these results, the bodyweight of the litter of the first study (Breslin *et al.*, 1994) will be used for the BMD analysis to obtain a more accurate value for risk assessment.

A. Data description

The BMD will only be performed for the most affected parameter so that the lowest BMDL can be selected for the risk assessment as a conservative approach. The results of the study are described in the spinosad DAR (2001) as follows:

Input data:

Dose levels (mg/kg bw/d)	Generation	Sex	Weight
0	F1a	F	56.7
0	F1a	M	59.6
0	F1b	F	55.6
0	F1b	M	57.4
0	F2	F	55.9
0	F2	M	58.0
3	F1a	F	56.1
3	F1a	M	59.3
3	F1b	F	55.9
3	F1b	M	59.3
3	F2	F	57.7
3	F2	M	60.9
10	F1a	F	58.1
10	F1a	M	60.7
10	F1b	F	55.7
10	F1b	M	58.7
10	F2	F	57
10	F2	M	59.7
100	F1a	F	51.5
100	F1a	M	52.9
100	F1b	F	50.5
100	F1b	M	51.3
100	F2	F	52.3
100	F2	M	53.4

From this data, three BMD analysis were conducted:

- 1) Considering all data (with generation or sex as covariates)
- 2) Considering only F1 generations (with generation or sex as covariates)
- 3) Considering only F2 generation (with sex as covariate)

A. Selection of the BMR

A small change was made in the parameters in order to be more conservative: the BMR was changed from the default value of 0.1 to 0.05 which indicates that only a 5% effect on the population is considered acceptable instead of the default 10%.

B. Software used

The software used to run the BMD was the PROAST, available at <https://proastweb.rivm.nl/>. This was chosen especially because and as it says in the PROAST website: “Examples of useful functionalities in PROAST are the possibility of statistically comparing dose-response relationships among subgroups (covariate analysis), and the larger flexibility in plotting. PROAST closely follows the EFSA guidance on the BMD approach, while BMDS does not allow for performing BMD analyses according to EFSA guidance in all respects.”

C. Specification of deviations from default assumptions

No changes were made to the default settings with the exception of the BMR value.

Removed data	No
Dose column	Dose levels (mg/kg bw/d)
Response column	Weight (g)
Type of response data	Continuous, value for each individual
Covariate	Sex/Generation
BMR	0.05
Model averaging	No
AIC criterion	2

D. Results

Fitted models

model	converged	loglik	npar	AIC
Considering the entire dataset and sex as covariate				
Hill m3-ab	1	64.46	6	-116.92
Considering the entire dataset and generation as covariate				
Hill m3-a	1	54.88	6	-97.76
Considering the F1 dataset and sex as covariate				
Hill m3-ab	1	44.19	6	-76.38
Considering the F1 dataset and generation as covariate				
Hill m3-a	1	37.04	5	-64.08
Considering the F2 dataset and sex as covariate				
Hill m3-a	1	21.93	5	-33.86

BMD Confidence Interval

Considering the entire dataset and sex as covariate		
Sex	Lowest BMDL	Highest BMDU
CED-F	58.2	92.5
CED-M	48.3	87.1
Considering the entire dataset and generation as covariate		
Generation	Lowest BMDL	Highest BMDU
all	43.7	89.8
Considering the F1 dataset and sex as covariate		
Sex	Lowest BMDL	Highest BMDU
CED-F	53.9	91.9
CED-M	44.5	86.5
Considering the F1 dataset and generation as covariate		
Generation	Lowest BMDL	Highest BMDU
all	36.9	89.4
Considering the F2 dataset and sex as covariate		

Sex	Lowest BMDL	Highest BMDU
all	42.8	93.1

E. Plots of fitted models

The plots of the fitted models can be found in the BMD reports (5 in total) sent in the KCP, B9 folder of this dossier.

F. Conclusions

The lowest BMDL₅ found is **36.9** mg/kg bw/d which will be used in the refined risk assessment below.

Table 9.3-4: First-tier assessment of the long-term/reproductive risk for mammals due to the use of SAP09I in all intended uses

Intended use		all intended uses				
Active substance/product		spinosad				
Reprod. toxicity (mg/kg bw/d)		36.9				
TER criterion		5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days BBCH 10-49	Small herbivorous mammal “vole” BBCH 40-49	72.3	2.0 x 0.53	7.36	5.01	
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small herbivorous mammal “vole” BBCH ≥40	43.4	1.8 x 0.53	3.97	9.29	
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days BBCH 10-59 & >70	Small herbivorous mammal “vole” BBCH 10- 19	57.8	1.5 x 0.53	6.62	5.57	
	Small herbivorous mammal “vole” BBCH 20-40	43.4	1.5 x 0.53	4.97	7.42	
	Small herbivorous mammal “vole” BBCH ≥40	21.7	1.5 x 0.53	2.48	14.88	
	Frugivorous mammal “dormouse” BBCH 71-79	22.7	1.5 x 0.53	2.6	14.19	

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.

Refinement of Deposition Factor (DF/IF) &

In the refined risk assessment interception/deposition factors according to Generic Guidance for Tier 1 FOCUS Ground Water Assessments, 2021 were used.

Crop	Stage				
	BBCH [#] 0–9	BBCH [#] 10–69		BBCH [#] 71–75	BBCH [#] 76–89
Apples	without leaves 50	flowering 60		early fruit development 65	full canopy 65
	BBCH [#] 0–9	BBCH [#] 10–69			BBCH [#] 71–89
Bushberries	without leaves 40	flowering 60		flowering 60	full foliage 75
Citrus	all stages 80				
	BBCH [#] 0–9	BBCH [#] 11–13	BBCH [#] 14–19	BBCH [#] 53–69	BBCH [#] 71–89
Vines	without leaves 40	first leaves 50	leaf development 60	flowering 60	ripening 75

[#]The BBCH code is indicative (Meier, 2001).

Figure: 1 Interception factors according to the Generic Guidance for Tier 1 FOCUS Ground Water Assessments (2021)

Crop	Bare – emergence	Leaf development	Stem elongation		Flowering		Senescence Ripening
	BBCH [#]						
	0– 09	10–19	20–39		40–89		90–99
Beans (field + vegetable)	0	25	40		70		80
Cabbage	0	25	40		70		90
Carrots	0	25	60		80		80
Cotton	0	30	60		75		90
Grass ^{##}	0	40	60		90		90
Linseed	0	30	60		70		90
Maize	0	25	50		75		90
Oil seed rape (summer)	0	40	80		80		90
Oil seed rape (winter)	0	40	80		80		90
Onions	0	10	25		40		60
Peas	0	35	55		85		85
Potatoes	0	15	60		85		50
Soybean	0	35	55		85		65
Spring cereals	0	0	BBCH 20–29*	BBCH 30–39*	BBCH 40–69	BBCH 70–89	80
			20	80	90	80	
Strawberries	0	30	50		60		60
Sugar beets	0	20	70 (rosette)		90		90
Sunflower	0	20	50		75		90
Tobacco	0	50	70		90		90
Tomatoes	0	50	70		80		50
Winter cereals	0	0	BBCH 20–29*	BBCH 30–39*	BBCH 40–69	BBCH 70–89	80
			20	80	90	80	

[#] The BBCH code is indicative (Meier, 2001).

^{##} A value of 90 is used for applications to established turf

* BBCH code of 20-29 for tillering and 30-39 for elongation

Figure: 2 Interception factors according to the Generic Guidance for Tier 1 FOCUS Ground Water Assessments (2021)

Review Comments:

According to “Working document on Risk Assessment of Plant Protection Products in the Central Zone – Ecotoxicology” Version 3.0 (December 2024), point 3.2.15, the interception values following EFSA Guidance Document to obtain DegT50 values, can be use in the Tier 2 risk assessment. It should be noted that this rule applies only to the later stages of crop growth.

In Appendix E of EFSA B&M guidance the following recommendation is given:

“It was concluded that estimation of residues on undergrowth vegetation using FOCUS interception factors would become increasingly uncertain with decreasing soil cover of the crop and increasing height of weeds in relation to the crop. Thus reliable predictions are only deemed possible where the largest part of the soil surface is actually covered by the crop from a bird’s eye view and undergrowth vegetation is clearly smaller than the crop plants. Weeds or grasses overgrowing the crop at those stages are deemed unlikely to occur in intensive agriculture, but would anyway not form a part of the diet of small to medium herbivores.”

Therefore, the refined DF values are considered acceptable for those crops type and BBCH stages:

1. Bulbs and onion like crops

In Table 1 Appendix E for onions BBCH ≥ 41 growth stages are recognized satisfactorily high soil coverage by crop plants.

In Table 1.5 of EFSA Guidance Document to obtain DegT50 values for BBCH 40-89 growth stages of onions the interception is 40%. Therefore, DF of 0.6 is considered acceptable.

2. Leafy vegetables

In Table 1 Appendix E for leafy vegetables BBCH ≥ 51 growth stages are recognized satisfactorily high soil coverage by crop plants.

In Table 1.5 of EFSA Guidance Document to obtain DegT50 values for BBCH 40-89 growth stages of cabbage the interception is 70%.

There is no justification for the different approach to IF for BBCH stages ≥ 40 and ≥ 51 . Therefore, in zRMS opinion, for BBCH growth stages of leafy vegetables ≥ 40 , the DF of 0.3 can be used in the risk assessment.

3. Orchards

zRMS disagree with using DF in fruits. The residue data were collected from fruits found within a canopy of leaves. Therefore, these RUD values already include interception and for 100% frugivorous animals the DF in EFSA B&M is 1. Therefore, this refined is crossed out.

Table 9.3-5: Higher-tier assessment of the long-term/reproductive risk for mammals due to the use of SAP09I in all uses – refined parameters (*) are further described and justified in the text

Intended use		all intended uses						
Active substance/product		spinosad						
Reprod. toxicity (mg/kg bw/d)		10						
TER criterion		5						
Focal species	Food category, % in diet	FIR/bw	RUD _m × DF (*) (mg/kg food)	MAF _m × TWA	PT	DDD _m (mg/kg bw/d)	TER _{lt}	
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days BBCH 10-49	Small herbivorous mammal “vole” BBCH 40-49 100% grass	1.33	54.2 x 0.3	2.0 x 0.53	1	2.20	4.55	
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days BBCH 10-49	Small herbivorous mammal “vole” BBCH 40-49 100% grass	1.33	54.2 x 0.3	1.8 x 0.53	1	1.98	5.05	
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small herbivorous mammal “vole” BBCH ≥40 100% grass	1.33	54.2 x 0.6	1.8 x 0.53	1	3.96	2.53	
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days BBCH 10-59 & >70	Small herbivorous mammal “vole” BBCH 10-19 100% grass	1.33	54.2 x 0.4	1.5 x 0.53	1	3.30	3.03	
	Small herbivorous mammal “vole” BBCH 20-40 100% grass	1.33	54.2 x 0.4	1.5 x 0.53	1	3.30	3.03	
	Small herbivorous mammal “vole” BBCH ≥40 100% grass	1.33	54.2 x 0.4	1.5 x 0.53	1	3.30	3.03	
	Frugivorous mammal “dormouse” BBCH 71-79 100% fruit	1.16	19.5 x 0.65	1.5 x 0.53	1	1.68	5.95	

Intended use		all intended uses					
Active substance/product		spinosad					
Reprod. toxicity (mg/kg bw/d)		10					
TER criterion		5					
Focal species	Food category, % in diet	FIR/bw	RUD_m × DF (*) (mg/kg food)	MAF_m × TWA	PT	DDD_m (mg/kg bw/d)	TER_{It}
Orchards (use no. 5, 6) Appl. rate: 1 x 144 g as/ha BBCH < 40	Small herbivorous mammal “vole”	1.33	54.2 x 0.4	1.0 x 0.53	1	2.20	4.55
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha BBCH > 70	Small herbivorous mammal “vole” BBCH ≥ 40 100% grass	1.33	47.2* x 0.35	1.5 x 0.53	0.8**	2.01	4.97

FIR/bw: Food intake rate per body weight; RUD: residue unit dose; DF: deposition factor (considering possible interception by the crop); MAF: multiple application factor; DDD: daily dietary dose; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger. *RUD from B&M guidance Table J.2**Considering CZ Evaluation Manual Dec.2024 The risk for voles on population level could be lower than for other mammalian species at the same calculated TER so a default PT 0.8 is proposed for a more realistic estimation of the exposure

Review Comments:

Orchards

zRMS disagree with using PT of 0.8 without any field data. Therefore the proposed refinement was not accepted by zRMS.

Refinement of Residue per Unit Dose (RUD)

There is an unprotected field study (McCormick, 2005) which was conducted in pasture grass in the Southern and in the Northern European Zones, namely in France, Italy, United Kingdom and Germany.

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies considered in the first registration in 2003 may be used to evaluate SAP09I. These data were included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, made available to the assessor for the purposes of this evaluation. The study by McCormick, 2005 is not included in the provided documentation and, therefore, cannot be used in higher-tier assessment for mammals.

In addition, the residue data collected from the substance-specific studies currently need to be merged with the generic data given in EFSA guidance. The replacement is possible only than the number of new measured residue data is higher than these used to generate the existing RUDs. For food categories like non-grass weeds, grasses and cereals it is very unlikely.

~~The product GF-976, an SC formulation containing 480 g a.s./L, was applied to pasture at 4x130 g a.s./ha with a 7-day interval (worst-case application in relation to the GAP here defended for SAP09I). From this study, a total of four trials are available (one/country) with the following results:~~

Sample/Soil	Days after treatment	Residue total Spinosad (spin A+D, mg/kg)
France	0	16.28
	1	14.22
	2	10.07
	3	9.42
	5	2.26
	7	0.54
	10	0.25
Italy	0	11.45
	1	8.93
	2	3.96
	3	2.69
	5	0.95
	7	0.7
	10	0.35
United Kingdom	0	5.91
	1	4.81
	2	2.43
	3	1.86
	5	0.27
	7	0.18
	10	0.03
Germany	0	9.65
	1	5.43
	2	3.02
	3	1.95
	5	0.33
	7	0.2
	10	0.25

From all these trials, the worst case residue found was 16.28 mg/kg.
Since vole diet is constituted by 100% grass and cereals, these residue results can then be used to further refine the mean residue per unit dose figure in the long term risk assessment.

Looking at the maximum residue values/study, the following data is considered:

Location	Residue
France	16.28
Italy	11.45
UK	5.91
Germany	9.65
RUD _m	10.8225
Application Rate	4 x 130 g a.s./ha (max. 520 g a.s./ha/season)
RUD _m (normalized for 1 kg a.s./ha)	20.8125

The normalized RUD_m is thus applied in the refined risk assessment provided below.

Table 9.3-6: Higher tier assessment of the long-term/reproductive risk for mammals due to the use of SAP09I in all uses – refined parameters (*) are further described and justified in the text

Intended use		all intended uses					
Active substance/product		spinosad					
Reprod. toxicity (mg/kg bw/d)		10					
TER criterion		5					
Focal species	Food category, % in diet	FIR/bw	$RUD_m(*) \times DF(*)$ (mg/kg food)	$MAF_m \times TWA$	PT	DDD_m (mg/kg bw/d)	TER_{tt}
Leafy vegetables (use no. 1, 2) Appl. rate: 3 x 96 g as/ha Min. interval: 7 days BBCH 10-49	Small herbivorous mammal "vole" BBCH 40-49 100% grass	1.33	20.8125 x 0.3	2.0 x 0.53	1	0.85	11.76
Bulbs and onion like crops (use no. 3, 4, 7) Appl. rate: 3 x 96 g as/ha Min. interval: 10 days	Small herbivorous mammal "vole" BBCH ≥40 100% grass	1.33	20.8125 x 0.6	1.8 x 0.53	1	1.52	6.58
Orchards (use no. 5, 6) Appl. rate: 2 x 144 g as/ha Min. interval: 10 days BBCH 10-59 & >70	Small herbivorous mammal "vole" BBCH 10-19 100% grass	1.33	20.8125 x 0.4	1.5 x 0.53	1	1.27	7.87
	Small herbivorous mammal "vole" BBCH 20-40 100% grass	1.33	20.8125 x 0.4	1.5 x 0.53	1	1.27	7.87
	Small herbivorous mammal "vole" BBCH ≥40 100% grass	1.33	20.8125 x 0.4	1.5 x 0.53	1	1.27	7.87

FIR/bw: Food intake rate per body weight; RUD: residue unit dose; DF: deposition factor (considering possible interception by the crop); MAF: multiple application factor; DDD: daily dietary dose; TER: toxicity to exposure ratio. TER values shown in bold fall below the relevant trigger.

Review Comments:

The refined risk assessment for frugivorous mammal performed by the Applicant was not accepted. In higher tier risk assessment performed by zRMS, RUD values for stone fruits given in new EFSA B&M Guidance were used. The default RUD values given in EFSA (2009) had several shortcomings (e.g., only few data were available for some matrices, including fruits). Nowadays available information on residues in pome/stone fruits has been evaluated according to current standards and using a larger dataset (the database developed by CropLife and Lahr et al., 2018; 238 samples for stone fruits (RUD_m = 1.22) and 102 for pome fruits (RUD_m = 0.97)). In z RMS opinion, the residue data evaluated for the purposes of revision B&M guidance are the most reliable for the regulatory use. The WG considered that a geometric mean value was preferable to the arithmetic mean which was used in EFSA (2009) as there is less influence of the extreme values in the distribution.

Intended use		Orchards					
Active substance/product		Spinosad					
Application rate (g/ha)		2 x 144					
Reprod. toxicity (mg/kg bw/d)		NOEL = 10					
TER criterion		5					
Crop scenario	Indicator/generic focal species	FIR/bw	RUD _m *	MAF _m	TWA	DDD _m (mg/kg bw/d)	TER _{it}
Growth stage							
Fruit stage (BBCH 71-79)	Frugivorous mammal "doremouse"	1.16	1.22	1.5	0.53	0.82	31.7

*RUD geomean for stone fruits from Table J.6 of new EFSA B&M Guidance (draft version; 2023); page 297.

Overall conclusion

~~The risk assessment to mammals was conducted according to the EFSA Guidance Document for the Risk Assessment for Birds and Mammals.~~

~~An acceptable risk was obtained for SAP09I at the screening phase for acute as well as the first tier and higher tier phase for long term exposure. The risk assessment for drinking water is also acceptable as well as for the secondary poisoning scenarios. Overall, mammals present an acceptable risk towards SAP09I when used according to the proposed application patterns.~~

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 (K_{oc} < 500 L/kg) or 3000 in the case of more sorptive substances (K_{oc} ≥ 500 L/kg).

With a K(f)_{oc} of 8281.6 (geomean), spinosyn A belongs to the group of more sorptive substances. Spinosyn D K(f)_{oc} is not available and it is assumed that it has sorption characteristics equal to spinosyn A.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 5) covers the risk for mammals from all intended uses (see 9.1.2).

Effective application rate (g/ha) =	144		
Acute toxicity (mg/kg bw) =	2000	quotient =	0.072 < 3000
Reprod. toxicity (mg/kg bw/d) =	10	quotient =	14.4 < 3000

Review Comments:

In the risk assessment for birds via drinking water the MAF value should be calculated based on DT₅₀ in soil. Nevertheless, the Applicant not included in the evaluation any MAF value. Thus, zRMS corrected assessment accordingly.

The effective application rate should be calculated using following equations:

$$AR_{\text{eff}} = \text{Application rate (g/ha)} \times \text{MAF}_{\text{mean}}$$

where

$\text{MAF}_m = (1 - e^{-nki}) / (1 - e^{-ki})$ with $k = \ln(2)/\text{DT}_{50}$ (rate constant), n = number of applications and i = application interval [d]

Correct MAF_m for spinosad for use in orchards (2 x 0.144 kg a.s./ha with 10 days interval) is 1.895.

Effective application rate (g/ha) =	272.91		
Acute toxicity (mg/kg bw) =	2000	quotient =	0.14
Reprod. toxicity (mg/kg bw/d) =	10	quotient =	27.29

*mean MAF(Spinosyn D DT_{50soil} = 62.6 d)

Correct MAF_m for spinosad for use in leafy vegetables (3 x 0.096 kg a.s./ha with 7 days interval) is 2.782.

Effective application rate (g/ha) =	267		
Acute toxicity (mg/kg bw) =	2000	quotient =	0.13
Reprod. toxicity (mg/kg bw/d) =	10	quotient =	26.7

*mean MAF(Spinosyn D DT_{50soil} = 62.6 d)

9.3.2.4 Effects of secondary poisoning

The max log P_{ow} of spinosyn A and D amounts to 5.16 and 5.21, respectively, and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

Risk assessment for earthworm-eating mammals via secondary poisoning

According to EFSA/2009/1438, the risk for vermivorous mammals is assessed for a small mammal of 10 g body weight with a daily food consumption of 12.8 g. Bioaccumulation in earthworms is estimated based on predicted concentrations in soil.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for bulb and onion like vegetables (use no. 3) covers the risk for mammals from all other intended uses (see 9.1.2).

Table 9.3-7: Assessment of the risk for earthworm-eating mammals due to exposure to spinosad via bioaccumulation in earthworms (secondary poisoning) for the intended use in bulb and onion like vegetables (use no. 3) – risk envelope approach

Parameter	Spinosad	Comments
PEC _{soil} (twa = 21 d) (mg/kg soil)	0.237	Tier I PECs initial (worst case) for onion, leek, garlic (uses no. 3, 4, 7) – risk envelope approach
log P _{ow} / P _{ow}	5.185 / 153109	Mean of spinosyn A (5.16) and D (5.21)
Koc	8281.6	Geomean (n = 5)
foc	0.02	Default
BCF _{worm}	11.0932	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	2.6291	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	3.3652	$DDD = PEC_{worm} \times 1.28$
NOEL (mg/kg bw/d)	10	
TER _{lt}	2.97	> 5, no further refinement needed

TER values shown in bold fall below the relevant trigger.

Table 9.3-8: Higer tier assessment of the risk for earthworm-eating mammals due to exposure to spinosad via bioaccumulation in earthworms (secondary poisoning) for the intended use in bulb and onion like vegetables (use no. 3) – risk envelope approach

Parameter	Spinosad		Comments
PEC _{soil} (twa = 21 d) (mg/kg soil)	0.205 (21-d TWA PECs)	0.119 (Tier II PECs initial)	21d-TWA PECs /Tier II PECs initial (worst case) for onion, leek, garlic (uses no. 3, 4, 7) – risk envelope approach
log P _{ow} / P _{ow}	5.185 / 153109	5.185 / 153109	Mean of spinosyn A (5.16) and D (5.21)
Koc	8281.6	8281.6	Geomean (n = 5)
foc	0.02	0.02	Default
BCF _{worm}	11.0932	11.0932	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	2.2741	1.3201	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	2.9108	1.6897	$DDD = PEC_{worm} \times 1.28$
NOEL (mg/kg bw/d)	10	10	
TER _{lt}	3.44	5.92	> 5, no further refinement needed

TER values shown in bold fall below the relevant trigger.

Risk assessment for fish-eating mammals via secondary poisoning

According to EFSA/2009/1438, the risk for piscivorous mammals is assessed for a mammal of 3000 g body weight with a daily food consumption of 425 g. Bioaccumulation in fish is estimated based on predicted concentrations in surface water as a limit value for admissible concentrations of spinosad in water.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 6) covers the risk for mammals from all intended uses (see 9.1.2).

Table 9.3-9: Assessment of the risk for fish-eating mammals due to exposure to spinosad via bioaccumulation in fish (secondary poisoning) for the intended use in orchards (use no. 6) – risk envelope approach

Parameter	Spinosad	Comments
PEC _{sw} (mg/L)	0.0092 0.00956	Step 1 PEC _{sw} (worst case) for orchards late application (uses no. 6) onion (uses no. 3, 4, 7) - risk envelope approach
BCF _{fish}	114.5	Mean of spinosyn A (114 L/kg) and D (115 L/kg)
BMF	-	biomagnification factor (relevant for BCF ≥ 2000)
PEC _{fish}	1.0534 1.095	PEC _{fish} = PEC _{water} × BCF _{fish}
Daily dietary dose (mg/kg bw/d)	0.1496 0.155	DDD = PEC _{fish} × 0.142
NOEL (mg/kg bw/d)	10	
TER _{lt}	66.85 64.5	> 5, no further refinement needed

TER values shown in bold fall below the relevant trigger.

9.3.2.5 Biomagnification in terrestrial food chains

Not relevant.

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

Not relevant.

9.3.4 Overall conclusions

The risk assessment to mammals was conducted according to the EFSA Guidance Document for the Risk Assessment for Birds and Mammals.

An acceptable risk was obtained for SAP09I at the screening phase for acute exposure. ~~After refinement of endpoint or deposition factor or residue per unit dose with more ecologically relevant figure, an acceptable risk is also observed for the long term exposure in all uses.~~ The risk assessment for drinking water is also acceptable as well as for the secondary poisoning scenarios. ~~Overall, mammals present an acceptable risk towards SAP09I when used according to the proposed application patterns.~~

Review Comments:

The performed risk assessment for mammals demonstrates acceptable risk for following uses of SAP09I:

- Leafy vegetables (use no. 1, 2) – BBCH 10-49, 3 x 96 g a.s./ha with min. interval of 10 days;
- Bulbs and onion like crops (use no. 3, 4, 7) – BBCH 10-39, 3 x 96 g a.s./ha with min. interval of 10 days;
- Bulbs and onion like crops - 1 x 76.8 g a.s./ha and BBCH $\geq$ 40;
- Orchards (uses no. 5, 6) – BBCH $\geq$ 40, 1-2 x 144 g a.s./ha with min. interval of 21 days
- Orchards – BBCH $\geq$ 40, 2 x 108 g a.s./ha with min. interval of 10 days

An unacceptable chronic risk to mammals (voles) has been identified for the following uses:

- Leafy vegetables (use no. 1, 2) – BBCH 10-49, 3 x 96 g a.s./ha with min. interval of 7 days;
- Bulbs and onion like crops (use no. 3, 4, 7) – BBCH $\geq$ 40, 3 x 96 g a.s./ha with min. interval of 10 days;
- Orchards (uses no. 5, 6) – BBCH $<$ 40, 1-2 x 144 g a.s./ha with min. interval of 10 days
- Orchards (uses no. 5, 6) – BBCH $>$ 70, 2 x 144 g a.s./ha with min. interval of 10 days
- Orchards – BBCH $<$ 40, 2 x 108 g a.s./ha with min. interval of 10 days

Evaluation of exposing to mammals through the drinking water demonstrated the acceptable risk. The risk to earthworm- and fish-eating animals from secondary poisoning is acceptable.

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

No available data.

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 spinosad and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents, as well as in Appendix 2 of this document (new studies).

Effects on aquatic organisms of SAP09I were not evaluated as part of the EU assessment of spinosad. 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 – spinosad and relevant metabolites

Species	Substance	Exposure System	Results	Reference
<i>Cyprinus carpio</i>	Spinosad	96 h	LC ₅₀ = 4.0 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Cyprinus carpio</i>	NAF-85	96 h	LC ₅₀ > 49 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006) IIIA 10.2.1/0,1MJ16
<i>Oncorhynchus mykiss</i>	Spinosad	80 d (ELS)	NOEC = 0.50 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	Spinosad	48 h	EC ₅₀ > 1.0 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	NAF-85	48 h	EC ₅₀ = 9.1 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006) IIIA 10.2.1/03, MJ06
<i>Daphnia magna</i>	Spinosyn B	48 h	EC ₅₀ = 6.5 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	N-demethylated spinosyn D	48 h	EC ₅₀ = 3.8 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	β-13,14-dihydropseudoglycone of spinosyn A	48 h	EC ₅₀ > 197 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	β-13,14-dihydropseudoglycone of spinosyn D	48 h	EC ₅₀ = 65.8 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	Spinosad	21 d	NOEC (flow-through) = 0.0012 mg a.s./L _{mm} NOEC (semi-static) = 0.0080 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	Spinosyn B	21 d	NOEC (flow-through) = 0.00095 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	N-demethylated spinosyn D	21 d	NOEC (flow-through and semi-static) = 0.001 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	β-13,14-dihydropseudoglycone of spinosyn A	21 d	NOEC (flow-through and semi-static) = 1.250 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Daphnia magna</i>	β-13,14-dihydropseudoglycone of spinosyn D	21 d	NOEC (flow-through and semi-static) = 4.850 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Anabaena flos-aquae</i>	Spinosad	120 h	EC ₅₀ = 6.1 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Selenastrum capricornutum</i>	NAF-85	120 h	EC ₅₀ > 48 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006) IIIA 10.2.1/04, MJ18

Species	Substance	Exposure System	Results	Reference
<i>Navicula pelliculosa</i>	Spinosad	120 h	EC ₅₀ = 0.079 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Navicula pelliculosa</i>	NAF-85	120 h	EC ₅₀ = 0.35 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006) IIIA 10.2.1/05, MJ17
<i>Navicula pelliculosa</i>	Spinosyn B	120 h	EC ₅₀ = 0.077 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Navicula pelliculosa</i>	N-demethylated spinosyn D	120 h	EC ₅₀ = 0.25 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Navicula pelliculosa</i>	β-13,14-dihydropseudoglycone of spinosyn A	72 h	EC ₅₀ = 38.8 mg a.s./L _{nom}	SANCO/1428/2001 – rev. final (2006)
<i>Navicula pelliculosa</i>	β-13,14-dihydropseudoglycone of spinosyn D	96 h	EC ₅₀ = 28 mg a.s./L _{mm}	SANCO/1428/2001 – rev. final (2006)
<i>Chironomus riparius</i>	Spinosad	28 d	NOEC (initial concentration in overlying water) = 0.0016 mg a.s./L _{im}	SANCO/1428/2001 – rev. final (2006)
<i>Chironomus riparius</i>	Spinosyn B	28 d	NOEC (initial concentration in overlying water) = 0.0032 mg a.s./L _{im}	SANCO/1428/2001 – rev. final (2006)
<i>Chironomus riparius</i>	N-demethylated spinosyn D	28 d	NOEC (initial concentration in overlying water) = 0.0024 mg a.s./L _{im}	SANCO/1428/2001 – rev. final (2006)
<i>Chironomus riparius</i>	β-13,14-dihydropseudoglycone of spinosyn A	28 d	NOEC (initial concentration in overlying water) ≥ 1.120 mg a.s./L _{im}	SANCO/1428/2001 – rev. final (2006)
<i>Chironomus riparius</i>	β-13,14-dihydropseudoglycone of spinosyn D	28 d	NOEC (initial concentration in overlying water) ≥ 0.731 mg a.s./L _{im}	SANCO/1428/2001 – rev. final (2006)
Higher-tier studies (micro- or mesocosm studies)				
Not provided.				
Bioconcentration factor (BCF)				
spinosyn A: 114 L/kg spinosyn D: 115 L/kg				

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

For SAP09I only one aquatic study was conducted. The intention of this study was to understand if SAP09I and NAF-85 would have the same ecotoxicological profile. For further details on the comparison of the ecotoxicological profile of both products, please refer to part C of this dossier. A complete data package is available for NAF-85 (as shown in the table above) and since both formulations are ecotoxicologically similar, the Applicant understanding is that the data requirements have been fulfilled and no more testing shall be needed.

