

FINAL REGISTRATION REPORT

Part B

Section 9

Ecotoxicology

Detailed summary of the risk assessment

Product code: CHR/F/PROTAZO

Product name(s): CLARO 375 SC, KAJMAN 375 SC

Chemical active substance(s):

Prothioconazole, 175 g/L

Azoxystrobin, 200 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

(authorization)

Applicant: Innvigo Sp. z o.o.

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Version history

When	What
May 2021	Dossier sent for evaluation
December 2021	Applicant updated dRR on the zRMS request
December 2021	Applicant updated dRR on the zRMS request
January 2022	zRMS finalised evaluation
April 2022	Final version prepared by zRMS after Commenting period

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9 Ecotoxicology (KCP 10)

Review Comments:

This document describes the acceptable use conditions required for registration of CHR/F/PROTAZO 375 SC (CLARO 375 SC, KAJMAN 375 SC), a suspension concentrate formulation (SC) containing 175 g/L prothioconazole and 200 g a.s./ha azoxystrobin for use as a fungicide in winter wheat, winter triticale, spring barley and winter oilseed rape.

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. Any incorrect data or text not evaluated by the zRMS has been crossed out.

9.1 Critical GAP and overall conclusions

Table 9.1-1: Table of critical GAPs

PPP (product name/code):	CHR/F/PROTAZO	Formulation type:	EC ^(a, b)
Active substance 1:	prothioconazol	Conc. of as 1:	175g ^(c)
Active substance 2:	azoxystrobin	Conc. of as 2:	200 ^(c)
Safener:	n/a	Conc. of safener:	conc. ^(c)
Synergist:	n/a	Conc. of synergist:	conc. ^(c)
Applicant:	PUH Chemirol Sp. z o.o.	Professional use:	<input checked="" type="checkbox"/>
Zone(s):	northern/central/southern/interzonal ^(d)	Non professional use:	<input type="checkbox"/>
Verified by MS:	yes/ no		

Field of use: fungicide

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Use -No. *	Membe r state(s)	Crop and/ or situation (crop destination / purpose of crop)	F, Fn, Fn G, Gn, Gn p or I **	Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)	Application				Application rate			PHI (days)	Remarks: e.g. g safener/ synergist per ha, other dose rate expression , dose range (min-max)	zRMS Conclusion (efficacy)						
					Metho d / Kind	Timing / Growth stage of crop & season	Max. number a) per use b) per crop/ season	Min. interval between applicatio ns (days)	L/kg product / ha a) max. rate per appl. b) max. total rate per crop/season	kg as/ha a) max. rate per appl. b) max. total rate per crop/season	Wate r L/ha min / max			Birds	Mammals	Aquatic organisms	Bees	Non-target invertebrates	Soil organisms	Non-target plants
Zonal uses (field or outdoor uses, certain types of protected crops)																				
1	PL	Winter wheat/ Pszenica ozima,, triticum	F	diseases	Spray	Spring BBCH 25-69	a) 1 b) 2	14-28	a) 1.0 b) 2.0	c) 0.200 AZX + 0.175 PROTIO d) 0.400	200- 400	35								

		<i>aestivum</i> (TRZAW), Spring barley/ Jęczmień jary <i>hordeum vulgare</i> (HORVS), Winter Triticale/ Pszenżyto ozimy triticale (TTLWI)								AZX + 0.350 PROTIO									
2	PL	Winter oilseed rape / Rzepak ozimy (BRSNW)	F	Diseases	Spray	Spring BBCH 59-69, the risk of infection , warning	c) 1 d) 1	N/A	e) 1.0 f) 1.0	g) 0.200 AZX + 0.175 PROTIO h) 0.200 AZX + 0.175 PROTIO	200- 400	56							
Interzonal uses (use as seed treatment, in greenhouses (or other closed places of plant production), as post-harvest treatment or for treatment of empty storage rooms)																			
3																			
Minor uses according to Article 51 (field uses)																			
5	PL	Spring Rye	F	diseases	Spray	Spring BBCH 25-69	e) 1 f) 2	14-28	i) 1.0 j) 2.0	k) 0.200 AZX + 0.175 PROTIO l) 0.400 AZX + 0.350 PROTIO	200- 400	35							
6	PL	Spring oilseed rape	F	Diseases	Spray	Spring BBCH 59-69, the risk	g) 1 h) 1	N/A	m) 1.0 n) 1.0	o) 0.200 AZX + 0.175 PROTIO	200- 400	56							

						of infection , warning				p) 0.200 AZX + 0.175 PROTIO									
	PL	Common Sunflower	F	Diseases	Spray	Spring BBCH 18-69, the risk of infection , warning	i) 1 j) 1	N/A	q) 1.0 r) 1.0	s) 0.200 AZX + 0.175 PROTIO t) 0.200 AZX + 0.175 PROTIO	200- 400	56							
	PL	Soya	F	Diseases	Spray	Spring BBCH 12-39 or 50-69, the risk of infection , warning	k) 1 l) 1	N/A	u) 1.0 v) 1.0	w) 0.200 AZX + 0.175 PROTIO x) 0.200 AZX + 0.175 PROTIO	200- 400	56							
	PL	Breadseed poppy	F	Diseases	Spray	Spring BBCH 59-69, the risk of infection , warning	m) 1 n) 1	N/A	y) 1.0 z) 1.0	aa) 0.200 AZX + 0.175 PROTIO bb) 0.200 AZX + 0.175 PROTIO	200- 400	56							
	PL	Mustard	F	Diseases	Spray	Spring BBCH 59-69, the risk of infection , warning	o) 1 p) 1	N/A	cc) 1.0 dd) 1.0	ee) 0.200 AZX + 0.175 PROTIO ff) 0.200 AZX + 0.175 PROTIO	200- 400	56							
	PL	Tobacco (NIOTA)	F	diseases	Spray	Spring BBCH 10-30-89	q) 1 r) 2	14-28	gg) 1.0 hh) 2.0	ii) 0.200 AZX + 0.175	200- 400	n/a							

										jj) 0.400 AZX + 0.350 PROTIO									
	PL	Coniferous / deciduous forest nurseries, Ornamenta l shrubs	F	diseases	Spray	Spring BBCH 10-39 or 50-89, the risk of infection , warning	s) 1 t) 2	14-28	kk) 1.0 ll) 2.0	mm) 0.200 AZX + 0.175 PROTIO nn) 0.400 AZX + 0.350 PROTIO	200- 400	n/a							
	PL	<i>Salix viminalis</i> (SAXVI) Wicker (1SAXG)	F	diseases	Spray	BBCH 10-39 or 50-89, the risk of infection , warning	u) 1 v) 2	14-28	oo) 1.0 pp) 2.0	qq) 0.200 AZX + 0.175 PROTIO rr) 0.400 AZX + 0.350 PROTIO	200- 400	n/a							
	PL	<i>Ornamenta l</i>	F	diseases	Spray	BBCH 10-39 or 50-89, the risk of infection , warning	w) 1 x) 2	14-28	ss) 1.0 tt) 2.0	uu) 0.200 AZX + 0.175 PROTIO vv) 0.400 AZX + 0.350 PROTIO	200- 400	n/a							
Minor uses according to Article 51 (interzonal uses)																			
7																			

* 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

Explanation for column 15 – 21 “Conclusion”

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

**Remarks
table:**

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

9.1.1 Overall conclusions

Table 9.1.4 Metabolites of Azoxystrobin

Metabolite	Molar mass	Chemical structure	Maximum-observed occurrence in compartments	Exposure assessment required due to
R234886	389.4		Soil: 28.8% Water/Sediment: 17.7%	PEC _{gw} PEC _{soil} PEC _{sw}
R401553	213.2		Soil: 17% Water/Sediment: 8.9%	PEC _{gw} PEC _{soil} PEC _{sw}
R402173	333.3		Soil: 17% Water/Sediment: 2.4%	PEC _{gw} PEC _{soil} PEC _{sw}

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)

CHR/F/PROTAZO 375 SC pose no unacceptable risk to birds and mammals used according to the label.

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to Prothioconazole-desthio in pulses (soya) BBCH 40-49 and ornamentals (forestry tree, Salix, Wicker) BBCH 40-49 and tobacco BBCH 10-29.

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to azoxystrobin in pulses (soya) BBCH 40-49 and ornamentals BBCH 40-49 (forestry tree, Salix, Wicker) and tobacco BBCH 10-29. Thus, further assessment is required.

The all other TER values exceed the relevant triggers indicating that CHR/F/PROTAZO does not pose an unacceptable risk to mammals following applications according to recommended and accepted use pattern.

9.1.1.2 Effects on aquatic organisms (KCP 10.2)

CHR/F/PROTAZO 375 SC pose no unacceptable risk to aquatic species according to the label with appropriate buffer zone.

9.1.1.3 Effects on bees (KCP 10.3.1)

CHR/F/PROTAZO 375 SC pose no unacceptable risk to bees according to the label

9.1.1.4 Effects on arthropods other than bees (KCP 10.3.2)

CHR/F/PROTAZO 375 SC pose no unacceptable risk to bees according to the label

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

CHR/F/PROTAZO 375 SC pose no unacceptable risk to bees according to the label

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

CHR/F/PROTAZO 375 SC pose no unacceptable risk to bees according to the label

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

Not relevant

9.1.2 Grouping of intended uses for risk assessment

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

Table 9.1-2: Critical use pattern of CHR/F/PROTAZO grouped according to crop, application rate, number of applications, timing criterion

Grouping according to crop, application rate, number of applications, timing criterion			
Group	Intended-uses	relevant-use-parameters for grouping	relevant parameter or value for sorting
	cereals BBCH 25-69 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	oilseed rape BBCH 59-69 1x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Spring rye (BBCH 25-69) 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Spring oil seed rape (BBCH 59-69) 1x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Sunflower (BBCH 18-69) 1x 813.04 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Soya (BBCH 12-69) 1x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Breadseed poppy (BBCH 59-69) 1x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Mustard	crop, application rate, number	crop, application rate, number

Grouping according to crop, application rate, number of applications, timing criterion			
Group	Intended uses	relevant use parameters for grouping	relevant parameter or value for sorting
	(BBCH 59-69) 1 x 1121 g [prod]/ha	of applications, timing criterion	of applications, timing criterion
	Tobacco (BBCH 10-89) 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Coniferous/ deciduous forest nurseries, Ornamental shrubs (BBCH 10-89) 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	<i>Salix viminalis</i> , Wicker (BBCH 10-89) 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
	Ornamental (BBCH 10-89) 2 x 1121 g [prod]/ha	crop, application rate, number of applications, timing criterion	crop, application rate, number of applications, timing criterion
Review comments: The grouping of the intended uses of CHR/F/PROTAZO provided by the Applicant in Table 9.1-2 was to very general, therefore for clarity of the assessment zRMS updated critical GAP. As application of product to fruiting vegetables is limited to greenhouses, additional assessment was not required.			
Group	Intended uses	relevant use parameters for grouping	relevant parameter or value
Terrestrial vertebrates (Birds and Mammals; 9.2 and 9.3)	According to GAP	Scenarios according to EFSA Birds and Mammals Guidance (2009)	Crop, application rate, number of applications, timing criterion
Aquatic organisms (9.5)	According to GAP	Crops according to FOCUS surface water guidance (2015) ¹	FOCUS modelling, for details see Part B 8
Bees (9.6)	Generic risk envelope covering all product uses	Risk assessments are based on the maximum single application rate	Maximum single application rate
Terrestrial non-target arthropods other than bees (9.7)	According to GAP In-field	In-field and off-field risk assessments are based on the maximum application rate for each type of crops	Application rate and number of uses
	According to GAP Off-field		Crop type (height), application rate and number of uses
Soil meso- and macrofauna / soil microorganisms (9.8 and 9.9)	Generic risk envelope covering all product uses	Risk assessments are based on maximum PECsoil	Worst case PECsoil value taken from Section 8 (Environmental Fate)
Non-target terrestrial plants (9.10)	According to GAP	Risk assessments are based on the maximum single application rate for each type of crops	Application rate and drift rate

¹ FOCUS (2015): Generic guidance for FOCUS surface water Scenarios. Version 1.4.

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 CHR/F/PROTAZO is indicated in the table.

Table 9.1-3 Metabolites of prothioconazole

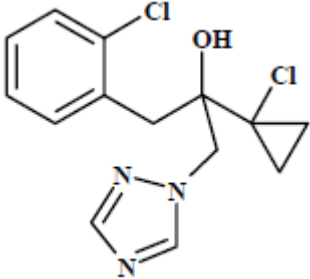
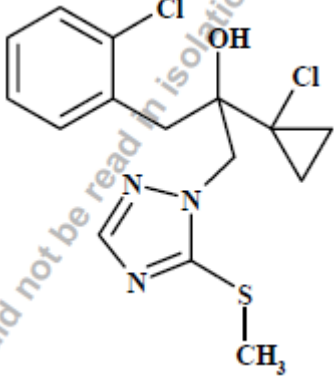
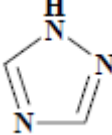
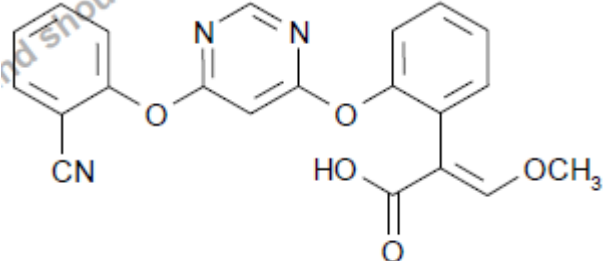
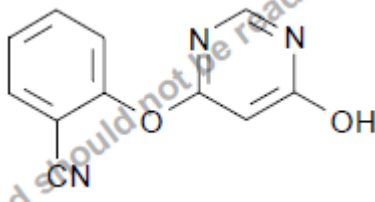
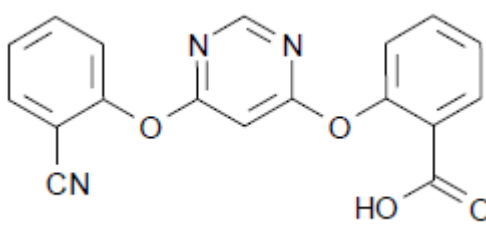
Metabolite	Chemical structure	Molar mass	Maximum occurrence in compartments	Risk assessment required?
Prothioconazole-desthio		312.2	Soil: 14.6%	PECgw PECsoil PECsoil
Prothioconazole-S-methyl		358.8	Soil: 57.1% Water: 32.2% Sediment: 26.9%	PECgw PECsoil PECsw
1,2,3-triazole		69.065	Water/Sediment: 32.7%	PECsw

Table 9.1-4 Metabolites of Azoxystrobin

Metabolite	Molar mass	Chemical structure	Maximum observed occurrence in compartments	Exposure assessment required due to
R234886	389.4		Soil: 28.8% Water/Sediment: 17.7%	PECgw PECsoil PECsw

Metabolite	Molar mass	Chemical structure	Maximum observed occurrence in compartments	Exposure assessment required due to
R401553	213.2		Soil: 17% Water/Sediment: 8/9%	PEC _{gw} PEC _{soil} PEC _{sw}
R402173	333.3		Soil: 17% Water/Sediment: 2.4%	PEC _{gw} PEC _{soil} PEC _{sw}

9.2 Effects on birds (KCP 10.1.1)

9.2.1 Toxicity data

Avian toxicity studies have been carried out with Prothioconazole, azoxystrobin and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents. Effects on birds of CHR/F/PROTAZO were not evaluated as part of the EU assessment of prothioconazole and azoxystrobin.