Table 9.5-2: Endpoints and effect values relevant for the risk assessment for aquatic organisms – SAP09I

Species	Substance	Exposure System	Results	Reference
<i>Navicula pelliculosa</i>	SAP09I	72 h, s	E _r C ₅₀ = 0.674 mg/L _{mm} (equivalent to 0.31 mg a.s./L) E _y C ₅₀ = 0.515 mg/L _{mm} NOEC = 0.326 mg/L _{mm}	KCP 10.2.1/01, Schuler, L., 2021, S21-04361
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

Not relevant.

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 spinosad for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in cabbage, broccoli, cauliflower (uses no. 1, 2)

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 4000	NOEC 500	EC ₅₀ 1000	NOEC 1.2	EC ₅₀ 79	NOEC 1.6
AF		100	10	100	10	10	10
RAC (µg/L)		40	50	10	0.12	7.9	0.16
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	7.9655	0.20	0.16	0.80	66.38	1.01	49.78
Step 2– multiple application							
N-Europe	0.7500	0.02	0.02	0.08	6.25	0.09	4.69
Step 2– single application							
N-Europe	0.6621	0.02	0.01	0.07	5.52	0.08	4.14
Step 3 – multiple application							
D3/ditch 1st	0.32853	0.01	0.01	0.03	2.74	0.04	2.05
D3/ditch 2nd	0.32712	0.01	0.01	0.03	2.73	0.04	2.04
D4/pond	0.026371	0.00	0.00	0.00	0.22	0.00	0.16
D4/stream	0.26151	0.01	0.01	0.03	2.18	0.03	1.63
R1/pond 1st	0.022135	0.00	0.00	0.00	0.18	0.00	0.14
R1/pond 2nd	0.026797	0.00	0.00	0.00	0.22	0.00	0.17
R1/stream 1st	0.21529	0.01	0.00	0.02	1.79	0.03	1.35
R1 stream 2nd	0.21541	0.01	0.00	0.02	1.80	0.03	1.35

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Step 3 – single application							
D3/ditch 1st	0.44944	0.01	0.01	0.04	3.75	0.06	2.81
D3/ditch 2nd	0.44743	0.01	0.01	0.04	3.73	0.06	2.80
D4/pond	0.01539	0.00	0.00	0.00	0.13	0.00	0.10
D4/stream	0.35218	0.01	0.01	0.04	2.93	0.04	2.20
R1/pond 1st	0.01539	0.00	0.00	0.00	0.13	0.00	0.10
R1/pond 2nd	0.015462	0.00	0.00	0.00	0.13	0.00	0.10
R1/stream 1st	0.29597	0.01	0.01	0.03	2.47	0.04	1.85
R1 stream 2nd	0.29702	0.01	0.01	0.03	2.48	0.04	1.86

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

For the intended uses (use no. 1, 2), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates and sediment dwelling organisms as characterised by a NOEC of 1.2 and 1.6 µg/L, respectively, 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 9.5-4: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for spinosad based on FOCUS Step 4 calculations and toxicity data for invertebrates and sediment dwelling organisms with mitigation of spray drift and run-off for the use of SAP09I in cabbage, broccoli, cauliflower (uses no. 1, 2)

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Chironomus riparius</i>		<i>Daphnia magna</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		NOEC 1.2	NOEC 1.6		NOEC 1.2	NOEC 1.6
AF		10	10		10	10
RAC (µg/L)		0.12	0.16		0.12	0.16
FOCUS Scenario	Multiple application			Single application		
	PEC _{gl-max} (µg/L)			PEC _{gl-max} (µg/L)		
Step 4 – 5m buffer zone						
D3/ditch 1st	0.08672	0.72	0.54	0.12158	1.01	0.76
D3/ditch 2nd	0.08636	0.72	0.54	0.12099	1.01	0.76
D4/stream	0.09355	0.78	0.58	0.1284	1.07	0.80
R1/stream 1st	0.07704	0.64	0.48	0.10794	0.90	0.67
R1 stream 2nd	0.07704	0.64	0.48	0.1083	0.90	0.68
Step 4 – 10m buffer zone						
D3/ditch 1st	0.045381	0.38	0.28	0.064395	0.54	0.40
D3/ditch 2nd	0.045194	0.38	0.28	0.0641	0.53	0.40
D4/stream	0.048982	0.41	0.31	0.06803	0.57	0.43
R1/stream 1st	0.061412	0.51	0.38	0.057166	0.48	0.36
R1 stream 2nd	0.053545	0.45	0.33	0.057365	0.48	0.36

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 9.5-5: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for spinosyn B for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in cabbage, broccoli, cauliflower (uses no. 1, 2)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 6500	NOEC 0.95	EC ₅₀ 77	NOEC 3.2
AF		100	10	10	10
RAC (µg/L)		65	0.095	7.7	0.32
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	19.9061	0.31	209.54	2.59	62.21
Step 2 – multiple application					
N-Europe	2.3496	0.04	24.73	0.31	7.34
Step 2 – single application					
N-Europe	0.9396	0.01	9.89	0.12	2.94
Step 3– multiple application					
D3/ditch 1st	< 1e-6	0.00	0.00	0.00	0.00
D3/ditch 2nd	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.001462	0.00	0.02	0.00	0.00
D4/stream	0.01146	0.00	0.12	0.00	0.04
R1/pond 1st	0.0571	0.00	0.60	0.01	0.18
R1/pond 2nd	0.04678	0.00	0.49	0.01	0.15
R1/stream 1st	0.1049	0.00	1.10	0.01	0.33
R1 stream 2nd	0.0695	0.00	0.73	0.01	0.22

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Step 3 – single application					
D3/ditch 1st	< 1e-6	0.00	0.00	0.00	0.00
D3/ditch 2nd	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000324	0.00	0.00	0.00	0.00
D4/stream	0.002724	0.00	0.03	0.00	0.01
R1/pond 1st	0.01603	0.00	0.17	0.00	0.05
R1/pond 2nd	0.01743	0.00	0.18	0.00	0.05
R1/stream 1st	0.03261	0.00	0.34	0.00	0.10
R1 stream 2nd	0.0283	0.00	0.30	0.00	0.09

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

For the intended uses (use no. 1, 2), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates as characterised by a NOEC of 0.95 µg/L, in connection with an assessment factor of 10 in one 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 9.5-6: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for spinosyn B based on FOCUS Step 4 calculations and toxicity data for invertebrates organisms with mitigation of spray drift and run-off for the use of SAP09I in cabbage, broccoli, cauliflower (uses no. 1, 2)

Group		Inverteb. prolonged		Inverteb. prolonged
Test species		<i>Daphnia magna</i>		<i>Daphnia magna</i>
Endpoint (µg/L)		NOEC		NOEC
AF		0.95		0.95
RAC (µg/L)		10		10
		0.095		0.095
FOCUS Scenario	Multiple application		Single application	
	PEC _{gl-max} (µg/L)		PEC _{gl-max} (µg/L)	
Step 4 – 5m buffer zone				
R1/stream 1st	0.1049	1.10	0.03261	0.34
Step 4 – 10m buffer zone				
R1/stream 1st	0.1049	1.10	0.03261	0.34
Step 4 – 10m buffer zone with 5m vegetated buffer zone (VFSmod)				
R1/stream 1st	0.01884	0.20	0.007257	0.08

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 9.5-7: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for N-demethylated spinosyn D for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in cabbage, broccoli, cauliflower (uses no. 1, 2)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 3800	NOEC 1	EC ₅₀ 250	NOEC 2.4
AF		100	10	10	10
RAC (µg/L)		38	0.1	25	0.24
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	3.5141	0.09	35.14	0.14	14.64
Step 2 – multiple application					
N-Europe	0.4399	0.01	4.40	0.02	1.83
Step 2 – single application					
N-Europe	0.1699	0.00	1.70	0.01	0.71
Step 3 – multiple application					
D3/ditch 1st	< 1e-6	0.00	0.00	0.00	0.00
D3/ditch 2nd	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000448	0.00	0.00	0.00	0.00
D4/stream	0.003181	0.00	0.03	0.00	0.01
R1/pond 1st	0.01101	0.00	0.11	0.00	0.05
R1/pond 2nd	0.009807	0.00	0.10	0.00	0.04
R1/stream 1st	0.0185	0.00	0.19	0.00	0.08
R1 stream 2nd	0.01357	0.00	0.14	0.00	0.06

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Step 3 – single application					
D3/ditch 1st	< 1e-6	0.00	0.00	0.00	0.00
D3/ditch 2nd	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000103	0.00	0.00	0.00	0.00
D4/stream	0.000786	0.00	0.01	0.00	0.00
R1/pond 1st	0.003312	0.00	0.03	0.00	0.01
R1/pond 2nd	0.003307	0.00	0.03	0.00	0.01
R1/stream 1st	0.005791	0.00	0.06	0.00	0.02
R1 stream 2nd	0.005109	0.00	0.05	0.00	0.02

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 9.5-8: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for spinosad for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in onion, leek, garlic (uses no. 3, 4, 7)

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 4000	NOEC 500	EC ₅₀ 1000	NOEC 1.2	EC ₅₀ 79	NOEC 1.6
AF		100	10	100	10	10	10
RAC (µg/L)		40	50	10	0.12	7.9	0.16
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	9.5586	0.24	0.19	0.96	79.66	1.21	59.74
Step 2– multiple application							
N-Europe	0.9593	0.02	0.02	0.10	7.99	0.12	6.00
Step 2– single application							
N-Europe	0.7946	0.02	0.02	0.08	6.62	0.10	4.97
Step 3 – multiple application							
D3/ditch	0.39438	0.01	0.01	0.04	3.29	0.05	2.46
D4/pond	0.029285	0.00	0.00	0.00	0.24	0.00	0.18
D4/stream	0.30009	0.01	0.01	0.03	2.50	0.04	1.88
R1/pond	0.027649	0.00	0.00	0.00	0.23	0.00	0.17
R1/stream	0.25833	0.01	0.01	0.03	2.15	0.03	1.61
Step 3 – single application							
D3/ditch	0.53962	0.01	0.01	0.05	4.50	0.07	3.37
D4/pond	0.018481	0.00	0.00	0.00	0.15	0.00	0.12

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
D4/stream	0.41391	0.01	0.01	0.04	3.45	0.05	2.59
R1/pond	0.018482	0.00	0.00	0.00	0.15	0.00	0.12
R1/stream	0.35501	0.01	0.01	0.04	2.96	0.04	2.22

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

For the intended uses (~~use no. 1, 2~~) (uses no. 3, 4, 7), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates and sediment dwelling organisms as characterised by a NOEC of 1.2 and 1.6 µg/L, respectively, 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 9.5-9: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for spinosad based on FOCUS Step 4 calculations and toxicity data for invertebrates and sediment dwelling organisms with mitigation of spray drift and run-off for the use of SAP09I in onion, leek, garlic (uses no. 3, 4, 7)

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Chironomus riparius</i>		<i>Daphnia magna</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		NOEC 1.2	NOEC 1.6		NOEC 1.2	NOEC 1.6
AF		10	10		10	10
RAC (µg/L)		0.12	0.16		0.12	0.16
FOCUS Scenario	Multiple application			Single application		
	PEC _{gl-max} (µg/L)			PEC _{gl-max} (µg/L)		
Step 4 – 5m buffer zone						
D3/ditch	0.1041	0.87	0.65	0.14592	1.22	0.91
D4/stream	0.10735	0.89	0.67	0.15097	1.26	0.94
R1/stream	0.09245	0.77	0.58	0.12952	1.08	0.81
Step 4 – 10m buffer zone						
D3/ditch	0.054506	0.45	0.34	0.07731	0.64	0.48
D4/stream	0.056202	0.47	0.35	0.07994	0.67	0.50
R1/stream	0.08514	0.71	0.53	0.06856	0.57	0.43

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 9.5-10: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Spinosyn B for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in onion, leek, garlic (uses no. 3, 4, 7)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 6500	NOEC 0.95	EC ₅₀ 77	NOEC 3.2
AF		100	10	10	10
RAC (µg/L)		65	0.095	7.7	0.32
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	23.8874	0.37	251.45	3.10	74.65
Step 2 – multiple application					
N-Europe	3.1517	0.05	33.18	0.41	9.85
Step 2 – single application					
N-Europe	1.3245	0.02	13.94	0.17	4.14
Step 3– multiple application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.001982	0.00	0.02	0.00	0.01
D4/stream	0.01508	0.00	0.16	0.00	0.05
R1/pond	0.04163	0.00	0.44	0.01	0.13
R1/stream	0.1454	0.00	1.53	0.02	0.45
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000451	0.00	0.00	0.00	0.00
D4/stream	0.003703	0.00	0.04	0.00	0.01

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
R1/pond	0.01097	0.00	0.12	0.00	0.03
R1/stream	0.04267	0.00	0.45	0.01	0.13

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

For the intended uses (~~use no. 1, 2~~) (uses no. 3, 4, 7), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates as characterised by a NOEC of 0.95 µg/L, in connection with an assessment factor of 10 in one 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 9.5-11: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for Spinosyn B based on FOCUS Step 4 calculations and toxicity data for invertebrates organisms with mitigation of spray drift and run-off for the use of SAP09I in onion, leek, garlic (uses no. 3, 4, 7)

Group		Inverteb. prolonged		Inverteb. prolonged
Test species		<i>Daphnia magna</i>		<i>Daphnia magna</i>
Endpoint		NOEC		NOEC
(µg/L)		0.95		0.95
AF		10		10
RAC (µg/L)		0.095		0.095
FOCUS Scenario	Multiple application		Single application	
	PEC _{gl-max} (µg/L)		PEC _{gl-max} (µg/L)	
Step 4 – 5m buffer zone				
R1/stream	0.1454	1.53	0.04267	0.45
Step 4 – 10m buffer zone				
R1/stream	0.1454	1.53	0.04267	0.45
Step 4 – 10m buffer zone with 5m vegetated buffer zone (VFSmod)				
R1/stream	0.02035	0.21	0.005346	0.06

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 9.5-12: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for N-demethylated spinosyn D for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I in onion, leek, garlic (uses no. 3, 4, 7)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 3800	NOEC 1	EC ₅₀ 250	NOEC 2.4
AF		100	10	10	10
RAC (µg/L)		38	0.1	25	0.24
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	4.2170	0.11	42.17	0.17	17.57
Step 2 – multiple application					
N-Europe	0.5974	0.02	5.97	0.02	2.49
Step 2 – single application					
N-Europe	0.2396	0.01	2.40	0.01	1.00
Step 3 – multiple application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000617	0.00	0.01	0.00	0.00
D4/stream	0.004176	0.00	0.04	0.00	0.02
R1/pond	0.009310	0.00	0.09	0.00	0.04
R1/stream	0.02564	0.00	0.26	0.00	0.11
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000140	0.00	0.00	0.00	0.00
D4/stream	0.001035	0.00	0.01	0.00	0.00

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
R1/pond	0.002604	0.00	0.03	0.00	0.01
R1/stream	0.007571	0.00	0.08	0.00	0.03

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 9.5-13: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for spinosad for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – early application (BBCH <53)

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 4000	NOEC 500	EC ₅₀ 1000	NOEC 1.2	EC ₅₀ 79	NOEC 1.6
AF		100	10	100	10	10	10
RAC (µg/L)		40	50	10	0.12	7.9	0.16
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	7.2002	0.18	0.14	0.72	60.00	0.91	45.00
Step 2– single application							
N-Europe	5.6059	0.14	0.11	0.56	46.72	0.71	35.04
Step 3 – single application							
D3/ditch	4.4195	0.11	0.09	0.44	36.83	0.56	27.62
D4/pond	0.26752	0.01	0.01	0.03	2.23	0.03	1.67
D4/stream	4.5065	0.11	0.09	0.45	37.55	0.57	28.17
R1/pond	0.2674	0.01	0.01	0.03	2.23	0.03	1.67
R1/stream	3.5727	0.09	0.07	0.36	29.77	0.45	22.33

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

For the intended uses (~~use no. 1, 2~~) (uses no. 5, 6, BBCH<53), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates and sediment dwelling organisms as characterised by a NOEC of 1.2 and 1.6 µg/L, respectively, 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 9.5-14: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for spinosad based on FOCUS Step 4 calculations and toxicity data for invertebrates and sediment dwelling organisms with mitigation of spray drift and run-off for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – early application (BBCH <53)

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Chironomus riparius</i>		<i>Daphnia magna</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		NOEC 1.2	NOEC 1.6		NOEC 1.2	NOEC 1.6
AF		10	10		10	10
RAC (µg/L)		0.12	0.16		0.12	0.16
FOCUS Scenario	Single application			Single application		
	PEC _{gl-max} (µg/L)			PEC _{gl-max} (µg/L)		
-	Step 4 – 30m buffer zone			Step 4 – 40m buffer zone		
D3/ditch	0.18554	1.55	1.16	0.09329	0.78	0.58
D4/pond	0.025726	0.21	0.16	0.014965	0.12	0.09
D4/stream	0.20683	1.72	1.29	0.10404	0.87	0.65
R1/pond	0.025726	0.21	0.16	0.014953	0.12	0.09
R1/stream	0.16402	1.37	1.03	0.08248	0.69	0.52
-	Step 4 – 20m buffer zone + 75% DRN			Step 4 – 20m buffer zone + 95% DRN		
D3/ditch	0.12122	1.01	0.76	0.02415	0.20	0.15
D4/pond	0.013272	0.11	0.08	0.002641	0.02	0.02
D4/stream	0.13511	1.13	0.84	0.026935	0.22	0.17
R1/pond	0.013262	0.11	0.08	0.00264	0.02	0.02

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
R1/stream	0.10712	0.89	0.67	0.021346	0.18	0.13

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 9.5-15: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Spinosyn B for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – early application (BBCH <53)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 6500	NOEC 0.95	EC ₅₀ 77	NOEC 3.2
AF		100	10	10	10
RAC (µg/L)		65	0.095	7.7	0.32
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	9.5406	0.15	100.43	1.24	29.81
Step 2 – single application					
N-Europe	4.6739	0.07	49.20	0.61	14.61
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000014	0.00	0.00	0.00	0.00
D4/stream	0.000201	0.00	0.00	0.00	0.00
R1/pond	0.000275	0.00	0.00	0.00	0.00
R1/stream	0.01692	0.00	0.18	0.00	0.05

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

For the intended uses (use no. 1, 2) (uses no. 5, 6 BBCH <53), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates as characterised by a NOEC of 0.95 µg/L, in connection with an assessment factor of 10 in one 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 9.5-16: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for N-demethylated spinosyn D for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – early application (BBCH <53)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 3800	NOEC 1	EC ₅₀ 250	NOEC 2.4
AF		100	10	10	10
RAC (µg/L)		38	0.1	25	0.24
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	1.6842	0.04	16.84	0.07	7.02
Step 2 – single application					
N-Europe	0.8251	0.02	8.25	0.03	3.44
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000004	0.00	0.00	0.00	0.00
D4/stream	0.000056	0.00	0.00	0.00	0.00
R1/pond	0.000066	0.00	0.00	0.00	0.00
R1/stream	0.003439	0.00	0.03	0.00	0.01

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 9.5-17: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for spinosad for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – late application (BBCH >70)

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 4000	NOEC 500	EC ₅₀ 1000	NOEC 1.2	EC ₅₀ 79	NOEC 1.6
AF		100	10	100	10	10	10
RAC (µg/L)		40	50	10	0.12	7.9	0.16
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	9.2272	0.23	0.18	0.92	76.89	1.17	57.67
Step 2– multiple application							
N-Europe	2.5983	0.06	0.05	0.26	21.65	0.33	16.24
Step 2– single application							
N-Europe	3.0192	0.08	0.06	0.30	25.16	0.38	18.87
Step 3 – multiple application							
D3/ditch	1.6628	0.04	0.03	0.17	13.86	0.21	10.39
D4/pond	0.12892	0.00	0.00	0.01	1.07	0.02	0.81
D4/stream	1.6393	0.04	0.03	0.16	13.66	0.21	10.25
R1/pond	0.12325	0.00	0.00	0.01	1.03	0.02	0.77
R1/stream	1.2583	0.03	0.03	0.13	10.49	0.16	7.86
Step 3 – single application							
D3/ditch	2.0898	0.05	0.04	0.21	17.42	0.26	13.06
D4/pond	0.0931	0.00	0.00	0.01	0.78	0.01	0.58

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
D4/stream	2.0474	0.05	0.04	0.20	17.06	0.26	12.80
R1/pond	0.09309	0.00	0.00	0.01	0.78	0.01	0.58
R1/stream	1.5712	0.04	0.03	0.16	13.09	0.20	9.82

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

For the intended uses (~~use no. 1, 2~~) (uses no. 5, 6 BBCH>70), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates and sediment dwelling organisms as characterised by a NOEC of 1.2 and 1.6 µg/L, respectively, 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 9.5-18: Aquatic organisms: PEC calculation and acceptability of risk (PEC/RAC < 1) for spinosad based on FOCUS Step 4 calculations and toxicity data for invertebrates and sediment dwelling organisms with mitigation of spray drift and run-off for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – late application (BBCH >70)

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Chironomus riparius</i>		<i>Daphnia magna</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		NOEC	NOEC		NOEC	NOEC
AF		1.2	1.6		1.2	1.6
RAC (µg/L)		10	10		10	10
		0.12	0.16		0.12	0.16
FOCUS Scenario	Multiple application			Single application		
	PEC _{gl-max} (µg/L)			PEC _{gl-max} (µg/L)		
Step 4 – 20m buffer zone						
D3/ditch	0.15697	1.31	0.98	0.19378	1.61	1.21
D4/pond	0.033303	0.28	0.21	0.026794	0.22	0.17
D4/stream	0.1772	1.48	1.11	0.21954	1.83	1.37
R1/pond	0.031882	0.27	0.20	0.02683	0.22	0.17

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
R1/stream	0.13604	1.13	0.85	0.16848	1.40	1.05
Step 4 – 30m buffer zone						
D3/ditch	0.07231	0.60	0.45	0.09615	0.80	0.60
D4/pond	0.018778	0.16	0.12	0.016058	0.13	0.10
D4/stream	0.08162	0.68	0.51	0.10895	0.91	0.68
R1/pond	0.018017	0.15	0.11	0.016103	0.13	0.10
R1/stream	0.062641	0.52	0.39	0.08359	0.70	0.52
Step 4 – 10m buffer zone + 75% DRN						
D3/ditch	0.13816	1.15	0.86	0.15697	1.31	0.98
D4/pond	0.02006	0.17	0.13	0.014696	0.12	0.09
D4/stream	0.15592	1.3	0.97	0.17779	1.48	1.11
R1/pond	0.01925	0.16	0.12	0.014742	0.12	0.09
R1/stream	0.11972	1.00	0.75	0.13651	1.14	0.85
Step 4 – 10m buffer zone + 90% DRN						
D3/ditch	0.055164	0.46	0.34	0.062665	0.52	0.39
D4/pond	0.007995	0.07	0.05	0.005866	0.05	0.04
D4/stream	0.06226	0.52	0.39	0.07098	0.59	0.44
R1/pond	0.007728	0.06	0.05	0.005918	0.05	0.04
R1/stream	0.047789	0.40	0.30	0.054486	0.45	0.34
Step 4 – 20m buffer zone + 50% DRN						
D3/ditch	0.02319	0.19	0.14	0.09676	0.52	0.39
D4/pond	0.006128	0.05	0.04	0.013378	0.05	0.04
D4/stream	0.02619	0.22	0.16	0.10963	0.59	0.44
R1/pond	0.005937	0.05	0.04	0.013424	0.05	0.04

Group		Inverteb. prolonged	Sed. dwell. prolonged		Inverteb. prolonged	Sed. dwell. prolonged
R1/stream	0.020051	0.17	0.13	0.08415	0.45	0.34

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 9.5-19: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Spinosyn B for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – late application (BBCH >70)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 6500	NOEC 0.95	EC ₅₀ 77	NOEC 3.2
AF		100	10	10	10
RAC (µg/L)		65	0.095	7.7	0.32
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	14.7679	0.23	155.45	1.92	46.15
Step 2 – multiple application					
N-Europe	2.3418	0.04	24.65	0.30	7.32
Step 2 – single application					
N-Europe	2.5173	0.04	26.50	0.33	7.87
Step 3– multiple application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000119	0.00	0.00	0.00	0.00
D4/stream	0.001539	0.00	0.02	0.00	0.00
R1/pond	0.002346	0.00	0.02	0.00	0.01
R1/stream	0.04779	0.00	0.50	0.01	0.15
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000040	0.00	0.00	0.00	0.00
D4/stream	0.000522	0.00	0.01	0.00	0.00

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
R1/pond	0.001128	0.00	0.01	0.00	0.00
R1/stream	0.02313	0.00	0.24	0.00	0.07

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

For the intended uses (~~use no. 1, 2~~) (uses no. 5, 6 BBCH>70), calculated PEC/RAC ratios did not indicate an acceptable risk for the most sensitive group of aquatic organisms (risk for invertebrates as characterised by a NOEC of 0.95 µg/L, in connection with an assessment factor of 10 in one 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 9.5-20: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for N-demethylated spinosyn D for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of SAP09I to apple, pear, wild apple, quince (uses no. 5, 6) – late application (BBCH >70)

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
Test species		<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		EC ₅₀ 3800	NOEC 1	EC ₅₀ 250	NOEC 2.4
AF		100	10	10	10
RAC (µg/L)		38	0.1	25	0.24
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	2.6070	0.07	26.07	0.10	10.86
Step 2 – multiple application					
N-Europe	0.4121	0.01	4.12	0.02	1.72
Step 2 – single application					
N-Europe	0.4444	0.01	4.44	0.02	1.85
Step 3 – multiple application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000030	0.00	0.00	0.00	0.00
D4/stream	0.000369	0.00	0.00	0.00	0.00
R1/pond	0.000469	0.00	0.00	0.00	0.00
R1/stream	0.008646	0.00	0.09	0.00	0.04
Step 3 – single application					
D3/ditch	< 1e-6	0.00	0.00	0.00	0.00
D4/pond	0.000011	0.00	0.00	0.00	0.00
D4/stream	0.000135	0.00	0.00	0.00	0.00

Group		Inverteb. acute	Inverteb. prolonged	Algae/Diatom	Sed. dwell. prolonged
R1/pond	0.000226	0.00	0.00	0.00	0.00
R1/stream	0.004209	0.00	0.04	0.00	0.02

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

Spinosad metabolites β -13,14-dihydropseudoglycone of spinosyn A and β -13,14-dihydropseudoglycone of spinosyn D are clearly less sensitive than the active substance and the remaining metabolites - spinosyn B and N-demethylated spinosyn D (please see table below). Therefore, the risk assessment performed above as well as the mitigation measures provided below are considered to cover the risk of these two metabolites towards aquatic organisms and no further calculations will be presented.

Species	Reported endpoints (lowest bounded endpoint signaled in bold for each organism)					
	Spinosad	NAF-85	Spinosyn B	N-demethylated spinosyn D	β -13,14-dihydropseudoglycone of spinosyn A	β -13,14-dihydropseudoglycone of spinosyn D
<i>Daphnia magna</i> (acute)	EC ₅₀ > 1.0 mg a.s./L _{nom}	EC ₅₀ = 9.1 mg a.s./L _{nom}	EC ₅₀ = 6.5 mg a.s./L _{mm}	EC₅₀ = 3.8 mg a.s./L_{mm}	EC ₅₀ > 197 mg a.s./L _{mm}	EC ₅₀ = 65.8 mg a.s./L _{mm}
<i>Daphnia magna</i> (chronic)	NOEC = 0.0012 mg a.s./L _{mm}	-	NOEC = 0.00095 mg a.s./L_{mm}	NOEC = 0.001 mg a.s./L _{mm}	NOEC = 1.250 mg a.s./L _{nom}	NOEC = 4.850 mg a.s./L _{mm}
<i>Navicula pelliculosa</i>	EC ₅₀ = 0.079 mg a.s./L _{mm}	EC ₅₀ = 0.35 mg a.s./L _{mm}	EC₅₀ = 0.077 mg a.s./L_{mm}	EC ₅₀ = 0.25 mg a.s./L _{mm}	EC ₅₀ = 38.8 mg a.s./L _{nom}	EC ₅₀ = 28 mg a.s./L _{mm}
<i>Chironomus riparius</i>	NOEC = 0.0016 mg a.s./L_{im}	-	NOEC = 0.0032 mg a.s./L _{im}	NOEC = 0.0024 mg a.s./L _{im}	NOEC ≥ 1.120 mg a.s./L _{im}	NOEC ≥ 0.731 mg a.s./L _{im}

9.5.3 Overall conclusions

The risk assessment to aquatic organisms was conducted according to EFSA Journal 2013; 11(7):3290. An acceptable risk is obtained for SPA09I application according to the intended uses when the following mitigation measures are implemented:

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 10m with vegetated buffer of 5m to surface water bodies in applications to leafy vegetables (uses no. 1, 2) and bulb and onion like vegetables (uses no. 3, 4, 7).

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 40m or 20 m with 95% DRN to surface water bodies in early applications to orchards (uses no. 5, 6).

SPe3: To protect aquatic organisms, respect an unsprayed buffer zone of 30m or 10 m with 90% DRN or 20 m with 50% DRN to surface water bodies in late applications to orchards (uses no. 5, 6).

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_{sw} values and the results of laboratory toxicity testing. The PEC_{sw} FOCUS Steps 1-4 for spinosad (A+D), spinosyn B and Steps 1-3 for N-demethylated spinosyn D. Additional evaluation for formulation and metabolites: β -13,14-dihydropseudoglycone of spinosyn A and β -13,14-dihydropseudoglycone was not required.

SAP09I applications close to surface water pose acceptable risk to aquatic organisms with appropriate risk mitigation measures provided above.

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 spinosad. Full details of these studies are provided in the respective EU DAR and related documents as well as in Appendix 2 of this document (new studies).