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

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

Species	Substance	Exposure System	Results	Reference
Bobwhite quail	Prothioconazole	Acute	LD ₅₀ > 2000 mg a.s./kg bw	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Prothioconazole	5 d dietary	LC ₅₀ > 5000 mg a.s./kg diet calc. LD ₅₀ > 1413 mg a.s./kg bw/day**	EFSA Scientific Report (2007) 106, 1-98
Mallard duck	Prothioconazole	5d dietary	LC ₅₀ > 5000 mg a.s./kg diet calc. LD ₅₀ > 2547 mg a.s./kg bw/day	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Prothioconazole	Reproduction 22 w dietary	NOEC ≥ 1000 mg a.s./kg diet calc. NOEL ≥ 86 mg a.s./kg bw/day	EFSA Scientific Report (2007) 106, 1-98

Species	Substance	Exposure System	Results	Reference
Mallard duck	Prothioconazole	Reproduction 21 w dietary	NOEC= 700 mg a.s./kg diet calc. NOEL = 78 mg a.s./kg bw/day	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Prothioconazole- desthio	Acute	LD50 > 2000 mg p.m./kg b.w	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Prothioconazole- desthio	5 d dietary	LC50 = 4090 mg p.m./kg diet calc. LD50 > 297 mg p.m./kg bw/d	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Prothioconazole- desthio	Reproduction 20 w dietary	NOEC = 173 mg p.m./kg diet calc. NOEL= 14.8 mg p.m./kg bw/day	EFSA Scientific Report (2007) 106, 1-98
Mallard duck	Prothioconazole- desthio	Reproduction 20 w dietary	NOEC ≥ 500 mg p.m./kg diet calc. NOEL = 63 mg p.m./kg bw/day	EFSA Scientific Report (2007) 106, 1-98
Bobwhite quail	Azoxystrobin	Acute oral	LD50 > 2000 mg a.s./kg/bw	EFSA Journal 2010; 8(4):1542
Bobwhite quail	Azoxystrobin	Short-term	LC50> 5200 1179 mg a.s./kg bw/d*	EFSA Journal 2010; 8(4):1542
Bobwhite quail	Azoxystrobin	Long-term toxicity and reproduction	NOEL= 1200 117 mg a.s/ka bw/d*	EFSA Journal 2010; 8(4):1542

*there is an error in the EFSA conclusion (2010)

** Endpoint used in the RA as represent worst case

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). To achieve a concise risk assessment, the risk envelope approach is applied.

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

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

Table 9.2-2: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in cereals, tobacco

Intended use	Prothioconazole
Active substance/product	
Application rate (g/ha)	
Acute 5 d dietary toxicity (mg/kg bw)	

TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.2	33.35	42.4	
Reprod. toxicity (mg/kg bw/d)		78				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	64.8	0.74	8.41	9.3	

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 9.2-3: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use		Prothioconazole				
Active substance/product						
Application rate (g/ha)						
Acute 5 d dietary toxicity (mg/kg bw)						
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀	TER _a	
Growth stage				(mg/kg bw/d)		
Screening step	Small omnivorous bird	158.8	1.0	27.79	50.8	
Reprod. toxicity (mg/kg bw/d)		78				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m ×	DDD _m	TER _{lt}	
Growth stage			TWA	(mg/kg bw/d)		
Screening step	Small omnivorous bird	64.8	0.53	6.01	13	

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 9.2-4: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in sunflower, soya

Intended use						
Active substance/product		Prothioconazole				
Application rate (g/ha)		1 x 175				
Acute 5 d dietary toxicity (mg/kg bw)		1413				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV₉₀	MAF₉₀	DDD₉₀ (mg/kg bw/d)	TER_a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.0	27.79	50.8	

Reprod. toxicity (mg/kg bw/d)		78			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small omnivorous bird	64.8	0.53	6.01	13

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 9.2-5: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in soya

Intended use					
Active substance/product		Prothioconazole			
Application rate (g/ha)		1 x 175			
Acute 5 d dietary toxicity (mg/kg bw)		1413			
TER criterion		10			
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small omnivorous bird	158.8	1.0	27.79	50.8
Reprod. toxicity (mg/kg bw/d)		78			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small omnivorous bird	64.8	0.53	6.01	13

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 9.2-6: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in ornamental, tobacco, forestry tree, Salix, Wicker

Intended use					
Active substance/product		Prothioconazole			
Application rate (g/ha)		2 x 175			
Acute 5 d dietary toxicity (mg/kg bw)		1413			
TER criterion		10			
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small insectivorous bird	46.8	1.2	9.83	143.8
Reprod. toxicity (mg/kg bw/d)		78			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}

Screening step	Small insectivorous bird	18.2	0.64	2.36	33.0
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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 9.2-7: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in winter cereals, tobacco

Intended use					
Active substance/product		Prothioconazole-desthio			
Application rate (g/ha)		2 × 175			
Acute 5 d dietary toxicity (mg/kg bw)		297			
TER criterion		10			
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small omnivorous bird	158.8	1.2	33.35	8.9
Cereals BBCH ≥ 40	Small omnivorous bird “lark” Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	7.2			196.4
Cereals BBCH 10 - 29	Small omnivorous bird “lark” Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	24.0			58.9
Cereals BBCH 30 -39	Small omnivorous bird “lark” Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	12.0			117.9
Cereals Early (shoots) autumn-winter BBCH 10-29	Large herbivorous bird "goose" Grass + cereals 100% cereal shoots	30.5			46.4
Tobacco BBCH 10-29	Medium granivorous bird “gamebird”	6.6	1.2	1.39	214
Tobacco BBCH 30-39	Medium granivorous bird “gamebird”	3.3		6.93	42.6
Tobacco BBCH ≥ 40	Medium granivorous bird “gamebird”	1.6		0.34	874
Tobacco BBCH 10-19	Small insectivorous bird “thrust”	10.5		2.21	134
Tobacco BBCH 10-29	Small omnivorous bird “lark”	24.0		5.04	58.9
Tobacco BBCH 30-39	Small omnivorous bird “lark”	12.0		2.52	118
Tobacco BBCH ≥ 40	Small omnivorous bird “lark”	6.0		1.26	236

Tobacco BBCH 10-29	Medium herbivorous bird "pigeon"	55.6		11.68	25.4
Tobacco BBCH 30-39	Medium herbivorous bird "pigeon"	27.8		5.84	50.9
Tobacco BBCH ≥ 40	Medium herbivorous bird "pigeon"	13.9		2.92	102
Tobacco BBCH 10-19	Small insectivorous bird "wagtail"	26.8		5.63	52.8
Tobacco BBCH ≥ 20	Small insectivorous bird "wagtail"	12.6		2.65	112
Reprod. toxicity (mg/kg bw/d)		14.8			
TER criterion		5			
Crop scenario	Indicator/generic focal species	SV_m	MAF_m × TWA	DDD_m (mg/kg bw/d)	TER_{lt}
Screening step	Small omnivorous bird	64.8	0.74	8.41	1.8
Cereals BBCH ≥ 40	Small omnivorous bird "lark" Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	3.3			34.5
Cereals BBCH 10 - 29	Small omnivorous bird "lark" Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	10.9			10.5
Cereals BBCH 30 -39	Small omnivorous bird "lark" Combination (invertebrates with interception) 25% crop leaves 25% weed seeds 50% ground arthropods	5.4			21.1
Cereals Early (shoots) autumn-winter BBCH 10-29	Large herbivorous bird "goose" Grass + cereals 100% cereal shoots	16.2		14.8	7.0
Tobacco BBCH 10-29	Medium granivorous bird "gamebird"	3.0	0.74	0.39	37.9
Tobacco BBCH 30-39	Medium granivorous bird "gamebird"	1.5		0.19	77.9
Tobacco BBCH ≥ 40	Medium granivorous bird "gamebird"	0.8		0.10	148
Tobacco BBCH 10-19	Small insectivorous bird "thrush"	5.7		0.74	20
Tobacco BBCH 10-29	Small omnivorous bird "lark"	10.9		1.40	10.6
Tobacco BBCH 30-39	Small omnivorous bird "lark"	5.4		0.70	21.1
Tobacco BBCH ≥ 40	Small omnivorous bird "lark"	2.7		0.35	42.3
Tobacco BBCH 10-29	Medium herbivorous bird "pigeon"	22.7		2.94	5.03
Tobacco BBCH 30-	Medium herbivorous bird	11.4		1.47	10.1

39	“pigeon”				
Tobacco BBCH \geq 40	Medium herbivorous bird “pigeon”	5.7		0.74	20.1
Tobacco BBCH 10-19	Small insectivorous bird “wagtail”	11.3		1.47	10.1
Tobacco BBCH \geq 20	Small insectivorous bird “wagtail”	4.8		0.62	23.9

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 9.2-8: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use		Prothioconazole-desthio				
Active substance/product						
Application rate (g/ha)						
Acute 5 d dietary toxicity (mg/kg bw)		1 x 175				
TER criterion		297				
		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.0	27.79	10.7	
Reprod. toxicity (mg/kg bw/d)		14.8				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	64.8	0.53	6.01	2.5	
Oilseed rape BBCH ≥ 40	medium herbivorous/granivorous bird "pigeon" Comby to be calculated 50 % crop leaves 50 % weed seeds	0.9			177.3	
Oilseed rape BBCH ≥ 40	Small omnivorous bird “lark” Combination (invertebrates without interception) 25% crop leaves 25% weed seeds 50% ground arthropods	2.7			59.1	
Oilseed rape late – late (with seeds) (BBCH 30-99)	Small insectivorous bird "dunnock) ground invertebrates with interception 100% soil dwelling invertebrates	2.7			59.1	

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 9.2-9: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in sunflower, soya

Intended use						
Active substance/product		Prothioconazole-desthio				

Application rate (g/ha)		1 x 175				
Acute 5 d dietary toxicity (mg/kg bw)		297				
TER criterion		10				
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Screening step	Small omnivorous bird	158.8	1.0	27.79	10.7	
Reprod. toxicity (mg/kg bw/d)		14.8				
TER criterion		5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Screening step	Small omnivorous bird	64.8	0.53	6.01	2.5	
Sunflower Early (Germination/ leaf development) BBCH 00-19	Small insectivorous bird “wagtail” Combination (ground invertebrates without interception) 50% ground arthropods, 50% foliar arthropods	11.3			14.1	
Sunflower Early (Germination/ leaf development) BBCH 00-19	Small omnivorous bird “lark” Combination (invertebrates without interception) 25% crop leaves 25% weed seeds 50% ground arthropods	10.9			14.6	
Sunflower Late (Flowering, seed ripening) BBCH 61- 92	Small granivorous/insectivorous bird “bunting” Small seeds 100% crop seeds	10.0			16	

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 9.2-10: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in soya

Intended use		Prothioconazole-desthio				
Active substance/product						
Application rate (g/ha)						
Acute 5 d dietary toxicity (mg/kg bw)						
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.0	27.79	10.7	
Reprod. toxicity (mg/kg bw/d)		14.8				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	64.8	0.53	6.01	2.5	

Pulses BBCH ≥ 20	Small insectivorous bird "wagtail" ground invertebrates with interception 50% ground arthropods, 50% foliar arthropods	9.7			16.5
Pulses BBCH ≥ 50	Small granivorous bird "finch" Small seeds 100% weed seeds	3.4			46.9
Pulses BBCH ≥ 50	Small omnivorous bird "lark" Combination (invertebrates without interception) 25% crop leaves 25% weed seeds 50% ground arthropods	3.3			48.4
Pulses BBCH 10 - 19	Small insectivorous bird "wagtail" ground invertebrates without interception 50% ground arthropods, 50% foliar arthropods	11.3			14.1
Pulses BBCH 10 - 49	Small granivorous bird "finch" Small seeds 100% weed seeds	11.4			14.0
Pulses BBCH 10 - 49	Small omnivorous bird "lark" Combination (invertebrates without interception) 25% crop leaves 25% weed seeds 50% ground arthropods	10.9			14.6
Pulses Leaf development BBCH 10-19	medium herbivorous/granivorous bird "pigeon" Non-grass herbs 100% leaves	22.7			7.0

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 9.2-11: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in ornamental, tobacco, forestry tree, Salix, Wicker

Intended use		Prothioconazole-desthio				
Active substance/product						
Application rate (g/ha)						
Acute 5 d dietary toxicity (mg/kg bw)						
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small insectivorous bird	46.8	1.2	9.83	30.2	
Reprod. toxicity (mg/kg bw/d)		14.8				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	18.2	0.74	2.36	6.3	

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 9.2-12: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in winter cereals, tobacco

Intended use						
Active substance/product		Azoxystrobin				
Application rate (g/ha)		2 × 200				
Acute toxicity (mg/kg bw)		2000				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.2	38.11	52.5	
Reprod. toxicity (mg/kg bw/d)		2000 117				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	64.8	0.74	9.62	20.8 12.2	

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 9.2-13: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppu, sunflower, soya

Intended use						
Active substance/product		Azoxystrobin				
Application rate (g/ha)		1 x 200				
Acute toxicity (mg/kg bw)		2000				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small omnivorous bird	158.8	1.0	31.76	63	
Reprod. toxicity (mg/kg bw/d)		2000 117				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small omnivorous bird	54.8 64.8	0.53	6.87	29.4 17.0	
Oilseed rape BBCH ≥ 40	medium herbivorous/granivorous bird "pigeon" Comby to be calculated 50 % crop leaves 50 % weed seeds	0.9			2096.4	
Oilseed rape BBCH ≥ 40	Small omnivorous bird "lark" Combination (invertebrates without interception) 25% crop	2.7			698.8	

	leaves 25% weed seeds 50% ground arthropods				
Oilseed rape late late (with seeds) (BBCH 30-99)	Small insectivorous bird "duncock" ground invertebrates with interception 100% soil dwelling invertebrates	2.7			698.8

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 9.2-14: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in sunflower, soya

Intended-use					
Active substance/product		Azoxystrobin			
Application rate (g/ha)		1 x 200			
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
Screening step	Small omnivorous bird	158.8	1.0	31.76	63
Reprod. toxicity (mg/kg bw/d)		1200			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m x TWA	DDD _m (mg/kg bw/d)	TER _t
Screening step	Small omnivorous bird	64.8	0.53	6.87	29.1
Sunflower Early (Germination/leaf development) BBCH 00-19	Small insectivorous bird "wagtail" Combination (ground invertebrates without interception) 50% ground arthropods 50% foliar arthropods	14.3			167.0
Sunflower Early (Germination/leaf development) BBCH 00-19	Small omnivorous bird "lark" Combination (invertebrates without interception) 25% crop leaves 25% weed seeds 50% ground arthropods	10.9			173.1
Sunflower Late (Flowering, seed ripening) BBCH 61- 92	Small granivorous/insectivorous bird "bunting" Small seeds 100% crop seeds	10.0			188.7

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 9.2-15: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in soya

Intended-use	
Active substance/product	Azoxystrobin

Application rate (g/ha)	1 x 200				
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
Screening step	Small omnivorous bird	158.8	1.0	31.76	63
Reprod. toxicity (mg/kg bw/d)	1200				
TER criterion	5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small omnivorous bird	64.8	0.53	6.87	39.1

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 9.2-16: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in ornamental, tobacco, forestry tree, Salix, Wicker

Intended use					
Active substance/product	Azoxystrobin				
Application rate (g/ha)	2 x 200				
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
Screening step	Small insectivorous bird	46.8	1.2	11.23	178.1
Reprod. toxicity (mg/kg bw/d)	1200 117				
TER criterion	5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small insectivorous bird	18.2	0.74	2.70	74 43.3

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.

Combined risk assessment for CHR/F/PROTAZO mixture for prothioconazole and azoxystrobin (worst case)

A TER_{mix} was calculated with the following formula:

$$TER(mix) = \left(\sum_i \frac{1}{TER(a.s._i)} \right)^{-1}$$

where:

TER_(a.s._i) = calculated TER for the active substance *i*

	TER _{prothioconazole}	TER _{azoxystrobin}	TER _{mix-birds-acute}	Trigger-value
Acute				
Winter-cereals	42.4	52.5	13	10
Oilseedrape, mustard, breadseed, poppy	50.8	62	21	10
Sunflower	50.8	62	21	10
Soya	50.8	62	21	
Ornamental, forestry—tree, tobacco, Salix, Wicker	142.8	178.1	160	10
Long-term				
Winter-cereals	9.3	20.8	13	12
Oilseed—rape, mustard, breadseed, poppy	12	29.1	13	12
Sunflower	12	29.1	13	12
Soya	12	29.1	13	
Ornamental, Forestry—tree, Tobacco, Salix, Wicker	12	72	12	12

The combined toxicity was based on a concentration addition approach (screening assessment)

Combined toxicity, using the TER value for active substances (the lowest TER)		
Acute: 2 x 0.175 kg PRO /ha + 2 x 0.2 kg AZO/ha	TER(mix) 1/ ((1/42.4) + (1/52.5))	23.5
Chronic: 2 x 0.175 kg PRO /ha + 2 x 0.2 kg AZO/ha	TER(mix) 1/ ((1/9.3) + (1/12.2))	5.3

Conclusion

The calculated TER_{mix} value is higher than the trigger value, indicating CHR/F/PROTAZO does not posses unacceptable acute and long term risk for birds. No further risk refinement is needed.

9.2.2.2 Higher-tier risk assessment

Not required

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 CHR/F/PROTAZO is not a product for spray applications / not intended to be applied on leafy vegetables forming heads or crop plants with comparable water collecting structures at principal growth stage 4 or later, the leaf scenario does not have to be considered.

Puddle scenario

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

With a $K(f)_{oc}$ of 1765, prothioconazole belongs to the group of less/more sorptive substances. To achieve a concise risk assessment, the risk envelope approach is applied. (see 9.1.2).

Prothioconazole			
Effective application rate (g/ha)=	350*		
Acute toxicity (mg/kg bw) =	>1413	quotient =	0.25
Reprod. toxicity (mg/kg bw/d) =	78	quotient =	4.48
Prothioconazole-desthio			
Effective application rate (g/ha)=	350*		
Acute toxicity (mg/kg bw) =	>297	quotient =	1.18
Reprod. toxicity (mg/kg bw/d) =	>14.8	quotient =	23.65

*worst case value

With a $K(f)_{oc}$ of 423, azoxystrobin belongs to the group of less/more sorptive substances. To achieve a concise risk assessment, the risk envelope approach is applied. (see 9.1.2).

Azoxystrobin			
Effective application rate (g/ha)=	400*		
Acute toxicity (mg/kg bw) =	2000	quotient =	0.2
Reprod. toxicity (mg/kg bw/d) =	1200 117	quotient =	0.33 3.4

*worst case value

9.2.2.4 Effects of secondary poisoning

The log P_{ow} of Prothioconazole amounts to 3.82 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Prothioconazole-desthio amounts to 3.04 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Prothioconazole-S-methyl amounts to 4.19 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Azoxystrobin amounts to 2.5 and thus do not exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is not required.

Risk assessment for earthworm-eating birds via secondary poisoning

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 measured/predicted concentrations in soil/porewater / is based on experimental data. To achieve a concise risk assessment, the risk envelope approach is applied.

Table 9.2-17: Assessment of the risk for earthworm-eating birds due to exposure to Prothioconazole via bioaccumulation in earthworms (secondary poisoning) for the intended use in cereals (worse case use from GAP table)

Parameter	Prothioconazole	comments
PEC _{soil} (twa = 21-d) (mg/kg soil)	0.2905 0.3397	Cereals PEC _{accu}
log P_{ow} / P_{ow}	3.82/6606.93	
Koc	1765	Mean
foc	0.02	Default
BCF _{worm}	2.27	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	0.6594 0.77	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	0.6924 0.81	DDD = $PEC_{worm} \times 1.05$
NOEL (mg/kg bw/d)	78	
TER _{lt}	412.65 96.3	

TER values shown in bold fall below the relevant trigger.

Table 9.2-18: Assessment of the risk for earthworm-eating birds due to exposure to Prothioconazole-desthio via bioaccumulation in earthworms (secondary poisoning) for the intended use in cereals (worse case use from GAP table)

Parameter	Prothioconazole-desthio	comments
PEC _{soil} (twa = 21-d) (mg/kg soil)	0.0871 0.0944	Cereals PEC _{accu}
log P_{ow} / P_{ow}	3.04/1096.5	
Koc	575.4	Mean
foc	0.02	Default
BCF _{worm}	1.22	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	0.01045 0.115	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	0.01097 0.12	DDD = $PEC_{worm} \times 1.05$
NOEL (mg/kg bw/d)	14.8	
TER _{lt}	4349 123	

Table 9.2-19: Assessment of the risk for earthworm-eating birds due to exposure to Prothioconazole-S-methyl via bioaccumulation in earthworms (secondary poisoning) for the intended use in cereals (worse case use from GAP table)

Parameter	Prothioconazole-S-methyl	comments
PEC _{soil} (twa = 21 d) (mg/kg soil)	0.0207 0.0214	Cereals PEC _{accu}
log P _{ow} / P _{ow}	4.19/15448.2	
Koc	2556.3	Mean
foc	0.02	Default
BCF _{worm}	3.64	BCF _{worm/soil} = (PEC _{worm,ww} /PEC _{soil,dw}) = (0.84 + 0.012 × P _{ow}) / foc × Koc
PEC _{worm}	0.07535 0.078	PEC _{worm} = PEC _{soil} × BCF _{worm/soil}
Daily dietary dose (mg/kg bw/d)	0.07912 0.082	DDD = PEC _{worm} × 1.05
NOEL (mg/kg bw/d)	7.8	NOEL of the parent was divided by a factor of 10
TER _{lt}	98.58 95.1	

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 / is based on the regulatory acceptable concentration for aquatic organisms as a limit value for admissible concentrations of prothioconazole in water.

Table 9.2-20: Assessment of the risk for fish-eating birds due to exposure to Prothioconazole via bioaccumulation in fish (secondary poisoning) for the intended use in forestry tree (worse case use from GAP table in surface water)

Parameter	Prothioconazole	comments
PEC _{sw} (twa = 21 d) (mg/L)	0.03443 0.00459	Step 1 worst case value
BCF _{fish}	19.7	
BMF	-	biomagnification factor (relevant for BCF ≥ 2000)
PEC _{fish}	0.6783 0.09	PEC _{fish} = PEC _{water} × BCF _{fish}
Daily dietary dose (mg/kg bw/d)	0.1078 0.0144	DDD = PEC _{fish} × 0.159
NOEL (mg/kg bw/d)	78	
TER _{lt}	723 5417	

TER values shown in bold fall below the relevant trigger.

Table 9.2-21: Assessment of the risk for fish-eating birds due to exposure to Prothioconazole via bioaccumulation in fish (secondary poisoning) for the intended use in forestry tree (worse case use from GAP table in surface water)

Parameter	Prothioconazole-desthio	comments
PEC _{sw} (twa = 21 d) (mg/L)	0.06349 0.05884	Step 1 worst case value
BCF _{fish}	65	
BMF	-	biomagnification factor (relevant for BCF ≥ 2000)

Parameter	Prothioconazole-desthio	comments
PEC _{fish}	4.12 0.825	PEC _{fish} = PEC _{water} × BCF _{fish}
Daily dietary dose (mg/kg bw/d)	0.6562 0.61	DDD = PEC _{fish} × 0.159
NOEL (mg/kg bw/d)	14.8	
TER _{lt}	23 24.2	

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

In conclusion, the acute, short term risk and long term to bird from the proposed uses of prothioconazole and azoxystrobin was found acceptable.

Review Comments:

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

All TER values exceed the relevant triggers indicating that CHR/F/PROTAZO 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 of 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 prothioconazole and its relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on mammals of CHR/F/PROTAZO were not evaluated as part of the EU assessment of Prothioconazole.

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

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

Species	Substance	Exposure System	Results	Reference
Rat	Prothioconazole	Acute Oral	LD50 > 6200 mg a.s./kg bw/d	EFSA Scientific Report (2007) 106, 1-98,
Rat	Prothioconazole	Long-term (2-generation), gavage	NOEL _{parental} = 9.7 mg a.s./kg bw/d NOEL _{reproduction} = 95.6 mg a.s./kg bw/d	EFSA Scientific Report (2007) 106, 1-98,
Rat	Prothioconazole-desthio	Acute Oral	LD50 _(female) = 2506 mg p.m./kg bw/d LD50 _(male) = 2806 mg p.m./kg bw/d	EFSA Scientific Report (2007) 106, 1-98,
Mouse	Prothioconazole-desthio	Acute Oral	LD50 _(female) = 3459 mg p.m./kg bw/d LD50 _(male) = 2235 mg p.m./kg bw/d	EFSA Scientific Report (2007) 106, 1-98,
Rat	Prothioconazole-desthio	long-term (2-generation) oral	NOEL _(parental) = 2.5 mg p.m./kg bw/d NOEL _(male) = 10 mg p.m./kg bw/d	EFSA Scientific Report (2007) 106, 1-98,
Rat	Azoxystrobin	Acute	LD50 >5000 mg/kg bw	EFSA Journal 2010; 8(4):1542
Rat	Azoxystrobin	Long term	NOEL= 32 mg a.s/kg bw/d	EFSA Journal 2010; 8(4):1542

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). To achieve a concise risk assessment, the risk envelope approach is applied.

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

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

Table 9.3-2: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in cereals, tobacco (as maize)

Intended use						
Active substance/product		Prothioconazole				
Application rate (g/ha)		2 × 175				
Acute toxicity (mg/kg bw)		6200				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	118.4	1.2	24.86	249.4	
Reprod. toxicity (mg/kg bw/d)		95.6				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small herbivorous mammal	48.3	0.74	6.27	15.24	

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 acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use		Prothioconazole				
Active substance/product						
Application rate (g/ha)						
Acute toxicity (mg/kg bw)		6200				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	118.4	1.0	20.72	299.2	
Reprod. toxicity (mg/kg bw/d)		95.6				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small herbivorous mammal	48.3	0.53	4.48	21.34	

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 9.3-4: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in sunflower, soya

Intended use						
Active substance/product		Prothioconazole				
Application rate (g/ha)		1 × 175				
Acute toxicity (mg/kg bw)		6200				

TER criterion		10				
Crop scenario Growth stage	Indicator/generic focal species		SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small herbivorous mammal		118.4	1.0	20.72	299.2
Reprod. toxicity (mg/kg bw/d)		95.6				
TER criterion		5				
Crop scenario Growth stage	Indicator/generic focal species		SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small herbivorous mammal		48.3	0.53	4.48	21.34

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 9.3-5: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in soya

Intended use						
Active substance/product		Prothioconazole				
Application rate (g/ha)		1 × 175				
Acute toxicity (mg/kg bw)		6200				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	118.4	1.0	20.72	299.2	
Reprod. toxicity (mg/kg bw/d)		95.6				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small herbivorous mammal	48.3	0.53	4.48	21.34	

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 9.3-6: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in ornamental, tobacco, forestry tree, Salix, Wicker

Intended use						
Active substance/product		Prothioconazole				
Application rate (g/ha)		2 × 175				
Acute toxicity (mg/kg bw)		6200				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	136.4	1.2	28.64	216.5	
Reprod. toxicity (mg/kg bw/d)		95.6				
TER criterion		5				

Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small herbivorous mammal	72.3	0.74	9.39	10.18
Ornamentals and nursery BBCH ≥ 50*	Small herbivorous mammal "vole Grass + cereals 100% grass	36.1	0.74	4.69	20.4

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

*calculated for combitox evaluation

Table 9.3-7: First-tier assessment of the acute and long-term/reproductive risk for **birds **mammals** due to the use of CHR/F/PROTAZO in cereals **tobacco** (as maize)**

Intended use					
Active substance/product		Prothioconazole-desthio			
Application rate (g/ha)		2 × 175			
Acute toxicity (mg/kg bw)		2235			
TER criterion		10			
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small herbivorous mammal	118.4	1.2	24.86	89.9
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}
Screening step	Small herbivorous mammal	48.3	0.74	6.27	1.59
Cereals, tobacco BBCH ≥ 20	Small insectivorous mammal "shrew" ground dwelling invertebrates with interception 100% ground arthropods	1.9			40.5
Cereals BBCH ≥ 40	Small herbivorous mammal "vole Grass + cereals 100% grass	21.7			3.5
Cereals, tobacco BBCH ≥ 40	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	2.3			33.5
Cereals, tobacco BBCH 30 - 39	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9			19.7
Cereals, tobacco BBCH 10-29	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			9.9
Tobacco BBCH 10- 19	Small insectivorous mammal "shrew"	4.2		0.55	18.2

Tobacco BBCH 10-29	Small herbivorous mammal "vole"	72.3		9.39	1.06
Tobacco BBCH 30-39	Small herbivorous mammal "vole"	36.1		4.69	2.13
Tobacco BBCH ≥ 40	Small herbivorous mammal "vole"	18.1		2.35	4.25

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 9.3-8: First-tier assessment of the acute and long-term/reproductive risk for **mammals** due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use		Prothioconazole-desthio				
Active substance/product						
Application rate (g/ha)						
Acute toxicity (mg/kg bw)						
TER criterion						
		1 × 175				
		2235				
		10				
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Screening step	Small herbivorous mammal	118.4	1.0	20.72	107.9	
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}	
Screening step	Small herbivorous mammal	48.3	0.74	4.48	2.23	
Oilseed rape All season	Large herbivorous mammal “lagomorph” Non-grass herbs 100% crop leaves	14.3	-	-	7.5	
Oilseed rape BBCH ≥ 20	Small insectivorous mammal "shrew" ground dwelling invertebrates with interception 100% ground arthropods	1.9	-	-	56.7	
Oilseed rape BBCH ≥ 40	Small herbivorous mammal "vole Grass + cereals 100% grass	18.1	-	-	6.0	
Oilseed rape BBCH ≥ 40	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	1.9	-	-	56.7	

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 9.3-9: First-tier assessment of the acute and long-term/reproductive risk for **mammals** due to the use of CHR/F/PROTAZO in sunflower, **soya**

Intended use	
Active substance/product	Prothioconazole-desthio

Application rate (g/ha)		1 × 175				
Acute toxicity (mg/kg bw)		2235				
TER criterion		10				
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Screening step	Small herbivorous mammal	118.4	1.0	20.72	107.9	
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}	
Screening step	Small herbivorous mammal	48.3	0.53	4.48	2.23	
Sunflower BBCH ≥ 20	Small insectivorous mammal “shrew” ground dwelling invertebrates with interception 100% ground arthropods	1.9			56.7	
Sunflower BBCH ≥ 40	Large herbivorous mammal “lagomorph” Non-grass herbs 100% Non-grass herbs	3.6			29.9	
Sunflower BBCH ≥ 40	Small herbivorous mammal "vole Grass + cereals 100% grass	18.1			6.0	
Sunflower BBCH ≥ 40	Small omnivorous mammal “mouse” Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	1.9			56.7	
Sunflower BBCH 10 - 19	Small insectivorous mammal “shrew” ground dwelling invertebrates without interception 100% ground arthropods	4.2			25.7	
Sunflower BBCH 10 - 19	Large herbivorous mammal “lagomorph” Non-grass herbs 100% Non-grass herbs	14.3			7.5	
Sunflower BBCH 10 - 19	Small omnivorous mammal “mouse” Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			13.8	
Sunflower BBCH 20 - 39	Large herbivorous mammal “lagomorph” Non-grass herbs 100% Non-grass herbs	7.2			15.0	
Sunflower BBCH 20 - 39	Small omnivorous mammal “mouse” Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9			27.6	

Table 9.3-10: First-tier assessment of the acute and long-term/reproductive risk for ~~birds~~ mammals due to the use of CHR/F/PROTAZO in soya

Intended use						
Active substance/product		Prothioconazole-desthio				
Application rate (g/ha)		1 × 175				
Acute toxicity (mg/kg bw)		2235				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	118.4	1.0	20.72	107.9	
Reprod. toxicity (mg/kg bw/d)		10				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						
Screening step	Small herbivorous mammal	48.3	0.53	4.48	2.23	
Pulses BBCH ≥ 20	Small insectivorous mammal “shrew” ground dwelling invertebrates with interception 100% ground arthropods	1.9			56.7	
Pulses BBCH ≥ 50	Large herbivorous mammal “lagomorph” Non-grass herbs 100% Non-grass herbs	4.3			25.1	
Pulses BBCH ≥ 50	Small herbivorous mammal "vole Grass + cereals 100% grass	21.7			5.0	
Pulses BBCH ≥ 50	Small omnivorous mammal “mouse” Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	2.3			46.9	
Pulses BBCH 10 - 19	Small insectivorous mammal “shrew” ground dwelling invertebrates without interception 100% ground arthropods	4.2			25.7	
Pulses BBCH 10 - 49	Large herbivorous mammal “lagomorph” Non-grass herbs 100% Non-grass herbs	14.3			7.5	
Pulses BBCH 10 - 49	Small omnivorous mammal “mouse” Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			13.8	
Pulses BBCH 40 -	Small herbivorous mammal	72.3			1.5	

49	"vole Grass + cereals 100% grass			
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Table 9.3-11: First-tier assessment of the acute and long-term/reproductive risk for **birds** **mammals** due to the use of CHR/F/PROTAZO in ornamental, ~~tobacco~~, forestry tree, Salix, Wicker

Intended use		Prothioconazole-desthio				
Active substance/product						
Application rate (g/ha)						
Acute toxicity (mg/kg bw)						
TER criterion		10				
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Screening step	Small herbivorous mammal	136.4	1.2	28.64	78	
Reprod. toxicity (mg/kg bw/d)	10					
TER criterion						
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Screening step	Small omnivorous bird	72.3	0.74	9.39	1.07	
Ornamentals and nursery Application crop directed BBCH ≥ 50	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9			19.7	
Ornamentals and nursery Application crop directed BBCH 10 - 49	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			9.9	
Ornamentals and nursery Application to plant – exposure to underlying ground	Small insectivorous mammal “shrew” Ground dwelling invertebrates with interception 100% ground arthropods	1.9			40.5	
Ornamentals and nursery BBCH ≥ 50	Small herbivorous mammal "vole Grass + cereals 100% grass	36.1			2.1	
Ornamentals and nursery BBCH 40 - 49	Small herbivorous mammal "vole Grass + cereals 100% grass	72.3			1.1	