Effects on bees of SAP09I were not evaluated as part of the EU assessment of spinosad. 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 9.6-1: Endpoints and effect values relevant for the risk assessment for bees

Species	Substance	Exposure System	Results	Reference
<i>Apis mellifera</i>	Spinosad	Oral	LD ₅₀ = 0.057 µg a.s./bee	SANCO/1428/2001 – rev. final (2006)
<i>Apis mellifera</i>	NAF-85	Oral	LD₅₀ = 0.049 µg a.s./bee	SANCO/1428/2001 – rev. final (2006) IIIA 10.4.1/01, MJ14
<i>Apis mellifera</i>	Spinosad	Contact	LD₅₀ = 0.036 µg a.s./bee	SANCO/1428/2001 – rev. final (2006)
<i>Apis mellifera</i>	NAF-85	Contact	LD ₅₀ = 0.050 µg a.s./bee	SANCO/1428/2001 – rev. final (2006) IIIA 10.4.1/01, MJ14
<i>Bombus terrestris</i>	NAF-85	Oral	LD₅₀ = 0.37 µg a.s./bee	Section B9 of DAR Spinosad
<i>Bombus terrestris</i>	NAF-85	Contact	LD₅₀ = 17.2 µg a.s./bee	Section B9 of DAR Spinosad
<i>Apis mellifera</i>	SAP09I	Chronic, oral	LDD₅₀ > 0.01492 µg a.s./bee	KCP 10.3.1.2/01, Aguilar-Alberola, 2021, S21-04335
<i>Apis mellifera</i>	SAP09I	Chronic, larvae	NOED = 0.0062 µg a.s./bee	KCP 10.3.1.3/01, Aguilar-Alberola, 2021, S21-04336
Higher-tier studies (tunnel test, field studies)				
Cage test in <i>Apis mellifera</i> (IIIA 10.4.3, MJ41): Effects on foraging behaviour were observed, probably caused by repellency, as well as some effects on brood survival in two out of three hives in the higher application rate. The information on the dead broods was however only qualitative. The following results are used for risk assessment: • Early morning applications of NAF-85 at rates of 540 and 144 g as/ha (before bees were active) on flowering <i>Phacelia tanacetifolia</i> caused no mortality in bees. • NAF-85 at 540 g as/ha has an effect on foraging behaviour, indications for effects on brood survival are present. Semi-field study with <i>Bombus terrestris</i> (IIIA 10.4.3/04, MJ44): Control corrected mortality was -1% for the spinosad treatment and 14 and 4% for the drench and the foliar application of the toxic reference, respectively. Reductions were not significant. Foraging activity in the NAF-85 treatment was reduced with 65% after 2 days, 19% after 4 days, and 13% after 6 days. The differences were not statistically significant. Foliar application of NAF-85 caused a reduction in brood development (new adults plus larvae and pupae) of 27%. This reduction was not statistically significant. Ref: Section B9 DAR Spinosad				

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies considered in the first registration in 2003 may be used to evaluate SAP09I. These data were included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, made available to the assessor for the purposes of this evaluation. The acute oral and contact toxicity of NAF-85 to bumblebees was not included in provided documentation and, therefore, cannot be used in current evaluation.

9.6.1.1 Justification for new endpoints

No acute studies were performed with the formulation. The studies performed with the active substance and the reference product are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Additionally, the risk assessment was performed with the lowest endpoints available (either coming from the active substance data or the reference product). Therefore, no further studies are considered necessary.

9.6.2 Risk assessment

9.6.2.1 Hazard quotients for bees

Risk assessment acc. to SANCO/10329/2002 rev.2 (final), October 17, 2002

The evaluation of the acute 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) as well as “Guidance on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees)” (EFSA Journal 2013;11(7):3295).

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment orchards (use no. 5) cover the risk for bees and bumble bees from all intended uses (see 9.1.2).

Table 9.6-2: Risk assessment of the risk for bees due to the use of SAP09I in orchards (use no. 5) – risk envelope approach

Intended use		orchards (use no. 5) – risk envelope approach	
Active substance/Product		spinosad/SAP09I	
Application rate (g/ha)		1 x 144	
Test design	LD₅₀ (lab.) (µg a.s./bee)	Single application rate (g a.s./ha)	Q_{HO}, Q_{HC} criterion: Q_H ≤ 50
Oral toxicity	0.049	144	2939
Contact toxicity	0.036		4000
Test design	LD₅₀ (lab.) (µg a.s./bumble-bee)	Single application rate (g a.s./ha)	Q_{HO}, Q_{HC} criterion: Q_H ≤ 50
Oral toxicity	0.37	144	389.2
Contact toxicity	17.2		8.37

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

Since an unacceptable risk is found for bees and bumble bees exposure to SAP09I, a restriction must be imposed in the application of SAP09I according to the application patterns:

SPe8: Dangerous to bees. To protect bees and other pollinating insects do not apply on flowering crops. Do not use where bees are actively foraging. Do not apply when flowering weeds are present. Remove weeds before flowering.

Risk assessment acc. to EFSA Journal 2013;11(7):3295

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the screening assessment for orchards (use no. 5) covers the risk for bees and bumble bees from all intended uses (see 9.1.2).

Table 9.6-3: Screening risk assessment of the risk for bees due to the use of SAP09I in orchards (use no. 5) – risk envelope approach

Intended use	orchards (use no. 5) – risk envelope approach				
Active substance/Product	spinosad/SAP09I				
Application rate (g/ha)	1 x 144				
Test design	LD ₅₀ (µg a.s./bee) LDD ₅₀ (µg a.s./bee/day) NOED (µg a.s./larva)	Single application rate (g a.s./ha)	SV	HQ/ ETR	Trigger
Contact exposure					
Bees - acute contact toxicity	0.036	144	1	4000	42
Bumble bees – acute contact toxicity	17.2		1	8.4	7
Oral exposure					
Bees - acute oral toxicity	0.049	144	10.6	31.15	0.2
Bumble bees – acute oral toxicity	0.37		13.3	5.18	0.036
Bees - chronic adult oral toxicity	0.01492		10.6	102.306	0.03
Bees - larval development oral toxicity	0.0062		6.1	141.68	0.2

HQ (hazard quotients) and ETR (exposure toxicity ratio) for oral and contact exposure. HQ/ETR values shown in bold breach the relevant trigger

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the Tier-1 assessment for the use no. 1 (cabbage), use no. 3 (onion) and use no. 5, 6 (pear, wild apple, quince) cover the risk for bees and bumble bees from all intended uses (see 9.1.2).

Table 9.6-4: Tier-1 assessment of the risk for bees and bumble bees due to the use of SAP09I in cabbage (use no. 1) and onion (use no. 3) – risk envelope approach

Category	Scenario	BBCH	cabbage (use no. 1) crop category: leafy vegetables 96 g a.s./ha		onion (use no. 3) crop category: bulb vegetables 96 g a.s./ha		Trigger bees/ bumble bees
			HQ bees	HQ bumble bees	HQ bees	HQ bumble bees	
Acute dermal toxicity, adult (DW spray)	treated crop	<50	0.0	0.0	-	-	42/7
	treated crop	<40	-	-	0.0	0.0	42/7
	treated crop	≥40	-	-	0.0	0.0	42/7
	weeds	<50	2666.7	5.6	-	-	42/7
	weeds	<40	-	-	2666.7	5.6	42/7
	weeds	≥40	-	-	1600.0	3.3	42/7
	field margin	<50	74.7	0.2	-	-	42/7
	field margin	<40	-	-	74.7	0.2	42/7
	field margin	≥40	-	-	74.7	0.2	42/7

Category	Scenario	BBCH	cabbage (use no. 1) crop category: leafy vegetables 96 g a.s./ha		onion (use no. 3) crop category: bulb vegetables 96 g a.s./ha		Trigger bees/ bumble bees
			HQ bees	HQ bumble bees	HQ bees	HQ bumble bees	
Acute oral toxicity, adult (DW spray)	treated crop	10 - 39	-	-	14.89	2.9059	0.2/0.036
	treated crop	10 - 49	14.89	2.9059	-	-	0.2/0.036
	treated crop	40 - 69	-	-	14.89	2.9059	0.2/0.036
	weeds	10 - 39	-	-	7.25	1.6865	0.2/0.036
	weeds	10 - 49	7.25	1.6865	-	-	0.2/0.036
	weeds	40 - 69	-	-	4.35	1.0119	0.2/0.036
	field margin	10 - 39	-	-	0.07	0.0155	0.2/0.036
	field margin	10 - 49	0.07	0.0155	-	-	0.2/0.036
	field margin	40 - 69	-	-	0.07	0.0155	0.2/0.036
	adjacent crop	10 - 39	-	-	0.05	0.0096	0.2/0.036
	adjacent crop	10 - 49	0.05	0.0096	-	-	0.2/0.036
	adjacent crop	40 - 69	-	-	0.05	0.0096	0.2/0.036
	next crop	10 - 39	-	-	1.37	0.2335	0.2/0.036
	next crop	10 - 49	1.37	0.2335	-	-	0.2/0.036
	next crop	40 - 69	-	-	1.37	0.2335	0.2/0.036
Chronic oral toxicity, adult (DW spray)	treated crop	10 - 39	-	-	26.870	-	0.03/NR
	treated crop	10 - 49	26.870	-	-	-	0.03/NR
	treated crop	40 - 69	-	-	26.870	-	0.03/NR
	weeds	10 - 39	-	-	13.435	-	0.03/NR
	weeds	10 - 49	13.435	-	-	-	0.03/NR
	weeds	40 - 69	-	-	8.061	-	0.03/NR
	field margin	10 - 39	-	-	0.124	-	0.03/NR
	field margin	10 - 49	0.124	-	-	-	0.03/NR
	field margin	40 - 69	-	-	0.124	-	0.03/NR
	adjacent crop	10 - 39	-	-	0.089	-	0.03/NR
	adjacent crop	10 - 49	0.089	-	-	-	0.03/NR
	adjacent crop	40 - 69	-	-	0.089	-	0.03/NR
	next crop	10 - 39	-	-	2.502	-	0.03/NR
	next crop	10 - 49	2.502	-	-	-	0.03/NR
	next crop	40 - 69	-	-	2.502	-	0.03/NR
Larvae toxicity (DW spray)	treated crop	10 - 39	-	-	57.91	-	0.2/NR
	treated crop	10 - 49	57.91	-	-	-	0.2/NR
	treated crop	40 - 69	-	-	57.91	-	0.2/NR
	weeds	10 - 39	-	-	28.95	-	0.2/NR
	weeds	10 - 49	28.95	-	-	-	0.2/NR
	weeds	40 - 69	-	-	17.37	-	0.2/NR
	field margin	10 - 39	-	-	0.27	-	0.2/NR
	field margin	10 - 49	0.27	-	-	-	0.2/NR
	field margin	40 - 69	-	-	0.27	-	0.2/NR
	adjacent crop	10 - 39	-	-	0.19	-	0.2/NR
	adjacent crop	10 - 49	0.19	-	-	-	0.2/NR
	adjacent crop	40 - 69	-	-	0.19	-	0.2/NR
	next crop	10 - 39	-	-	5.26	-	0.2/NR
	next crop	10 - 49	5.26	-	-	-	0.2/NR
	next crop	40 - 69	-	-	5.26	-	0.2/NR

ETR: Exposure toxicity ratio; **Bold**: ETR values that exceed the trigger value (need a refinement)

Table 9.6-5: Tier-1 assessment of the risk for bees and bumble bees due to the use of SAP09I in orchards (use no. 5, 6) – risk envelope approach

Category	Scenario	BBCH	pear, wild apple, quince (use no. 5, 6) crop category: orchards 1 144 g a.s./ha*		Trigger bees/ bumble bees
			HQ bees	HQ bumble bees	
Acute dermal toxicity, adult (SUW spray)	treated crop	10 - 19	0.0	0.0	85/14
	treated crop	20 - 39	0.0	0.0	85/14
	treated crop	≥40	0.0	0.0	85/14
	weeds	10 - 19	3200	6.7	42/7
	weeds	20 - 39	2400	5.0	42/7
	weeds	≥40	1200	2.5	42/7
	field margin	10 - 19	628	1.3	42/7
	field margin	20 - 39	628	1.3	42/7
	field margin	≥40	628	1.3	42/7
Acute oral toxicity, adult (SUW spray)	treated crop	10 - 19	31.15	5.1762	0.2/0.036
	treated crop	20 - 39	31.15	5.1762	0.2/0.036
	treated crop	40 - 69	31.15	5.1762	0.2/0.036
	treated crop	≥70	0.0	0.0000	0.2/0.036
	weeds	10 - 19	8.70	2.0238	0.2/0.036
	weeds	20 - 39	6.52	1.5178	0.2/0.036
	weeds	40 - 69	3.26	0.7589	0.2/0.036
	weeds	≥70	3.26	0.7589	0.2/0.036
	field margin	10 - 19	0.57	0.1315	0.2/0.036
	field margin	20 - 39	0.57	0.1315	0.2/0.036
	field margin	40 - 69	0.57	0.1315	0.2/0.036
	field margin	≥70	0.57	0.1315	0.2/0.036
	adjacent crop	10 - 19	0.69	0.1351	0.2/0.036
	adjacent crop	20 - 39	0.69	0.1351	0.2/0.036
	adjacent crop	40 - 69	0.69	0.1351	0.2/0.036
	adjacent crop	≥70	0.69	0.1351	0.2/0.036
	next crop	10 - 19	2.06	0.3503	0.2/0.036
	next crop	20 - 39	2.06	0.3503	0.2/0.036
	next crop	40 - 69	2.06	0.3503	0.2/0.036
	next crop	≥70	2.06	0.3503	0.2/0.036
Chronic oral toxicity, adult (SUW spray)	treated crop	10 - 19	56.982	-	0.03/NR
	treated crop	20 - 39	56.982	-	0.03/NR
	treated crop	40 - 69	56.982	-	0.03/NR
	treated crop	≥70	0.0	-	0.03/NR
	weeds	10 - 19	16.122	-	0.03/NR
	weeds	20 - 39	12.091	-	0.03/NR
	weeds	40 - 69	6.046	-	0.03/NR
	weeds	≥70	6.046	-	0.03/NR
	field margin	10 - 19	1.048	-	0.03/NR
	field margin	20 - 39	1.048	-	0.03/NR
	field margin	40 - 69	1.048	-	0.03/NR
	field margin	≥70	1.048	-	0.03/NR
	adjacent crop	10 - 19	1.249	-	0.03/NR
	adjacent crop	20 - 39	1.249	-	0.03/NR
	adjacent crop	40 - 69	1.249	-	0.03/NR
	adjacent crop	≥70	1.249	-	0.03/NR

Category	Scenario	BBCH	pear, wild apple, quince (use no. 5, 6) crop category: orchards 1 144 g a.s./ha*		Trigger bees/ bumble bees
			HQ bees	HQ bumble bees	
Larvae toxicity (SUW spray)	next crop	10 - 19	3.752	-	0.03/NR
	next crop	20 - 39	3.752	-	0.03/NR
	next crop	40 - 69	3.752	-	0.03/NR
	next crop	≥70	3.752	-	0.03/NR
	treated crop	10 - 39	120.43	-	0.2/NR
	treated crop	10 - 49	120.43	-	0.2/NR
	treated crop	40 - 69	120.43	-	0.2/NR
	treated crop	≥70	0.00	-	0.2/NR
	weeds	10 - 39	34.75	-	0.2/NR
	weeds	10 - 49	26.06	-	0.2/NR
	weeds	40 - 69	13.03	-	0.2/NR
	weeds	≥70	13.03	-	0.2/NR
	field margin	10 - 39	2.26	-	0.2/NR
	field margin	10 - 49	2.26	-	0.2/NR
	field margin	40 - 69	2.26	-	0.2/NR
	field margin	≥70	2.26	-	0.2/NR
	adjacent crop	10 - 39	2.69	-	0.2/NR
	adjacent crop	10 - 49	2.69	-	0.2/NR
	adjacent crop	40 - 69	2.69	-	0.2/NR
	adjacent crop	≥70	2.69	-	0.2/NR
	next crop	10 - 39	7.90	-	0.2/NR
	next crop	10 - 49	7.90	-	0.2/NR
	next crop	40 - 69	7.90	-	0.2/NR
	next crop	≥70	7.90	-	0.2/NR

ETR: Exposure toxicity ratio; **Bold:** ETR values that exceed the trigger value (need a refinement)

*application is not done during flowering but orchards are sprayed in “late” stage

Since an unacceptable risk is found for bees and bumble bees exposure to SAP09I, a restriction must be imposed in the application of SAP09I according to the application patterns:

SPe8: Dangerous to bees. To protect bees and other pollinating insects do not apply on flowering crops. Do not use where bees are actively foraging. Do not apply when flowering weeds are present. Remove weeds before flowering.

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

Two higher tier studies are available: one with honeybee and another with bumble bees. The study with honeybee was conducted with application of the product when bees were not foraging. Thus, restricting the application of the product to when bees are not foraging, results in an acceptable risk up to 144 g a.s./ha.

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies considered in the first registration in 2003 may be used to evaluate SAP09I. These data were included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, made available to the assessor for the purposes of this evaluation. The study by Vinall, S. (2000), GHE-P-7111 (MJ41), was found in the dossier mentioned above in point 10.4.3/01.

Apis mellifera – semi study (RAR March 2017, Volume 3 – Annex B (PPP))

Vinall, S., 2000. A semi-field test to evaluate the effects of spinosad 480 SC (NAF-85), a suspension concentrate formulation containing 480 g/L DE-105, on the honeybee, *Apis mellifera*; Report No. GHE-P-7111 – unprotected study used for the 1st approval of spinosad

The aim of this study was to assess the effect of NAF-85 (a 480 g/L formulation of DE-105) on honeybees (*Apis mellifera*) in a cage test according to EPPO 170 requirements. Bee hives were placed in plots of flowering *Phacelia tanacetifolia*, one hive per plot of 4 x 4.5 m. NAF-85 was applied in the morning on a flowering crop at a rate of 144 g as/ha and 540 g as/ha. Plots were sprayed when bees were not foraging. All treatments were in triplicate. The sprays were applied at a rate approximately 1500 L/ha. The test was initiated on August 11th, 1999 in Southampton, UK, between 6:30 – 7:30 h in the morning. Cages were 4 x 4.5 m in area. The number of dead bees in each cage and bee behaviour were assessed at -2, -1, 0, 1, 2, 3, 5, and 7 days after treatment. The condition of bee brood and food reserves was assessed at the start and the end of the test. The results were analysed using ANOVA ($p < 0.05$).

At -2, -1 and 0 days after treatment bee mortality and behaviour were similar in all treatments. Mortality in positive control was significantly higher than in control: 1222 dead bees versus 359 in control were found in 7 days. In the low and high NAF-85 treatments, these numbers were 329 and 344 bees, respectively. Bee movement in and out the hives were relatively uniform, except for 14:00 h days after treatment, where movements in the control were significantly greater than in the treatments. Foraging activity however was not uniform. Activity in the positive control was significantly lower from 12:00 h at day of treatment onwards. The 540 g as/ha treatment resulted likewise in significantly reduced foraging activity at most assessments made. Only a small decline in foraging activity was observed in the 144 g as/ha treatment, significant at 10.00, 14.00 and 16.00 h on the first day after treatment. No obvious differences in brood and food reserves were noted between treatments. There was a mean reduction of brood cells of 33% in the control, 56% in the 9.6 g a.s./ha treatment rate, 60% in the 36 g a.s./ha treatment rate and 55% in the toxic reference treatment. Considering the time of year in which the test was performed (August) and the limited pollen availability within the caged plots, this was not unexpected. However, some dead brood was observed in two of the three hives in the 540 g as/ha treatment and not in any other hives. The authors conclude that an early-morning application of Spinosad 480 SC at a rate of 540 g a.s./ha had some effects on hives of the honey bee. Most notably it caused a reduction in the foraging activity of bees over the freshly-treated crop, although this may have been due to repellency. There was no significant increase in worker mortality related to these effects, but some minor effects on the survival of brood were detected for two out of three hives in this treatment. For the lower treatment rate of 144 g a.s./ha, no significant harmful effects on bee behaviour or survival were detected.

Remarks by RMS: Increases of the number of dead bees as large as 341% at 1 day pm after application were not significant. Variability within the data was probably too high to detect significant differences. With 84 dead bees at day 1 for the control the mortality was 2.8% (assuming a total of 3000 bees), while on this day the mortality in the 36 g as/hL (540 g a.s./ha) treatment was still only 4.4% with 132 dead bees. The positive control had a mortality of 11% on day 1. It is agreed that there was no adverse effect on mortality. Effects on foraging behaviour were observed, probably caused by repellency, as well as some effects on brood survival in two out of three hives in the higher application rate. The information on the dead broods was however only qualitative and the observation period does not cover even one full

brood cycle. The following results are used for risk assessment:

- Early morning applications of NAF-85 at rates of 540 and 144 g as/ha (before bees were active) on flowering *Phacelia tanacetifolia* caused no mortality in bees.
- NAF-85 at 540 g as/ha has an effect on foraging behaviour, indications for effects on brood survival are present.
- NAF-85 at 144 g as/ha has no effect on foraging behaviour (but see remark below). Effects on brood were not seen but are not conclusive due to a too short study duration.

Remarks RMS for Renewal: In accordance with a comment from the co-RMS, it is now considered that NAF-85 at 144 g as/ha had a slight effect on foraging behaviour (small decline on the day of application only). The other conclusions are still considered valid.

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies considered in the first registration in 2003 may be used to evaluate SAP09I. These data were included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, made available to the assessor for the purposes of this evaluation. The study by Aldershof, S. (1999), GHE-P-8398 (MJ44), was found in the dossier mentioned above in point 10.4.3/04.

Bombus terrestris – semi field study (RAR March 2017, Volume 3 – Annex B (PPP))

Aldershof, S., 1999. Effect of spinosad (formulated as 480 g/L SC, NAF-85) on the bumblebee *Bombus terrestris* determined under greenhouse conditions – unprotected study used for the 1st approval of spinosad

The effect of NAF-85 (a nominally 480 g/L SC formulation of DE-105 (spinosad), batch no. MC31160P11) applied as a foliar application to tomato plants on adult mortality and foraging activity of the bumblebee *Bombus terrestris* was determined in a semi-field study in accordance with the general recommendations from SETAC/ESCORT (Barrett et al. 1994). Test location was Naaldwijk, The Netherlands. NAF-85 was applied as a foliar spray at a rate of 0.36 g as/L (497-583 g as/ha in the four replicates, mean 542 g as/ha). Imidacloprid was used as a toxic reference and was applied foliarly or as a drench, at a rate of 148 (drench) and 156-168 (foliar, mean 162) g as/ha. Untreated plants were used as the negative control. NAF-85 and the negative control were tested in four replicates (four compartments). Each greenhouse compartment was 58 m² and contained 12 rows of 8 tomato plants of about 6 months old, growing on rockwool. Imidacloprid was tested in two replicates (two for the drench and two for the foliar treatment). Bumblebee colonies were obtained from a commercial supplier. One colony was placed in one compartment. Colonies were queen-right and manipulated such that each colony contained 25 workers when introduced into the test compartments. Pupae were removed before introduction. Applications took place in the late afternoon on the day before the exposure phase started. Hives were opened in the early afternoon after the day of application to allow bees to forage. Mortality was determined 1 week after application. Foraging was expressed as proportion of flowers observed with bite marks. Flowers were counted 2, 4 and 6 days after exposure started. Effect on development of the colony was determined during an additional observation period in the laboratory of 8 days, after the end of the exposure period. Mortality was analysed statistically using Fisher's exact test. Proportions of flowers observed with bite marks were examined in a repeated measures ANOVA, followed by linear contrasts for pairwise comparisons. Brood development data were examined statistically using a one-way ANOVA, followed by linear contrasts for pairwise comparisons. Brood development data were examined statistically using a one-way ANOVA, followed by Fisher's LSD test for pairwise comparisons. Temperature and relative humidity were measured throughout the test. Control corrected mortality was - 1% for the spinosad treatment and 14 and 4% for the drench and the foliar application of the toxic reference, respectively. The proportion of flowers observed with bite marks in untreated control compartments was rather constant during the observation period (36-44%). Foliar application of NAF-85

caused a reduction in brood development (new adults plus larvae and pupae) of 27%. This reduction was not statistically significant. See the table below, copied from the report. The only statistically significant reduction were found for imidacloprid applied as drench (for larvae + pupae and for total offspring).

Remarks by RMS: The results that NAF-85 at an application rate of 0.36 g as/L (542 g as/ha, sprayed on tomato plants) causes 1% mortality after 7 days, a 65% reduction in foraging activity after 6 days and a 27% reduction in colony development after 16 days, are used for risk assessment. Although the reduced foraging activity and brood development in the treatment were not statistically significant, they are considered relevant.

9.6.3 Effects on bumble bees

Please refer to point 9.6.2.1.

9.6.4 Effects on solitary bees

No data available.

9.6.5 Overall conclusions

An unacceptable risk is found for bees exposure to SAP09I. Therefore, a restriction must be imposed in the application of this product according to the proposed application patterns:

SPe8: Dangerous to bees. To protect bees and other pollinating insects do not apply on flowering crops. Do not use where bees are actively foraging. Do not apply when flowering weeds are present. Remove weeds before flowering.

Review Comments:

The evaluation of the acute 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.

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

Furthermore, the evaluation of the acute and chronic risk for bees was performed by the applicant in accordance with the recommendations of the “EFSA Guidance Document on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees)” (EFSA Journal 2013;11(7):3295; hereafter referred to as EFSA/2013/3295). As the HQ and ETR values are above than the relevant triggers, therefore, an unacceptable acute and chronic risk to bees is concluded.

The SPe8 restriction must be respected.

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 spinosad. Full details of these studies are provided in the respective EU DAR and related documents as well as in Appendix 2 of this document (new studies).

Effects on non-target arthropods of SAP09I were not evaluated as part of the EU assessment of spinosad. 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 9.7-1: Endpoints and effect values relevant for the risk assessment for non-target arthropods

Species	Substance	Exposure System	Results (effect)	Reference
Laboratory studies on glass				
<i>Typhlodromus pyri</i>	Spinosad	Laboratory test glass plates, 14 days	100% mortality at 31 g a.s./ha	SANCO/1428/2001 – rev. final (2006)
<i>Typhlodromus pyri</i>	NAF-85	Laboratory test glass plates, 7 days	100% mortality at 40-320 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/01, MJ11)
<i>Typhlodromus pyri</i>	SAP09I	Laboratory test glass plates, 7 days	100% mortality at 40-320 g a.s./ha	KCP 10.3.2.1/01, Varela, S., 2022, S21-04370
<i>Aphidius rhopalosiphi</i> (protected luife stage)	NAF-85	lab, direct spray of mummified aphids on glass, 9 days	20% mortality at 38 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/07, MJ19)
<i>Aphidius rhopalosiphi</i>	NAF-85	lab, residue on glass, 24 hour	100% mortality at 40-320 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/03, MJ12)
<i>Coccinella septempunctata</i>	Spinosad	lab, residue on glass, 10 weeks (fecundity)	17 g a.s./L, until run-off Mortality: +1.1% Fecundity: +9.3%	SANCO/1428/2001 – rev. final (2006)
<i>Coccinella septempunctata</i>	NAF-85	lab, residue on glass, 14 weeks (fecundity)	40 g a.s./ha Mortality 11% Fecundity: 43%	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/11, MJ10)
<i>Coccinella septempunctata</i>	NAF-85	lab, residue on glass, 14 weeks (fecundity)	160 g a.s./ha Mortality: 8% Fecundity: 40%	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/11, MJ10)

Species	Substance	Exposure System	Results (effect)	Reference
<i>Coccinella septempunctata</i>	NAF-85	lab, residue on glass, 14 weeks (fecundity)	320 g a.s./ha Mortality: 19% Fecundity: 20%	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/11, MJ10)
<i>Poecilus cupreus</i>	NAF-85	lab, direct spray of beetles and food on sand, 14 days	0-3.3% effect on food consumption (n.s.) at 4 applications 29-38-67-77 of 50-400 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/16, MJ13)
<i>Poecilus cupreus</i>	NAF-85	lab, direct spray of beetles and food on sand, 29 days	6.7% mortality and 4% effect on food consumption at 1 x 540 g a.s./ha	SANCO/1428/2001 – rev. final (2006) MJ23
<i>Poecilus cupreus</i>	NAF-85	lab, direct spray of beetles and food on sand, 29 days	63% mortality and 19% effect on food consumption at 2 x 540 g a.s./ha, interval 8 d	SANCO/1428/2001 – rev. final (2006) MJ23
<i>Poecilus cupreus</i>	NAF-85	lab, direct spray of beetles and food on sand, 29 days	81% mortality and 41% effect on food consumption at 3 x 540 g a.s./ha, interval 8 d	SANCO/1428/2001 – rev. final (2006) MJ23
<i>Episyrphus balteatus</i>	Spinosad	lab, residue on glass, 10 days	45 g a.s./L, until run-off Mortality: 80% Fecundity: 75%	SANCO/1428/2001 – rev. final (2006)
Extended laboratory studies				
<i>Aphidius rhopalosiphi</i>	NAF-85	extended lab, residue on plants, 13 days	Mortality: 13% at 4.8 g a.s./ha 40% at 9.6 g a.s./ha 67% at 19 g a.s./ha 80% at 38 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/07, MJ19)
<i>Aphidius rhopalosiphi</i> (protected luife stage)	NAF-85	extended lab, direct spray of mummified aphids on plants, 7 days	47% mortality at 38 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/07, MJ19)
<i>Aphidius colemani</i>	NAF-85	extended lab, residue on plants, 2 days	100% mortality at 540 g a.s./ha, aged 0-5 d 98% mortality at 540 g a.s./ha, aged 7 d 100% mortality at 1080 g a.s./ha, aged 0-7 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/05, MJ22)
<i>Aphidius colemani</i>	NAF-85	extended lab, residue on leaves, 2 days	100% mortality at 0.36 g a.s./L until run-off, aged 0-29 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/06, MJ39)
<i>Aphidius colemani</i> (protected luife stage)	NAF-85	extended lab, direct spray of mummified aphids on plants, 7 days	82% effect on emergende at 0.36 g a.s./L until run-off	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/07, MJ31)

Species	Substance	Exposure System	Results (effect)	Reference
<i>Phytoseiulus persimilis</i>	NAF-85	Extended laboratory test on plants, 6 days	93% mortality at 2 x 0.36 g a.s./L until runoff, interval 14 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/02, MJ29)
<i>Chrysoperla carnea</i>	NAF-85	Extended lab, residue on plants, 12 days	2 x 0.36 g a.s./L until run-off, interval 14 d Mortality: 25% Fecundity: 18%	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/13, MJ24)
<i>Coccinella septempunctata</i>	NAF-85	extended lab, residue on leaves, 8 weeks minimum	2 x 0.36 g a.s./L until run-off, interval 14 d Mortality: 16%	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/12, MJ30)
Field or semi-field tests				
<i>Typhlodromus pyri</i>	NAF-85	field, sprayed vineyard, ±7 weeks	<25% effect on abundance at 24 and 48 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/01, MJ27)
<i>Typhlodromus pyri</i>	NAF-85	field, sprayed vineyard, ±7 weeks	43% effect on abundance after 7 d; <25% from 16d at 96 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/01, MJ27)
<i>Typhlodromus pyri</i>	NAF-85	field, sprayed vineyard	6.8-34% effect <25% effect on abundance after 4 th treatment at 4 applications 29-38-67-77	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/05, MJ51 / IIIA 10.5.2/02, MJ64)
<i>Typhlodromus pyri</i>	NAF-85	field, sprayed vineyard	<25% effect on abundance at 4 applications 29-38-67-77	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/05, MJ51 / IIIA 10.5.2/02, MJ64)
<i>Typhlodromus pyri</i>	NAF-85	Field experiment, sprayed vineyard 75 days ³	23.5% maximum effect on abundance after 35 d at 2 application 3.8 g a.s./ha ¹ and 3 applications 3.2 g a.s./ha ²	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/03, MJ68)
<i>Typhlodromus pyri</i>	NAF-85	Field experiment, sprayed vineyard, 75 days	31.6% maximum effect on abundance after 14 d at 2 application 58 g a.s./ha ¹ and 3 applications 48 g a.s./ha ²	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/03, MJ68)

Species	Substance	Exposure System	Results (effect)	Reference
<i>Typhlodromus pyri</i>	NAF-85	Field experiment, apple orchard, 122 days ³	34.1% maximum effect on abundance after 93 d at 2 applications 39 g a.s./ha ¹ and 3 applications 18 g a.s./ha ²	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/04, MJ69)
<i>Typhlodromus pyri</i>	NAF-85	Field experiment, apple orchard, 122 days	26% positive effect maximum on abundance after 122 d at 4 applications 166 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/04, MJ69)
<i>Typhlodromus pyri</i>	NAF-85	Field sprayed vineyard, Freiburg	24.6% positive effect on abundance at 4 applications 29-39-68-78	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.2/02, MJ64)
<i>Chrysoperla carnea</i>	NAF-85	Semi-field, 75 d.	16.5% effect on mortality, 30% positive effect on fecundity and 0.4% positive effect on hatching success at 2 x 26 g a.s./ha and 2 x 11 g a.s./ha	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/14, MJ71)
<i>Aphidius colemani</i>	NAF-85	semi-field, direct spray of aphids on plants, 12 days	70% effect on parasitism at 0.096 g a.s./L until run-off and 88% effect on parasitism at 0.36 g a.s./L until run-off	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	98% effect on parasitism at 0.096 g a.s./L until run-off aged 1 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	41% effect on parasitism at 0.096 g a.s./L until run-off aged 7 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	46% effect on parasitism at 0.096 g a.s./L until run-off aged 14 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	98% effect on parasitism at 0.36 g a.s./L until run-off aged 1 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	36% effect on parasitism at 0.36 g a.s./L until run-off aged 7 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)

Species	Substance	Exposure System	Results (effect)	Reference
<i>Aphidius colemani</i>	NAF-85	semi-field, residue on plants, 12 days	27% effect on parasitism at 0.36 g a.s./L until run-off aged 14 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/08, MJ43)
<i>Aphidius colemani</i>	NAF-85	semi-field (glasshouse); extended lab	95% maximum mortality after 1 d at 4 x 109 g a.s./ha, 46d, fresh residue 0% mortality after 14 d at 4 x 109 g a.s./ha, 60 d, aged 14 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/10, MJ70)
<i>Aphidius colemani</i>	NAF-85	semi-field (glasshouse); extended lab ³	45% maximum mortality after 1 d at 2 x 26 g a.s./ha and 2 x 11 g a.s./ha, 46 d, fresh residue 0% maximum mortality after 14 d at 2 x 26 g a.s./ha and 2 x 11 g a.s./ha, 60 d, aged 14 d	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/10, MJ70)
<i>Macrolophus caliginosus</i>	NAF-85	semi-field, direct spray of bugs on plants, 14 days	23% (1-7 days) effect on abundance at 0.0096 g a.s./L until run-off	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/15, MJ42)
<i>Macrolophus caliginosus</i>	NAF-85	semi-field, direct spray of bugs on plants, 14 days	49% (1 day), 75% (3 days), 64% (7 days), 32% (14 days) effect on abundance at 0.36 g a.s./L until run-off	SANCO/1428/2001 – rev. final (2006) (IIIA 10.5.1/15, MJ42)
<i>Natural off-field NTA community</i>	SAP09I	Field study	Population NOER = 1.5 g a.s./ha NOEAER = 24 g a.s./ha	KCP 10.3.2.4/01, Roig, 2022, S20-09667

1 first and second application

2 third, fourth and fifth application

3 lower application rate to simulate exposure due to drift

Review Comments:

In accordance with the request of the Polish Ministry of Agriculture and Rural Development DHR.rs1.8208.1.5.2025, dated 09 April 2025, only SpinTor 480 SC (GF-976/NAF-85) studies considered in the first registration in 2003 may be used to evaluate SAP09I. These data were included in document M-III: GF-976 (SPINOSAD, 480 G/L SUSPENSION CONCENTRATE) Section 6: Ecotoxicology, made available to the assessor for the purposes of this evaluation. The study by Miles, M (1999), GHE-P-7888 (MJ23, *Poecilus cupreus*), was not found in the dossier mentioned above.

9.7.1.1 Justification for new endpoints

No other non-target arthropod studies were performed with the formulation, with the exception of the *Typhlodromus pyri* glass plate study. The studies performed with the active substance and the reference product are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Additionally, the risk assessment was performed with the lowest endpoints available (either coming from the active substance data or the reference product). Therefore, no further studies are considered necessary.

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

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 6) covers the risk for non-target arthropods from all intended uses (see 9.1.2).

PER_{in-field} values according to ESCORT 2 as:

Application rate × MAF

Table 9.7-2: First- and higher-tier assessment of the in-field risk for non-target arthropods due to the use of SAP09I in orchards (use no. 6) – risk envelope approach

Intended use	orchards (use no. 6) – risk envelope approach		
Active substance/product	spinosad		
Application rate (g/ha)	2 × 144		
MAF	1.7		
Tier I			
Test species	LR ₅₀ (lab.) (g/ha)	PER _{in-field} (g/ha)	HQ _{in-field} criterion: HQ ≤ 2
<i>Typhlodromus pyri</i>	<31	244.8	>7.90
<i>Aphidius rhopalosiphi</i>	<40		>6.12
<i>Coccinella septempunctata</i>	>320		<0.77
<i>Poecilus cupreus</i>	>540		<0.45
Test species	Concentration with ≤ 50 % effect* (g/L)	PER _{in-field} (g/L)	HQ _{in-field} criterion: HQ ≤ 2
<i>Episyrphus balteatus</i>	<45	0.48 (assuming 144 g a.s./ha at 300 L water/ha)	>0.01, the HQ result is as unbounded value. However, there is a high margin of safety so considering an unacceptable risk would be overconservative and probably not ecologically relevant.

Higher-tier (extended lab)			
Test species	Rate with ≤ 50 % effect* (g/ha)	PER _{in-field} (g/ha)	PER _{in-field} below rate with ≤ 50 % effect?
<i>Aphidius rhopalosiphi</i>	9.6	244.8	No (please see below the results of the semi-field studies for <i>A. colemani</i> , a close relative of <i>A. rhopalosiphi</i>).
<i>Aphidius rhopalosiphi</i> (protected life stage)	>38		
Test species	Concentration with ≤ 50 % effect* (g/L)	PER _{in-field} (g/L)	HQ _{in-field} criterion: HQ ≤ 2
<i>Aphidius colemani</i>	<0.36 g/L	0.48 (assuming 144 g a.s./ha at 300 L water/ha)	No
<i>Phytoseiulus persimilis</i>	<2x0.36 g/L (xMAF1.7 = 0.612 g/L)		Yes
<i>Chrysoperla carnea</i>	>2x0.36g/L (xMAF1.7 = 0.612 g/L)		Yes
<i>Coccinella septempunctata</i>	>2x0.36g/L (xMAF1.7 = 0.612 g/L)		Yes
Higher-tier (field/semi-field studies)			
Test species	Rate with ≤ 50 % effect (g/ha)	PER _{in-field} (g/ha)	PER _{in-field} below rate with ≤ 50 % effect?
<i>Typhlodromus pyri</i>	>96	244.8	No
<i>Chrysoperla carnea</i>	>2x26 (xMAF1.7 = 44.2)		No
<i>Aphidius colemani</i>	4 x 109 (xMAF2.7 = 294.3)		Yes
Test species	Concentration with ≤ 50 % effect* (g/L)	PER _{in-field} (g/L)	HQ _{in-field} criterion: HQ ≤ 2
<i>Macrolophus caliginosus</i>	0.36 g/L after 14 d	0.48 (assuming 144 g a.s./ha at 300 L water/ha)	No, at 0.36g/L possible recolonization was seen after 14 days. Based on this results, it is unlikely that at 0.48 g/L possible recolonization of the in-field area will take longer than 1 year (the maximum time allowed according to ESCORT2). Therefore, it is likely that possible recolonization of the in-field area will occur for this species within an acceptable time frame.

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 LR₅₀ or ER₅₀ from a relevant extended laboratory test is available, it should be considered in place of the rate with ≤ 50 % effect.

A. rhopalosiphi is mainly affected by spinosad on mortality, as reproduction seems to be the least sensitive parameter (evidenced also by the low sensitivity of the protected life stages). Semi-field studies are available for a closely related species, *A. colemani*. The laboratory studies mentioned above showed that this species seems to be even more sensitive than *A. rhopalosiphi*. One of the semi-field studies conducted with *A. colemani* showed that 4 x 109 g a.s./ha lead to a high mortality, directly after

applications, but no effects are found after 14 days aging. This indicates that recovery can take place within an ecological relevant period.

Several field studies are also available for *T. pyri* which show slight effects (but not significant) with no long-term effects infield.

The information described above shows a lower or the same sensitivity of other arthropods to *T. pyri* and *A. rhopalosiphi*. Hence, also for these species it can be concluded that the in-field risk is acceptable.

Review Comments:

Based on all available laboratory, semi-field and field studies with NAF–85 the following conclusion was drawn in DAR - SPINOSAD – VOLUME 3 – ANNEX B:

In-crop assessment: The proposed maximum dosage is lowered to a maximum of 144 g a.s./ha with a maximum frequency of three times. In the submitted field studies with *Aphidius spec* and *T.pyri* recovery is shown, also at the highest dose. Because these species are belonging to the most sensitive species for spinosad the risk in-crop is considered acceptable for all non-target arthropods.

No further assessment is required.

9.7.2.2 Risk assessment for off-field exposure

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for orchards (use no. 6) covers the risk for non-target arthropods from all intended uses (see 9.1.2).

PER_{off-field} values according to ESCORT 2 as:

Application rate × MAF × (drift factor/vegetation distribution factor)

Corrected PER_{off-field} values according to ESCORT 2 as:

corr. PER_{off-field} = PER_{off-field} / correction factor

Table 9.7-3: ~~First~~ and higher-tier assessment of the off-field risk for non-target arthropods due to the use of SAP09I in orchards (use no. 6) – risk envelope approach

Intended use		orchards (use no. 6) – risk envelope approach			
Active substance/product		Spinosad (SAP09I)			
Application rate (g/ha)		2 × 144			
MAF		1.7			
VDF		5 ⁺ N/A			
Test species Higher-tier	Rate with ≤ 50 % effect* NOER (g/ha)	Drift rate (%)	CF	corr. PER_{off-field} (g/ha)	corr. PER_{off-field} below rate with ≤ 50 % effect? PER_{off-field} below NOER?
Off-field field study	1.5	25.53 (fruit crops, 2 appl.)	5 N/A	4.920 62.50	No

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 LR₅₀ or ER₅₀ from a relevant extended laboratory test is available, it should be considered in place of the rate with ≤ 50 % effect.

⁺ VDF of 5 for all the tiers of the assessment was agreed in Working document on Risk Assessment of Plant Protection Products in the Central Zone, Ecotoxicology, Version 1.0, May 2021

9.7.2.3 Additional higher-tier risk assessment

~~Not relevant.~~ Please see above.

9.7.2.4 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 (DRN). The results of the risk assessment using typical mitigation measures (no-spray buffer zones of 5 or 10 m; drift-reducing nozzles (DRN) with reduction by 50 %, 75 %, or 90 %) are summarised in the following tables.

Table 9.7-4: Assessment of the off-field risk for non-target arthropods due to the use of SAP09I in leafy vegetables (use no. 1, 2) and bulb and onion like vegetables (use no. 3, 4, 7) considering risk mitigation (in-field no-spray buffer zones, and drift-reducing nozzles)

Intended use		leafy vegetables (use no. 1, 2), bulb and onion like vegetables (use no. 3, 4, 7)			
Active substance/product		Spinosad (SAP09I)			
Application rate (g/ha)		3 × 96			
MAF		2.3			
VDF		5 ⁺ N/A			
Buffer strip (m)	Drift rate (%)	corr. PER_{off-field} (g/ha)	corr. PER_{off-field} 50% drift red. (g/ha)	corr. PER_{off-field} 75% drift red. (g/ha)	corr. PER_{off-field} 90% drift red. (g/ha)
1	2.01	4.438	2.219	1.11	0.444
5	0.41	0.905	0.453	0.226	0.091
10	0.20	0.442	0.221	0.111	0.044
15	0.14	0.309	0.155	0.077	0.031
20	0.10	0.221	0.111	0.055	0.022
Higher-tier toxicity value					
NOER = 0.3 1.5 g/ha		corr. PER_{off-field} below rate with ≤ 50 % effect PER_{off-field} below NOER?			
1		No	No	No Yes	No Yes
5		No Yes	No Yes	Yes	Yes
10		No	Yes	Yes	Yes
15		No	Yes	Yes	Yes
20		Yes	Yes	Yes	Yes

MAF: Multiple application factor; PER: Predicted environmental rates; HQ: Hazard quotient; Criteria values shown in bold breach the relevant trigger.

⁺ VDF of 5 for all the tiers of the assessment was agreed in Working document on Risk Assessment of Plant Protection Products in the Central Zone, Ecotoxicology, Version 1.0, May 2021

Table 9.7-5: Assessment of the off-field risk for non-target arthropods due to the use of SAP09I in orchards (use no. 5, 6) considering risk mitigation (in-field no-spray buffer zones, and drift-reducing nozzles)

Intended use		orchards (use no. 5, 6)			
Active substance/product		spinosad			
Application rate (g/ha)		2 × 144			
MAF		1.7			
VDF		5 [†]			
Buffer strip (m)	Drift rate (%)	corr. PER_{off-field} (g/ha)	corr. PER_{off-field} 50 % drift red. (g/ha)	corr. PER_{off-field} 75 % drift red. (g/ha)	corr. PER_{off-field} 90 % drift red. (g/ha)
3	25.53	4.920	2.460	1.230	0.492
5	16.87	1.004	0.502	0.251	0.100
10	9.61	0.490	0.245	0.123	0.049
15	5.61	0.343	0.172	0.086	0.034
20	2.59	0.245	0.123	0.061	0.025
Higher tier toxicity value					
NOER = 0.3 g/ha		corr. PER _{off-field} below rate with ≤ 50 % effect			
3		No	No	No	No
5		No	No	Yes	Yes
10		No	Yes	Yes	Yes
15		No	Yes	Yes	Yes
20		Yes	Yes	Yes	Yes

MAF: Multiple application factor; PER: Predicted environmental rates; HQ: Hazard quotient; Criteria values shown in bold breach the relevant trigger.

[†] VDF of 5 for all the tiers of the assessment was agreed in Working document on Risk Assessment of Plant Protection Products in the Central Zone, Ecotoxicology, Version 1.0, May 2021

Intended use		Orchards (use no. 5, 6)			
Active substance/product		Spinosad (SAP09I)			
Application rate (g/ha)		2 × 144			
MAF		1.7			
Buffer strip (m)	Drift rate (%)	corr. PER_{off-field} (g/ha)	corr. PER_{off-field} 50 % drift red. (g/ha)	corr. PER_{off-field} 75 % drift red. (g/ha)	corr. PER_{off-field} 90 % drift red. (g/ha)
3	25.53	62.50	31.25	15.62	6.25
5	16.87	41.30	20.65	10.32	4.13
10	9.61	23.53	11.76	5.88	2.35
15	5.61	13.73	6.87	3.43	1.37
20	2.59	6.34	3.17	1.59	0.63
30	0.87	2.13	1.06	-	-
40	0.40	0.98	-	-	-

Intended use	Orchards (use no. 5, 6)			
Active substance/product	Spinosad (SAP09I)			
Application rate (g/ha)	2 × 144			
MAF	1.7			
Higher-tier toxicity value				
NOER = 1.5 g/ha	PER_{off-field} below NOER?			
3	No	No	No	No
5	No	No	No	No
10	No	No	No	No
15	No	No	No	Yes
20	No	No	No	Yes
30	No	Yes	-	-
40	Yes	-	-	-

MAF: Multiple application factor; PER: Predicted environmental rates; HQ: Hazard quotient; Criteria values shown in bold breach the relevant trigger.

9.7.3 Overall conclusions

The risk assessment for non-target arthropods was conducted according to SANCO/10329/2002 rev 2 final. No unacceptable risk is expected when SAP09I is applied according to the proposed application patterns when the following mitigation measures are in place:

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 20m or 10m with 50% DRN or 5m with 75 % DRN to adjacent crops/non-agricultural land in applications to vegetables.

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 20m or 10m with 50% DRN or 5m with 75 % DRN to adjacent crops/non-agricultural land in applications to orchards.

Review Comments:

Based on the results of field studies used in the risk assessment, it can be concluded that SAP09I does not pose a risk to non-target arthropods when applied according to proposed use pattern. Particular precautions to reduce the environmental concentrations applications are required.

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 5m or 1m with 75% DRN to non-agricultural land in applications to vegetables.

SPE3: To protect non-target arthropods, respect an unsprayed buffer zone of 40m or 30m with 50% DRN or 15m with 90 % DRN to non-agricultural land in applications to orchards.

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 spinosad and its relevant metabolites. Full details of these studies are provided in

the respective EU DAR and related documents as well as in Appendix 2 of this document (new studies).

Effects on earthworms and other non-target soil organisms (meso- and macrofauna) of SAP09I were not evaluated as part of the EU assessment of spinosad. 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 9.8-1: 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
<i>Eisenia fetida</i>	Spinosad (NAF-85)	Mixed into substrate Acute	LC ₅₀ > 916 mg/kg dw LC _{50,corr} > 458 mg/kg dw*	SANCO/1428/2001 – rev. final (2006)
<i>Eisenia fetida</i>	Spinosyn B	Mixed into substrate Acute	LC ₅₀ > 1000 mg/kg dw LC _{50,corr} > 500 mg/kg dw*	SANCO/1428/2001 – rev. final (2006)
<i>Eisenia fetida</i>	N-demethylated spinosyn D	Mixed into substrate Acute	LC ₅₀ > 1000 mg/kg dw LC _{50,corr} > 500 mg/kg dw*	SANCO/1428/2001 – rev. final (2006)
<i>Eisenia fetida</i>	Spinosad (NAF-85)	Mixed into substrate Chronic	NOEC > 2700 mg/kg dw NOEC _{corr} > 1350 mg/kg dw*	SANCO/1428/2001 – rev. final (2006) (IIIA 10.6.1.2/01, MJ45)
<i>Eisenia fetida</i>	Spinosyn B	Mixed into substrate Chronic	NOEC ≥ 3.582 mg/kg dw NOEC _{corr} ≥ 1.791 mg/kg dw*	SANCO/1428/2001 – rev. final (2006)
<i>Eisenia fetida</i>	N-demethylated spinosyn D	Mixed into substrate Chronic	NOEC ≥ 1.928 mg/kg dw NOEC _{corr} ≥ 0.964 mg/kg dw*	SANCO/1428/2001 – rev. final (2006)
<i>Folsomia candida</i>	SAP09I	Mixed into substrate 21 d, chronic 5 % peat content	NOEC _{MORTALITY} > 42.59 mg a.s./kg soil NOEC = 0.70 mg a.s./kg dw EC ₁₀ = 0.98 mg a.s./kg soil	KCP 10.4.2.1/01, Antón, 2021, S21-04368
<i>Hypoaspis aculeifer</i>	SAP09I	Mixed into substrate 14 d, chronic 5 % peat content	NOEC _{MORTALITY} > 45.35 mg a.s./kg soil NOEC = 1.33 mg a.s./kg dw EC ₁₀ = 1.50 mg a.s./kg soil	KCP 10.4.2.1/02, Lozano, 2022, S21-04369
Field studies				
-				
Litter bag test				
-				

* 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

No earthworm studies were performed with the formulation. The studies performed with the active substance and the reference product are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Therefore, no further studies are considered necessary.

Two studies were conducted with the formulation SAP09I, one on the collembolan *Folsomia candida* and another on the mite *Hypoaspis aculeifer*. These two studies were conducted according to the respective OECD guidelines, which means that they were conducted using the artificial soil OECD with 5 % peat (reduced organic matter content). Since Spinosad has a $\log Pow > 2$, the correction of the endpoints is an issue to consider.

In the EPPO scheme of 2002, it is stated that “*For chemicals with $\log Pow > 2$, sorption is expected to be linearly related to soil organic matter content. A factor of 2 (division of all toxicity end-points) is used to correct the toxicity data from earthworm studies conducted in artificial soil. This derives from the fact that the artificial soil of OECD guideline 207 has a peat content of 10%, whereas atypical agricultural soil has not more than 5% organic matter.*” This regulation implies the correction of the endpoints obtained in studies conducted with earthworms and only when a peat content of 10% would be used, which is not the case. In 2015, the Outcome on recurring issues in ecotoxicology brought another light on this issue by stating that “*it was agreed to retain the factor of 2 for all the first tier studies, when relevant (i.e. $\log Pow$ of > 2)*”. This would mean that studies conducted with all different soil organisms with either peat content (5 or 10%) would need to be corrected. In the same document it is also stated that “*Some experts considered the factor of 2 as not scientifically underpinned and, in some cases, not even sufficiently protective. It was agreed that there is a clear lack of data regarding the bioavailability of test substances in soils with different amounts of organic matter. It was also noted that this is a complex topic and there could be more influential parameters than just the organic matter used in the test soil affecting the toxicity*” and more specifically for collembolan and mites “*It was noted that the application of the correction factor of 2 may not be totally appropriate for collembolan and soil mite studies where 5% OM is used in the test guideline. (...)*”. The Applicant agrees that the application of the correction factor without further evidence to endpoints of collembolans and mites is quite arbitrary and overly conservative and would like to provide further arguments for this point.

All these soil organisms (earthworms, collembolans and mites) have as one of the main functions in soil the decomposition of organic matter. However, their contribution to such process is quite different; with earthworms feeding directly on soil and its organic matter, and collembolans and mites being saprophytes (thus mainly feeding on decomposing items). Consequently, their exposure to soil organic matter (and possible substances mixed in it) will be much different with earthworms being the most exposed ones.

Awhile the study conduction, all standard species are being fed with specific food items that are placed on top of the soil. However, for earthworms the added food item is organic matter (namely cow/horse manure) which they easily mix into soil and they additionally still feed on the soil organic content. Collembolans and mites are fed with yeast or cheese mites, respectively, thus not so exposed to the soil organic matter. Contact exposure is also very different between the soil organisms: earthworms are soft-bodied organisms, only protected by a tegument; while collembolans and mites are protected by a rigid exoskeleton that protects them from exposure through contact to substances present in soil. The most relevant exposure pathway for both collembolans and mites is expected to be the pore water ingestion which would be similar for all soil organisms. In summary, in the Applicant's point of view:

- earthworms are highly dependent and exposed to soil organic matter (and substances mixed in it):
 - o through feeding of the organic matter and also through the soil pore water
 - o and contact exposure
- collembolans and mites are less dependent on soil organic matter and will be mainly exposed to substances in the soil pore water (exposure pathway shared with earthworms).

This point of view is further supported by scientific literature. Jänsch *et al.* (2005) is a review paper which analyzed all available published data to date and tried to infer on the most important soil

characteristics for standard soil species, i.e. the ecological requirements of these species (the paper is sent in the KCP folder B9 of this dossier). In this paper, the pH, temperature, moisture, soil (organic matter and texture) and food preferences are analyzed for the earthworms *Eisenia fetida* and *Eisenia andrei*, the collembolans *Folsomia candida* and the mites *Hypoaspis aculeifer*. Please find in the table below the main findings of the study related to organic matter and food.

Table 9.8-2: Ecological requirements preference of the standard species usually tested in the risk assessment of plant protection products according to Jänsch *et al.* (2005).

Species	Soil property			
	Organic matter		Food	
	range tested	preference	range tested	preference
<i>Eisenia fetida</i> & <i>Eisenia andrei</i>	wide	Very high	variable	Nutrient-rich dead organic matter
<i>Folsomia candida</i>	1-11.5	Unknown	variable	fungi
<i>Hypoaspis aculeifer</i>	wide	none	Wide (predatory)	<i>Tyrophagus putrescentiae</i>

Namely for collembolans the following information is stated in the scientific paper that “*recently these endpoints were studied in 11 natural soils (plus OECD artificial soil and the standard LUFA 2.2 soil as controls), covering a wide range of soil properties (Table 6; Amorim et al. 2005c). With the exception of a negative correlation between adult survival and soil cation exchange capacity (CEC) values there was minimal influence of soil properties on the survival and reproduction of collembolans (Fig. 2)*” and “*Owing to their broad range of ecological requirements, without specific preferences in terms of organic matter and pH, different soil types can be successfully inhabited by this species*”. Also for mites, the authors specified that “*Hypoaspis aculeifer is not affected by soil type as long as there is sufficient moisture and prey. It occurs in almost every kind of soil (Glockemann 1992; Ignatowicz 1974)*”. These clearly reinforce the arguments provided above by the Applicant.

Accounting for all the abovementioned arguments, the Applicant believes that since both studies were conducted with artificial soil OECD 5% peat content, the endpoints obtained for collembolans and mites should not be corrected.

Review Comments:

According to Outcome of the pesticides peer review meeting on general recurring issues in ecotoxicology - EFSA Supporting publication 2015:EN-924 point 5, the factor of 2 should be applied for all the first tier studies, when relevant (i.e. log Pow of >2). Therefore, the NOEC values for collembola and soil mite were corrected accordingly.

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. According to the assessment of environmental-fate data, multi-annual accumulation in soil does not need to be considered for spinosad but needs to be considered for both spinosad metabolites (spinosyn B and N-demethylated spinosyn D).

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for bulb

and onion like vegetables (use no. 3) covers the risk for earthworms from all other intended uses (see 9.1.2).

Table 9.8-3: First-tier assessment of the acute and chronic risk for earthworms due to the use of SAP09I in bulb and onion like vegetables (use no. 3) – risk envelope approach

Intended use	bulb and onion like vegetables (use no. 3) – risk envelope approach		
Acute effects on earthworms – no longer a data requirement			
Chronic effects on earthworms			
Product/active substance	NOEC (mg/kg dw)	PEC _{soil} (mg/kg dw)	TER _{It} (criterion TER ≥ 5)
Spinosad	1350	0.237 ¹	5696
Spinosyn B	1.791	0.187 ²	9.58
N-demethylated spinosyn D	0.964	0.031 ²	31.10

TER values shown in bold fall below the relevant trigger.

¹PECs Tier I modelling

²PECs,accumulation

Table 9.8-4: 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 SAP09I in all intended uses

Intended use	all intended uses		
Acute effects on earthworms – no longer a data requirement			
Chronic effects on other soil macro- and mesofauna			
Product/active substance	NOEC _{CORR.} (mg/kg dw)	PEC _{soil} (mg/kg dw)	TER _{It} (criterion TER ≥ 5)
Cabbage, Broccoli, Cauliflower (uses no. 1, 2) – application rate 3 x 0.2 L/ha (3 x 96 g a.s./ha), interval: 7d, BBCH 10-49			
SAP09I – <i>Folsomia candida</i>	0.70 0.35	0.186 ¹	3.76 1.88
		0.079 ²	8.86 4.43
SAP09I – <i>Hypoaspis aculeifer</i>	1.33 0.665	0.186 ¹	7.15 3.58
		0.079	8.42
Cabbage, Broccoli, Cauliflower (uses no. 1, 2) – application rate 2 x 0.16 L/ha (2 x 76.8 g a.s./ha), interval: 10d, BBCH 20-49			
SAP09I – <i>Folsomia candida</i>	0.35	0.065 ²	5.38
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.065 ²	10.23
Cabbage, Broccoli, Cauliflower (uses no. 1, 2) – application rate 3 x 0.1 L/ha (3 x 48 g a.s./ha), interval: 10d, BBCH 10-49			
SAP09I – <i>Folsomia candida</i>	0.35	0.089 ¹	3.93
		0.040 ²	8.71
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.089 ¹	7.46
Onion, leek, garlic (uses no. 3, 4, 7) – application rate 3 x 0.2 L/ha (3 x 96 g a.s./ha), interval: 10d, BBCH 10-49			
SAP09I – <i>Folsomia candida</i>	0.70 0.35	0.237 ¹	2.95 1.48
		0.119 ²	5.88 2.94

Intended use	all intended uses		
Acute effects on earthworms – no longer a data requirement			
Chronic effects on other soil macro- and mesofauna			
Product/active substance	NOEC _{CORR.} (mg/kg dw)	PEC _{soil} (mg/kg dw)	TER _{It} (criterion TER ≥ 5)
SAP09I – <i>Hypoaspis aculeifer</i>	1.33 0.665	0.237 ¹	5.61 2.81
		0.119	5.59
Onion, leek, garlic (uses no. 3, 4, 7) – application rate 1 x 0.16 L/ha (1x 76.8 g a.s./ha), BBCH 40-49			
SAP09I – <i>Folsomia candida</i>	0.35	0.061 ²	5.74
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.061 ²	10.9
Pear, wild apple, quince (uses no. 5, 6) – application rate 2 x 0.3 L/ha (2 x 144 g a.s./ha), interval: 10d, BBCH 10-59 / BBCH >70			
SAP09I – <i>Folsomia candida</i>	0.70 0.35	0.133 ¹	5.26 2.63
		0.093	3.76
SAP09I – <i>Hypoaspis aculeifer</i>	1.33 0.665	0.133 ¹	10.0 5.0
Pear, wild apple, quince (uses no. 5, 6) – application rate 2 x 0.225 L/ha (2 x 108 g a.s./ha), interval: 10d → max. orchard 9780 m ² I.W.A., BBCH 10 /40-59 (1 st appl.) / BBCH >70 (2 nd appl.)			
SAP09I – <i>Folsomia candida</i>	0.35	0.069 ²	5.07
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.100 ³ 0.069 ²	6.65 9.64
Pear, wild apple, quince (uses no. 5, 6) – application rate 2 x 0.225 L/ha (2 x 108 g a.s./ha), interval: 10d BBCH >70			
SAP09I – <i>Folsomia candida</i>	0.35	0.067 ²	5.22
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.067 ²	9.9
Pear, wild apple, quince (uses no. 5, 6) – application rate 2 x 0.225 L/ha (2 x 108 g a.s./ha), interval: 21d BBCH ≥ 40 / BBCH >70			
SAP09I – <i>Folsomia candida</i>	0.35	0.059 ²	5.91
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.093 ²	7.14
Pear, wild apple, quince (uses no. 5a, 6b) – application rate 1 x 0.3 L/ha (1 x 144 g a.s./ha), BBCH >70			
SAP09I – <i>Folsomia candida</i>	0.35	0.067 ¹	5.22
SAP09I – <i>Hypoaspis aculeifer</i>	0.665	0.067 ¹	9.93

TER values shown in bold fall below the relevant trigger.