Table 9.3-12: First-tier assessment of the acute and long-term/reproductive risk for **mammals** due to the use of CHR/F/PROTAZO in cereals, tobacco (as maize)

Intended use						
Active substance/product		Azoxystrobin				
Application rate (g/ha)		2 × 200				
Acute toxicity (mg/kg bw)		5000				

TER criterion		10				
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Screening step	Small herbivorous mammal	118.4	1.2	28.42	176	
Reprod. toxicity (mg/kg bw/d)		32				
TER criterion		5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Screening step	Small herbivorous mammal	48.3	0.74	7.17	4.46	
Cereals, tobacco BBCH ≥ 20	Small insectivorous mammal "shrew" ground dwelling invertebrates with interception 100% ground arthropods	1.9	-	-	113.5	
Cereals BBCH ≥ 40	Small herbivorous mammal "vole Grass + cereals 100% grass	21.7	-	-	9.9	
Cereals, tobacco BBCH ≥ 40	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	2.3	-	-	93.8	
Cereals, tobacco BBCH 30 - 39	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9	-	-	55.3	
Cereals, tobacco BBCH 10-29	Small omnivorous mammal “mouse” Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8	--	-	27.6	
Tobacco BBCH 10-19	Small insectivorous mammal "shrew"	4.2		0.62	51.4	
Tobacco BBCH 10-29	Small herbivorous mammal "vole”	72.3		10.73	3.0	
Tobacco BBCH 30-39	Small herbivorous mammal "vole”	36.1		5.36	6.0	
Tobacco BBCH ≥ 40	Small herbivorous mammal "vole”	18.1		2.69	11.9	

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 9.3-13: First-tier assessment of the acute and long-term/reproductive risk for mammals due to the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use	
Active substance/product	Azoxystrobin
Application rate (g/ha)	1 × 200

Acute toxicity (mg/kg bw)		5000			
TER criterion		10			
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small herbivorous mammal	118.4	1.0	23.68	211.1
Reprod. toxicity (mg/kg bw/d)		32			
TER criterion		5			
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Screening step	Small herbivorous mammal	48.3	0.53	5.12	6.25
Oilseed rape All season	Large herbivorous mammal "lagomorph" Non grass herbs 100% crop leaves	14.3	-	-	21.4
Oilseed rape BBCH ≥ 20	Small insectivorous mammal "shrew" ground dwelling invertebrates with interception 100% ground arthropods	1.9	-	-	158.9
Oilseed rape BBCH ≥ 40	Small herbivorous mammal "vole" Grass + cereals 100% grass	18.1	-	-	16.7
Oilseed rape BBCH ≥ 40	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	1.9	-	-	158.9

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 9.3-14: First-tier assessment of the acute and long-term/reproductive risk for birds mammals due to the use of CHR/F/PROTAZO in sunflower, soya

Intended use		Azoxystrobin			
Active substance/product					
Application rate (g/ha)					
Acute toxicity (mg/kg bw)					
TER criterion		10			
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Growth stage					
Screening step	Small herbivorous mammal	118.4	1.0	23.68	211.1
Reprod. toxicity (mg/kg bw/d)		10			
TER criterion		5			
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}
Growth stage					
Screening step	Small herbivorous mammal	48.3	0.53	5.12	6.25
Sunflower BBCH ≥ 20	Small insectivorous mammal “shrew” ground dwelling invertebrates with interception 100% ground arthropods	1.9			158.9

Sunflower BBCH \geq 40	Large herbivorous mammal "lagomorph" Non-grass herbs 100% Non-grass herbs	3.6			83.9
Sunflower BBCH \geq 40	Small herbivorous mammal "vole" Grass + cereals 100% grass	18.1			16.7
Sunflower BBCH \geq 40	Small omnivorous mammal "mouse" Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	1.9			158.9
Sunflower BBCH 10–19	Small insectivorous mammal "shrew" ground dwelling invertebrates without interception 100% ground arthropods	4.2			71.9
Sunflower BBCH 10–19	Large herbivorous mammal "lagomorph" Non-grass herbs 100% Non-grass herbs	14.3			21.1
Sunflower BBCH 10–19	Small omnivorous mammal "mouse" Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			38.7
Sunflower BBCH 20–39	Large herbivorous mammal "lagomorph" Non-grass herbs 100% Non-grass herbs	7.2			41.9
Sunflower BBCH 20–39	Small omnivorous mammal "mouse" Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9			77.4

Table 9.3-15: First-tier assessment of the acute and long-term/reproductive risk for birds mammals due to the use of CHR/F/PROTAZO in soya

Intended use						
Active substance/product		Azoxystrobin				
Application rate (g/ha)		1 × 200				
Acute toxicity (mg/kg bw)		2235				
TER criterion		10				
Crop scenario	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a	
Growth stage						
Screening step	Small herbivorous mammal	136.4	1.0	27.78	183.3	
Reprod. toxicity (mg/kg bw/d)		10				
TER criterion		5				
Crop scenario	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}	
Growth stage						

Screening step	Small herbivorous mammal	72.3	0.53	7.66	4.18
Pulses BBCH ≥ 20	Small insectivorous mammal "shrew" ground dwelling invertebrates with interception 100% ground arthropods	1.9			158.9
Pulses BBCH ≥ 50	Large herbivorous mammal "lagomorph" Non-grass herbs 100% Non-grass herbs	4.3			70.2
Pulses BBCH ≥ 50	Small herbivorous mammal "vole" Grass + cereals 100% grass	21.7			13.9
Pulses BBCH ≥ 50	Small omnivorous mammal "mouse" Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	2.3			131.3
Pulses BBCH 10 - 19	Small insectivorous mammal "shrew" ground dwelling invertebrates without interception 100% ground arthropods	4.2			71.9
Pulses BBCH 10 - 49	Large herbivorous mammal "lagomorph" Non-grass herbs 100% Non-grass herbs	14.3			21.1
Pulses BBCH 10 - 49	Small omnivorous mammal "mouse" Combination (invertebrates without interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			38.7
Pulses BBCH 40 - 49	Small herbivorous mammal "vole" Grass + cereals 100% grass	72.3			4.2

Table 9.3-16: First-tier assessment of the acute and long-term/reproductive risk for birds due to the use of CHR/F/PROTAZO in ornamental, tobacco, forestry tree, Salix, Wicker

Intended use	Azoxystrobin 2 × 200 5000 10				
Active substance/product					
Application rate (g/ha)					
Acute toxicity (mg/kg bw)					
TER criterion					
Crop scenario Growth stage	Indicator/generic focal species	SV ₉₀	MAF ₉₀	DDD ₉₀ (mg/kg bw/d)	TER _a
Screening step	Small herbivorous mammal	136.4	1.2	32.74	152.7
Reprod. toxicity (mg/kg bw/d)	32				
TER criterion	5				
Crop scenario Growth stage	Indicator/generic focal species	SV _m	MAF _m × TWA	DDD _m (mg/kg bw/d)	TER _{lt}

Screening step	Small herbivorous mammal	72.3	0.64	10.73	2.98
Ornamentals and nursery Application crop directed BBCH ≥ 50	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	3.9			20.2
Ornamentals and nursery Application crop directed BBCH 10 - 49	Small omnivorous mammal "mouse" Combination (invertebrates with interception) 25% weeds 50% weed seeds 25% ground arthropods	7.8			10.1
Ornamentals and nursery Application to plant – exposure to underlying ground	Small insectivorous mammal "shrew" Ground dwelling invertebrates with interception 100% ground arthropods	1.9			41.4
Ornamentals and nursery BBCH ≥ 50	Small herbivorous mammal "vole" Grass + cereals 100% grass	36.1			6.0
Ornamentals and nursery BBCH 40 - 49	Small herbivorous mammal "vole" Grass + cereals 100% grass	72.3			3.0

The combined toxicity was based on a concentration addition approach (screening assessment)

Combined toxicity , using the TER value for active substances (the lowest TER, without pulses BBCH 40-49 and ornamentals BBCH 40-49 and tobacco BBCH 10-29)		
Acute: 2 x 0.175 kg PRO /ha + 2 x 0.2 kg AZO/ha	TER(mix) $1/((1/216.5) + (1/152.7))$	89.5
Chronic: 2 x 0.175 kg PRO /ha + 2 x 0.2 kg AZO/ha	TER(mix) $1/((1/20.4^*) + (1/6^*))$	4.63

*Tier 1 (ornamentals)

Combined risk assessment for CHR/F/PROTAZO mixture for prothioconazole and azoxystrobin (worst case)

A TER_{mix} was calculated with the following formula:

$$TER_{(mix)} = \left(\sum_i \frac{1}{TER_{(a.s._i)}} \right)^{-1}$$

where:

TER_(a.s._i) = calculated TER for the active substance *i*

	TER _{Aprothioconazole}	TER _{Aazoxystrobin}	TER _{mix birds acute}	Trigger value
Acute				
Winter-cereals	219.4	176.0	204	10
Oilseedrape-mustard-broadseed	299.2	211.4	245	40

poppy				
Sunflower	299.2	244.4	245	10
Soya	299.2	244.4	245	
Ornamental, forestry tree, tobacco, Salix, Wicker	216.5	152.2	172	10
Long term				
Winter cereals	15.24	113.5	28	5
		9.9	12	5
		93.8	28	5
		52.3	25	5
		27.6	20	5
Oilseed rape, mustard, breadseed poppy	21.34	6.25	9	5
Sunflower	21.34	6.25	9	5
Soya	21.34	158.9	40	5
		70.2	34	5
		13.9	17	5
		131.3	39	5
		71.9	34	5
		21.1	21	5
		38.7	28	5
		4.2	7	5
Ornamental, Forestry tree, Tobacco, Salix, Wicker	10.18	20.2	6	5
		10.1	10.0	5
		41.4	17	5

		27.4	12	5
		15	9	5

Conclusion

The calculated TER_{mix} value is higher than the trigger value, indicating CHR/F/PROTAZO does not posses unacceptable acute and longterm risk for mammals. No further risk refinement is needed

9.3.2.2 Higher-tier risk assessment

According to DAR Prothioconazole Vol.3 Annex B.9 the field residue studies provided by notifier – 8 trialas- show realistic value of residue unite dose, dissipation time in wheat for Prothioconazole-desthio.. According to this studies, DT₅₀ was reduced to 3.2 days. Therefore, the risk assessment has been refined by applying a new value of MAF and fTWA uses formulas presented below:

- MAF for multiple applications = $(1 - e^{-nki}) / (1 - e^{-ki})$

$$k = \ln 2 / DT_{50}$$

n = number of applications

i = interval between applications (days)

- fTWA = $(1 - e^{-kt}) / kt$

$$k = \ln 2 / DT_{50}$$

t = averaging time (21 days used)

(Default fTWA of 0.53 is used at first tier)

This higher tier risk assessment is presented in the table below.

Crop group	species	SV	App. Rate (kg a.s./ha)	Interval	MAF	fTWA	DDD (mg a.s./kg bw/day)	NOEL (mg a.s./kg bw/day)	TER _{LT}
Cereals, covers Tobacco BBCH ≥40	Small herbivorous mammal "vole Grass + cereals 100% grass	21.7	2 x 0.175	14	1.01	0.22	0.84	10	12
Ornamentals and nursery BBCH 40-49, Tobacco BBCH 10-29	Small herbivorous mammal "vole Grass + cereals 100% grass	72.3	2 x 0.175	14	1.01	0.22	2.81	10	3.56
Ornamentals and nursery BBCH > 50, Tobacco BBCH 30-39	Small herbivorous mammal "vole Grass + cereals 100% grass	36.1	2 x 0.175	14	1.01	0.22	1.4	10	7.14
Pulses 40-	Small	72.3	1x 0.175	14	1	0.22	2.78	10	3.6

49	herbivorous mammal "vole Grass + cereals 100% grass								
Pulses > 50	Small herbivorous mammal "vole Grass + cereals 100% grass	21.7	1 x 0.175	14	1	0.22	0.83	10	12

The refined TER values are above the Regulation (EU) 546/2011 trigger of 5, indicating that long-term risk to herbivorous mammal is acceptable following use of CHR/F/PROTAZO according to the proposed use pattern.

Review Comments:

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to Prothioconazole-desthio in pulses BBCH 40-49 and ornamentals BBCH 40-49 and tobacco BBCH 10-29.

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to azoxystrobin in pulses BBCH 40-49 and ornamentals BBCH 40-49 and tobacco BBCH 10-29. Thus, further assessment is required.

Furthermore, although voles are listed as relevant focal species, it is widely acknowledged that voles are not relevant for arable crops and orchards.

- Gurney, *et al.* (1998) reports the feeding habit of field voles (*Microtus agrestis*) to be mainly rough, ungrazed grassland, including thick grass ground cover. In a two year study of small mammals on Scottish arable land and set-aside (Rodgers 1993) 159 field voles were caught, which were reported to have an almost exclusive preference for rough grassland and were completely absent from the wood and also infrequent in set-aside and crops.
- In a three year study of small mammals on an arable farm in Oxfordshire Tew (1994) failed to capture any field voles away from hedgerows around cereal fields. In the Boxworth project, field voles were occasionally caught in the fields but this was restricted to areas with dense ground cover, such as patches infested with blackgrass (Johnson *et al.*, 1992).
- No data are available from radio tracking studies for the bank vole or the field vole. Radio tracking has been tried unsuccessfully in both species (Plesner-Jensen 1993). Trapping studies have shown that although both species do not use arable fields as main habitat, they are common in hedgerows and woods adjacent to arable fields (Pollard & Relton 1970; Jefferies *et al* 1973; Green 1979; Loman 1991; Johnson *et al.* 1992). The preference of the common vole for non-cropped areas are discussed in Jacob *et al* (2014), in which it states: "The common vole is primarily a grassland species that is well adapted to steppe habitats. Primary habitats are meadows, set-aside land, flower strips, grassy field verges and alfalfa and clover fields. It prefers to inhabit undisturbed short vegetation and can be found in grass leys in forests after clear cuts and other grassy habitats."

Furthermore, information from DEFRA's research project on "Estimating wildlife exposure to pesticides in crops: additional scenarios and data" (2009) supports the non-relevance of the vole. The aim of this work was to provide further information on use of crops by wildlife by extensive surveying and by review of public literature. The following table taken from this report shows the number of captures of small mammals in the various habitat types.

Table 9.2.2.2-1: Captures of small mammals during 11,000 trap-events in different agricultural habitats (Table 3 from Report PS2328)

Captures per 100 trap events

	Potatoes	Arable hedge	Cereal	Sugar beet	Other non- crop	Orchard hedge	Orchard crop
Field vole	0	0.15	0.08	0	0	1.52	1.31
Pygmy shrew	0.02	0.53	0.23	0	0.34	1.82	0.51
Common shrew	0.38	6.43	1.36	1.00	6.38	3.33	1.85
Bank vole	0.02	6.43	1.44	0	1.55	4.24	0.27
Woodmouse	0.82	8.06	7.04	0.50	2.76	7.88	2.49
Total	1.24	21.6	10.15	1.5	11.03	18.79	6.43

Trap events in this habitat	5020	2630	2570	200	580	330	2970
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Although the study did not specifically include the intended crops, the results clearly show that wood mice are much more prevalent in arable crops (including potatoes, cereal and sugar beet) than voles. A follow on research project by DEFRA, on “Small mammal activity in soft fruit, cane fruit and top fruit orchards” (2012), focused only on the activity of wood mice as the key focal species. The report stated that “Although a number of species of small mammals occurred in orchards in PS2328 [DEFRA 2009], wood mice were caught in the greatest numbers and are probably at greatest risk from pesticides applied there. They are omnivorous and forage above ground, while shrews and voles tend to forage beneath thatch and litter layers”.

Additionally, in a study conducted in southern Moravia, Czech Republic, small mammal communities were snap trapped for six years in agricultural landscapes including vineyards (Heroldova et al., 2007) which is a comparable habitat to modern orchards. The dominant species in vineyards was also the wood mouse (*Apodemus sylvaticus*), which comprised 93% of all small mammals trapped in this habitat. The wood mouse was also regularly present in different types of vineyards in southwest Germany (Pedall et al., 2003). In contrast to the common vole, the wood mouse is a ubiquitous and euryoecious non-specialist (Tattersall et al., 1997), inhabiting a wide range of landscapes (Montgomery, 1999), and is found in arable habitats throughout the year (e.g. (Tew & Macdonald, 1994); (Loman, 1991); (Green, 1979); (Kikkawa, 1964); (Bergstedt, 1965)). This species has no specific habitat requirements, yet it tends to avoid habitats with a dense herb layer (Braun & Dieterlen, 2005). Tew et al. (2000) suggests that wood mice, due to their bouncing locomotion and granivorous habits, even favour habitats with partly bare ground over which they can easily travel and find fallen seed.