¹PECs Tier I modelling

²PECs Tier II modelling

9.8.2.2 Higher-tier risk assessment

The use of the derived EC_{10/2} for the *Folsomia* endpoint is proposed by the applicant over the use of the NOEC_{corr} as a Higher-tier.

An EC₁₀ is considered a more robust endpoint than the NOEC as it considers the effects of multiple dose responses rather than simply relying on a single concentration which may or may not lie close to the actual no effect concentration for multiple testing parameters. Additionally, in this specific case, there are eight experimental doses with responses for mortality and reproduction with tight spacing near the derived EC₁₀ endpoint. The fitted model appears to adequately represent a correct approximation of the real dose-response relation, and the NOEC is bracketed within the Coefficient of Variation. In summary,

the EC10 estimate can be considered as a valid science-based endpoint, allowing for a better direct comparison to other ecotoxicological endpoints, and a sufficient level of protection is observed.

This proposed approach is supported by the figure E11, as recommended by the external scientific report¹ and an EFSA technical report². Accounting for all the abovementioned arguments, it is noted EC₁₀ is the preferred estimate for chronic risk assessment according to the CZ Evaluation Manual Ecotoxicology version 3.0 December 2024.

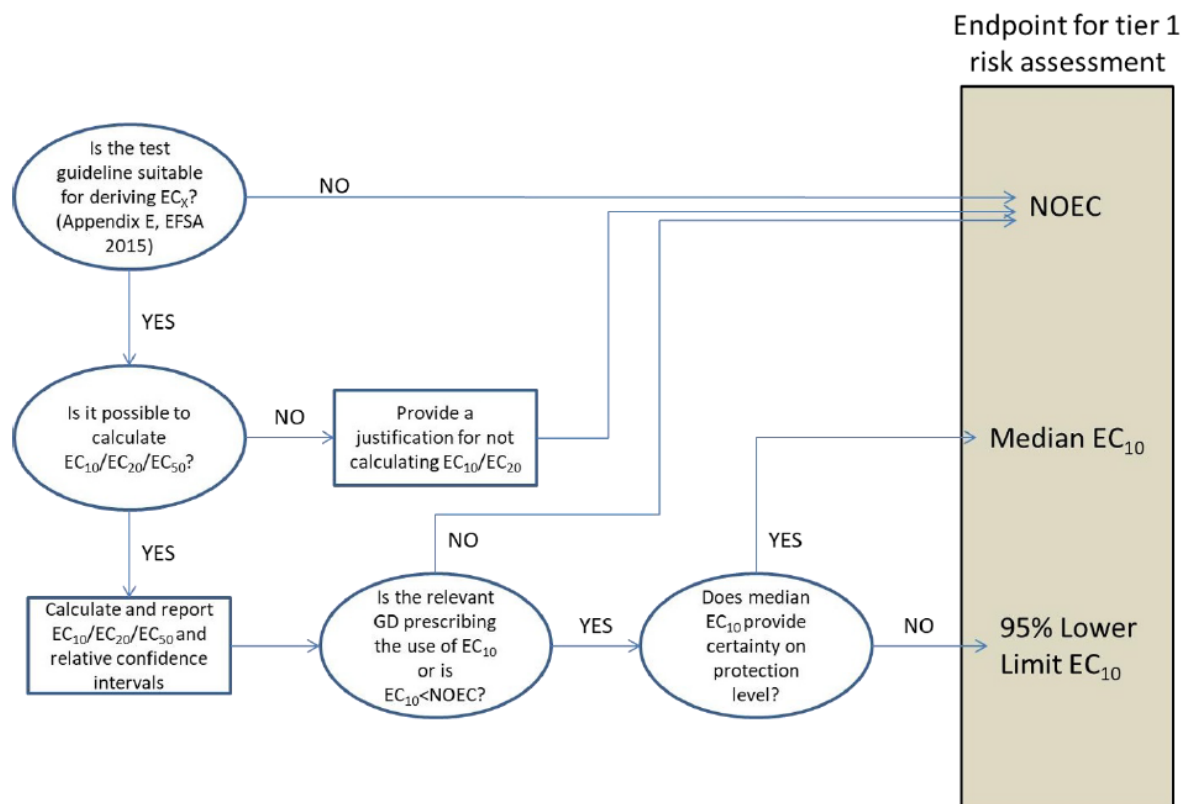


Figure E11: Flowchart for reporting EC_x and using EC₁₀ vs NOEC

¹ Reference 1 EXTERNAL SCIENTIFIC REPORT APPROVED: 26 November 2015 PUBLISHED: 03 December 2015 Comparison of NOEC values to EC10/EC20 values, including confidence intervals, in aquatic and terrestrial ecotoxicological risk assessment

² Reference 2 EFSA Technical Report [APPROVED: 28 June 2019 (doi:10.2903/sp.efsa.2019.EN-1673)] Outcome of the Pesticides Peer Review Meeting on general recurring issues in ecotoxicology

Review Comments:

According to EFSA Supporting publication 2015:EN-924 and EFSA Supporting publication 2019:EN-1673 regarding the use of EC_x in the risk assessment, the experts agreed that the lower between the median EC₁₀ and the NOEC should be used in the TER calculations. Consequently, the applicant's proposal to refine the risk assessment was not accepted by zRMS.

Table 9.8.5: 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 SAP09I in all intended uses

Intended use	all-intended uses		
Acute effects on earthworms—no longer a data requirement			
Chronic effects on other soil macro- and mesofauna			
Product/active substances	EC10 _{corr} (mg/kg dw)	PEC _{soil} (mg/kg dw)	TER _i (criterion-TER > 5)
Cabbage, Broccoli, Cauliflower (uses no. 1, 2)—application rate 3 x 0.2 L/ha (3 x 96 g a.s./ha), interval: 7d, BBCH 10-49			
SAP09I— <i>Folsomia candida</i>	0.39	0.079	5.2
Onion, leek, garlic (uses no. 3, 4, 7)—application rate 1 x 0.15 L/ha (1 x 72 g a.s./ha), BBCH 10-49			
SAP09I— <i>Folsomia candida</i>	0.39	0.086	5.70
Pear, wild apple, quince (uses no. 5, 6)—application rate 2 x 0.3 L/ha (2 x 144 g a.s./ha), interval: 10d, BBCH 10-59 / BBCH >70			
SAP09I— <i>Folsomia candida</i>	0.39	0.003	5.22

TER values shown in bold fall below the relevant trigger.

¹PECs Tier I modelling

²PECs Tier II modelling

9.8.3 Overall conclusions

The risk assessment for soil macro- and mesofauna was conducted according to SANCO/10329/2002 rev 2 final. No unacceptable risk is expected for soil macro- and mesofauna when exposed to SAP09I according to the proposed application patterns.

In accordance with risk assessment for soil organisms acceptable risk is identified when endpoints from Tier I (NOEC_{corr} *Folsomia* 0.35 mg/kg) and additional PECs modelling are considered for:

- Use no. 1, 2 (cabbage, broccoli, cauliflower) application rate 2 x 0.1 L/ha (2 x 48 g a.s./ha), interval: 10d, BBCH 10-49, application rate 2 x 0.16 L/ha (2 x 76.8 g a.s./ha), interval: 10d, BBCH 20-49,
- Uses no. 3, 4, 7 (onion, leek, garlic)—application rate 1 x 0.15-0.16 L/ha (1 x 72-76.8 g a.s./ha), BBCH 40-49
- Uses no. 5, 6 (pear, wild apple, quince) application rate 1 x 0.3 L/ha (1 x 144 g a.s./ha) BBCH >70, application rate 2 x 0.225 L/ha (2 x 108 g a.s./ha) BBCH >40, BBCH >70, interval: 10d,

However additional recalculated risk assessment from Tier 2 has been conducted to demonstrate the safe use on other non-target soil organisms (*Folsomia candida*) due to the use of SAP09I in all intended uses according the refined endpoint EC₁₀ (assumed) as recommended by Outcome of the pesticides peer review

~~meeting on general recurring issues in ecotoxicology – EFSA Supporting publication 2019 and CZ Evaluation Manual Ecotoxicology version 3.0 December 2024 resulting in acceptance for uses according to the proposed applications patterns.~~

~~Use no. 1, 2 (cabbage, broccoli, cauliflower) application rate 3 x 0.2 L/ha (3 x 96 g a.s./ha), interval: 7d, BBCH 10-49~~

~~Uses no. 3, 4, 7 (onion, leek, garlic) application rate 1 x 0.15 L/ha (1 x 72 g a.s./ha), BBCH 10-49~~

~~Uses no. 5, 6 (pear, wild apple, quince) application rate 2 x 0.2 L/ha (2 x 144 g a.s./ha), interval: 10d, BBCH 10-59 / BBCH >70~~

Review Comments:

The chronic TER values for earthworms and *H. aculeifer* exposed to spinosad and its relevant metabolites were greater than the Commission Regulation (EU) No. 546/2011 trigger of 5, indicating that the risk to earthworms and *H. aculeifer* is acceptable following use of SAP09I according to the proposed use pattern.

For *F. candida* the chronic TER value are lower than 5, therefore further assessment is required. The refined PEC_{SOIL} or additional higher tier data should be provided.

Currently, available data are considered as not sufficient and the unacceptable risk assessment for soil organisms is identified.

During the commenting process, the Applicant introduced modifications to the proposed scope of application of SAP09I (Volkete Max). Additional risk assessment was performed. Based on it, the acceptable risk was demonstrated for following uses:

- Use no. 1, 2 (cabbage, broccoli, cauliflower) application rate:

- 3 x 0.1 L/ha (3 x 48 g a.s./ha), interval: 10d, BBCH 10-49
- 2 x 0.16 L/ha (2 x 76.8 g a.s./ha), interval: 10d, BBCH 20-49

- Uses no. 3, 4, 7 (onion, leek, garlic) application rate:

- 1 x 0.15-0.16 L/ha (1 x 72-76.8g a.s./ha), BBCH 40-49

- Uses no. 5, 6 (pear, wild apple, quince) application rate:

- 1 x 0.3 L/ha (1x 144 g a.s./ha) BBCH >70
- 2 x 0.225 L/ha (2 x 108 g a.s./ha) BBCH >70, interval: 10d
- 2 x 0.225 L/ha (2 x 108 g a.s./ha) BBCH 10/40-59 (1st appl.) / BBCH>70 (2nd appl.), interval: 10d

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 spinosad and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on soil microorganisms of SAP09I were not evaluated as part of the EU assessment of spinosad.

However, the provision of further data on the formulation SAP09I is not considered essential, because the studies performed with the active substance and the reference product are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Therefore, SAP09I toxicity towards soil microbial activity can be safely and reliably predicted on the basis of the data already available.

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

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

Endpoint	Substance	Exposure System	Results	Reference
N-mineralisation	Spinosad	70 d, aerobic silt loam and sandy loam	Negative effects on nitrification (tested at 0.72 and 7.2 mg/kg) were maximally -55% (after 14 days at 7.2 mg/kg). At test termination, all effects (positive and negative) were <25% compared to the control.	SANCO/1428/2001 – rev. final (2006)
N-mineralisation	Spinosyn B	-	No unacceptable effects at 0.7164 and 3.582 mg/kg soil	SANCO/1428/2001 – rev. final (2006)
N-mineralisation	N-demethylated spinosyn D	-	No unacceptable effects at 0.3855 and 1.928 mg/kg soil	SANCO/1428/2001 – rev. final (2006)

9.9.1.1 Justification for new endpoints

Not relevant.

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 and were already used in the risk assessment for earthworms and other non-target soil organisms (meso- and macrofauna) (see 9.8).

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for bulb and onion like vegetables (use no. 3) covers the risk for soil micro-organisms from all other intended uses (see 9.1.2).

Table 9.9-2: Assessment of the risk for effects on soil micro-organisms due to the use of SAP09I in bulb and onion like vegetables (use no. 3) – risk envelope approach

Intended use	bulb and onion like vegetables (use no. 3) – risk envelope approach		
N-mineralisation			
Product/active substance	Max. conc. with effects ≤ 25 % (mg/kg dw)	PEC _{soil} (mg/kg dw)	Risk acceptable?
Spinosad	7.2 (at 70 d)	0.237 ¹	Yes
Spinosyn B	3.582	0.187 ²	Yes
N-demethylated spinosyn D	1.928	0.031 ²	Yes

¹PECs Tier I modelling

²PECs_aaccumulation

9.9.3 Overall conclusions

The risk assessment for microorganisms was conducted according to SANCO/10329/2002 rev 2 final. An acceptable risk is proven for microorganisms when exposed to the active substance and its metabolites and consequently to the formulated product according to the proposed application patterns.

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 spinosad. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on non-target terrestrial plants of SAP09I were not evaluated as part of the EU assessment of spinosad.

However, the provision of further data on the formulation SAP09I is not considered essential, because the studies performed with the active substance and the reference product are already unprotected and SAP09I is ecotoxicologically similar to the reference product. Therefore, SAP09I toxicity towards non-target terrestrial plants can be safely and reliably predicted on the basis of the data already available.

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

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

Species	Substance	Exposure System	Results	Reference
<i>Daucus carota</i> _d <i>Cucumis sativus</i> _d <i>Raphanus sativus</i> _d <i>Glycine max</i> _d <i>Helianthus annuus</i> _d <i>Lycopersicon esculentum</i> _d <i>Zea mays</i> _m <i>Avena sativa</i> _m	XDE-105 (88% spinosyn A and D)	Seed germination, seedling emergence and vegetative vigour	¹) ER ₅₀ emergence > 200 g spinosyn A and D/ha ²) ER ₅₀ plant weight > 200 g spinosyn A and D/ha ³) ER ₅₀ plant height > 200 g spinosyn A and D/ha	Schwab (1994) in DAR of Spinosad (2001)

Species	Substance	Exposure System	Results	Reference
<i>Allium cepa</i> _m <i>Triticum aestivum</i> _m				
<i>Daucus carota</i> _d <i>Zea mays</i> _m <i>Cucumis sativus</i> _d <i>Avena sativa</i> _m <i>Allium cepa</i> _m <i>Raphanus sativus</i> _d <i>Glycine max</i> _d <i>Helianthus annuus</i> _d <i>Lycopersicon esculentum</i> _d <i>Triticum aestivum</i> _m	NAF-85	Seedling emergence and vegetative vigour	¹⁾ ER ₅₀ emergence > 579 g a.s./ha ²⁾ ER ₅₀ plant weight > 579 g a.s./ha ³⁾ ER ₅₀ plant height > 579 g a.s./ha	McCormick and Schwab (1997) in DAR of Spinosad (2001) (IIIA 10.8/01, MJ33)

m: monocotyledonous; d: dicotyledonous

9.10.1.1 Justification for new endpoints

Not relevant.

9.10.2 Risk assessment

9.10.2.1 Tier-1 risk assessment (based screening data)

Since SAP09I is an insecticide (i.e. not an herbicide), the data available for the reference product will be used in the tier 1 risk assessment.

To achieve a concise risk assessment, the risk envelope approach is applied. Here, the assessment for leafy vegetables (use no. 1) covers the risk for non-target terrestrial plants from all intended uses (see 9.1.2).

Tests at rates up to 579 g a.s./ha were conducted with the reference formulation and effects were below the critical threshold as defined by the “Guidance Document on Terrestrial Ecotoxicology”, (SANCO/10329/2002 rev.2 final, 2002). The test rates exceed the highest field application rate 3 x 96 g a.s./ha and are thus considered an indicator for an acceptable risk.

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

Not relevant.

9.10.2.3 Higher-tier risk assessment

Not relevant.

9.10.2.4 Risk mitigation measures

No risk mitigation needed.

9.10.3 Overall conclusions

The risk assessment for non-target terrestrial plants was conducted according to SANCO/10329/2002 rev 2 final. Since spinosad is an insecticide, a tier 1 risk assessment was conducted, and an acceptable risk is proven when SAP09I is applied at the proposed application rates.

Review Comments:

According to the risk assessment for the intended uses, SAP09I does not pose any unacceptable risk for non-target plants. No mitigation measures required.

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

No available data.

9.12 Monitoring data (KCP 10.8)

No available data.

9.13 Classification and Labelling

For both SAP09I and NAF-85, experimentally data was derived for the algae *Navicula pelliculosa* as presented above.

From the data available for NAF-85 presented in the Spinosad Review Report (2006), *N. pelliculosa* is the most sensitive aquatic species to spinosad.

Table 9.13-1: Endpoints and effect values relevant for the risk assessment for aquatic organisms – NAF-85

Species	Substance	Exposure System	Results	Reference
<i>Cyprinus carpio</i>	NAF-85	96h	LC ₅₀ > 49 mg/L	Spinosad Review Report (2006)
<i>Daphnia magna</i>	NAF-85	48h	EC ₅₀ = 9.1 mg/L	Spinosad Review Report (2006)
<i>Selenastrum capricornutum</i>	NAF-85	120h	EC ₅₀ > 48 mg/L	Spinosad Review Report (2006)
<i>Navicula pelliculosa</i>	NAF-85	120h	EC ₅₀ = 0.35 mg/L	Spinosad Review Report (2006)

This means that the ecotoxicological classification of NAF-85 is driven by the *Navicula pelliculosa* endpoint. Since for SAP09I a similar endpoint was obtained for this organism, then attributing the same classification to both formulated products is a safe and reliable procedure.

Looking at the results obtained above (endpoints higher than 1 mg/L for fish and invertebrates but lower than 1 mg/L for algae), NAF-85 is ecotoxicologically classified as Aquatic Acute Category 1 (H400:

Very toxic to aquatic life) and ~~Aquatic Chronic Category 1 (H410: Very toxic to aquatic life with long lasting effects)~~. The same classification should therefore be applied to SAP09I.

Review Comments:

For chronic classification, the summation method in accordance with EU Regulation 1272/2008 (CLP labelling) was applied. Since the content of spinosad classified as category 1 for chronic toxicity (H410) is above the limit of 25% (being 45% w/w), the product SAP09I should be classified as category 1 for chronic aquatic toxicity.

Conclusion: SAP09I should be classified for ecotoxicity to aquatic organisms as “Aquatic Acute 1 and Chronic 1” with the Hazard Statements “H400 – Very toxic to aquatic life” and “H410 – Very toxic to aquatic life with long lasting effects”.

Classification and labelling in accordance with Regulation (EC) No 1272/2008

Hazard class(es), categories:	Aquatic Acute 1 Aquatic Chronic 1
Hazard pictograms:	 GHS09
Signal word:	Warning
Hazard statement(s):	H400 Very toxic to aquatic life. H410 Very toxic to aquatic life with long lasting effects.
Precautionary statement(s):	P273 Avoid release to the environment. P391 Collect spillage.
Additional labelling phrases:	To avoid risks to man and the environment, comply with the instructions for use. [EUH401]

Appendix 1 Lists of data considered in support of the evaluation

Tables considered not relevant can be deleted as appropriate.

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

List of data submitted by the applicant and relied on

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 10.2.1/01	Schuler, L.	2021	Spinosad 480 g/L SC: Toxicity to the Diatom <i>Navicula pelliculosa</i> under Laboratory Conditions Eurofins Report No S21-04361 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA
KCP 10.3.1.2/01	Aguilar-Alberola, J.	2021	Spinosad 480 g/L SC (SAP09I): Honey Bee (<i>Apis mellifera</i> L.) Chronic Oral Toxicity Test (10-Day Feeding) under Laboratory Conditions Eurofins Report No S21-04335 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA
KCP 10.3.1.3/01	Aguilar-Alberola, J.	2021	Spinosad 480 g/L SC (SAP09I): Honey Bee (<i>Apis mellifera</i> L.) Larval Toxicity Test following Repeated Exposure under laboratory conditions Eurofins Report No S21-04336 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA
KCP 10.3.2.1/01	Varela, S.	2022	Spinosad 480 g/L SC: Toxicity to the Predatory Mite, <i>Typhlodromus pyri</i> Scheuten (Acari, Phytoseiidae) under Standard Laboratory Conditions Eurofins Report No S21-04370 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA

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 10.3.2.4/01	Roig, J.	2022	A field trial to determine the effects of Spinosad 480SC on the non-target arthropod fauna of a meadow after one application in Southern Europe Zone (Fnace) in 2021 Eurofins Report No S20-09667 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA
KCP 10.4.2.1/01	Antón, B.	2021	Spinosad 480 g/L, SC: Effects on the Reproductive Output of the Springtail <i>Folsomia candida</i> Willem (Collembola, Isotomidae) in Artificial Soil Eurofins Report No S21-04368 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA
KCP 10.4.2.1/02	Lozano, J.	2021	Spinosad 480 g/L SC: Effects on the Reproductive Output of the Predatory Soil Mite <i>Hypoaspis (Geolaelaps) aculeifer</i> Canestrini (Acari: Laelapidae) in Artificial Soil Eurofins Report No S21-04369 GLP Unpublished	N	Albaugh Europe Sàrl Ascenza Agro SA Lainco SA

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

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
Higher tier risk assessment for mammals	xxxxxxx	1994	XDE-105: Two Generation Dietary Reproduction Study in Sprague-Dawley Rats Report No.: DR-0323-1194-008 GLP Unpublished	Y	Out of data protection
	xxxxxxx	1993	XDE-105: Oral Gavage Teratology Study in Sprague-Dawley Rats Report No.: DR-0323- 1194-003	Y	Out of data protection

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP Unpublished		
	xxxxxxx	1994	XDE-105: Oral Gavage Teratology Study in New Zealand White Rabbits Report No.: DECO-HET DR-0323-1194-011 GLP Unpublished	Y	Out of data protection
	McCormick	2005	Estimation of HalfLife in Decline of Spinosad Residues in Pasture Grasses, EU studies 2004 Dow AgroSciences, LLC, Report No.: GH-C-5827 GLP Unpublished	N	Out of data protection
Non-target terrestrial plants risk assessment	McCormick, Schwab	1997	Evaluating the Effects of Spinosad (XDE-105) on the Emergence and Vegetative Vigor of NonTarget Terrestrial Plants Report No.: RES96153, A43507, ABC Laboratories Inc., Columbia, Missouri, USA, GLP Unpublished	N	Out of data protection
	Schwab	1994	Evaluating the Effects of XDE-105 on the Germination, Emergence and Vegetative Vigor of Non-Target Terrestrial Plants Report No.: RES94062, ABC Laboratories Inc., Columbia, Missouri, USA, GLP Unpublished	N	Out of data protection
Bees risk assessment	Aldershof, S.	1999	Determination of the acute contact LD50 of spinosad (formulated as the 480 g/L SC, NAF-85) for the bumble bee Bombus Terrestris L Mitox Stichting Bevordering Duurzame Plaagbestrijding, Report No. GHE-P-7874R. GLP Not published	N	Out of data protection
	Aldershof, S.	1999	Determination of the acute oral LD50 of spinosad (formulated as the 480 g/L SC, NAF-85) for the bumble bee Bombus Terrestris L Mitox Stichting Bevordering Duurzame Plaagbestrijding, Report No. GHE-P-7875.	N	Out of data protection

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP Not published		
	Aldershof, S.	1999	Effect of spinosad (formulated as 480 g/L SC, NAF-85) on the bumblebee <i>Bombus terrestris</i> determined under greenhouse conditions GLP Not published	N	Out of data protection
	Goodwin, R.M.	1998	Effect of applying Spinosad to staminate kiwifruit flowers on the survival of foraging honey bees The horticulture & food Research Institute of New Zealand Ltd. GLP Not published	N	Out of data protection

The following tables are to be completed by MS

List of data submitted by the applicant and not relied on

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner

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

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner

Appendix 2 Detailed evaluation of the new studies

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

A 2.1.1.2 KCP 10.1.1.2 Higher tier data on birds

A 2.1.2 KCP 10.1.2 Effects on terrestrial vertebrates other than birds

A 2.1.2.1 KCP 10.1.2.1 Acute oral toxicity to mammals

A 2.1.2.2 KCP 10.1.2.2 Higher tier data on mammals

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

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

A 2.2.1.1.1 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 201 and according to the principles of GLP. No deviations to the guideline were noted. All validity criteria were met. The study is considered to be reliable and suitable for the risk assessment.
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Reference: KCP 10.2.1/01

Report Spinosad 480 g/L SC: Toxicity to the Diatom *Navicula pelliculosa* under Laboratory Conditions, Schuler, L., 2021, Report No S21-04361

Guideline(s): OECD 201 (2006, corrected 2011).

Deviations: No

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) N/A

Objective

The objective of this study was to determine the effects of the test item on the growth of the diatom *Navicula pelliculosa*, to determine the no observed effect concentration (NOEC), to determine the lowest observed effect concentration (LOEC) and to determine the effect concentration (EC_{10, 20, 50}), where possible.

Materials and methods

Test item:	Spinosad 480 g/L SC, batch number: D-CEA,, batch number: D-CEA, active ingredient (a.i.): Spinosad, content of a.i. (analysed): 460 g/L.
Test species:	<i>Navicula pelliculosa</i> .
Test design:	Initial target cell densities of 1.0×10^4 cells/mL were employed for the individual replicates. The increase of cell numbers was assessed over a test period of 72 hours.
Endpoints:	Where possible inhibition of growth was assessed by the determination of NOEC/LOEC and EC _{10, 20, 50} for growth rate and yield after 72 hours.
Test rates:	A static test with nominal test item concentrations of 1.00, 0.455, 0.207, 0.0939, 0.0427 mg/L and control was performed.
Test conditions:	Six replicates were employed for the control and three for each test item concentration. The test was performed in 100 mL Erlenmeyer flasks each containing ~ 50 mL test solution. The pH was recorded at test start and test end. Temperature was measured continuously over the whole test period and recorded daily. Light intensity of the continuous illumination was measured at test start.
Analysis:	Analytical samples were taken and analysed from control and all test item concentrations at 0 hours (initial value) from fresh test solutions and after 72 hours from aged test solutions.
Statistics:	The NOEC and LOEC were determined by using a multiple comparison method (Williams multiple sequential t-test, left sided). The EC _{10, 20, 50} -values for growth rate and yield were determined by logit analysis using linear maximum likelihood regression and probit analysis using linear maximum likelihood regression, respectively.
Dates of work:	16 Aug 2021 – 27 Aug 2021

Results and discussions

Validity criteria:	Biomass: Cell numbers, measured in the controls between 0 and 72 hours, were found to increase by a factor of 58.23 (corresponding to a specific growth rate of 1.35228 d^{-1}), which exceeds the threshold of 16 (corresponding to a specific growth rate of 0.92 d^{-1}). Coefficient of variation (section by section): The mean coefficient of variation for the section-by-section specific growth rates (hours 0 - 24, 24 - 48 and 48 - 72) in the control cultures was 28 % and did not exceed 35 %.
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Coefficient of variation (average growth rate): The coefficient of variation of average growth rate in replicate control cultures was 2.9 % and did not exceed 7 % for the whole test period.

Test conditions: The pH-value of the control ranged from 7.49 to 8.78 during the test period, the temperature was measured to be 22.6 – 23.8 °C during the test and the mean light intensity was 94.7 $\mu\text{Em}^{-2}\text{s}^{-1}$ at cell culture level.

Analytical Results: The measured initial concentrations of Spinosad ranged from 85 % to 105 % of nominal. In the aged samples the measured concentrations were between 26 % and 73 % of nominal. Therefore, toxicological endpoints were evaluated using the nominal and the mean measured concentrations (based on the geometric mean of the analytical recoveries for each concentration level) of the test item.

Statistical Results: Toxicological endpoints for the test item

	Test item [mg/L]	
	nominal	mean measured ⁴⁾
E_rC_{10} (Growth rate) ¹⁾	0.630	0.490
95 % confidential limits	0.511 – 0.702	0.378 – 0.563
E_rC_{20} ¹⁾	0.691	0.551
95 % confidential limits	0.585 – 0.754	0.447 – 0.616
E_rC_{50} ¹⁾	0.810	0.674
95 % confidential limits	0.737 – 0.852	0.598 – 0.718
E_yC_{10} (Yield) ²⁾	0.513	0.379
95 % confidential limits	0.480 – 0.546	0.348 – 0.410
E_yC_{20} ²⁾	0.558	0.421
95 % confidential limits	0.524 – 0.595	0.389 – 0.457
E_yC_{50} ²⁾	0.655	0.515
95 % confidential limits	0.612 – 0.711	0.473 – 0.572
NOEC ³⁾	0.455	0.326
LOEC ³⁾	1.00	0.878

¹⁾ Logit analysis using linear maximum likelihood regression

²⁾ Probit analysis using linear maximum likelihood regression

³⁾ Following Williams multiple sequential t-test (left-sided, $p \leq 0.05$) for growth rate and for yield

⁴⁾ Based on geometric mean of the analytical recoveries for each concentration level

Conclusion

Significant inhibitory effects were determined for growth rate and yield at test item concentrations of 1.00 mg/L (nominal) and 0.878 mg/L (mean measured). The overall LOEC was therefore determined to be

1.00 mg/L (nominal) and 0.878 mg/L (mean measured), the corresponding NOEC was set at 0.455 mg/L (nominal) and 0.326 mg/L (mean measured).

The EC₁₀-value for growth rate (E_rC₁₀) was determined to be 0.630 mg/L (nominal) and 0.490 mg/L (mean measured). The EC₁₀-value for yield (E_yC₁₀) was 0.513 mg/L (nominal) and 0.379 mg/L (mean measured).

The EC₂₀-value for growth rate (E_rC₂₀) was determined to be 0.691 mg/L (nominal) and 0.551 mg/L (mean measured). The EC₂₀-value for yield (E_yC₂₀) was 0.558 mg/L (nominal) and 0.421 mg/L (mean measured).

The EC₅₀-value for growth rate (E_rC₅₀) was determined to be 0.810 mg/L (nominal) and 0.674 mg/L (mean measured). The EC₅₀-value for yield (E_yC₅₀) was 0.655 mg/L (nominal) and 0.515 mg/L (mean measured).

Validity criteria

Biomass	Cell numbers, measured in the controls between 0 and 72 hours, were found to increase by a factor of 58.23 (corresponding to a specific growth rate of 1.35228 d ⁻¹), which exceeds the threshold of 16 (corresponding to a specific growth rate of 0.92 d ⁻¹).
Coefficient of variation (section by section)	The mean coefficient of variation for the section-by-section specific growth rates (hours 0 - 24, 24 - 48 and 48 - 72) in the control cultures was 28 % and did not exceed 35 %.
Coefficient of variation (average growth rate)	The coefficient of variation of average growth rate in replicate control cultures was 2.9 % and did not exceed 10 % for the whole test period.

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

A 2.2.3 KCP 10.2.3 Further testing on aquatic organisms

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

A 2.3.1.1.2 KCP 10.3.1.1.2 Acute contact toxicity to bees

A 2.3.1.2 KCP 10.3.1.2 Chronic toxicity to bees

A 2.3.1.2.1 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 245 and according to the principles of GLP. No significant deviations to the guideline were noted. All validity criteria were met. The study is considered to be reliable and suitable for the risk assessment.
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Reference:	KCP 10.3.1.2/01
Report	Spinosad 480 g/L SC (SAP09I): Honey Bee (<i>Apis mellifera</i> L.) Chronic Oral Toxicity Test (10-Day Feeding) under Laboratory Conditions, Aguilar-Alberola, J.A., 2021, Report No S21-04335
Guideline(s):	OECD 245 (2017) and SANTE/2020/12830, Rev.1 (2021).
Deviations:	Behavioural abnormalities in the reference item treatment were not recorded since the reference item is known to be toxic to honeybees and therefore effects are expected. Moreover, the validity criterion for reference item was met.
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	N/A

Objective

To determine the effects of Spinosad 480 g/L SC (SAP09I) on the honey bee (*Apis mellifera* L.) from chronic feeding exposure. To determine the No Observed Effect Concentration (NOEC), the No Observed Effect Dietary Dose (NOEDD), the median Lethal Concentration (LC₅₀), the median Lethal Dietary Dose (LDD₅₀) and any LC_x/LDD_x (i.e.: LC₁₀/LDD₁₀ and LC₂₀/LDD₂₀) values on exposure at day 10, where possible.

Materials and methods

Test item: Spinosad 480 g/L SC (SAP09I); batch code: D-CEA; active ingredient: spinosad; content of a.i.: 460 g/L (content of active ingredient according to certificate of analysis); density: 1.08 g/mL; expiry date: March 2023.

Reference item: BAS 152 65 I; batch code: 10248664A; active ingredient: dimethoate; content of a.i. analysed: 40.9 % w/v; density: 1.062 g/mL; expiry date: 16 Feb 2022.

Test organisms: Honey bee (*Apis mellifera* L.), young adult worker bees not older than 48 hours.

Source: Commercial beehives from the in-house Test Facility stock, adequately fed, healthy and as far as possible disease-free and queen-right. The hives from which the test organisms were obtained were not previously exposed to any chemical treatments within four weeks of test initiation.

Preparation of test organisms: Two days before the beginning of the test, brood frames containing capped cells from which adults were expected to emerge within 24 hours were taken from the honey bee hives to an incubator, transported to Eurofins Trialcamp S.L.U. facilities and put in a bioclimatic chamber. One day prior to test start, the bees were randomly collected directly from the frames, introduced into test units and kept under test conditions until the start of the test. Acclimatisation period comprised from bee collection to the start of the test. During this period, bees were fed ad libitum with a 50 % (w/v) aqueous sucrose solution. Dead and moribund bees were rejected and replaced by healthy bees before starting the test.

Test design: Dose-response test with an exposure phase duration of 10 days. One control group was used. Five different concentrations of SAP09I 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 reference item group. Five replicates of 10 bees each were used for each group. Mortality and behavioural abnormalities were assessed daily over the 10-day test period.

Test concentrations: Control: C: Control group (untreated feeding solution).

Test item: 0.0601, 0.1503, 0.3757, 0.9391 and 2.3478 mg test item/kg feeding solution.

Reference item: R: 0.90 mg dimethoate/kg feeding solution.

Test conditions:

Air Temperature: 31.9 – 33.7 °C

Relative humidity: 53.4 – 63.3 %

Exposure to light: constant darkness except during feeding and assessments.

Analytical verification: Samples of the control, the stock solution and the highest and lowest concentration of the test item treatments were taken on D9, directly after preparation, and placed in the freezer at < -18 °C until shipment. Analytical Phase was performed to verify the concentration of the samples taken. A method was validated and samples of feeding solution were analysed for concentration determination of spinosad. Quantification was performed by liquid chromatography with tandem mass spectrometry (LC-MS/MS). The limit of quantification (LOQ) of the analytical method was 0.00101 mg spinosad/kg (0.00236 mg test item/kg) with a limit of detection (LOD) set at 0.000240 mg spinosad/kg.

Statistics: In order to determine the NOEC/NOEDD, the step-down Cochran-Armitage test procedure (one-sided greater, $\alpha = 0.05$) was used. The $LC_{10,20}/LDD_{10,20}$ values with 95 % confidence limits were calculated by using the Weibull analysis using linear maximum likelihood regression. Due to the wide range between the confidence limits of the $LDD_{10,20}$ values, the calculated values were considered non valid and they were empirically estimated from the results. The LDD_{50}/LC_{50} values were empirically estimated from the results since no greater response than 50 % resulted in any test item group. Statistical evaluation was made using Microsoft Office Excel® v.16.0 and the statistical software ToxRatPro® v. 3.3.0.

Dates of work: 30 Jun 2021 – 02 Aug 2021.

Results and discussions

In the feeding solution of the control group no spinosad was detectable. The measured concentration in the stock solution and the test item treated samples was higher than 120 % of nominal test concentration used (141 – 156 %), but it was decided not to recalculate (increase) the endpoints accordingly and report them based on nominal concentrations as worst case scenario.

After 10 days of continuous exposure, the cumulative mortality was 8.00 % for the control group. The mortality in the reference item group was 100.00 % for the same period.

Therefore, the validity criteria for the study were met: the control mortality was ≤ 15 % and the reference item mortality was ≥ 50 %, across all replicates.

At the concentrations of 0.0601, 0.1503, 0.3757, 0.9391 and 2.3478 mg test item/kg feeding solution, cumulative mortalities of 6.00, 10.00, 6.00, 16.00 and 50.00 % (-2.17, 2.17, -2.17, 8.70 and 45.65 % corrected for the control) were observed, respectively, at the final assessment after 10 days of exposure.

Symptoms of intoxication (affected bees) were observed sporadically for a few bees, mainly in the test item groups with the highest concentrations. At the end of the test, in the final assessment on day 10 (D10), 2, 1 and 3 affected bees were observed in treatments T2, T4 and T5, respectively. Also, one moribund bee was observed in treatment T4. No affected bees were observed throughout the test in the control group.

Main results are shown in the table below.

Concentration (treatment group) [mg t.i./kg f.s.]	10 day cumulative mortality [%]	Corrected mortality ^a [%]	Overall mean consumption of f.s. [mg/bee/day]	Actual dietary dose [µg t.i./bee/day]	Cumulative doses [µg t.i./bee]
-- (C)	8.00	--	19.1	--	--
0.0601 (T1)	6.00	-2.17	16.4	0.00098	0.00983
0.1503 (T2)	10.00	2.17	16.7	0.00251	0.02510
0.3757 (T3)	6.00	-2.17	16.7	0.00626	0.06265
0.9391 (T4)	16.00	8.70	15.9	0.01492	0.14918
2.3478 (T5)	50.00 ^b	45.65	14.9	0.03503	0.35028
0.90 (R) ^d	100.00	100.00	15.2	0.014	0.093 ^c

t.i.: test item; f.s.: feeding solution.

^a Corrected for control according to Abbott's formula (1925) modified by Schneider-Orelli (1947).

^b Significantly increased compared to the Control group step-down Cochran-Armitage test procedure, one sided greater, $\alpha = 0.05$).

^c Over 8 days of exposure.

^d For the reference item, the concentration/dose values are expressed in active ingredient (dimethoate).

Conclusion

The chronic toxicity of the test item Spinosad 480 g/L SC (SAP09I) to honey bees was tested under laboratory conditions over a 10-day period.

All validity criteria were met and sensitivity of the test organisms was confirmed. Accordingly, the study was deemed valid.

The measured concentrations of the test item solutions were higher than nominal (141 – 156 %), but it was decided not to recalculate (increase) the endpoints accordingly and report them based on nominal concentrations as worst case scenario.

The No Observed Effect Concentration (NOEC) for mortality after 10 days of continuous exposure was determined to be 0.9391 mg test item/kg feeding solution (corresponding to 0.4000 mg active ingredient/kg feeding solution according to the certificate of analysis). The corresponding No Observed Effect Dietary Dose (NOEDD), based on the actual consumption of the feeding solutions, was determined to be 0.01492 µg test item/bee/day (corresponding to 0.00635 µg active ingredient/bee/day according to the certificate of analysis).

The 10-day LC₅₀ value (Lethal Concentration that kills 50 % of exposed individuals) was empirically determined to be higher than 2.3478 mg test item/kg feeding solution (corresponding to 1.0000 mg active ingredient/kg feeding solution according to the certificate of analysis). The corresponding 10-day LDD₅₀ value (Lethal Dietary Dose that kills 50 % of exposed individuals), based on the actual consumption of the feeding solutions, was determined to be higher than 0.03503 µg test item/bee/day (corresponding to 0.01492 µg active ingredient/bee/day according to the certificate of analysis).

The 10-day LC₁₀ value with 95 % confidence limits was determined to be 0.9362 [0.5738 – 1.2084] mg test item/kg feeding solution (corresponding to 0.3988 [0.2444 – 0.5147] mg active ingredient/kg feeding solution according to the certificate of analysis). The 10-day LC₂₀ value with 95 % confidence limits was determined to be 1.3984 [1.0415 – 1.7090] mg test item/kg feeding solution (corresponding to 0.5956 [0.4436 – 0.7279] mg active ingredient/kg feeding solution according to the certificate of analysis). The corresponding 10-day LDD_{10,20} values based on the actual consumption of the feeding solutions, were empirically determined to be between 0.01492 and 0.03503 µg test item/bee/day (corresponding to 0.00635 and 0.01492 µg active ingredient/bee/day according to the certificate of analysis).

At the end of the test, in the final assessment on day 10 (D10), 2, 1 and 3 affected bees were observed in treatments T2, T4 and T5, respectively. Also, one moribund bee was observed in treatment T4.

Main endpoints of the study are shown in the following table.

Concentration [mg/kg feeding solution]		
Endpoints (D10)	Test item	active ingredient: spinosad
NOEC a	0.9391	0.4000
LC ₁₀ b	0.9362	0.3988
[95 % confidence limits]	[0.5738 – 1.2084]	[0.2444 – 0.5147]
LC ₂₀ b	1.3984	0.5956
[95 % confidence limits]	[1.0415 – 1.7090]	[0.4436 – 0.7279]
LC ₅₀ c	> 2.3478	> 1.0000
Dose [µg/bee/day]		
Endpoints (D10)	Test item	active ingredient: spinosad
NOEDD a	0.01492	0.00635
LDD _{10,20} c	0.01492 < LDD _{10,20} < 0.03503	0.00635 < LDD _{10,20} < 0.01492
LDD ₅₀ c	> 0.03503	> 0.01492

a step-down Cochran-Armitage test procedure (one sided greater, $\alpha = 0.05$).

b Weibull analysis using linear maximum likelihood regression.

c empirically estimated from the results.

Validity criteria

The study was considered valid since validity criteria for both control and reference item groups mortality were met.

Control Mean mortality at the end of the test was ≤ 15 % (actual mean value 8.00 %).

Reference Mean mortality was ≥ 50 % at the end of the test (actual mean value 100.00 %).

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

A 2.3.1.3.1 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 245 and according to the principles of GLP. No significant deviations to the guideline were noted. All validity criteria were met. The study is considered to be reliable and suitable for the risk assessment.
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Reference:	KCP 10.3.1.3/01
Report	Spinosad 480 g/L SC (SAP09I): Honey Bee (<i>Apis mellifera</i> L.) Larval Toxicity Test following Repeated Exposure under laboratory conditions, Aguilar-Alberola, J. A., 2021, Report No S21-04336
Guideline(s):	OECD 239 (2021) and SANTE/2020/12830, Rev.1 (2021).
Deviations:	The reduction of the relative humidity conditions from $95 \pm 5\%$ to $80 \pm 5\%$ was done on day 7 (D7) of the test instead of on day 8 (D8). The reported deviation to the guidance has no impact on the outcome of the study since validity criteria for the control were met.
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	N/A

Objective

To determine the effects of Spinosad 480 g/L SC on honey bee (*Apis mellifera* L.) larvae, after a repeated exposure test. To determine the No Observed Effect Concentration/Dose (NOEC/NOED) and the Lowest Observed Effect Concentration/Dose (LOEC/LOED) for adult emergence (from D3 to D22). To determine the Median Effect Concentration/Dose (EC₅₀/ED₅₀) and any EC_x/ED_x (i.e., EC₁₀/ED₁₀ and EC₂₀/ED₂₀) for adult emergence (from D3 to D22), where possible.

Materials and methods

Test item:	Spinosad 480 g/L SC (SAP09I); Batch code: D-CEA; active ingredient: spinosad; content of a.i.: 460 g/L (content of active ingredient according to certificate of analysis); density: 1.08 g/mL; expiry date: March 2023.
Reference item:	BAS 152 I; Batch code: COD-002332; active ingredient: dimethoate; content of a.i. analysed: 98.2 % w/w (content of active ingredient according to certificate of analysis); expiry date: 23 Jan 2022.
Test organisms:	Honey bee (<i>Apis mellifera</i> L.), synchronized first instar (L1) larvae not older than 30 hours at grafting time.
Source:	Commercial beehives from the in-house Test Facility stock, adequately fed, healthy and as far as possible disease-free and queen-right. The hives from which the larvae were obtained were not previously exposed to any chemical treatments within four weeks of test initiation.
Preparation of test organisms and larvae collection:	On D-3, the queens from at least three colonies was isolated for one day within a queen excluder placed on a single frame with empty cells in their own hive, to provide known-aged eggs and subsequent larvae. On D-2, maximum 30 hours after isolation, the queens were released. Frames containing eggs were left in the excluder cages until hatching (D1). Three frames from

	different hives, containing the highest number of synchronized larvae, were selected for grafting in the laboratory.
Test design:	Dose response test with duration of 21 days from grafting on day 1 to the final assessment on day 22. From day 3 until day 6 of the test, 5 different concentrations of Spinosad 480 g/L SC were applied to the larvae of the test item groups and one single concentration of the reference item was applied to the larvae of the reference item group. Both, test and reference item, were supplied in diet B (day 3) and C (days 4, 5 and 6). The daily feeding volume increased from 20 µL to 50 µL diet per larva over the application period. The cumulative feeding volume from day 3 until day 6 of 140 µL diet per larva was considered for the calculation of the cumulative doses per larva. One control group was included in the test and exposed for the same period of time under identical exposure conditions to the treatments. Each treatment group consisted of 48 larvae; 16 from each of three different colonies (each colony representing one replicate). Larval mortality assessments were on days 4, 5, 6, 7, and 8. The presence of uneaten food was qualitatively recorded on day 8. Assessment of mortality during pupation phase was on day 15 and assessment of emergence on day 22 was recorded.
Test concentrations and doses:	Controls: C: Control group (untreated diet). Test item: 0.0215, 0.0464, 0.1042, 0.2370 and 0.5255 mg test item/L diet (equivalent to 0.0195, 0.0422, 0.0948, 0.2155 and 0.4778 mg test item/kg diet and 0.0030, 0.0065, 0.0146, 0.0332 and 0.0736 µg test item/larva). Values corrected according to analytical recoveries. Reference item: R: 48.00 mg dimethoate/kg diet (equivalent to 7.39 µg dimethoate/larva).
Test conditions:	Air Temperature: 33.5* – 34.9 °C Relative humidity: 43.4* – 98.3 % *Short term deviation.
Analytical verification:	Exposure to light: Constant darkness except during feeding and assessments. Samples of all the concentrations of the test item treated larval diets were taken from D3 to D6, directly after preparation, and placed in the freezer at < -18 °C until shipment. Analytical Phase was performed to verify the concentration of the samples taken. A method was validated and samples of diet were analysed for concentration determination of spinosad. Quantification was performed by liquid chromatography with tandem mass spectrometry (LC-MS/MS). The limit of quantification (LOQ) of the analytical method was 0.00101 mg spinosad/kg with a limit of detection (LOD) set at 0.000300 mg spinosad/kg.
Statistics:	In order to determine the NOEC, the step-down Cochran-Armitage test procedure (one-sided greater, $\alpha = 0.05$) was used. A probit analysis using linear maximum likelihood regression was used to determine the EC_{x}/ED_{x} values. Statistical calculations were made with MS Excel 2016 v.16.0 and the statistical program ToxRatPro® Version 3.3.0.
Dates of work:	17 May 2021 – 12 Nov 2021.

Results and discussions

The mean measured concentration of spinosad in the diets of the three treatments with the lowest concentration were lower than nominal (mean recovery value of 60, 65 and 73 % of nominal for treatments T1, T2 and T3, respectively). Therefore, the concentration/dose values, as well as endpoints, are shown adjusted for the mean analysed concentration of each treatment, including treatments T4 and T5.

On day 8, the cumulative larval mortality was 2.08 % for the control group. On day 22, the adult emergence rate of the initial grafted larvae was 87.50 % for the control group. Therefore, the validity criteria for the control group were met for both test periods: the D8 mortality was lower than 15 % and the D22 days emergence rate was greater than 70 %, across all replicates. Cumulative mortality in the Reference Item group also met the validity criteria (> 50 % at day 8, actual value 85.42 %).

Mean corrected cumulative larval mortality on day 8 (D8) of the test item treated groups was 0.00, 0.00, 4.26, 21.28 and 68.09 % in 0.0195, 0.0422, 0.0948, 0.2155 and 0.4778 mg test item/kg diet, respectively. Mean corrected pupal mortality on day 15 (D15) of the test item treated groups was -2.33, 2.33, 6.98, 27.91 and 72.09 % in 0.0195, 0.0422, 0.0948, 0.2155 and 0.4778 mg test item/kg diet, respectively. Mean corrected mortality at the end of the test (D22) of the test item treated groups was -2.38, 0.00, 4.76, 26.19 and 88.10 % in 0.0195, 0.0422, 0.0948, 0.2155 and 0.4778 mg test item/kg diet, respectively. On day 8, no individuals were observed with uneaten food or other affections. At the end of the test, in the final assessment of the emergence on day 22, no emerged bees were recorded as being affected (i.e. malformation).

Main results are shown in the table below.

Treatment Group		Cumulative Mortality [%]						
[mg t.i./kg diet] a		D4	D5	D6	D7	D8	D15	D22
C	[-]	0.00	0.00	0.00	0.00	2.08	10.42	12.50
T1	[0.0195]	0.00	2.08	2.08	2.08	2.08	8.33	10.42
T2	[0.0422]	0.00	2.08	2.08	2.08	2.08	12.50	12.50
T3	[0.0948]	2.08	2.08	4.17	4.17	6.25	16.67	16.67
T4	[0.2155]	10.42	12.50	18.75	18.75	22.92	35.42	35.42 b
T5	[0.4778]	35.42	47.92	62.50	66.67	68.75	75.00	89.58 b
R	[48.00] c	33.33	52.08	79.17	83.33	85.42	97.92	100.00

t.i.: test item (Spinosad 480 g/L SC).

a Values adjusted according analytical recoveries of the samples.

b Significantly increased compared to the control group (step-down Cochran-Armitage test procedure, one sided greater, $\alpha = 0.05$).

c mg dimethoate/kg diet.

Treatment Group		Corrected mortality [%] a						
[mg t.i./kg diet] b		D4	D5	D6	D7	D8	D15	D22
T1	[0.0195]	0.00	2.08	2.08	2.08	0.00	-2.33	-2.38
T2	[0.0422]	0.00	2.08	2.08	2.08	0.00	2.33	0.00
T3	[0.0948]	2.08	2.08	4.17	4.17	4.26	6.98	4.76
T4	[0.2155]	10.42	12.50	18.75	18.75	21.28	27.91	26.19
T5	[0.4778]	35.42	47.92	62.50	66.67	68.09	72.09	88.10

t.i.: test item (Spinosad 480 g/L SC).

a Corrected for control according to Abbott's formula (1925) modified by Schneider-Orelli (1947).

b Values adjusted according analytical recoveries of the samples.

Conclusion

The repeated exposure of the test item 'Spinosad 480 g/L SC (SAP09I)' to honey bee (*Apis mellifera* L.) was tested under laboratory conditions over a 21-day period.

All validity criteria were met and sensitivity of the test organisms was confirmed. Accordingly, the study was deemed valid.

The low mean analytical recoveries of some treatments have caused all the concentration / dose values to have been adjusted according to the mean values analyzed in each treatment.

On day 8, no individuals were observed with uneaten food or other affections. At the end of the test, in the final assessment of the emergence on day 22, no emerged bees were recorded as being affected (i.e. malformation).

Main endpoints of the study are shown in the following tables.

Endpoints.

Endpoints a	Concentration		Dose
	[mg t.i./L diet]	[mg t.i./kg diet] b	[µg t.i./larva] c
NOEC/NOED	0.1042	0.0948	0.0146
LOEC/LOED	0.2370	0.2155	0.0332
EC10 / ED10	0.1509	0.1372	0.0211
[95 % c.l.]	[0.1138 – 0.1817]	[0.1035 – 0.1652]	[0.0159 – 0.0254]
EC20 / ED20	0.1912	0.1738	0.0268
[95 % c.l.]	[0.1539 – 0.2234]	[0.1399 – 0.2031]	[0.0216 – 0.0313]
EC50 / ED50	0.3006	0.2733	0.0421
[95 % c.l.]	[0.2604 – 0.3495]	[0.2367 – 0.3177]	[0.0365 – 0.0489]

t.i.: test item (Spinosad 480 g/L SC); c.l.: confidence limits.

^a Values adjusted according analytical recoveries of the samples.

^b Based on the diet density (1.1 g/mL).

^c Based on the cumulative application volume of 140 µL/larva.

Endpoints a	Concentration		Dose
	[mg a.i./L diet]	[mg a.i./kg diet] b	[µg a.i./larva] c
NOEC/NOED	0.0444	0.0404	0.0062
LOEC/LOED	0.1010	0.0918	0.0141
EC ₁₀ / ED ₁₀	0.0643	0.0584	0.0090
[95 % c.l.]	[0.0485 – 0.0774]	[0.0441 – 0.0704]	[0.0068 – 0.0108]
EC ₂₀ / ED ₂₀	0.0814	0.0740	0.0114
[95 % c.l.]	[0.0656 – 0.0952]	[0.0596 – 0.0865]	[0.0092 – 0.0133]
EC ₅₀ / ED ₅₀	0.1280	0.1164	0.0179
[95 % c.l.]	[0.1109 – 0.1489]	[0.1008 – 0.1353]	[0.0155 – 0.0208]

a.i.: active ingredient (spinosad; 460 g/L); c.l.: confidence limits.

^a Values adjusted according analytical recoveries of the samples.

^b Based on the diet density (1.1 g/mL).

^c Based on the cumulative application volume of 140 µL/larva.

Validity criteria

The study was considered valid since validity criteria for both control and reference item groups mortality were met.

Control The cumulative larval mortality from day 3 (D3) until day 8 (D8) was ≤ 15 % across all replicates (actual mean value 2.08 % for the control group).

On day 22 (D22) the adult emergence rate was ≥ 70 % across all replicates (actual mean value 87.50 % for the control group).

Reference The cumulative larval mortality was ≥ 50 % across all replicates on day 8 (D8) (actual mean value 85.42 %).

A 2.3.1.4 KCP 10.3.1.4 Sub-lethal effects

A 2.3.1.5 KCP 10.3.1.5 Cage and tunnel tests

A 2.3.1.6 KCP 10.3.1.6 Field tests with honeybees

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

A 2.3.2.1.1 Study 1

Comments of zRMS:	The study was conducted to the guideline and according to the principles of GLP. No significant deviations to the guideline were noted. All validity criteria were met.
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Reference: KCP 10.3.2.1/01

Report Spinosad 480 g/L SC:
Toxicity to the Predatory Mite, *Typhlodromus pyri* Scheuten (Acari, Phytoseiidae) under Standard Laboratory Conditions, Varela, S., 2022, report No S21-04370

Guideline(s): IOBC/WPRS Guidelines to evaluate side-effects of plant protection products to non-target arthropods (Blümel *et al.*, 2000)
Use of standard toxicity tests with *Typhlodromus pyri* and *Aphidius ropalosiphi* to establish a dose-response relationship (Grimm C. *et al.*, 2001).

Deviations: No

GLP: Yes

Acceptability: Yes

Duplication N/A
(if vertebrate study)

Objective

The objective of the study was to determine the effects of Spinosad 480 g/L SC on mortality and reproduction of the predatory mite *Typhlodromus pyri* Scheuten under standard laboratory conditions (exposure on glass plates); to determine the rate causing 50 % mortality (LR₅₀) and 50 % reduction in reproduction (ER₅₀), and to determine the No Observed Effect Rate (NOER) where possible.

Typhlodromus pyri was selected because it is a recommended sensitive indicator species for testing the side effects of plant protection products on non-target arthropods and it is one of the two standard species required for EU registration.

Materials and methods

Test item: Spinosad 480 g/L SC
Batch No. D-CEA
Content of active ingredients (a.i.) (analysed): Spinosad 460.0 g/L

Reference item: Dimethoate 40 % w/v EC (BAS 152 65 I)
Batch No. 10248664A
Content of a.i. (analysed): Dimethoate: 409 g /L

Test species:	<i>Typhlodromus pyri</i> Scheuten, Life stage at test start: protonymphs (≤ 24 hours old)
Endpoints:	LR ₅₀ (median Lethal Rate) and ER ₅₀ (median Effect Rate), where possible. The No Observed Effect Rate (NOER), where possible.
Test design:	Test and reference items were diluted in deionised water and applied with a laboratory track sprayer to glass plates. A control group applied with deionised water was included in the study. All applications were performed with a spray volume of 200 L/ha. After assembling the test units, twenty protonymphs of <i>Typhlodromus pyri</i> were introduced into each test unit (5 replicates per treatment). Direct treatment effects (mortality) and any change in behaviour, with respect to the control, were assessed after 1, 3 and 7 days. Reproduction was not possible to be assessed since the corrected mortality was higher than 50 % at every tested rates of the test item.
Test rates:	Spinosad 480 g/L SC: 40.00, 160.00 and 320.00 g spinosad/ha equivalent to 0.087, 0.348 and 0.696 L test item /ha (according to the analytical concentration). Dimethoate 40 % w/v EC: 3.681 g dimethoate /ha equivalent to 0.009 L of reference item/ha (according to the analytical concentration).
Test conditions:	Temperature: 24.5 – 25.0° C Relative humidity: 78.9 – 85.1 % Light regime: 16 h light / 8 h darkness Light intensity: 1296 - 1387 lux during exposure
Statistics:	The LR ₅₀ and their 95%-confidence limits was not possible to be calculated with survival at 7 d since the corrected mortality was higher than 50 % at every tested rates of the test item. Therefore, the LR ₅₀ was estimated as lower than the lowest rate of the test item tested. Step-down Cochran-Armitage Test with survival at 7 d (one-sided greater, $\alpha=0.05$) was used to detect significant differences between mortality data of the test item groups and the control. Reproduction was not possible to be assessed since the corrected mortality was higher than 50 % at every tested rates of the test item.
Dates of work:	10 May – 17 May 2021 [Experimental Phase]

Results and discussions

Treatment group	Rates [g active ingredient/ha]	Mean mortality [%] ^a	Corrected mortality [%]	Reproduction [eggs/female]	Reduction in reproduction rate [%]
Control (deionised water)	--	14.00	--	Not studied	--
Test item (Spinosad 480 g/L SC)	40.00	100.00 ^{sd}	100.00	Not studied	--
	160.00	100.00 ^{sd}	100.00	Not studied	--
	320.00	100.00 ^{sd}	100.00	Not studied	--
Reference item Dimethoate 40% w/v EC	3.681	91.00	89.53	--	--

^a sd: statistically significantly increased compared to control (Step-down Cochran-Armitage Test, one-sided greater, ($\alpha = 0.05$))

Endpoint	Rate	
	[g spinosad/ha] ^a	[L test item/ha] ^b
LR ₅₀ (lower –upper 95 % cl)	< 40.00	< 0.087
ER ₅₀	Not studied	--
NOER (mortality)	< 40.00	< 0.087
NOER (reproduction)	Not studied	--

^a Rate in g of active ingredient /ha

^b content of formulated product according to the certificates of analysis (Spinosad 460.0 g/L)

Conclusion

The study was conducted as a rate response test under standard laboratory test conditions with five treatment groups on *Typhlodromus pyri*, including the test item Spinosad 480 g/L SC at three application rates, the reference item (Dimethoate 40 % w/v EC) at a single application rate and the control applied with deionised water.

Mortality below 20% (14.0 %) was achieved 7 days after the application meeting the validity criteria. Reproduction was not possible to be assessed since the corrected mortality was higher than 50 % at every tested rates of the test item. The toxic reference product caused 89.53 % mortality (corrected relative to control) and confirmed the sensitivity of the test species and the test conditions.

Under these extended laboratory test conditions, LR₅₀ (rate producing 50 % mortality) was estimated as lower than the lowest rate of the test item tested, 40 g spinosad/ha (equivalent to 0.087 L formulated/ha according to the analysed content).

The NOER for lethal effects was estimated as lower than 40.00 g spinosad/ha (equivalent to 0.087 L formulated/ha according to the analysed content) since significant effects on mortality of *Typhlodromus pyri* when compared to the control at the tested rates of the test item from 40.00 to 320.00 g spinosad/ha were obtained (Step-down Cochran-Armitage Test, one-sided greater, ($\alpha = 0.05$)).

Reproduction was not possible to be assessed since the corrected mortality was always above the 50 % at every tested rates of the test item.

Validity criteria

Control mortality	The mean mortality (dead and escaped individuals) in the control should be $\leq 20\%$ on day 7 of exposure (actual: 14.0 % mortality).
Reference item mortality	The corrected cumulative mean mortality in the reference item group should range between 50 % and 100 % on day 7 after application (actual: 89.53 % corrected mortality).
Control reproduction	The cumulative mean number of eggs per female in the control (from day 7 to day 14) should be ≥ 4.0 eggs/female (actual: reproduction phase not performed).