Moreover, EFSA’s Bird and Mammal Guidance Document (2009) identifies the European rabbit (*Oryctolagus cuniculus*) as the representative species for large herbivorous mammals. This species is abundant across Europe and may be associated with arable crops. Gurney *et al.* (1998) reports the feeding habitat of the rabbit to be areas of short grass; naturally occurring, dry heaths or closely grazed agricultural pastures with secure refuge nearby. The brown hare (*Lepus europaeus*) is also widespread and abundant across Europe.

Taking all of the above into consideration (high fecundity and population recuperation of the vole; primary source of food outside crops fields for the vole; necessity of population control measures since the vole is considered a crop pest when high population levels are reached; other agricultural techniques being also means of population control), voles are not considered to be a relevant focal species. Therefore, the risk assessment for small herbivorous mammals can be concluded to be acceptable (even with TER lower than the trigger value) if it is acceptable for other small omnivorous mammals (wood mouse and brown hare) which are considered as a relevant focal species.

Overall the following risk assessment focuses on the wood mouse (omnivorous) and the brown hare (herbivorous) as relevant focal species for the proposed uses.

Generic focal	FIR/bw	RUD	App. Rate (kg	MAF	Deposition factor	fTWA	DDD (mg	NOEL (mg	TERLT
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species			a.s./ha)				a.s./kg bw/day)	a.s./kg bw/day)	
Ornamental, forestry tree, Salix, Wicker Application: 2 x 175 g prothioconazole-desthio/ha									
Wood mouse Diet: 25% weeds50% weed seeds25% ground arthropods	0.27	29.2	0.175	1.4	1.0	0.53	1.02	10	9.8
Rabbit (100 % plant material)	0.50	28.7	0.175	1.4	1.0	0.53	1.86	10	5.4
Ornamental, forestry tree, Salix, Wicker Application: 2 x 200 g azoxystrobin/ha									
Wood mouse Diet: 25% weeds50% weed seeds25% ground arthropods	0.27	29.2	0.200	1.4	1.0	0.53	1.17	32	27.4
Rabbit (100 % plant material)	0.50	28.7	0.200	1.4	1.0	0.53	2.13	32	15

Review Comments:

The relevance of voles should be considered at the national level.
 The voles are relevant species for Poland. Thus, further assessment is required.

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

With a K(f)oc of 1765, Prothioconazole belongs to the group of less sorptive substances. To achieve a concise risk assessment, the risk envelope approach is applied. (see 9.1.2).

Prothioconazole				
Effective application rate (g/ha)=	350			
Acute toxicity (mg/kg bw) =	6200	quotient	=	0.06
Reprod. toxicity (mg/kg bw/d) =	95.6	quotient	=	3.66
Prothioconazole-desthio				

Effective application rate (g/ha)=	350		
Acute toxicity (mg/kg bw) =	2235	quotient =	0.17
Reprod. toxicity (mg/kg bw/d) =	10	quotient =	35

With a K(f)oc of 483, Azoxystrobin belongs to the group of less sorptive substances. To achieve a concise risk assessment, the risk envelope approach is applied. (see 9.1.2).

Azoxystrobin			
Effective application rate (g/ha)=	400		
Acute toxicity (mg/kg bw) =	5000	quotient =	0.08
Reprod. toxicity (mg/kg bw/d) =	32	quotient =	12.5

9.3.2.4 Effects of secondary poisoning

The log P_{ow} of Prothioconazole amounts to 3.82 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Prothioconazole-desthio amounts to 3.04 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Prothioconazole-S-methyl amounts to 4.19 and thus exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is required.

The log P_{ow} of Azoxystrobin amounts to 2.5 and thus does not exceeds the trigger value of 3. A risk assessment for effects due to secondary poisoning is not required.

Risk assessment for earthworm-eating mammals via secondary poisoning

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 measured/predicted concentrations in soil/porewater / is based on experimental data. To achieve a concise risk assessment, the risk envelope approach is applied. (see 9.1.2).

Table 9.3-17: Assessment of the risk for earthworm-eating mammals due to exposure to Prothioconazole via bioaccumulation in earthworms (secondary poisoning) for the intended use in winter cereals (worse case use from GAP table)

Parameter	Prothioconazole	comments
PEC _{soil} (twa = 21 d) (mg/kg soil)	0.2905 0.3397	Cereals PEC _{accu}
log P _{ow} / P _{ow}	3.82/6606.93	
Koc	1765	Mean
foc	0.02	Default
BCF _{worm}	2.27	BCF _{worm/soil} = (PEC _{worm,ww} /PEC _{soil,dw}) = (0.84 + 0.012 × P _{ow}) / foc × Koc
PEC _{worm}	0.6594 0.77	PEC _{worm} = PEC _{soil} × BCF _{worm/soil}
Daily dietary dose (mg/kg bw/d)	0.8441 0.99	DDD = PEC _{worm} × 1.28
NOEL (mg/kg bw/d)	95.6	
TER _{lt}	44.3 96.6	

TER values shown in bold fall below the relevant trigger.

Table 9.3-18: Assessment of the risk for earthworm-eating mammals due to exposure to Prothioconazole-desthio via bioaccumulation in earthworms (secondary poisoning) for the intended use in winter cereals (worse case use from GAP table)

Parameter	Prothioconazole-desthio	comments
PEC _{soil} ($t_{wa} = 21\text{ d}$) (mg/kg soil)	0.0871 0.0944	Cereals PEC _{accu}
log P _{ow} / P _{ow}	3.04/1096.5	
Koc	575.4	Mean
foc	0.02	Default
BCF _{worm}	1.22	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	0.1063 0.115	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	0.1360 0.147	$DDD = PEC_{worm} \times 1.28$
NOEL (mg/kg bw/d)	10	
TER _{lt}	73 68	

TER values shown in bold fall below the relevant trigger.

Table 9.3-19: Assessment of the risk for earthworm-eating mammals due to exposure to Prothioconazole-S-methyl via bioaccumulation in earthworms (secondary poisoning) for the intended use in winter cereals (worse case use from GAP table)

Parameter	Prothioconazole-S-methyl	comments
PEC _{soil} ($t_{wa} = 21\text{ d}$) (mg/kg soil)	0.0207 0.0214	Cereals PEC _{accu}
log P _{ow} / P _{ow}	4.19/14338.2	
Koc	2556.3	Mean
foc	0.02	Default
BCF _{worm}	3.65	$BCF_{worm/soil} = (PEC_{worm,ww}/PEC_{soil,dw}) = (0.84 + 0.012 \times P_{ow}) / foc \times Koc$
PEC _{worm}	0.07535 0.078	$PEC_{worm} = PEC_{soil} \times BCF_{worm/soil}$
Daily dietary dose (mg/kg bw/d)	0.0967 0.1	$DDD = PEC_{worm} \times 1.28$
NOEL (mg/kg bw/d)	9.56	NOEL of the parent was divided by a factor of 10
TER _{lt}	99 95.6	

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 / is based on the regulatory acceptable concentration for aquatic organisms as a limit value for admissible concentrations of Prothioconazole in water.

Table 9.3-20: Assessment of the risk for fish-eating mammals due to exposure to Prothioconazole via bioaccumulation in fish (secondary poisoning) for the intended use in cereals (worse case use from GAP table)

Parameter	Prothioconazole	comments
PEC _{sw} (tw = 21 d) (mg/L)	0.03443 0.00459	Step 1 worst case value
BCF _{fish}	19.7	
BMF		biomagnification factor (relevant for BCF ≥ 2000)
PEC _{fish}	0.06783 0.09	PEC _{fish} = PEC _{water} × BCF _{fish}
Daily dietary dose (mg/kg bw/d)	0.009631 0.0128	DDD = PEC _{fish} × 0.142
NOEL (mg/kg bw/d)	95.6	
TER _{lt}	9926 7469	

TER values shown in bold fall below the relevant trigger.

Table 9.3-21: Assessment of the risk for fish-eating mammals due to exposure to Prothioconazole-desthio via bioaccumulation in fish (secondary poisoning) for the intended use in cereals (worse case use from GAP table)

Parameter	Prothioconazole-desthio	comments
PEC _{sw} (tw = 21 d) (mg/L)	0.06349 0.05884	Step 1 worst case value
BCF _{fish}	65	
BMF		biomagnification factor (relevant for BCF ≥ 2000)
PEC _{fish}	4.12 3.825	PEC _{fish} = PEC _{water} × BCF _{fish}
Daily dietary dose (mg/kg bw/d)	0.586 0.543	DDD = PEC _{fish} × 0.142
NOEL (mg/kg bw/d)	10	
TER _{lt}	47 18.4	

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

In conclusion, the acute, short term risk and long term to mammals from the proposed uses of prothioconazole was found acceptable.

Review Comments:

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

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to Prothioconazole-dethio in pulses (soya) BBCH 40-49 and ornamentals (forestry tree, Salix, Wicker) BBCH 40-49 and tobacco BBCH 10-29.

The risk assessment conducted at Tier 1 indicates an unacceptable chronic risk to small herbivorous mammal vole exposed to azoxystrobin in pulses (soya) BBCH 40-49 and ornamentals BBCH 40-49 (forestry tree, Salix, Wicker) and tobacco BBCH 10-29. Thus, further assessment is required.

The all other TER values exceed the relevant triggers indicating that CHR/F/PROTAZO does not pose an unacceptable risk to mammals following applications according to recommended and accepted use pattern.

Evaluation of exposing to mammals through the drinking water demonstrated the acceptable risk. The risk of secondary poisoning is low.

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

Not required

9.5 Effects on aquatic organisms (KCP 10.2)

9.5.1 Toxicity data

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

Effects on aquatic organisms of CHR/F/PROTAZO were not evaluated as part of the EU assessment of prothioconazole. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

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

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

Species	Substance	Exposure System	Results	Reference
Oncorhynchus mykiss	Prothioconazole	acute	LC50 = 1.83 mg a.s/L	EFSA Scientific Report (2007) 106,
Lepomis macrochirus	Prothioconazole	acute	LC50= 4.59 mg a.s/L	EFSA Scientific Report (2007) 106,
Cyprinus carpio	Prothioconazole	acute	LC50= 6.91 mg a.s./L	EFSA Scientific Report (2007) 106,

Species	Substance	Exposure System	Results	Reference
Oncorhynchus mykiss	Prothioconazole	chronic	NOEC= 0.308 mg a.s./L	EFSA Scientific Report (2007) 106,
Daphnia magna	Prothioconazole	Acute	EC50= 1.3 mg a.s/L	EFSA Scientific Report (2007) 106,
Daphnia Magna	Prothioconazole	Chronic	NOEC= 0.56 mg a.s./L	EFSA Scientific Report (2007) 106,
Pseudokirchneriella subcapitata	Prothioconazole	Sub-chronic	EC50: 2.18 mg as/ha NOEC= 0.56 mg a.s/L	EFSA Scientific Report (2007) 106,
Chironomus riparius	Prothioconazole	Chronic	NOEC= 9.14 mg a.s/L	EFSA Scientific Report (2007) 106,
Oncorhynchus mykiss	Prothioconazole-desthio	Acute	LC50= 6.63 mg p.m/L	EFSA Scientific Report (2007) 106,
Leuciscus idus melanotus	Prothioconazole-desthio	Acute	LC50= 13.2 mg p.m/L	EFSA Scientific Report (2007) 106,
Oncorhynchus mykiss	Prothioconazole-desthio	Chronic	NOEC= 3.34 µg p.m./L	EFSA Scientific Report (2007) 106,
Daphnia magna	Prothioconazole-desthio	Acute	EC50 > 10 mg p.m./L	EFSA Scientific Report (2007) 106,
Daphnia magna	Prothioconazole-desthio	Chronic	NOEC= 0.10 mg p.m./L	EFSA Scientific Report (2007) 106,
Scenedesmus subspicatus	Prothioconazole-desthio	Sub-chronic	EbC50= 0.073 mg p.m./L ErC50= 0.55 mg p.m./L	EFSA Scientific Report (2007) 106,
Chironomus riparius	Prothioconazole-desthio	Chronic	NOEC= 2.0 mg p.m./L	EFSA Scientific Report (2007) 106,
Oncorhynchus mykiss	1,2,4-triazole	Acute	LC50= 498 mg p.m/L	EFSA Scientific Report (2007) 106,
Oncorhynchus mykiss	1,2,4-triazole	Chronic	NOErC= 3.2 mg a.s./L	EFSA Scientific Report (2007) 106,
Daphnia magna	1,2,4-triazole	Acute	EC50= 900 mg p.m./L	EFSA Scientific Report (2007) 106,
Daphnia magna	1,2,4-triazole	Acute	EC50= 900 mg p.m./L	PRAPER
Pseudokirchneriella subcapitata	1,2,4-triazole	Sub-chronic	EbC50= 8.2 mg p.m./L ErC50= 22.5 mg p.m./L	EFSA Scientific Report (2007) 106,
Oncorhynchus mykiss	Prothioconazole-S-methyl	Acute	LC ₅₀ = 1.8 mg metab/L	EFSA Journal (2007) 106, 1-98
Daphnia magna	Prothioconazole-S-methyl	Acute	EC ₅₀ = 2.8 mg met/L	EFSA Journal (2007) 106, 1-98
Pseudokirchneriella subcapitata	Prothioconazole-S-methyl	72h	EbC ₅₀ = 3.77 mg metab/L ErC ₅₀ = 47.4 mg	EFSA Scientific Report (2007) 106,

Species	Substance	Exposure System	Results	Reference
			metab./L	
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; im: based on initial measured concentrations

Test item	Test species ¹	Exposure system	Results	Reference
Acute toxicity to fish				
Azoxystrobin	trout	Acute	96 h EC ₅₀ = 0.47 mg as/L	EFSA Journal 2010; 8(4):1542
R234886	trout	Acute	96 h EC ₅₀ > 150 mg/L	EFSA Journal 2010; 8(4):1542
R401553	trout	Acute	96 h EC ₅₀ > 120 mg a.s/L	EFSA Journal 2010; 8(4):1542
R402173	trout	Acute	96 h LC ₅₀ = 62 mg a.s/L	EFSA Journal 2010; 8(4):1542
Chronic toxicity to fish				
Azoxystrobin	<i>Pimephales promelas</i>	33 d , chronic	NOEC = 0.147 mg as/L	EFSA Journal 2010; 8(4):1542
Acute toxicity to aquatic invertebrates				
Azoxystrobin	daphnia	acute	48 h EC ₅₀ = 0.23 (mm, as) mg /L	EFSA Journal 2010; 8(4):1542
Azoxystrobin	<i>Mysidopsis bahia</i>	acute	96h EC ₅₀ > = (nom, pm) 0.055 mg/L	EFSA Journal 2010; 8(4):1542
R234886	daphnia	acute	48h EC ₅₀ > 180 mg/L	EFSA Journal 2010; 8(4):1542
R401553	daphnia	acute	48h EC ₅₀ > 120 mg/L	EFSA Journal 2010; 8(4):1542
R402173	daphnia	acute	48h EC ₅₀ > 100 mg/L	EFSA Journal 2010; 8(4):1542
Chronic toxicity to aquatic invertebrates				
Azoxystrobin	daphnia	chronic	21d NOEC= 0.044 mg a.s/L	EFSA Journal 2010; 8(4):1542
Azoxystrobin	<i>Mysidopsis bahia</i>	chronic	28 d NOEC= 0.00954 mg a.s/L	EFSA Journal 2010; 8(4):1542
Sediment dwellers				
Azoxystrobin	<i>C. riparius</i>	chronic	28 d NOEC=0.8 mg/L	EFSA Journal 2010; 8(4):1542
Toxicity to green algae				
Azoxystrobin	<i>Navicula pelliculosa</i>	chronic	EbC ₅₀ = 251 µg/L ErC ₅₀ = 0.146 mg/L	geometric mean algal EbC ₅₀ from the 4 available studies DAR for azoxystrobin (May 2009) EFSA Journal 2010; 8(4):1542
R234886	<i>Selenastrum capricornutum</i>	chronic	72 h EC ₅₀ = 47 mg/L	EFSA Journal 2010; 8(4):1542
R402173	<i>Selenastrum capricornutum</i>	chronic	72 h EbC ₅₀ /ErC ₅₀ = 67 mg/L	EFSA Journal 2010; 8(4):1542
R401553	<i>Selenastrum capricornutum</i>	chronic	72 h EbC ₅₀ /ErC ₅₀ = 120 mg/L	EFSA Journal 2010; 8(4):1542

Test item	Test species ¹	Exposure system	Results	Reference
Toxicity to aquatic plants				
Azoxystrobin	<i>L. gibba</i>	chronic	Dry weight, EC50 >6.4mg/L Fronds, EC50=3.2 mg/L	EFSA Journal 2010; 8(4):1542
Mesocosm				
Azoxystrobin	<p>The mesocosm study is considered to be a well-conducted mesocosm with an appropriate diversity and abundance of species. It should be noted that azoxystrobin was only applied once, and concentrations were only measured 21 hours after application and not throughout the course of the study. Species/groups were present in sufficient numbers to allow appropriate statistical analysis.</p> <p>The Notifier proposed that the no observed ecologically adverse effects concentration (NOEAEC) is 10 µg/L with assessment factor of 3.</p>			

Table 9.5-2: Endpoints and effect values relevant for the risk assessment for aquatic organisms – CHR/F/PROTAZO

Species	Substance	Exposure System	Results	Reference
Daphnia magna	CHR/F/PROTAZO 375 SC	48 h, s	EC ₅₀ = 2.42 mg/L _{nom}	D. Janota, 2019, Study code: W/43/19
Raphidocelis subcapitata	CHR/F/PROTAZO 375 SC	72 h, s	E _r C ₅₀ = 3.05 mg/L _{nom} E _y C ₅₀ = 0.74 mg/L _{nom}	D. Janota, 2019, Study code: W/44/19
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

9.5.2 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. To achieve a concise risk assessment, the risk envelope approach is applied.