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

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

A 2.3.2.4 KCP 10.3.2.4 Field studies with non-target arthropods

A 2.3.2.4.1 Study 1

Comments of zRMS:	The applicant submitted a whole arthropod community field study. The product was applied once at 0.375, 1.5, 6, 24 and 96 g a.s./ha to grassland meadow. The study was designed as a NOER-type approach (Bakker and Miles, 2007, De Jong et al., 2010) and therefore makes the results applicable to any product use pattern. The number of taxa occurring in sufficiently high number to allow for a population level analysis was higher than the number of taxa usually evaluated in studies performed in commercial agricultural settings. This made the study more powerful. Sampling techniques: By using five different collecting methods (pitfall, Berlese Tullgren extraction from weed and from soil, aspirator and sweepnet sampling techniques) all strata of the arthropod community occurring in grasslands were comprehensively sampled (soil-dwelling, ground dwelling- and weed inhabiting arthropods). To analyse the technical reliability, the checklist provided by De Jong <i>et al.</i> (2010) was used. Based on this checklist the study and the study report are classified as reliable. The study was performed from spring to early summer because this is a period with high arthropod diversity and abundance. When the study took place, a large proportion of taxa included in univariate analyses were present in moderate to high numbers in the off-field habitat (90% abundant enough during application to be included in statistical analysis). This ensured that populations were exposed to the treatments, and potential treatment related adverse effects on population development could be evaluated. The direct overspray that was applied to mimic true spray drift should be considered a worst-case approach. The timing of the application agreed with the proposed GAP. Consequently, the phenological stages of off-field flora and fauna were representative for the proposed agronomical use. The field was surrounded by other grassland meadows and bordered by a forest on the southeast side. Part of the southern side of the field was adjacent to an agricultural field. For approximately five years preceding the test no pesticides or herbicides were used in the experimental field.
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	Four complete blocks of seven treatment plots each were set up in the experimental field. Assessments (arthropod sampling) were made five days before application, 3 days after application and then after 1 and 2 weeks, and approximately 1 and 2 months. From 7 April 2021 until the end of the trial climatic data were recorded with a calibrated weather station installed at the trial site. During applications, wind speed was measured on site with a calibrated hand-held anemometer. On the application day, all treatments were applied during dry weather conditions. During the application the temperature ranged between 10.60°C and 15.02°C. Wind speeds varied between 2.0 and 2.8 m/s. The vegetation of the experimental field was surveyed systematically in order to characterize the off-crop habitat, and assess the homogeneity of the foliage in the experimental field between blocks. The statistical significance of differences among treatments in population density was determined separately for all time points using the distribution free Mann-Whitney U test. This test is preferred over ANOVA. Large data heterogeneity is commonly observed in terrestrial arthropod community datasets, due to complex interactions between arthropod species and other biotic and abiotic factors. Type-1 error levels associated with parametric tests such as ANOVA are sensitive to departures from a set of assumptions. The vegetation was highly diverse with on average 43 plant species annotated in each plot but ranging from 33 to 59 plant species. Each plot contained at least 8 grass species and at least 24 flowering plant species. Twelve plant species were present in all 28 plots. There was no plant species which occupied more than 50% in a plot and in each plot at least 2 but mostly 3 or more plants were co-dominant of which in most plots at least 1 flowering plant. All four arthropod data sets (Acari from weed samples, Acari from soil samples, Collembola, other arthropods) showed a statistically significant correlation with block design and vegetation structure in the field. Soil moisture was included as co-variable in further community response analyses. Over 2 million arthropod specimens were attributed to a total of 1015 taxa. Taxa of low occurrence were either excluded from analyses, or they were pooled with other taxonomically related taxa. After this procedure, 67 taxa were retained for population level evaluations. This comprised 79 % of all specimens counted in all sample types. For the current uses, the application of the product is up to 3 times which means that in practice the frequency of application is substantially higher than what was tested in the field study. Therefore, according to the zRMS, it is appropriate to use the population NOER of 1.5 g a.s./ha for the risk assessment, because if no effects at all are observed after one application, it is not expected that unacceptable effects will occur after 3 applications. Currently no specific assessment factors are proposed for the mesocosm studies with arthropods. The study is considered valid and suitable for the risk assessment properties. In the opinion of zRMS, although the study was conducted in the southern zone, there are no contraindications to its usage in the central zone. A large population of arthropods was examined, which is important due to the high variability of the off-field habitats. The NOER of 1.5 g a.s./ha has been determined to be an appropriate endpoint for non-target arthropods risk assessment.
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Reference: KCP 10.3.2.4/01

Report A field trial to determine the effects of Spinosad 480SC on the non-target arthropod fauna of a meadow after one application in Southern Europe Zone (France) in 2021, Roig, J., 2022, Report No S20-09667

Guideline(s): Jong, F. De, F.M.W., Bakker, F.M., Brown, K., Jilesen, C.J.T.J., Posthuma-Doodeman, C.J.A.M., Smit, C.E., Van der Steen, J.J.M., Van Eekelen,

G.M.A. 2010. A guidance document of the Dutch platform for the assessment of higher tier studies. Rivm internal publication.

Deviations:

Deviation 1

Description of deviation: The pitfall traps in plot 6 were not placed at Sample 2. Reason for deviation: Due to a human error, the traps in this plot were not placed. Expected impact on the study: Data was extrapolated to 4 plots for this sample in order to obtain a data set comparable with the other plots for data analysis. No impact on the results expected.

Deviation 2

Description of deviation: Spray volume for treatment in some individual plots was 20 or 21 L/plot instead of 18 L $\pm$ 10% as described in study plan. Reason for deviation: The sprayer was stopped after the edge of some plots, resulting in total a higher volume sprayed. The extra volume was probably sprayed outside the plots. Expected impact on the study: No impact on the study expected. The study plots were sprayed with the correct rate.

Deviation 3

Description of deviation: The temperature of the freezer where the retain samples of the test solutions were stored raised above -18°C on 6 occasions during the storage. Samples, however, remain frozen. Reason for deviation: Punctual deviations due to opening of the door of the freezer or power failures. Expected impact on the study: No impact on the study expected. The samples remained frozen.

Deviation 4

Description of deviation: Prior to the taxonomic analysis the sweep-net samples were cleaned and separated into different fractions using a bottom sieve with an aperture of 200 μ m instead of 45 μ m. Reason for deviation: This was done to match the mesh size of the sampling nets. Expected impact on the study: No impact, all relevant taxa were counted.

GLP:

Yes

Acceptability:

Yes

Duplication

N/A

(if vertebrate study)

Objective

The test item Spinosad 480SC is an insecticide and is applied to protect commercial crops from a broad range of pests. This field study was designed to assess the potential acute adverse effects of non-intentional spray drift on non-target arthropods (NTA) in off-field habitats that might occur at various distances from a treated area for current and future use patterns of Spinosad 480SC. Potential effects were assessed after controlled exposure of a previously unexposed NTA community in a natural grassland habitat representative of agricultural landscapes in South-Western France. The plant community composition of the field was documented using a simplified Braun-Blanquet cover-abundance method (Westhoff and Maarel, 1978) as described by Aldershof and Bakker (2011). A broad range of NTA taxa was assessed in a rate-response design with multiple sampling events. Effects of exposure were determined both at the level of the NTA community and at the level of the population for individual taxa. The study was designed as a “NOER-type” (Bakker and Miles, 2007; De Jong et al., 2010). The choice for a NOER approach makes the results applicable to any product use pattern. At the same time the assessment of a NOEAER/LOEAER avoids the caveats of assessing the acceptability of certain effect levels at given drift rates (Bakker, 2012; Miles and Bakker, 2012). The finding that the NOER or NOEAER may be expected to occur at a certain distance from a treated area will be unambiguously interpretable. The approach was fully in line with Candolfi et al. (2000, 2001), De Jong et al. (2010) and ESCORT 3 (Alix et al., 2012). To enable a refined assessment of the NOEAER/LOEAER, sampling was

continued until approximately 2 months postapplication. Recovery within this time frame was interpreted as being ecologically acceptable, even when initial effects may have occurred.

The effects of the test item on the NTA fauna were expressed in terms of abundance changes relative to a water control and the statistical significance of any changes were assessed. This was done separately at the level of the NTA community and at the level of individual taxa occurring in sufficient abundance. Based on these analyses the following endpoints were determined: (1) NOER (No Observed Effect Rate), the highest rate where effects were not statistically significant at any time point; (2) LOER (Lowest Observed Effect Rate), the lowest rate resulting in statistically significant effects on at least one time point; (3) NOEAER (No Observed Ecologically Adverse Effect Rate) and (4) LOEAER (the Lowest Observed Ecologically Adverse Effect Rate). “Ecologically adverse” in this context is defined as statistically significant effects being observed at the end of the study period.

Materials and methods

This study is designed to assess the risk to off-field non-target arthropod (NTA) fauna potentially exposed around the time of application of Spinosad 480SC (active substance spinosad) through multiple exposure pathways (e.g. contact to spray droplets, residual contact, ingestion of contaminated food items). During the study, taxa with different ecological profiles were monitored using a combination of pitfall sampling, suction sampling, sweep-net sampling, soil cores and vegetation sampling followed by Berlese extraction. The test item Spinosad 480SC was tested according to the application of five different test item rates, 0.375 g a.s./ha, 1.5 g a.s./ha, 6 g a.s./ha, 24 g a.s./ha and 96 g a.s./ha. A water control treatment and a toxic reference treatment (Lambda cyhalothrin) were run in parallel to the application of the test item. The trial had a randomized block design with 4 replicates per treatment. The effects of the test item treatment and the reference treatment were expressed in terms of population changes relative to the water control.

Results and discussions

The experimental field was a cultivated hay land on sandy loam soil, with a high diversity of ground cover grass species and other plants. The field was entirely covered with vegetation except for a few molehills.

Vegetation within blocks was homogeneous, generally without structures potentially causing irregular microclimates.

The vegetation was highly diverse with on average 43 plant species annotated in each plot but ranging from 33 to 59 plant species. Each plot contained at least 8 grass species and at least 24 flowering plant species. No obvious spatial gradient in soil moisture was apparent in the field. The soil type was sandy loam, with a pH of 5.6 and 4.6% of organic matter. Over 2 million arthropod specimens were attributed to a total of 1015 taxa. Taxa of low occurrence were either excluded from the analyses, or they were pooled with other taxonomically related taxa. After this procedure, 67 taxa were retained for population level evaluations. This comprised 79 % of all specimens counted in all sample types.

Highly abundant taxa were springtails (85% of all specimens), juvenile Oribatida mites, juvenile Cicadellidae (Heteroptera) and Thysanoptera, both juvenile and adult. The highest diversity was found in spiders (Araneae) and beetles (Coleoptera). Highest diversity of taxa, which were abundant enough to be included in univariate analyses were mites and spiders, of which 13 and 12 taxa were included in univariate analyses. About one third of all taxa included in univariate analyses (36 %) could be evaluated at a taxonomic level below family (subfamily, genus- or species level).

Test performance

- Deviation rates per treatment (average of 4 plots) did not exceed 10%.
- A thorough survey of the entire arthropod community prevailing in the off-field habitat was established with a variety of sampling techniques, which yielded a comprehensive faunistic inventory of the experimental field.
- For several taxa no recovery was seen in the reference treatment within the time frame of the study, indicating that test design parameters, such as plot size, were adequate to demonstrate persistent adverse treatment related effects would these have occurred.

Summary results population analyses Spinosad 480SC applied at the lowest two test rates of 0.375 g a.s./ha and 1.5 g a.s./ha did not cause adverse effects (i.e., treatment related reductions on one or more

occasions) to any of the 67 taxa. Application at the middle dose rate of 6 g a.s./ha caused temporary adverse effects to two springtail taxa (Bourletiellidae, Sminthurinus sp.). At the second highest dose rate of 24 g a.s./ha, application of the test item negatively affected three springtail taxa (Entomobrya lanuginosa, Bourletiellidae, Sminthurinus sp.), and Mymaridae (Hymenoptera). At the highest dose rate of 96 g a.s./ha, application of the test item negatively affected 3 Acari taxa from soil samples (Pygmephoridae, Scutacaridae, Tarsonemidae), three Collembola taxa (Entomobrya lanuginosa, Bourletiellidae, Sminthurinus sp.), three Araneae taxa (juvenile Pachygnatha degeeri, adult Zelotinae, juvenile Thomisinae), one beetle (adult Latridiidae), two Hymenoptera (adult Mymaridae, brachypterous Chalcidoidea) and adult Thysanoptera. No recovery occurred for four taxa treated with the test item in the highest rate of 96 g a.s./ha (Tarsonemidae (Acari), Entomobrya lanuginosa, Sminthurinus sp. (both Collembola) and brachypterous Chalcidoidea (Hymenoptera)).

Summary table effect classifications

Effect classification (Based on De Jong <i>et al.</i> , 2010):						Effect class:	
<2 weeks	Adverse effect but duration less than 2 weeks, or only once more than 3 days after application					2	
<1 month	Adverse effects no longer apparent on the last two sampling moments					3a	
<2 months	Adverse effect no longer apparent on the last sampling moment					3b	
>2 months	No recovery from adverse effect within the study period (= 2 months)					8	
3a	Based on graphical trends (P>0.1 but effect >70% on two or more occasions)						
3a	Based on graphical trends and P<0.1 (one or more occasions)						
3a	Based on graphical trends and P<0.05 (one or more occasions)						
-	No effect						
Community level effects (multivariate analyses)							
(PRC/Monte-Carlo; 5% and 10% alpha level and trends)							
		0.375g	1.5g	6g	24g	96g	
M	Acari	-	-	-	-	-	
G	Acari	-	-	-	-	-	
C	Collembola	-	-	-	-	8	
PAS	Other arthropods	-	-	-	-	-	
Conclusion						Community NOER	Community LOEAE
Population level effects (univariate analyses)							
(Mann-Whitney U test; 5% and 10% alpha level and trends)							
	Class/Order	Taxon	0.375g	1.5g	6g	24g	96g
G	ACARI	Heterostigmata Scutacaroidae Scutacaridae					2
G	ACARI	Heterostigmata Tarsonemoidae Tarsonemidae					8
C	COLLEMBOLA	Entomobrya lanuginosa c.f.				3b	8
C	COLLEMBOLA	Bourletiellidae			2	2	2
C	COLLEMBOLA	Sminthurinus sp.			3a	3b	8
A	ARANEAE	Pachygnatha degeeri juvenile					3a
PAS	ARANEAE	Zelotinae adult					2
PAS	ARANEAE	Thomisinae juvenile					3a
PAS	COLEOPTERA	Cucujoidea Latridiidae adult					2
A	HYMENOPTERA	Chalcidoidea Mymaridae adult					3a
P	HYMENOPTERA	Chalcidoidea brachypterous adult					8
A	OTHER	Insecta Thysanoptera adult					3b
Conclusion			Population NOER	Population LOER	Population NOEAE	Population LOEAE	
C=aspirator Collembola; M= mites from weeds; G=mites from soil; A=aspirator other; P=pitfall; S=sweep net; combined letters=pooled							
Rates are in g.s./ha							

C=aspirator Collembola; M= mites from weeds; G=mites from soil; A=aspirator other; P=pitfall; S=sweep net; combined letters=pooled

Rates are in g a.s./ha

Table 3B Arthropod community composition and compliance with De Jong *et al.* (2010)

Order	Group	Minimum desired level of taxonomic precision (De Jong <i>et al.</i> , 2010)	Was taxon present/identified minimally at required taxonomic level?	Remark	Included	Conclusive (including trend evaluation)	Level A	Level B (cumulative)	Level C (cumulative)	Level D (cumulative)
ACARI (Weed)										
	Gamasida	family	yes		number of taxa*: 5	4	0	0	1	4
	Actinedida	family	yes		% from order: 80%	0%	0%	20%	80%	
	Onibatida	suborder	yes		% from community: 7%					
ACARI (Soil)										
	Gamasida	family	yes		number of taxa*: 8	8	0	3	4	8
	Actinedida	family	yes		% from order: 100.0%	0.0%	37.5%	50.0%	100.0%	
	Onibatida	suborder	yes		% from community: 12%					
COLLEMBOLA										
		suborder	yes		number of taxa*: 11	10	0	0	2	9
					% from order: 91%	0%	0%	18%	82%	
					% from community: 16%					
ARANEAE										
	Hunting spiders	family	yes		number of taxa*: 12	12	0	4	9	12
	Lycosidae	genus/species	yes		% from order: 100%	0%	33%	75%	100%	
	Thomisidae	genus/species	yes	R2	% from community: 18%					
	Web spiders	family	yes							
	Linyphiidae	genus/species	yes							
	Juveniles	family	yes							
COLEOPTERA										
	Carabidae	species	yes	R1	number of taxa*: 8	8	0	1	4	8
	Staphylinidae	genus/species	yes		% from order: 100%	0%	13%	50%	100%	
	Coccinellidae	genus/species	yes	R1	% from community: 12%					
	Lathridiidae	family	yes							
	Juveniles	family	yes							
DIPTERA										
	Adults	suborder	yes		number of taxa*: 4	3	0	0	1	3
	Juveniles	order	yes	R1	% from order: 75%	0%	0%	25%	75%	
					% from community: 6%					
HEMIPTERA										
	Aphidoidea	suborder/superfamily	yes		number of taxa*: 9	7	0	2	2	7
	Other Sternorrhyncha	suborder/superfamily	yes		% from order: 78%	0%	22%	22%	78%	
	Cicadellidae	family	yes		% from community: 13%					
	Other Auchenorrhyncha	suborder/superfamily	yes							
	Heteroptera	family	yes	R2						
HYMENOPTERA										
	Ichneumonidea	superfamily	yes		number of taxa*: 8	7	0	2	4	7
	Chalcidoidea	family	yes		% from order: 88%	0%	25%	50%	88%	
	Proctotrupoidea	superfamily	yes		% from community: 12%					
OTHER ARTHROPODS										
	THYSANOPTERA	order	yes		number of taxa*: 10	10	0	3	5	9
	LEPIDOPTERA	order	yes	R1	% from group: 100%	0%	30%	50%	90%	
	ORTHOPTERA	order	yes	R1	% from community: 15%					

R1 Several taxa were identified, but numbers were too low for inclusion in univariate analyses. Separate taxa were included in multivariate evaluations if totals over all samples and treatments exceeded 10.

R2 Several taxa were identified, but taxa were pooled to higher level for inclusion in univariate analyses due to low numbers. Separate taxa were included in multivariate evaluations.

*Number of taxa included in univariate analyses

Level A taxa: 50th percentile MSD below 50% for $\alpha=0.05$ (small effects)
 Level B taxa: 25th percentile MSD below 50% for $\alpha=0.05$ OR (moderately small effects)
 50th percentile MSD below 50% for $\alpha=0.1$ (moderately small effects)
 Level C taxa: 25th percentile MSD below 50% for $\alpha=0.1$ (moderate effects)
 Level D taxa: 25th percentile MSD below 70% for $\alpha=0.1$ (moderately large effects)
 Trend: 25th percentile MSD above 70% for $\alpha=0.1$ (large effects)

Taxa inconclusive: Minimum Significant detectable Difference (MSD) too high
 AND no adverse effect of the reference item and/or test item (no trend evaluation possible).

Table 3C Population data validity (MSD analyses combined with effect evaluations)

Fig	Method	Class/Order	Taxon	sample (weeks after application)	Level A	Level B	Level B	Level C	Level D	Sampled during application phase	Effect reference treatment	Effect test item	Evaluation level
					MSD (0.05) at 50 th percentile	MSD (0.05) at 25 th percentile	MSD (0.1) at 50 th percentile	MSD (0.1) at 25 th percentile					
5	M	ACARI	Monogynaspida all		86%	77%	71%	58%	●	yes	yes	no	Level D
6	M	ACARI	Oribatida adult		87%	82%	69%	61%	●	yes	no	no	Level D
7	M	ACARI	Sarcoptiformes Oribatida juvenile		✗ 95%	92%	84%	74%	●	yes	no	no	inconclusive
8	M	ACARI	Prostigmata Eupodidae Tydeidae		79%	74%	61%	52%	●	yes	no	no	Level D
9	M	ACARI	Heterostigmata Tarsonemoides Tarsonemidae		77%	64%	58%	47%	●	yes	no	no	Level C
10	G	ACARI	Gamasina all		65%	51%	48%	34%	●	yes	no	no	Level B
11	G	ACARI	Opilioidea Opitidae adult		90%	82%	71%	67%	●	yes	no	no	Level D
12	G	ACARI	Schelioribates sp. adult		69%	57%	48%	40%	●	yes	yes	no	Level B
13	G	ACARI	Oribatida other		88%	84%	74%	65%	●	yes	no	no	Level D
14	G	ACARI	Sarcoptiformes Oribatida juvenile		85%	60%	63%	44%	●	yes	no	no	Level C
15	G	ACARI	Prostigmata Heterostigmata Pymnophoroidea		93%	71%	75%	52%	●	yes	yes	yes	Level D
16	G	ACARI	Heterostigmata Scutacaroides Scutacaridae		90%	75%	72%	58%	●	yes	no	yes	Level D
17	G	ACARI	Heterostigmata Tarsonemoides Tarsonemidae		69%	64%	48%	43%	●	yes	yes	yes	Level B
18	C	COLLEMBOLA	Entomobrya laumigosa c.f.		90%	80%	76%	60%	●	yes	no	yes	Level D
19	C	COLLEMBOLA	Lepidocyrtus cynaeus all		88%	73%	71%	63%	●	yes	no	no	Level D
20	C	COLLEMBOLA	Isotomurus sp. all		✗ 99%	85%	93%	69%	●	yes	no	no	Level D
21	C	COLLEMBOLA	Isotomidae other		87%	83%	80%	65%	●	no	no	no	Level D
22	C	COLLEMBOLA	Entomobryomorpha Isotomidae small juv.		90%	68%	73%	56%	●	yes	no	no	Level D
23	C	COLLEMBOLA	Brachystomella parvula all		86%	72%	72%	61%	●	yes	yes	no	Level D
24	C	COLLEMBOLA	Xenylla humicola all		80%	70%	63%	49%	●	yes	no	no	Level C
25	C	COLLEMBOLA	Bourletellidae		✗ 100%	✗ 100%	✗ 99%	83%	●	yes	no	yes	trend
26	C	COLLEMBOLA	Smithurinus sp.		79%	61%	57%	45%	●	yes	no	yes	Level C
27	C	COLLEMBOLA	Smithurinus viridis all		✗ 100%	✗ 100%	✗ 99%	89%	●	yes	no	no	inconclusive
28	*C	COLLEMBOLA	Sphaeridia pumilis all		91%	69%	75%	54%	●	yes	no	no	Level D
29	PAS	ARANEAE	Linyphiidae Erigoninae juvenile		79%	72%	61%	55%	●	yes	yes	no	Level D
30	A	ARANEAE	Temiphanes sp. juvenile		55%	48%	42%	36%	●	yes	no	no	Level B
31	PAS	ARANEAE	Linyphiidae adult		78%	55%	51%	37%	●	yes	no	no	Level C
32	A	ARANEAE	Linyphiidae Linyphiinae juvenile		66%	58%	47%	39%	●	yes	yes	no	Level B
33	A	ARANEAE	Pachygnatha degeeri juvenile		66%	58%	51%	41%	●	yes	yes	yes	Level C
34	PAS	ARANEAE	Zelotinae adult		72%	66%	55%	43%	●	yes	yes	yes	Level C
35	PAS	ARANEAE	Zelotes sp. juvenile		74%	52%	55%	41%	●	yes	no	no	Level C
36	P	ARANEAE	Alopecosa cuneata adult		90%	74%	70%	56%	●	yes	yes	no	Level D
37	PA	ARANEAE	Lycosinae other		65%	57%	47%	43%	●	yes	no	no	Level B
38	PAS	ARANEAE	Pardosa sp. juvenile		81%	65%	58%	45%	●	yes	no	no	Level C
39	PA	ARANEAE	Pardosa sp. adult		60%	46%	43%	34%	●	yes	yes	no	Level B
40	PAS	ARANEAE	Thomisinae juvenile		81%	73%	61%	54%	●	yes	no	yes	Level D
41	PAS	COLEOPTERA	Chrysomelidae Chrysomelidae adult		76%	60%	56%	44%	●	yes	yes	no	Level C
42	PAS	COLEOPTERA	Cucujoides Latridiidae adult		72%	47%	57%	35%	●	yes	yes	yes	Level B
43	PAS	COLEOPTERA	Cucujoides adult		81%	66%	55%	44%	●	yes	no	no	Level C
44	PAS	COLEOPTERA	Elateroidea Elateridae adult		87%	80%	70%	67%	●	yes	no	no	Level D
45	P	COLEOPTERA	Helophorus sp. adult		85%	75%	64%	55%	●	yes	no	no	Level D
46	PAS	COLEOPTERA	Aleocharinae adult		93%	51%	87%	46%	●	no	no	no	Level C
47	PA	COLEOPTERA	Paederinae adult		83%	75%	61%	56%	●	yes	yes	no	Level D
48	A	COLEOPTERA	Tachyporinae sp. juvenile		91%	70%	78%	63%	●	no	no	yes	Level D
49	S	DIPTERA	Empidoidea Empididae adult		84%	74%	63%	53%	●	yes	yes	no	Level D
50	A	DIPTERA	Platypoda Phoridae adult		71%	56%	56%	36%	●	no	no	no	Level C
51	A	DIPTERA	Sciaroidea Cecidomyiidae adult		71%	64%	59%	53%	●	yes	yes	no	Level D
52	A	DIPTERA	Sciaroidea Sciaridae adult		✗ 98%	94%	85%	75%	●	yes	no	no	inconclusive
53	AS	HEMIPTERA	Membracidae Cicadellidae adult		60%	54%	46%	35%	●	yes	yes	no	Level B
54	AS	HEMIPTERA	Membracidae Cicadellidae juvenile		79%	72%	65%	55%	●	yes	yes	no	Level D
55	A	HEMIPTERA	Fulgoroidea Delphacidae juvenile		92%	78%	83%	62%	●	no	no	no	Level D
56	AS	HEMIPTERA	Miroidea Miridae adult		✗ 96%	72%	84%	58%	●	yes	no	no	Level D
57	A	HEMIPTERA	Miroidea Miridae juvenile		✗ 98%	86%	91%	65%	●	yes	no	no	Level D
58	PAS	HEMIPTERA	Lygaeoidea Lygaeidae adult		91%	89%	83%	81%	●	no	no	no	inconclusive
59	PAS	HEMIPTERA	Hemiptera Heteroptera other juv.		✗ 97%	86%	83%	67%	●	yes	no	no	Level D
60	A	HEMIPTERA	Sternorhyncha Aphidoidea all		58%	43%	43%	29%	●	yes	yes	no	Level B
61	*A	HEMIPTERA	Sternorhyncha Coccioidea juvenile		✗ 100%	✗ 99%	✗ 95%	91%	●	yes	no	no	inconclusive
62	A	HYMENOPTERA	Vespoidea Formicidae adult		93%	82%	80%	66%	●	yes	no	no	Level D
63	A	HYMENOPTERA	Ceraphronoidea Ceraphronidae adult		71%	64%	58%	47%	●	yes	no	no	Level C
64	A	HYMENOPTERA	Chalcidoidea Myrmecidae adult		61%	56%	43%	37%	●	yes	no	yes	Level B
65	A	HYMENOPTERA	Chalcidoidea Trichogrammatidae adult		✗ 99%	84%	80%	70%	●	yes	no	no	inconclusive
66	A	HYMENOPTERA	Chalcidoidea other		80%	50%	62%	32%	●	yes	no	no	Level C
67	P	HYMENOPTERA	Chalcidoidea brachypterous adult		83%	76%	66%	58%	●	yes	no	yes	Level D
68	AS	HYMENOPTERA	Braconidae adult		67%	59%	46%	37%	●	no	no	no	Level B
69	A	HYMENOPTERA	Platygastriidae Scelioninae adult		80%	73%	66%	54%	●	yes	no	no	Level D
70	A	OTHER	Insecta Thysanoptera adult		70%	63%	52%	43%	●	yes	yes	yes	Level C
71	A	OTHER	Insecta Thysanoptera juvenile		94%	90%	84%	74%	●	yes	yes	no	trend
Cumulative total					0	4	11	28	59	60	Taxa conclusive:		61
					0%	6%	16%	42%	88%	90%	% taxa conclusive:		91%

C=aspirator Collembola, M=mites (weeds), G=mites (soil), A=aspirator other, P=Pitfall, S=sweep net, combined letters=pooled

*=CV larger than 1.4 on more than 50% of the sampling moments with sufficient numbers.

See Table 3B for explanations of evaluation levels

✗ xxth percentile MSD>95%
● xxth percentile MSD<50%

Taxa inconclusive: Minimum Significant detectable Difference (MSD) too high (not within Level A - D) AND no adverse effect of the reference item and/or test item (no trend evaluation possible).

†: MSD sufficiently low (at least 25th percentile lower than 70% when calculated at alpha=0.1) BUT no effect of the reference item and/or test item, despite exposure. Sensitivity of this taxon is not demonstrated.

††: MSD sufficiently low (at least 25th percentile lower than 70% when calculated at alpha=0.1) BUT no effect of the reference item nor test item. Lack of effect may be due to insufficient exposure (this taxon was not or hardly sampled during the application phase).

◊: taxon sampled during application phase, but number of individuals too low for statistical analysis

Application phase: before and up to and including one week after treatment.