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 Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
Step 2							
N-Europe	1.42	0.07760	0.04610	0.10923	0.02536	0.00651	0.00155

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

Table 9.5-4: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	54.46	0.82142	163.05389	0.54460	5.44600	0.99018	0.27230
Step 2							
N-Europe	6.07	0.09155	18.17365	0.06070	0.60700	0.11036	0.03035
Step 3							
D3/ditch	0.05873	0.00089	0.17584	0.00059	0.00587	0.00107	0.00029
D4/pond	0.06139	0.00093	0.18380	0.00061	0.00614	0.00112	0.00031
D4/stream	0.188	0.0028356	0.56287	0.001880	0.018800	0.00342	0.000940
D5/pond	0.02556	0.0003855	0.07653	0.000256	0.002556	0.00046	0.000128
D5/stream	0.07038	0.0010615	0.21072	0.000704	0.007038	0.00128	0.000352
R1/pond	0.1388	0.0020935	0.41557	0.001388	0.013880	0.00252	0.000694
R1/stream	0.8144	0.0122836	2.43832	0.008144	0.081440	0.01481	0.004072
R3/stream	0.7908	0.0119276	2.36766	0.007908	0.079080	0.01438	0.003954

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
R4/ stream	1.604	0.0241931	4.80240	0.016040	0.160400	0.02916	0.008020
Step 4 – 10 meters vegetative buffer zone and 5 meters no-spray buffer zone							
R1/pond	0.04293	0.00065	0.12853	0.00043	0.00429	0.00078	0.00021
R1/stream	0.251	0.00379	0.75150	0.00251	0.02510	0.00456	0.00126
R3/stream	0.3557	0.0053650	1.06497	0.003557	0.035570	0.00647	0.001779
R4/ stream	0.7294	0.0110015	2.18383	0.007294	0.072940	0.01326	0.003647
Step 4 – 20 meters vegetative buffer zone and 5 meters no-spray buffer zone							
R3/stream	0.1855	0.00280	0.55539	0.00186	0.01855	0.00337	0.00093
R4/ stream	0.3821	0.00576	1.14401	0.00382	0.03821	0.00695	0.00191

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 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
	8.02	0.00161	0.02506	0.00802	0.00356

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 9.5-6: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						

Step 1

	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
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Step 2

N-Europe	1.61	0.08798	0.05227	0.12385	0.02875	0.00739	0.00176
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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 Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	27.73	0.41825	83.02395	0.27730	2.77300	0.50418	0.13865
Step 2							
N-Europe	1.51	0.02278	4.52096	0.01510	0.15100	0.02745	0.00755
Step 3							
D3/ditch	0.08018	0.00121	0.24006	0.00080	0.00802	0.00146	0.00040
D4/pond	0.01194	0.00018	0.03575	0.00012	0.00119	0.00022	0.00006
D4/stream	0.04127	0.0006225	0.12356	0.000413	0.004127	0.00075	0.000206
D5/pond	0.009430	0.0001422	0.02823	0.000094	0.000943	0.00017	0.000047
D5/stream	0.06362	0.0009596	0.19048	0.000636	0.006362	0.00116	0.000318
R1/pond	0.08236	0.0012422	0.24659	0.000824	0.008236	0.00150	0.000412
R1/stream	0.2881	0.0043454	0.86257	0.002881	0.028810	0.00524	0.001441
R3/stream	0.3048	0.0045973	0.91257	0.003048	0.030480	0.00554	0.001524

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 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	4.01	0.00081	0.01253	0.00401	0.00178

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 9.5-9: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
Step 2							
N-Europe	1.42	0.07760	0.04610	0.10923	0.02536	0.00651	0.00155

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 Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC ^{gl-max} (µg/L)						
Step 1							
	54.46	0.82142	163.05389	0.54460	5.44600	0.99018	0.27230
Step 2							

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
N-Europe	6.07	0.09155	18.17365	0.06070	0.60700	0.11036	0.03035
Step 3							
D3/ditch	0.05873	0.00089	0.17584	0.00059	0.00587	0.00107	0.00029
D4/pond	0.06157	0.00093	0.18434	0.00062	0.00616	0.00112	0.00031
D4/stream	0.1880	0.0028356	0.56287	0.001880	0.018800	0.00342	0.000940
D5/pond	0.02556	0.0003855	0.07653	0.000256	0.002556	0.00046	0.000128
D5/stream	0.07038	0.0010615	0.21072	0.000704	0.007038	0.00128	0.000352
R1/pond	0.1388	0.0020935	0.41557	0.001388	0.013880	0.00252	0.000694
R1/stream	0.8144	0.0122836	2.43832	0.008144	0.081440	0.01481	0.004072
R4/stream	1.153	0.0173906	3.45210	0.011530	0.115300	0.02096	0.005765
Step 4 – 10 meters vegetative buffer zone and 5 meteres no-spray buffer zone							
R1/pond	0.0429	0.00065	0.12844	0.00043	0.00429	0.00078	0.00021
R1/stream	0.2510	0.00379	0.75150	0.00251	0.02510	0.00456	0.00126
R4/stream	0.5246	0.0079125	1.57066	0.005246	0.052460	0.00954	0.002623
Step 4 – 20 meters vegetative buffer zone and 5 meteres no-spray buffer zone							
R4/stream	0.1803	0.00272	0.53982	0.00180	0.01803	0.00328	0.00090

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 9.5-11: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint		LC ₅₀	NOEC	EC ₅₀	E _r C ₅₀ /E _y C ₅₀

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
(µg/L)		498000	3200	100000	22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				

Step 1

	8.02	0.00161	0.02506	0.00802	0.00356
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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 Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Step 2							
N-Europe	1.61	0.08798	0.05227	0.12385	0.02875	0.00739	0.00176

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 Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						

Step 1							
	27.23	0.41071	81.52695	0.27230	2.72300	0.49509	0.13615

Step 2							
N-Europe	2.72	0.04103	8.14371	0.02720	0.27200	0.04945	0.01360

Step 3							
D3/ditch	0.03745	0.00056	0.11213	0.00037	0.00375	0.00068	0.00019
D4/pond	0.02473	0.00037	0.07404	0.00025	0.00247	0.00045	0.00012

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
D4/stream	0.08048	0.0012139	0.24096	0.000805	0.008048	0.00146	0.000402
D5/pond	0.008842	0.0001334	0.02647	0.000088	0.000884	0.00016	0.000044
D5/stream	0.04037	0.0006089	0.12087	0.000404	0.004037	0.00073	0.000202
R1/pond	0.05973	0.0009009	0.17883	0.000597	0.005973	0.00109	0.000299
R1/stream	0.4693	0.0070784	1.40509	0.004693	0.046930	0.00853	0.002347
Step 4 – 5 meters buffer zone							
R1/pond	0.007904	0.00012	0.02366	0.00008	0.00079	0.00014	0.00004
R1/stream	0.03392	0.00051	0.10156	0.00034	0.00339	0.00062	0.00017

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 9.5-14: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, ~~2 and 3~~ calculations for the use of CHR/F/PROTIO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	4.01	0.00081	0.01253	0.00401	0.00178

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 Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in ornamental

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
Step 2							
N-Europe	1.42	0.07760	0.04610	0.10923	0.02536	0.00651	0.00155

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 9.5-16: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in ornamental

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	54.46	0.82142	163.05389	0.54460	5.44600	0.99018	0.27230
Step 2							
N-Europe	5.73	0.08643	17.15569	0.05730	0.57300	0.10418	0.02865
Step 3							
D3/ditch	0.06106	0.00092	0.18281	0.00061	0.00611	0.00111	0.00031
D4/pond	0.1257	0.00190	0.37635	0.00126	0.01257	0.00229	0.00063
D4/stream	0.2809	0.0042368	0.84102	0.002809	0.028090	0.00511	0.001405
D5/pond	0.02556	0.0003855	0.07653	0.000256	0.002556	0.00046	0.000128
D5/stream	0.07038	0.0010615	0.21072	0.000704	0.007038	0.00128	0.000352
R1/pond	0.2131	0.0032142	0.63802	0.002131	0.021310	0.00387	0.001066
R1/stream	1.022	0.0154148	3.05988	0.010220	0.102200	0.01858	0.005110
R3/stream	1.136	0.0171342	3.40120	0.011360	0.113600	0.02065	0.005680
R4/stream	1.331	0.0200754	3.98503	0.013310	0.133100	0.02420	0.006655
Step 4 – 10 meters vegetative buffer zone and 5 meteres no-spray buffer zone							
R1/pond	0.03818	0.00058	0.11431	0.00038	0.00382	0.00069	0.00019

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
R1/stream	0.1208	0.00182	0.36168	0.00121	0.01208	0.00220	0.00060
Step 4 – 20 meters vegetative buffer zone and 10 meters buffer zone							
R3/pond	0.2719	0.00410	0.81407	0.00272	0.02719	0.00494	0.00136
R4/stream	0.3171	0.00478	0.94940	0.00317	0.03171	0.00577	0.00159

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 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in ornamental

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	8.02	0.00161	0.02506	0.00802	0.00356

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 9.5-18: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in tobacco

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
Step 2							
N-Europe	1.42	0.07760	0.04610	0.10923	0.02536	0.00651	0.00155

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 Prothioconazole-desithio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in tobacco

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint		LC ₅₀	NOEC	EC ₅₀	NOEC	E _r C ₅₀ /E _y C ₅₀	NOEC

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
(µg/L)		6630	3.34	10000	100	550	2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	54.46	0.82142	163.05389	0.54460	5.44600	0.99018	0.27230
Step 2							
N-Europe	6.07	0.09155	18.17365	0.06070	0.60700	0.11036	0.03035
Step 3							
D3/ditch	0.7674	0.01157	2.29760	0.00767	0.07674	0.01395	0.00384
D4/pond	0.2903	0.00438	0.86916	0.00290	0.02903	0.00528	0.00145
D4/stream	0.4913	0.0074103	1.47096	0.004913	0.049130	0.00893	0.002457
D5/pond	0.3139	0.0047345	0.93982	0.003139	0.031390	0.00571	0.001570
D5/stream	0.5888	0.0088808	1.76287	0.005888	0.058880	0.01071	0.002944
R1/pond	0.3045	0.0045928	0.91168	0.003045	0.030450	0.00554	0.001523
R1/stream	0.6112	0.0092187	1.82994	0.006112	0.061120	0.01111	0.003056
R3/stream	1.239	0.0186878	3.70958	0.012390	0.123900	0.02253	0.006195
R4/stream	0.7804	0.0117707	2.33653	0.007804	0.078040	0.01419	0.003902
Step 4 – 20 meters vegetative buffer zone and 5 meteres no-spray buffer zone							
R3/stream	0.2963	0.00447	0.88713	0.00296	0.02963	0.00539	0.00148

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Step 4 – 20 metres vegetative buffer zone and 20 meters no-spray buffer zone							
D3/ditch	0.09025	0.00136	0.27021	0.00090	0.00903	0.00164	0.00045
D4/pond	0.05420	0.00082	0.16228	0.00054	0.00542	0.00099	0.00027
D4/stream	0.06373	0.0009612	0.19081	0.000637	0.006373	0.00116	0.000319
D5/pond	0.05934	0.0008950	0.17766	0.000593	0.005934	0.00108	0.000297
D5/stream	0.07639	0.0011522	0.22871	0.000764	0.007639	0.00139	0.000382
R1/pond	0.05712	0.0008615	0.17102	0.000571	0.005712	0.00104	0.000286
R1/stream	0.1358	0.0020483	0.40659	0.001358	0.013580	0.00247	0.000679
R4/stream	0.1454	0.0021931	0.43533	0.001454	0.014540	0.00264	0.000727
Step 4 – 15 metres buffer zone (vfs mode) – only for Poland assesment							
D3/ditch	0.1915	0.00289	0.57335	0.00192	0.01915	0.00348	0.00096
D4/pond	0.09526	0.00144	0.28521	0.00095	0.00953	0.00173	0.00048
D4/stream	0.1352	0.0020392	0.40479	0.001352	0.013520	0.00246	0.000676
D5/pond	0.1036	0.0015626	0.31018	0.001036	0.010360	0.00188	0.000518
D5/stream	0.162	0.0024434	0.48503	0.001620	0.016200	0.00295	0.000810
R1/pond	0.1002	0.0015113	0.30000	0.001002	0.010020	0.00182	0.000501
R1/stream	0.1039	0.0015671	0.31108	0.001039	0.010390	0.00189	0.000520

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 9.5-20: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in tobacco

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				

Step 1

	8.02	0.00161	0.02506	0.00802	0.00356
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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 9.5-21: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 - 4 calculations for the use of CHR/F/PROTIO in forestry tree, Salix, Wicker

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						

Step 1

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
	34.43	1.82678	1.08539	2.57154	0.59696	0.15335	0.03658
Step 2							
N-Europe	14.91	0.81475	0.48409	1.14692	0.26625	0.06839	0.01631
Step 3							
D3/ditch	11.67	0.63770	0.37890	0.89769	0.20839	0.05353	0.01277
D4/pond	0.7203	0.03936	0.02339	0.05541	0.01286	0.00330	0.00079
D4/stream	11.81	0.6453552	0.38344	0.908462	0.210893	0.05417	0.012921
D5/pond	0.7437	0.0406393	0.02415	0.057208	0.013280	0.00341	0.000814
D5/stream	12.51	0.6836066	0.40617	0.962308	0.223393	0.05739	0.013687
R1/pond	0.7321	0.0400055	0.02377	0.056315	0.013073	0.00336	0.000801
R1/stream	9.362	0.5115847	0.30396	0.720154	0.167179	0.04294	0.010243
R3/stream	13.25	0.7240437	0.43019	1.019231	0.236607	0.06078	0.014497
R4/stream	9.417	0.5145902	0.30575	0.724385	0.168161	0.04320	0.010303
Step 4 - 10 meteres vegetative buffer zone and 10 meteres no-spray buffer zone							
R3/stream	6.643	0.36301	0.21568	0.51100	0.11863	0.03047	0.00727

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 9.5-22: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole-dethio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in forestry tree, Salix, Wicker

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	63.49	0.95762	190.08982	0.63490	6.34900	1.15436	0.31745
Step 2							
N-Europe	11.25	0.16968	33.68263	0.11250	1.12500	0.20455	0.05625
Step 3							
D3/ditch	0.7674	0.01157	2.29760	0.00767	0.07674	0.01395	0.00384
D4/pond	0.2903	0.00438	0.86916	0.00290	0.02903	0.00528	0.00145
D4/stream	0.4913	0.0074103	1.47096	0.004913	0.049130	0.00893	0.002457
D5/pond	0.3139	0.0047345	0.93982	0.003139	0.031390	0.00571	0.001570
D5/stream	0.5888	0.0088808	1.76287	0.005888	0.058880	0.01071	0.002944
R1/pond	0.3045	0.0045928	0.91168	0.003045	0.030450	0.00554	0.001523
R1/stream	0.6112	0.0092187	1.82994	0.006112	0.061120	0.01111	0.003056
R3/stream	0.7538	0.0113695	2.25689	0.007538	0.075380	0.01371	0.003769
R4/stream	0.6751	0.0101825	2.02126	0.006751	0.067510	0.01227	0.003376
Step 4 – 20 metres vegetative buffer zone and 20 meters no-spray buffer zone							
D3/ditch	0.09025	0.00136	0.27021	0.00090	0.00903	0.00164	0.00045

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
D4/pond	0.05420	0.00082	0.16228	0.00054	0.00542	0.00099	0.00027
D4/stream	0.06373	0.0009612	0.19081	0.000637	0.006373	0.00116	0.000319
D5/pond	0.05934	0.0008950	0.17766	0.000593	0.005934	0.00108	0.000297
D5/stream	0.07639	0.0011522	0.22871	0.000764	0.007639	0.00139	0.000382
R1/pond	0.05712	0.0008615	0.17102	0.000571	0.005712	0.00104	0.000286
R1/stream	0.1358	0.0020483	0.40659	0.001358	0.013580	0.00247	0.000679
R3/stream	0.1439	0.0021704	0.43084	0.001439	0.014390	0.00262	0.000720
R4/stream	0.1454	0.0021931	0.43533	0.001454	0.014540	0.00264	0.000727
Step 4 – 15 meteres buffer zone (vfs mode)- only for Poland assesment							
D3/ditch	0.1915	0.00289	0.57335	0.00192	0.01915	0.00348	0.00096
D4/pond	0.09526	0.00144	0.28521	0.00095	0.00953	0.00173	0.00048
D4/stream	0.1352	0.0020392	0.40479	0.001352	0.013520	0.00246	0.000676
D5/pond	0.1036	0.0015626	0.31018	0.001036	0.010360	0.00188	0.000518
D5/stream	0.162	0.0024434	0.48503	0.001620	0.016200	0.00295	0.000810
R1/pond	0.1002	0.0015113	0.30000	0.001002	0.010020	0.00182	0.000501
R1/stream	0.1039	0.0015671	0.31108	0.001039	0.010390	0.00189	0.000520

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 9.5-23: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in forestry tree, Salix, Wicker

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				

Step 1

	10.32	0.00207	0.03225	0.01032	0.00459
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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 9.5-24: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in sunflower

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						

Step 1

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080

Step 2

N-Europe	1.61	0.08798	0.05227	0.12385	0.02875	0.00739	0.00176
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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 9.5-25: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in sunflower

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						

Step 1

	27.23	0.41071	81.52695	0.27230	2.72300	0.49509	0.13615
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Step 2

N-Europe	3.52	0.05309	10.53892	0.03520	0.35200	0.06400	0.01760
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Step 3

D3/ditch	0.03745	0.00056	0.11213	0.00037	0.00375	0.00068	0.00019
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Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
D4/pond	0.02473	0.00037	0.07404	0.00025	0.00247	0.00045	0.00012
D4/stream	0.08048	0.0012139	0.24096	0.000805	0.008048	0.00146	0.000402
D5/pond	0.009864	0.0001488	0.02953	0.000099	0.000986	0.00018	0.000049
D5/stream	0.03994	0.0006024	0.11958	0.000399	0.003994	0.00073	0.000200
R1/pond	0.06504	0.0009810	0.19473	0.000650	0.006504	0.00118	0.000325
R1/stream	0.396	0.0059729	1.18563	0.003960	0.039600	0.00720	0.001980
R3/stream	0.4964	0.0074872	1.48623	0.004964	0.049640	0.00903	0.002482
R4/stream	0.7420	0.0111916	2.22156	0.007420	0.074200	0.01349	0.003710
Step 4 – 10 meter vegetative buffer zone and 10 meter no-spray buffer zone							
R1/pond	0.009465	0.00014	0.02834	0.00009	0.00095	0.00017	0.00005
R1/stream	0.03068	0.00046	0.09186	0.00031	0.00307	0.00056	0.00015
R3 stream	0.2211	0.0033348	0.66198	0.002211	0.022110	0.00402	0.001106
R4 stream	0.3361	0.0050694	1.00629	0.003361	0.033610	0.00611	0.001681
Step 4 – 20 meter vegetative buffer zone and 5 meter no-spray buffer zone 5 meters buffer zone							
R4/stream	0.1758	0.00265	0.52635	0.00176	0.01758	0.00320	0.00088
Step 3 for surrogate crop maize for sunflower according to Polish national requirements							
D3/ditch	0.05457	0.00082	0.16338	0.00055	0.00546	0.00099	0.00027
D4/pond	0.03392	0.00051	0.10156	0.00034	0.00339	0.00062	0.00017
D4/stream	0.1112	0.0016772	0.33293	0.001112	0.011120	0.00202	0.000556
D5/pond	0.01019	0.0001537	0.03051	0.000102	0.001019	0.00019	0.000051
D5/stream	0.04333	0.0006535	0.12973	0.000433	0.004333	0.00079	0.000217
R1/pond	0.04754	0.0007170	0.14234	0.000475	0.004754	0.00086	0.000238
R1/stream	0.3571	0.0053861	1.06916	0.003571	0.035710	0.00649	0.001786

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
R3/stream	0.5217	0.0078688	1.56198	0.005217	0.052170	0.00949	0.002609
R4/stream	0.6665	0.0100528	1.99551	0.006665	0.066650	0.01212	0.003333
Step 4 – 10 meter vegetative buffer zone and 10 meter no-spray buffer zone for surrogate crop maize for sunflower according to Polish national requirements							
R1/pond	0.02055	0.00031	0.06153	0.00021	0.00206	0.00037	0.00010
R1/stream	0.1622	0.00245	0.48563	0.00162	0.01622	0.00295	0.00081
R3 stream	0.2379	0.0035882	0.71228	0.002379	0.023790	0.00433	0.001190
R4 stream	0.3023	0.0045596	0.90509	0.003023	0.030230	0.00550	0.001512

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 9.5-26: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in sunflower

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	4.01	0.00081	0.01253	0.00401	0.00178

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 9.5-27: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in soya

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>
Endpoint (µg/L)		LC ₅₀ 1830	NOEC 308	EC ₅₀ 1300	NOEC 560	E _r C ₅₀ /E _y C ₅₀ 2180	NOEC 9140
AF		100	10	100	10	10	10
RAC (µg/L)		18.3	30.8	13	56	218	914
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	19.01	1.03880	0.61721	1.46231	0.33946	0.08720	0.02080
Step 2							
N-Europe	1.61	0.08798	0.05227	0.12385	0.02875	0.00739	0.00176

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 9.5-28: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Prothioconazole-desthio for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTIO in soya

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>	<i>Chironomus riparius</i>

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
Endpoint (µg/L)		LC ₅₀ 6630	NOEC 3.34	EC ₅₀ 10000	NOEC 100	E _r C ₅₀ /E _y C ₅₀ 550	NOEC 2000
AF		100	10	100	10	10	10
RAC (µg/L)		66.3	0.334	100	10	55	200
FOCUS Scenario	PEC _{gl-max} (µg/L)						
Step 1							
	27.23	0.41071	81.52695	0.27230	2.72300	0.49509	0.13615
Step 2							
N-Europe	3.52	0.05309	10.53892	0.03520	0.35200	0.06400	0.01760
Step 3							
D3/ditch	0.03745	0.00056	0.11213	0.00037	0.00375	0.00068	0.00019
D4/pond	0.02473	0.00037	0.07404	0.00025	0.00247	0.00045	0.00012
D4/stream	0.08048	0.0012139	0.24096	0.000805	0.008048	0.00146	0.000402
D5/pond	0.008842	0.0001334	0.02647	0.000088	0.000884	0.00016	0.000044
D5/stream	0.04037	0.0006089	0.12087	0.000404	0.004037	0.00073	0.000202
R1/pond	0.05973	0.0009009	0.17883	0.000597	0.005973	0.00109	0.000299
R1/stream	0.4693	0.0070784	1.40509	0.004693	0.046930	0.00853	0.002347
R3/stream	0.5146	0.0077617	1.54072	0.005146	0.051460	0.00936	0.002573
R4/stream	0.7971	0.0120226	2.38653	0.007971	0.079710	0.01449	0.003986
Step 4 – 10 meter vegetative buffer zone and 5 meteres no-spray buffer zone							
R1/pond	0.007904	0.00012	0.02366	0.00008	0.00079	0.00014	0.00004
R1/stream	0.03392	0.00051	0.10156	0.00034	0.00339	0.00062	0.00017

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. prolonged	Algae	Sed. dwell. prolonged
R3/stream	0.2329	0.0035128	0.69731	0.002329	0.023290	0.00423	0.001165
R4/stream	0.3581	0.0054012	1.07216	0.003581	0.035810	0.00651	0.001791
Step 4 – 20 meter vegetative buffer zone and 5 meteres no-spray buffer zone							
R4/stream	0.1867	0.00282	0.55898	0.00187	0.01867	0.00339	0.00093
Step 3 for surrogate crop legumes for soyabean according to Polish national requirements							
D3/ditch	0.02944	0.00044	0.08814	0.00029	0.00294	0.00054	0.00015
D4/pond	0.04164	0.00063	0.12467	0.00042	0.00416	0.00076	0.00021
D4/stream	0.1099	0.0016576	0.32904	0.001099	0.010990	0.00200	0.000550
D5/pond	0.008613	0.0001299	0.02579	0.000086	0.000861	0.00016	0.000043
D5/stream	0.03502	0.0005282	0.10485	0.000350	0.003502	0.00064	0.000175
R1/pond	0.03307	0.0004988	0.09901	0.000331	0.003307	0.00060	0.000165
R1/stream	0.4662	0.0070317	1.39581	0.004662	0.046620	0.00848	0.002331
R3/stream	0.3648	0.0055023	1.09222	0.003648	0.036480	0.00663	0.001824
R4/stream	0.6436	0.0097074	1.92695	0.006436	0.064360	0.01170	0.003218
Step 4 - 10 meter vegetative buffer zone and 5 meteres no-spray buffer zone- for surrogate crop legumes for soyabean according to Polish national requirements							
R1/pond	0.01560	0.00024	0.04671	0.00016	0.00156	0.00028	0.00008
R1/stream	0.2097	0.00316	0.62784	0.00210	0.02097	0.00381	0.00105
R3/stream	0.1600	0.0024133	0.47904	0.001600	0.016000	0.00291	0.000800
R4/stream	0.2869	0.0043273	0.85898	0.002869	0.028690	0.00522	0.001435

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 9.5-29: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for 1,2,4-triazole for each organism group based on FOCUS Steps 1, 2 and 3 calculations for the use of CHR/F/PROTIO in soya

Group		Fish acute	Fish prolonged	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Pseudokirchn. subcapitata</i>
Endpoint (µg/L)		LC ₅₀ 498000	NOEC 3200	EC ₅₀ 100000	E _r C ₅₀ /E _y C ₅₀ 22500
AF		100	10	100	10
RAC (µg/L)		4980	320	1000	2250
FOCUS Scenario	PEC _{gl-max} (µg/L)				
Step 1					
	4.01	0.00081	0.01253	0.00401	0.00178

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 9.5-30: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀ 470	NOEC 147	EC ₅₀ 55	NOEC 9.54	E _r C ₅₀ /E _y C ₅₀ 146	NOEC 800	EC50 3200
AF		100	10	100	10	10	10	10

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	88.93	18.92128	6.04966	161.69091	93.21803	6.09110	1.11163	0.2779
Step 2								
N-Europe	14.72	3.13191	1.00136	26.76364	15.42977	1.00822	0.18400	0.0460
Step 3								
D3/ditch	1.108	0.23574	0.07537	2.01455	1.16143	0.07589	0.01385	0.0035
D4/pond	0.8562	0.18217	0.05824	1.55673	0.89748	0.05864	0.01070	0.0027
D4/stream	0.9022	0.1919574	0.06137	1.640364	0.945702	0.06179	0.011278	0.0028
D5/pond	0.2721	0.0578936	0.01851	0.494727	0.285220	0.01864	0.003401	0.0009
D5/stream	0.9718	0.2067660	0.06611	1.766909	1.018658	0.06656	0.012148	0.0030
R1/pond	0.3801	0.0808723	0.02586	0.691091	0.398428	0.02603	0.004751	0.0012
R1/stream	3.72	0.7914894	0.25306	6.763636	3.899371	0.25479	0.046500	0.0116
R3/stream	4.051	0.8619149	0.27558	7.365455	4.246331	0.27747	0.050638	0.0127
R4/stream	4.192	0.8919149	0.28517	7.621818	4.394130	0.28712	0.052400	0.0131
Step 4		10 meters vegetative buffer zone and 5 meteres no-spray buffer zone						
R1/pond	0.1724	0.03668	0.01173	0.31345	0.18071	0.01181	0.00216	0.0005
R1/stream	1.69	0.35957	0.11497	3.07273	1.77149	0.11575	0.02113	0.0053
R3/stream	1.849	0.3934043	0.12578	3.361818	1.938155	0.12664	0.023113	0.0058
R4/stream	1.907	0.4057447	0.12973	3.467273	1.998952	0.13062	0.023838	0.0060

Table 9.5-31: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Inverteb.
Test species		<i>mesocosm</i>
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC (µg/L)		3.3
FOCUS Scenario	PEC_{gl-max} (µg/L)	
Step 1		
	88.93	26.9485
Step 2		
N-Europe	14.72	4.46061
Step 3		
D3/ditch	1.108	0.33576
D4/pond	0.8562	0.25945
D4/stream	0.9022	0.27339
D5/pond	0.2721	0.08245
D5/stream	0.9718	0.29448
R1/pond	0.3801	0.11518
R1/stream	3.72	1.12727

Group		Inverteb.
R3/stream	4.051	1.22758
R4/stream	4.192	1.27030
Step 4		
R1/pond	0.1724	0.05224
R1/stream	1.69	0.51212
R3/stream	1.849	0.56030
R4/stream	1.907	0.57788

Table 9.5-32: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-33: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	ErC ₅₀ /EyC ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-34: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter cereals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	ErC ₅₀ /EyC ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000

Group		Fish acute	Inverteb. acute	Algae
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-35: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀ 470	NOEC 147	EC ₅₀ 55	NOEC 9.54	E _r C ₅₀ /E _y C ₅₀ 146	NOEC 800	EC50 3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	44.47	9.46170	3.02517	80.85455	46.61426	3.04589	0.55588	0.1390
Step 2								
N-Europe	3.8	0.80851	0.25850	6.90909	3.98323	0.26027	0.04750	0.0119
Step 3								
D3/ditch	1.27	0.27021	0.08639	2.30909	1.33124	0.08699	0.01588	0.0040

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
D4/pond	0.1961	0.04172	0.01334	0.35655	0.20556	0.01343	0.00245	0.0006
D4/stream	1.07	0.2276596	0.07279	1.945455	1.121593	0.07329	0.013375	0.0033
D5/pond	0.1262	0.0268511	0.00859	0.229455	0.132285	0.00864	0.001578	0.0004
D5/stream	1.182	0.2514894	0.08041	2.149091	1.238994	0.08096	0.014775	0.0037
R1/pond	0.1911	0.0406596	0.01300	0.347455	0.200314	0.01309	0.002389	0.0006
R1/stream	1.456	0.3097872	0.09905	2.647273	1.526205	0.09973	0.018200	0.0046
R3/stream	1.401	0.2980851	0.09531	2.547273	1.468553	0.09596	0.017513	0.0044

Table 9.5-36: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Inverteb.
Test species		mesocosm
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC (µg/L)		3.3
FOCUS Scenario	PEC _{gl-max} (µg/L)	
Step 1		
	44.47	13.4758
Step 2		
N-Europe	3.8	1.15152

Group		Inverteb.
Step 3		
D3/ditch	1.27	0.38485
D4/pond	0.1961	0.05942
D4/stream	1.07	0.32424
D5/pond	0.1262	0.03824
D5/stream	1.182	0.35818
R1/pond	0.1911	0.05791
R1/stream	1.456	0.44121
R3/stream	1.401	0.42455

Table 9.5-37: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	29.42	0.0196	0.02	0.00626

Table 9.5-38: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	10.38	0.1674194	0.1038	0.015493

Table 9.5-39: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in winter oilseed rape

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10

Group		Fish acute	Inverteb. acute	Algae
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC_{gl-max} (µg/L)			
Step 1				
	7.38	0.0615	0.0615	0.000615