Table 4 Summary performance PRC analyses

A: Overall responses (entire sampling period)

Data set	Treatments included in analysis	% Variance explained by treatment regime in		P-value ax1	P-value ax2
		ax1	ax2		
Acari Weed	Test item rates	17.9	14.8	0.793	0.922
	0.375g	41	24.2	0.408	0.695
	1.5g	35.5	18.8	0.804	0.997
	6g	37.1	29.3	0.551	0.555
	24g	42	24.2	0.342	0.862
	96g	44.7	24.6	0.246	0.904
	Reference	47.1	23.3	0.143	0.635
Acari Soil	Test item rates	21.7	14.3	0.802	0.981
	0.375g	41.8	18.2	0.709	0.978
	1.5g	47.9	18.2	0.826	1.000
	6g	55.2	15.4	0.446	0.981
	24g	46	19.5	0.636	0.980
	96g	48	17.8	0.585	0.920
	Reference	51.5	18.3	0.539	0.916
Collembola	Test item rates	31.1	11.3	0.026	0.967
	0.375g	36.7	24.9	0.927	0.994
	1.5g	39.9	29	0.509	0.780
	6g	36.6	24.3	0.743	0.779
	24g	40.5	19.8	0.434	0.919
	96g	52.5	16.1	0.087	0.406
	Reference	42.1	25.9	0.519	0.660
Other arthropods	Test item rates	11.3	8.3	0.916	0.994
	0.375g	29.8	22.5	0.841	0.991
	1.5g	31.1	21.7	0.968	0.943
	6g	33.5	19.2	0.839	0.981
	24g	31.9	19.9	0.919	0.978
	96g	41.4	19.1	0.472	0.813
	Reference	58.6	14.2	0.030	0.658

B: P-values at individual sampling moments (Monte Carlo Permutation Test-pairwise comparison to control)

Data set	sample	0.375g	1.5g	6g	24g	96g	Reference
Acari Weed	pre	1	0.567	0.818	0.282	0.581	0.341
	week 0	2	0.582	0.238	0.454	0.211	0.200
	week 1	3	0.102	0.937	0.659	0.925	0.962
	week 2	4	0.595	0.857	0.549	0.380	0.100
	week 4	5	0.927	0.832	0.691	0.410	0.666
	week 8	6	0.184	0.823	0.247	0.795	0.365
							0.077
Acari Soil	pre	1	0.690	0.913	0.974	0.915	0.941
	week 0	2	0.827	0.833	0.346	0.573	0.343
	week 1	3	0.804	0.920	0.719	0.715	0.633
	week 2	4	0.375	0.668	0.304	0.596	0.473
	week 4	5	0.798	0.518	0.509	0.889	0.053
	week 8	6	0.294	1.000	0.276	0.738	0.772
							0.403
Collembola	pre	1	0.938	0.481	1.000	0.985	0.251
	week 0	2	0.986	0.976	0.778	0.640	0.402
	week 1	3	0.726	0.913	0.376	0.168	0.099
	week 2	4	0.464	0.260	0.520	0.163	0.130
	week 4	5	0.905	0.154	0.642	0.733	0.039
	week 8	6	0.708	0.747	0.685	0.436	0.100
							0.201
Other arthropods	pre	1	0.834	0.971	0.885	0.775	0.833
	week 0	2	0.771	0.972	0.165	0.343	0.551
	week 1	3	0.711	0.806	0.535	0.623	0.308
	week 2	4	0.602	0.803	0.801	0.914	0.402
	week 4	5	0.971	1.000	0.946	0.855	0.690
	week 8	6	0.771	0.915	0.919	0.919	0.551
							0.190

Test item rates All test item treatments analysed together
T1, T2, etc One test item treatment analysed separately
Reference Reference item analysed separately
Rates are in g a.s./ha

Statistically significant at alpha = 0.1
Statistically significant at alpha = 0.05
(Monte Carlo permutation test)

Effect classification (Based on De Jong *et al.*, 2010):

Effect classification (Based on De Jong et al., 2010)

<1 week

Adverse effect only once immediately after application

1

2

<2 weeks

Adverse effect but duration less than 2 weeks, or only once more than 3 days after application

2

3a

<1 month

Adverse effects no longer apparent on the last two sampling moments

3

3b

<2 months

Adverse effect no longer apparent on the last sampling moment

4

3b

>2 months

No recovery from adverse effect within the study period (= 2 months)

5

3b

Based on graphical trends (P<0.1 but effect >70% on two or more occasions)

3a

3a

Based on graphical trends and P<0.1 (one or more occasions)

3a

3a

Based on graphical trends and P<0.05 (one or more occasions)

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CV larger than 1.4 on more than 50% of sampling moments (with sufficient numbers allowing for robust evaluations)

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Table 7 Summary community and population effect classifications

Effect classification (Based on De Jong <i>et al.</i> , 2010):		Effect class:				
<2 weeks	Adverse effect but duration less than 2 weeks, or only once more than 3 days after application					2
<1 month	Adverse effects no longer apparent on the last two sampling moments					3a
<2 months	Adverse effect no longer apparent on the last sampling moment					3b
>2 months	No recovery from adverse effect within the study period (= 2 months)					8
3a	Based on graphical trends (P>0.1 but effect >70% on two or more occasions)					
3a	Based on graphical trends and P<0.1 (one or more occasions)					
3a	Based on graphical trends and P<0.05 (one or more occasions)					
-	No effect					
Community level effects (multivariate analyses)		0.375g	1.5g	6g	24g	96g
M	Acari	-	-	-	-	-
G	Acari	-	-	-	-	-
C	Collembola	-	-	-	-	8
PAS	Other arthropods	-	-	-	-	-
Conclusion		Community NOER		Community LOEER		
Population level effects (univariate analyses)		0.375g	1.5g	6g	24g	96g
Class/Order	Taxon					
G ACARI	Heterostigmata Scutacaroidae Scutacaridae					2
G ACARI	Heterostigmata Tarsonemoidae Tarsonemidae					8
C COLLEMBOLA	Entomobrya lanuginosa c.f.				3b	8
C COLLEMBOLA	Bourletiellidae		2		2	2
C COLLEMBOLA	Sminthurinus sp.		3a		3b	8
A ARANEAE	Pachygnatha degeeri juvenile					3a
PAS ARANEAE	Zelotinae adult					2
PAS ARANEAE	Thomisinae juvenile					3a
PAS COLEOPTERA	Cucujoidae Latridiidae adult					2
A HYMENOPTERA	Chalcidoidea Mymaridae adult					3a
P HYMENOPTERA	Chalcidoidea brachypterous adult					8
A OTHER	Insecta Thysanoptera adult					3b
Conclusion		Population NOER	Population LOER	Population NOEER	Population LOEER	

C=aspirator Collembola; M= mites from weeds; G=mites from soil; A=aspirator other; P=pitfall; S=sweep net; combined letters=pooled

Rates are in g a.s./ha

Taxa scored effect level 1 are not included in this conclusions table, as effect was observed only once directly after treatment. This adaption was made to have conclusions more in line with study designs that do not have a sampling moment 3 days after treatment included (and hence transient effects shorter than one week would not be detected).

Conclusion

Spinosad 480SC applied at a rate of 24 g a.s./ha is classified as the highest community NOER (No Observed Effect Rate) for arthropods prevailing in off-field grassland habitats.

- Spinosad 480SC applied at a rate of 96 g a.s./ha is the community LOEER (Lowest Ecologically Adverse Effect Rate) tested in this study.
- Spinosad 480SC applied at a rate of 1.5 g a.s./ha is classified as the highest population NOER (No Observed Effect Rate) for arthropods prevailing in off-field grassland habitats.
- Spinosad 480SC applied at a rate of 6 g a.s./ha is the population LOER (Lowest Observed Effect Rate) tested in this study. Two springtail taxa were adversely affected. Bourletiellidae were affected only once (Effect class 2), the adverse effect on Sminthurinus sp. lasted less than a month (Effect class 3a).
- Spinosad 480SC applied at a rate of 24 g a.s./ha is the population NOEER (No Ecologically Adverse Effect Rate) tested in this study.
- Spinosad 480SC applied at a rate of 96 g a.s./ha is the population LOEER (Lowest Ecologically Adverse Effect Rate) tested in this study.
- In the highest test item rate 96 g a.s./ha, 18% of all examined taxa at the population level were adversely affected (12 out of 67 taxa). Scutacaridae (Acari), Bourletiellidae (Collembola), Zelotinae (Araneae) and Latridiidae (Coleoptera) were adversely affected only once (Effect class 2). Juvenile Pachygnatha degeeri, juvenile Thomisinae and adult Mymaridae were adversely affected but recovered within a month (Effect class 3a). Adult Thysanoptera recovered within the two months of the study period (Effect class 3b). Adverse effects on Tarsonemidae (Acari), Entomobrya lanuginosa, Sminthurinus sp. (both Collembola) and brachypterous Chalcidoidea (Hymenoptera) lasted throughout the two-months sampling period of the study, and no recovery was observed (Effect class 8).

Validity criteria

- The arthropod community occurring in the study field was sufficiently diverse and sufficiently abundant to draw valid and generally applicable conclusions. Except for Lepidoptera and Orthoptera, the main insect orders expected in off-field habitats were represented (De Jong et al., 2010).
- Several techniques were used to examine validity and robustness of the data generated in this study. All datasets were considered appropriate for evaluation of potential treatment related effects, despite a higher variability observed in the Collembola- and the two Acari datasets. Separate community analyses of the datasets avoided masking of potential treatment related effects on arthropods by highly variable or abundant taxa.
- Up to date there are no techniques available to investigate the power of the multivariate analyses used in this study (ordination analyses: Van den Brink, personal communication). Sample scores obtained in Principal Response Curves analyses were used to present variability of arthropod community data per plot.
- Correlation analyses of environmental parameters and intrinsic arthropod distribution patterns prior to treatment (soil moisture, vegetation structure and spatial location), and empirical examination of reference data selected the best model with highest discriminative power to test for treatment related effects on arthropod community composition and abundance.
- At the population level a total of 67 taxa were considered for statistical analysis, based on their abundance in the control. The taxonomic diversity and spread of 91% of these taxa were considered sufficiently fit to be accurately used as indicators to evaluate the risk for potential exposure of off-field arthropod fauna.
- Results from analysis of six taxa were considered inconclusive due to high MSD values in combination with a lack of a clear adverse effect induced by the reference treatment or the test item treatment. These taxa were one mite taxon (juvenile Oribatida), one Collembola taxon (*Sminthurus viridis*), one Diptera taxon (adult Sciariidae), two Hemiptera taxa (adult Lygaeidae, juvenile Coccoidea) and one Hymenoptera taxon (adult Trichogrammatidae). The concerned taxa were taxonomically related to other taxa with MSD values that were considered sufficiently robust for potential effect evaluations.
- Data of Aranea taxa were most robust: out of the twelve taxa included in evaluations, nine (75%) had MSD values appropriate to detect moderate adverse effects on abundance in a statistically significant manner.
- Data of Collembola were least robust. MSD values indicated that only moderately large effects were detectable in a statistical manner for most taxa of these taxa, would these occur.
- A proportion of 47% of the taxa were reduced by more than 50% (Figure 77) on the sampling moment following the reference item application.
- Given the large number of comparisons on which evaluations were based (352; Figure A7-1), it is highly unlikely that effects would have been missed. In other words, the risk of drawing false negative conclusions (concluding there is no effect when in fact there is an effect) is negligible under a 10% false positive error rate. MSD analysis shows that in general the results of the statistical hypothesis testing should be considered under $\alpha=0.1$. Including trend evaluation and expert judgement further refined conclusions.

A 2.3.2.5 KCP 10.3.2.5 Other routes of exposure for non-target arthropods

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

A 2.4.1.2 KCP 10.4.1.2 Earthworms - field studies

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

A 2.4.2.1.1 Study 1

Comments of zRMS:	The study was conducted to OECD 232 the guideline and according to the principles of GLP. All validity criterions were met. The study is considered to be reliable and suitable for the risk assessment.
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Reference: KCP 10.4.2.1/01

Report Spinosad 480 g/L, SC: Effects on the Reproductive Output of the Springtail *Folsomia candida* Willem (Collembola, Isotomidae) in Artificial Soil, Antón, B., 2021, Report No S21-04368

Guideline(s): OECD Guideline for the Testing of Chemicals, No. 232 (2016): Collembola Reproduction Test in Soil; Adopted: 29th July 2016.

Deviations: More than 10 initial individuals were introduced in the replicate T4-1, since 18 adult individuals were found on day 28. Data from replicate T4-1 are not considered for the calculations. Calculations for T4 treatment are performed with 3 replicates instead of 4 replicates. The aforementioned deviation had no impact on the outcome of the study.

GLP: Yes

Acceptability: Yes

Duplication N/A
(if vertebrate study)

Objective

The objective of the study was to determine the effect of Spinosad 480 g/L, SC, on survival and the reproductive output of the springtail *Folsomia candida* Willem (Collembola, Isotomidae) under worst-case exposure conditions. For this purpose, the Lowest Observed Effect Concentration (LOEC) for mortality and reproductive output, the No Observed Effect Concentration (NOEC), the median Lethal Concentration (LC₅₀) for adult mortality, and the Effect Concentration on reproductive output (EC_{10, 20, 50}) were determined, where possible.

Materials and methods

Test item: 'Spinosad 480 g/L, SC – SAP09I; Batch code: D-CEA; expiry date: March 2023; active ingredient content: 460 g spinosad/L; relative density: 1.08.

Test species: *Folsomia candida* Willem (Collembola, Isotomidae), from in-house culture, 11 to 12 day old juveniles.

Test design: 28-day exposure in treated artificial soil; the test item solutions were mixed homogeneously into the soil, deionised water was added for the control group and the treated soil was filled in glass vessels before the springtails were introduced on top of the soil.

8 test item groups with 4 replicates each; 1 control group with 8 replicates; 10 juvenile springtails per replicate.

Assessment of mortality and reproductive output after 28 days of exposure.

Endpoints: LOEC (Lowest Observed Effect Concentration for reproductive output and additionally mortality) and NOEC (No Observed Effect Concentration for reproductive output and additionally mortality); LC₅₀ (Lethal Concentration for 50 % adults mortality); EC₁₀, EC₂₀ and EC₅₀ (Effect Concentration for 10, 20, 50 % effect on reproductive output), where possible.

Test item concentrations: Control (deionised water), 1.63, 2.94, 5.29, 9.53, 17.15, 30.86, 55.56 and 100.00 mg test item/kg soil dry weight, equivalent to 0.70, 1.25, 2.25, 4.06, 7.30, 13.15, 23.66 and 42.59 mg spinosad/kg soil dry weight.

Test conditions: Artificial soil with 5 % sphagnum peat content; soil pH 5.85 to 6.39 at test initiation (target: 6.0 ± 0.5) and pH 6.07 to 6.31 at test termination; water content 17.27 % of soil dry weight (45 % of WHC), temperature during exposure: 19.88 to 23.97 °C (target: 20 ± 2 °C). (A short peak above 22 °C was measured for a period < 1 hour, not affecting the outcome of the study). 16:08 light-dark cycles, 436.7 to 708.1 lux (target: 400 to 800 lux).

Statistics: Calculation of treatment means and standard deviations. Fisher's Exact Binomial Test with Bonferroni Correction test ($\alpha=0.05$, one-sided) was used for comparison of mortality data of the test item treatments and the control group. The LC₅₀ was estimated according to the observed mortalities. Reproductive output data were tested for normality and homoscedasticity using Shapiro-Wilk Test and Cochran's Test, respectively. The Williams multiple sequential t-test ($\alpha=0.05$, one-sided) was used for hypothesis testing, since a monotone response was obtained. The effective concentrations EC₁₀, EC₂₀ and EC₅₀ values were determined by Probit analysis, using maximum likelihood regression.

Results and discussions

Regarding physiological symptoms and behavioural abnormalities, a gradual size decrease of the adult individuals as well as symptoms of affectation to the movement were observed from treatment T5.

Compared to the control group, no statistically significant increase in mortality of *Folsomia candida* was detected at any of the test item concentrations after 28 days of exposure. Accordingly, the NOEC and LOEC for adult mortality were estimated as ≥ 100.00 mg test item/kg soil dry weight and as > 100.00 mg test item/kg soil dry weight, respectively. The LC₅₀ was determined to be > 100.00 mg test item/kg soil dry weight, according to the results.

Statistically significant reduction in the number of juveniles compared to the control group was found from the test item concentration 2.94 up to 100.00 mg/kg soil dry weight. Accordingly, the NOEC and LOEC for reproduction were determined as 1.63 mg test item/kg soil dry weight and 2.94 mg test item/kg soil dry weight, respectively.

The EC₁₀, EC₂₀ and EC₅₀ for reproductive output were determined as 2.31, 3.70 and 9.13 mg test item/kg soil dry weight, respectively.

Results of mortality and reproductive output of *Folsomia candida* summarised:

Treatment group	Test item concentration [mg/kg sdw]	Mean mortality [%]	Corrected mortality a [%]	Mean number of juveniles per replicate	Reproductive output CV [%]	% Reduction relative to Control [%]
Control	0.00	6.25	--	598.25	21.16	--
T1	1.63	10.00	4.00	553.00	14.38	7.56
T2	2.94	0.00	-6.67	498.75 b	7.11	16.63
T3	5.29	5.00	-1.33	460.75 b	15.19	22.98
T4	9.53	10.00	4.00	256.67 b	38.70	57.10
T5	17.15	15.00	9.33	148.75 b	13.77	75.14
T6	30.86	10.00	4.00	111.75 b	62.14	81.32
T7	55.56	20.00	14.67	7.50 b	93.97	98.75
T8	100.00	10.00	4.00	4.75 b	146.24	99.21

sdw: soil dry weight; CV: coefficient of variation, T: Treatment

a Corrected mortality according to the Abbott's formula according to Schneider-Orelli (1947). Negative values indicate lower mortality compared to the control.

b Statistically significantly different compared to the control group (Williams Multiple Sequential t-test procedure, $\alpha=0.05$, one-sided).

Endpoints	[mg test item/kg soil dry weight]	[mg spinosad/kg soil dry weight] a	NW 95% CL b
LOEC mortality	> 100.00	> 42.59	--
NOEC mortality	$\geq$ 100.00	$\geq$ 42.59	--
LC ₅₀ (lower / upper CL)	> 100.00	> 42.59	--
LOEC reproductive output	2.94	1.25	--
NOEC reproductive output	1.63	0.70	--
EC ₁₀ (lower / upper CL)	2.31 (1.61 / 2.99)	0.98 (0.69 / 1.27)	0.60
EC ₂₀ (lower / upper CL)	3.70 (2.83 / 4.52)	1.58 (1.21 / 1.93)	0.46
EC ₅₀ (lower / upper CL)	9.13 (7.85 / 10.61)	3.89 (3.34 / 4.52)	0.30

a Based on the analysed active ingredient content and density from the test item CoA: 460 g a.i./L; 1.08.

b 95 % confidence limits normalised width

Reference: The toxic reference item Boric Acid was tested in a separate study (S21-04874) done in June-July 2021. The EC₅₀ for reproductive output was determined as 91.40 mg Boric Acid/kg soil dry weight. This is within the target of about 100 mg/kg soil dry weight given by the OECD guideline 232 (2016) and also within the expected range according to historical facility data.

Conclusion

In the control group, 6.25 % mortality (≤ 20 %) and an acceptable reproductive capacity with a mean of 598.25 juveniles per replicate (≥ 100 juveniles per vessel) were observed at the end of the test. Furthermore, the coefficient of variation for the number of juveniles was 21.16 %, less than 30 %. Therefore, the validity criteria were fulfilled and the study is considered valid.

The NOEC and LOEC for adult mortality were determined as ≥ 100.00 mg test item/kg soil dry weight and > 100.00 mg test item/kg soil dry weight, respectively (equivalent to ≥ 42.59 and > 42.59 mg spinosad/kg soil dry weight, respectively).

The LC₅₀ for mortality was estimated to be > 100.00 mg test item/kg soil dry weight, according to the results (equivalent to > 42.59 mg spinosad/kg soil dry weight).

The NOEC and LOEC regarding reproductive output of *Folsomia candida* were determined as 1.63 mg test item/kg soil dry weight and as 2.94 mg test item/kg soil dry weight, (equivalent to 0.70 and 1.25 mg spinosad/kg soil dry weight, respectively).

The EC₁₀ was determined as 2.31 mg test item/kg soil dry weight (equivalent to 0.98 mg active ingredient/kg soil dry weight, 95 % confidence limits normalised width: 0.60).

The EC₂₀ was determined as 3.70 mg test item/kg soil dry weight (equivalent to 1.58 mg active ingredient/kg soil dry weight, 95 % confidence limits normalised width: 0.46).

The EC₅₀ was determined as 9.13 mg test item/kg soil dry weight (equivalent to 3.89 mg active ingredient/kg soil dry weight, 95 % confidence limits normalised width: 0.30).

Validity criteria

All required validity criteria were met. Accordingly, the study is regarded as valid.

Study validity check for parameters in the control group:

Parameter	Required	Observed
Mean mortality of adults	≤ 20 %	6.25 %
Mean number of juveniles per vessel	≥ 100	598.25
Coefficient of variation of reproductive output	< 30 %	21.16 %

A 2.4.2.1.2 Study 2

Comments of zRMS:	The study was conducted to OECD 226 the guideline and according to the principles of GLP. All validity criterions were met. The study is considered to be reliable and suitable for the risk assessment.
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Reference:	KCP 10.4.2.1/02
Report	Spinosad 480 g/L SC: Effects on the Reproductive Output of the Predatory Soil Mite <i>Hypoaspis (Geolaelaps) aculeifer</i> Canestrini (Acari: Laelapidae) in Artificial Soil, Lozano, J., 2021, Report No S21-04369
Guideline(s):	OECD 226 (2016): OECD Guidelines for the testing of chemicals, No. 226; Predatory mite (<i>Hypoaspis (Geolaelaps) aculeifer</i>) reproduction test in soil.
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	N/A

Objective

The objective of the study was to determine the effects of the test item Spinosad 480 g/L SC in soil on the reproductive output and, additionally, on the mortality of the predatory soil mite species *Hypoaspis (Geolaelaps) aculeifer* Canestrini (Acari: Laelapidae) under worst-case exposure conditions. For this purpose, the Lowest Observed Effect Concentration (LOEC) for mortality and reproductive output, the No Observed Effect Concentration (NOEC) for mortality and reproductive output, the median Lethal Concentration (LC₅₀) for adult mortality, and the Effect Concentration (EC_{10, 20, 50}) for reproductive output were determined, where possible.

Materials and methods

Test item: Spinosad 480 g/L SC; batch code: D-CEA; active ingredient: spinosad, content according to the certificate of analysis: 460 g/L; density: 1.08 g/cm³; expiry date: March 2023.

Test organisms: *Hypoaspis aculeifer* Canestrini (Acari, Laelapidae), from in-house culture, adult mites (32 days after the start of the egg-laying for synchronisation).

Test design: Adult females were exposed to the test item in artificial soil. After 14 days, the surviving individuals were extracted from the test units. The number of juveniles per test unit and, additionally, the number of surviving adult females were determined. The reproductive output and the mortality in each test item group were compared to that of the control group. A concentration-response test with 8 different test item concentrations and 4 replicates each as well as a water control (without test item) with 8 replicates was performed; 10 adult females were exposed per replicate.

Test item concentrations: 0 (control), 1.74, 3.13, 5.63, 10.14, 18.26, 32.86, 59.15 and 106.47 mg test item/kg soil dry weight. Equivalent to: 0 (control) 0.74, 1.33, 2.40, 4.32, 7.78, 14.00, 25.19 and 45.35 mg spinosad/kg soil dry weight.

Endpoints: LOEC (Lowest Observed Effect Concentration) and NOEC (No Observed Effect Concentration) for mortality and reproductive output; LC₅₀ (median Lethal Concentration) for adult mortality and EC_{10, 20, 50} (Effect Concentration of 10, 20, 50 %) for reproductive output, where possible.

Test conditions: Artificial soil with 5 % peat content; soil pH 5.55 to 5.81 at test initiation and pH 5.83 to 5.89 at test termination; water content at test initiation between 17.79 % and 18.37 % (corresponding to 46.34 and 47.87 % of the WHC_{max}), 16.75 and 17.51 % (corresponding to 43.65 and 45.62 % of the WHC_{max}) at test termination; temperature during exposure: 19.8 °C to 20.4 °C; 16:8 light:dark cycles (long day conditions), and light intensity 498.1 lux to 642.4 lux.

Statistics: Statistical calculations were performed with ToxRat Professional 3.3.0 and Microsoft Office Excel-2016® v.16.0. Calculation of treatment means and standard deviations.

Level of significance $\alpha = 0.05$ for the final statistical comparison tests.

After pre-testing for trend performing a qualitative trend analysis by contrasts using proportions (monotonicity of concentration/response, $\alpha = 0.05$); mortality data of the control group and the test item groups was compared using the Chi² 2 x 2 test with Bonferroni correction ($\alpha = 0.05$, one-sided greater). The median Lethal Concentration (LC₅₀) for adult mortality could not be calculated and was empirically estimated from the results.

After pre-testing for normality of data distribution with Shapiro-Wilk's test ($\alpha = 0.01$), homoscedasticity with Levene's test ($\alpha = 0.01$) and trend performing a trend analysis by contrasts (monotonicity of concentration/response, $\alpha = 0.05$); the reproductive output was analysed using Williams' multiple sequential t-test procedure (one-sided smaller, $\alpha = 0.05$). The EC₁₀, 20, 50 for reproductive output were calculated by Probit analysis using linear max. likelihood regression and their 95 %-confidence limits according to Fieller's theorem.

Dates of work: 08 Jul 2021 – 30 Sep 2021 (first application of non-valid test – last assessment valid test)

Results and discussions

In the control group, the mean mortality of adult females was 7.50 %, the mean number of juveniles per replicate was 308.50 and the coefficient of variation of reproductive output was 10.38 %. Therefore, the validity criteria for the control group were met.

The maximum mortality occurred at two intermediate concentrations, 5.63 and 32.86 mg test item/kg soil dry weight, with a 20 % mortality; 13.51 % mortality corrected for control group. The maximum reduction in reproductive output occurred at the highest concentration tested (106.47 mg test item/kg soil dry weight) with a 90.68 % reduction compared to the control group; the mean number of juveniles was 28.75.

No behavioural abnormalities or any pathological symptoms of the test organisms could be observed in the control group and in any of the test item groups.

The toxic reference item BAS 152 65 I (Dimethoate 400 g/L, EC), a.i. dimethoate, was tested in a separate study (S20-08626, dates of work: 18 Nov 2020 – 04 Dec 2020).

The EC₅₀ for reproductive output was determined to be 3.35 mg a.i./kg soil dry weight (95 %-confidence interval: 2.99 – 3.75 mg a.i./kg soil dry weight). This is within the target range of 3.0 to 7.0 mg a.i./kg soil dry weight given by the OECD test guideline No. 226 (2016) and, hence, acceptable sensitivity of the test system was assured.

Mortality and reproductive output of *Hypoaspis aculeifer* after 14 days of exposure to artificial soil treated with Spinosad 480 g/L SC.

Treatment group	Test item concentration [mg t.i./kg sdw]	Mean mortality [%]	Corrected mortality a [%]	Mean number of juveniles per replicate	CV [%]	Reduction in reproductive output b [%]
C	–	7.50	–	308.50	10.38	–
T1	1.74	2.50	-5.41	285.00	9.15	7.62
T2	3.13	15.00	8.11	291.25	12.92	5.59
T3	5.63	20.00	13.51	248.50 c	15.36	19.45
T4	10.14	5.00	-2.70	227.75 c	15.49	26.18
T5	18.26	0.00	-8.11	144.00 c	20.75	53.32
T6	32.86	20.00	13.51	109.25 c	24.81	64.59
T7	59.15	15.00	8.11	69.50 c	82.15	77.47
T8	106.47	15.00	8.11	28.75 c	36.74	90.68

C: control group; T: test item group; t.i.: test item (Spinosad 480 g/L SC); sdw: soil dry weight;

CV: coefficient of variation.

a Mortality corrected to control group according to Abbott's formula (1925) modified by Schneider-Orelli (1947). Negative values indicate lower mortality compared to control group.

b Reduction in reproductive output compared to control group.

c Significantly reduced compared to control group (Williams' multiple sequential t-test procedure, one-sided smaller, $\alpha = 0.05$).

Conclusion

In a 14-day *Hypoaspis aculeifer* reproduction test in artificial soil for the test item Spinosad 480 g/L SC, all validity criteria were met and the sensitivity of the test organisms was confirmed. Accordingly, the study was deemed valid.

No behavioural abnormalities or any pathological symptoms were observed.

Under the conditions of this study, the resulting endpoints are as presented below.

The LOEC for mortality could not be determined and was estimated to be greater than the highest concentration tested, 106.47 mg test item/kg soil dry weight; corresponding to 45.35 mg spinosad/kg soil dry weight according to the analysed content. Accordingly, the NOEC for mortality was determined to be equal or greater than the highest concentration tested 106.47 mg test item/kg soil dry weight; corresponding to 45.35 mg spinosad/kg soil dry weight according to the analysed content.

The LC₅₀ for adult mortality could not be calculated and was assumed to be greater than the highest concentration tested, 106.47 mg test item/kg soil dry weight; corresponding to 45.35 mg spinosad/kg soil dry weight according to the analysed content.

The LOEC for reproductive output was determined to be 5.63 mg test item/kg soil dry weight; corresponding to 2.40 mg spinosad/kg soil dry weight according to the analysed content. Accordingly, the NOEC for reproductive output was determined to be 3.13 mg test item/kg soil dry weight; corresponding to 1.33 mg spinosad/kg soil dry weight according to the analysed content.

The EC₁₀, EC₂₀ and EC₅₀ for the reproductive output with 95 %-confidence intervals were calculated to be 3.52 [2.36 – 4.71], 6.34 [4.73 – 7.90] and 19.52 [16.64 – 22.95] mg test item/kg soil dry weight; corresponding to 1.50 [1.01 – 2.01], 2.70 [2.01 – 3.36] and 8.31 [7.09 – 9.78] mg spinosad/kg soil dry weight according to the analysed content, respectively.

Endpoints	Concentration	
	Test item [mg t.i./kg sdw]	Active ingredient [mg spinosad/kg sdw] a
LOEC mortality b	> 106.47	> 45.35
NOEC mortality b	≥ 106.47	≥ 45.35
LC ₅₀ c (95 %-confidence interval)	> 106.47 n.d.	> 45.35 n.d.
LOEC reproductive output d	5.63	2.40
NOEC reproductive output d	3.13	1.33
EC ₁₀ e (95 %-confidence interval)	3.52 (2.36 – 4.71)	1.50 (1.01 – 2.01)
EC ₂₀ e (95 %-confidence interval)	6.34 (4.73 – 7.90)	2.70 (2.01 – 3.36)
EC ₅₀ e (95 %-confidence interval)	19.52 (16.64 – 22.95)	8.31 (7.09 – 9.78)

t.i.: test item (Spinosad 480 g/L SC); sdw: soil dry weight; n.d.: not determined.

a Based on the active ingredient content and density from the certificate of analysis (spinosad: 460 g/L; density: 1.08 g/cm³).

b Chi² 2 x 2 test with Bonferroni correction ($\alpha = 0.05$, one-sided greater).

c Empirically estimated from the results.

d Williams' multiple sequential t-test procedure (one-sided smaller, $\alpha = 0.05$).

e Probit analysis using linear max. likelihood regression (95 %-confidence limits according to Fieller's theorem).

Validity criteria

All required validity criteria were met. Accordingly, the study is regarded as valid.

Study validity check for parameters in the control group:

Parameter	Required	Observed
Mean mortality of adult females	≤ 20 %	7.50 %
Mean number of juveniles per replicate	≥ 50	308.50
Coefficient of variation of reproductive output	≤ 30 %	10.38 %

A 2.4.2.2	KCP 10.4.2.2	Higher tier testing
A 2.5	KCP 10.5	Effects on soil nitrogen transformation
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
A 2.6.3	KCP 10.6.3	Extended laboratory studies on non-target plants
A 2.7	KCP 10.7	Effects on other terrestrial organisms (flora and fauna)
A 2.8	KCP 10.8	Monitoring data

Appendix 3 Additional calculations considering the minimum proposed dose rate

Not relevant.