Table 9.5-40: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀	NOEC	EC ₅₀	NOEC	E _r C ₅₀ /E _y C ₅₀	NOEC	EC50
AF		470	147	55	9.54	146	800	3200
RAC (µg/L)		100	10	100	10	10	10	10
FOCUS Scenario	PEC _{gl-max} (µg/L)	4.7	14.7	0.55	0.954	14.6	80	320
Step 1								
	18.92128	6.04966	161.69091	93.21803	6.09110	1.11163	0.2779	18.92128
Step 2								
N-Europe	14.72	3.13191	1.00136	26.76364	15.42977	1.00822	0.18400	0.0460
Step 3								

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
D3/ditch	1.108	0.23574	0.07537	2.01455	1.16143	0.07589	0.01385	0.0035
D4/pond	0.8202	0.17451	0.05580	1.49127	0.85975	0.05618	0.01025	0.0026
D4/stream	0.9259	0.1970000	0.06299	1.683455	0.970545	0.06342	0.011574	0.0029
D5/pond	0.2727	0.0580213	0.01855	0.495818	0.285849	0.01868	0.003409	0.0009
D5/stream	0.9581	0.2038511	0.06518	1.742000	1.004298	0.06562	0.011976	0.0030
R1/pond	0.3801	0.0808723	0.02586	0.691091	0.398428	0.02603	0.004751	0.0012
R1/stream	3.72	0.7914894	0.25306	6.763636	3.899371	0.25479	0.046500	0.0116
R3/stream	6.332	1.3472340	0.43075	11.512727	6.637317	0.43370	0.079150	0.0198
Step 4		10 meters vegetative buffer zone and 5 meteres no-spray buffer zone						
R1/pond	0.1724	0.03668	0.01173	0.31345	0.18071	0.01181	0.00216	0.0005
R1/stream	1.69	0.35957	0.11497	3.07273	1.77149	0.11575	0.02113	0.0053
R4/stram	2.858	0.6080851	0.19442	5.196364	2.995807	0.19575	0.035725	0.0089

Table 9.5-41: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Inverteb.
Test species		mesocosm
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC		3.3
(µg/L)		

Group		Inverteb.
FOCUS Scenario	PEC _{gl-max} (µg/L)	
Step 1		
	88.93	26.9485
Step 2		
N-Europe	14.72	4.4606
Step 3		
D3/ditch	1.108	0.3358
D4/pond	0.8202	0.2485
D4/stream	0.9259	0.2806
D5/pond	0.2727	0.0826
D5/stream	0.9581	0.2903
R1/pond	0.3801	0.1152
R1/stream	3.72	1.1273
R4/stream	6.332	1.9188
Step 4		
R1/pond	0.1724	0.0522
R1/stream	1.69	0.5121
R4/stream	2.858	0.8661

Table 9.5-42: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Inverteb. acute	Algae
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Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-43: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			

Group		Fish acute	Inverteb. acute	Algae
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-44: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring cereals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-45: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint		LC ₅₀	NOEC	EC ₅₀	NOEC	E _r C ₅₀ /E _y C ₅₀	NOEC	EC50

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
(µg/L)		470	147	55	9.54	146	800	3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	44.47	9.46170	3.02517	80.85455	46.61426	3.04589	0.55588	0.1390
Step 2								
N-Europe	6.27	1.33404	0.42653	11.40000	6.57233	0.42945	0.07838	0.0196
Step 3								
D3/ditch	1.266	0.26936	0.08612	2.30182	1.32704	0.08671	0.01583	0.0040
D4/pond	0.4078	0.08677	0.02774	0.74145	0.42746	0.02793	0.00510	0.0013
D4/stream	1.038	0.2208511	0.07061	1.887273	1.088050	0.07110	0.012975	0.0032
D5/pond	0.1502	0.0319574	0.01022	0.273091	0.157442	0.01029	0.001878	0.0005
D5/stream	1.01	0.2148936	0.06871	1.836364	1.058700	0.06918	0.012625	0.0032
R1/pond	0.1576	0.0335319	0.01072	0.286545	0.165199	0.01079	0.001970	0.0005
R1/stream	1.649	0.3508511	0.11218	2.998182	1.728512	0.11295	0.020613	0.0052

Table 9.5-46: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in spring oilseed rape, mustard, breadseed poppy

Group		Inverteb.
Test species		<i>mesocosm</i>

Group		Inverteb.
Endpoint (µg/L)		NOEAEC 10
AF		3
RAC (µg/L)		3.3
FOCUS Scenario	PEC _{gl-max} (µg/L)	
Step 1		
	44.47	13.4758
Step 2		
N-Europe	6.27	1.9000
Step 3		
D3/ditch	1.266	0.3836
D4/pond	0.4078	0.1236
D4/stream	1.038	0.3145
D5/pond	0.1502	0.0455
D5/stream	1.01	0.3061
R1/pond	0.1576	0.0478
R1/stream	1.649	0.4997

Table 9.5-47: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Inverteb. acute	Algae
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Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	29.42	0.0196	0.02	0.00626

Table 9.5-48: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			

Group		Fish acute	Inverteb. acute	Algae
Step 1				
	10.38	0.1674194	0.1038	0.015493

Table 9.5-49: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in spring oilseed rape, mustard, breadseed poppy

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	7.38	0.0615	0.0615	0.000615

Table 9.5-50: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in soya

Group		Fish acute	Fish prolonged	Inverteb. acute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint		LC ₅₀	NOEC	EC ₅₀	NOEC	E _r C ₅₀ /E _y C ₅₀	NOEC	EC50

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
(µg/L)		470	147	55	9.54	146	800	3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	44.47	9.46170	3.02517	80.85455	46.61426	3.04589	0.55588	0.1390
Step 2								
N-Europe	7.91	1.68298	0.53810	14.38182	8.29140	0.54178	0.09888	0.0247
Step 3								
D3/ditch	1.266	0.26936	0.08612	2.30182	1.32704	0.08671	0.01583	0.0040
D4/pond	0.4078	0.08677	0.02774	0.74145	0.42746	0.02793	0.00510	0.0013
D4/stream	1.038	0.2208511	0.07061	1.887273	1.088050	0.07110	0.012975	0.0032
D5/pond	0.1502	0.0319574	0.01022	0.273091	0.157442	0.01029	0.001878	0.0005
D5/stream	1.01	0.2148936	0.06871	1.836364	1.058700	0.06918	0.012625	0.0032
R1/pond	0.1576	0.0335319	0.01072	0.286545	0.165199	0.01079	0.001970	0.0005
R1/stream	1.649	0.3508511	0.11218	2.998182	1.728512	0.11295	0.020613	0.0052
R3/stream	3.156	0.6714894	0.21469	5.738182	3.308176	0.21616	0.039450	0.0099
R4/stream	1.765	0.3755319	0.12007	3.209091	1.850105	0.12089	0.022063	0.0055
Step 3 for surrogate crop legumes for soyabean according to Polish national requirements								
D3/ditch	1.047	0.22277	0.07122	1.90364	1.09748	0.07171	0.01309	0.0033
D4/pond	0.4646	0.09885	0.03161	0.84473	0.48700	0.03182	0.00581	0.0015
D4/stream	0.8573	0.1824043	0.05832	1.558727	0.898637	0.05872	0.010716	0.0027

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
D5/pond	0.1304	0.0277447	0.00887	0.237091	0.136688	0.00893	0.001630	0.0004
D5/stream	0.8765	0.1864894	0.05963	1.593636	0.918763	0.06003	0.010956	0.0027
R1/pond	0.09557	0.0203340	0.00650	0.173764	0.100178	0.00655	0.001195	0.0003
R1/stream	1.609	0.3423404	0.10946	2.925455	1.686583	0.11021	0.020113	0.0050
R3/stream	1.025	0.2180851	0.06973	1.863636	1.074423	0.07021	0.012813	0.0032
R4/stream	3.216	0.6842553	0.21878	5.847273	3.371069	0.22027	0.040200	0.0101

Table 9.5-51: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in soya

Group		Inverteb.
Test species		mesocosm
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC (µg/L)		3.3
FOCUS Scenario	PEC _{gl-max} (µg/L)	
Step 1		
	44.47	13.4758
Step 2		
N-Europe	7.91	2.3970
Step 3		
D3/ditch	1.266	0.3836

Group		Inverteb.
D4/pond	0.4078	0.1236
D4/stream	1.038	0.3145
D5/pond	0.1502	0.0455
D5/stream	1.01	0.3061
R1/pond	0.1576	0.0478
R1/stream	1.649	0.4997
R3/stream	3.156	0.9564
R4/stream	1.765	0.5348
Step 3 for surrogate crop legumes for soyabean according to Polish national requirements		
D3/ditch	1.047	0.3173
D4/pond	0.4646	0.1408
D4/stream	0.8573	0.2598
D5/pond	0.1304	0.0395
D5/stream	0.8765	0.2656
R1/pond	0.09557	0.0290
R1/stream	1.609	0.4876
R3/stream	1.025	0.3106
R4/stream	3.216	0.9745

Table 9.5-52: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in soya

Group		Fish acute	Inverteb. acute	Algae
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Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-53: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, 2, ~~3~~ and 4 calculations for the use of CHR/F/PROTAZO in soya

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			

Group		Fish acute	Inverteb. acute	Algae
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-54: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in soya

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-55: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in sunflower

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint		LC ₅₀	NOEC	EC ₅₀	NOEC	E _r C ₅₀ /E _y C ₅₀	NOEC	EC50

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
(µg/L)		470	147	55	9.54	146	800	3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	44.47	9.46170	3.02517	80.85455	46.61426	3.04589	0.55588	0.1390
Step 2								
N-Europe	7.91	1.68298	0.53810	14.38182	8.29140	0.54178	0.09888	0.0247
Step 3								
D3/ditch	1.266	0.26936	0.08612	2.30182	1.32704	0.08671	0.01583	0.0040
D4/pond	0.4078	0.08677	0.02774	0.74145	0.42746	0.02793	0.00510	0.0013
D4/stream	1.038	0.2208511	0.07061	1.887273	1.088050	0.07110	0.012975	0.0032
D5/pond	0.4075	0.0867021	0.02772	0.740909	0.427149	0.02791	0.005094	0.0013
D5/stream	0.9421	0.2004468	0.06409	1.712909	0.987526	0.06453	0.011776	0.0029
R1/pond	0.1927	0.0410000	0.01311	0.350364	0.201992	0.01320	0.002409	0.0006
R1/stream	1.982	0.4217021	0.13483	3.603636	2.077568	0.13575	0.024775	0.0062
R3/stream	2.809	0.5976596	0.19109	5.107273	2.944444	0.19240	0.035113	0.0088
R4/stream	3.531	0.7512766	0.24020	6.420000	3.701258	0.24185	0.044138	0.0110
Step 4 - 10 meters vegetative buffer zone and 5 meters no-spray buffer zone								
R4/stream	1.600	0.34043	0.10884	2.90909	1.67715	0.10959	0.02000	0.0050

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Step 3 for surrogate crop maize for sunflower according to Polish national requirements								
D3/ditch	1.047	0.22277	0.07122	1.90364	1.09748	0.07171	0.01309	0.0033
D4/pond	0.5689	0.12104	0.03870	1.03436	0.59633	0.03897	0.00711	0.0018
D4/stream	0.9283	0.1975106	0.06315	1.687818	0.973061	0.06358	0.011604	0.0029
D5/pond	0.4302	0.0915319	0.02927	0.782182	0.450943	0.02947	0.005378	0.0013
D5/stream	0.9438	0.2008085	0.06420	1.716000	0.989308	0.06464	0.011798	0.0029
R1/pond	0.1752	0.0372766	0.01192	0.318545	0.183648	0.01200	0.002190	0.0005
R1/stream	2.297	0.4887234	0.15626	4.176364	2.407757	0.15733	0.028713	0.0072
R3/stream	2.011	0.4278723	0.13680	3.656364	2.107966	0.13774	0.025138	0.0063
R4/stream	3.932	0.8365957	0.26748	7.149091	4.121593	0.26932	0.049150	0.0123
Step 4 - 10 meters vegetative buffer zone and 5 meters no-spray buffer zone - for surrogate crop maize for sunflower according to Polish national requirements								
R4/stream	1.780	0.37872	0.12109	3.23636	1.86583	0.12192	0.02225	0.0056

Table 9.5-56: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in sunflower

Group		Inverteb.
Test species		<i>mesocosm</i>
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC		
(µg/L)		3.3

FOCUS Scenario	PEC_{gl-max} (µg/L)	
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Step 1

	44.47	13.4758
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Step 2

N-Europe	7.91	2.3970
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Step 3

D3/ditch	1.266	0.3836
D4/pond	0.4078	0.1236
D4/stream	1.038	0.3145
D5/pond	0.4075	0.1235
D5/stream	0.9421	0.2855
R1/pond	0.1927	0.0584
R1/stream	1.982	0.6006
R3/stream	2.809	0.8512
R4/stream	3.531	1.0700

R4/stream	1.600	0.4848
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**Step 3 for surrogate crop maize for
sunflower according to Polish
national requirements**

D3/ditch	1.047	0.3173
D4/pond	0.5689	0.1724
D4/stream	0.9283	0.2813
D5/pond	0.4302	0.1304
D5/stream	0.9438	0.2860

R1/pond	0.1752	0.0531
R1/stream	2.297	0.6961
R3/stream	2.011	0.6094
R4/stream	3.932	1.1915
Step 4 - 10 meters vegetative buffer zone and 5 meters no-spray buffer zone - for surrogate crop maize for sunflower according to Polish national requirements		
R4/stream	1.780	0.5394

Table 9.5-57: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in sunflower

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-58: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in sunflower

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-59: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in sunflower

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000

Group		Fish acute	Inverteb. acute	Algae
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-60: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in ornamentals

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀ 470	NOEC 147	EC ₅₀ 55	NOEC 9.54	E _r C ₅₀ /E _y C ₅₀ 146	NOEC 800	EC50 3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	88.93	18.92128	6.04966	161.69091	93.21803	6.09110	1.11163	0.2779
Step 2								
N-Europe	13.95	2.96809	0.94898	25.36364	14.62264	0.95548	0.17438	0.0436
Step 3								
D3/ditch	1.108	0.23574	0.07537	2.01455	1.16143	0.07589	0.01385	0.0035
D4/pond	1.106	0.23532	0.07524	2.01091	1.15933	0.07575	0.01383	0.0035

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
D4/stream	1.072	0.2280851	0.07293	1.949091	1.123690	0.07342	0.013400	0.0034
D5/pond	0.2721	0.0578936	0.01851	0.494727	0.285220	0.01864	0.003401	0.0009
D5/stream	0.9718	0.2067660	0.06611	1.766909	1.018658	0.06656	0.012148	0.0030
R1/pond	0.3997	0.0850426	0.02719	0.726727	0.418973	0.02738	0.004996	0.0012
R1/stream	4.287	0.9121277	0.29163	7.794545	4.493711	0.29363	0.053588	0.0134
R3/stream	3.356	0.7140426	0.22830	6.101818	3.517820	0.22986	0.041950	0.0105
R4/stream	5.039	1.0721277	0.34279	9.161818	5.281971	0.34514	0.062988	0.0157
Step 4		10 meters vegetative buffer zone and 5 meteres no-spraybuffer zone						
R1/pond	0.06137	0.01306	0.00417	0.11158	0.06433	0.00420	0.00077	0.0002
R1/stream	0.2674	0.05689	0.01819	0.48618	0.28029	0.01832	0.00334	0.0008
R3/stream	1.693	0.3602128	0.11517	3.078182	1.774633	0.11596	0.021163	0.0053
R4/stream	2.292	0.4876596	0.15592	4.167273	2.402516	0.15699	0.028650	0.0072

Table 9.5-61: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in ornamentals

Group		Inverteb.
Test species		mesocosm
Endpoint		NOEAEC
(µg/L)		10
AF		3
RAC (µg/L)		3.3

Group		Inverteb.
FOCUS Scenario	PEC _{gl-max} (µg/L)	

Step 1

	88.93	26.9485
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Step 2

N-Europe	13.95	4.2273
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Step 3

D3/ditch	1.108	0.3358
D4/pond	1.106	0.3352
D4/stream	1.072	0.3248
D5/pond	0.2721	0.0825
D5/stream	0.9718	0.2945
R1/pond	0.3997	0.1211
R1/stream	4.287	1.2991
R3/stream	3.356	1.0170
R4/stream	5.039	1.5270

Step 4

R1/pond	0.06137	0.0186
R1/stream	0.2674	0.0810
R3/stream	1.693	0.5130
R4/stream	2.292	0.6945

Table 9.5-62: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in ornamentals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	ErC ₅₀ /EyC ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-63: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in ornamentals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	ErC ₅₀ /EyC ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670

Group		Fish acute	Inverteb. acute	Algae
FOCUS Scenario	PEC_{gl-max} (µg/L)			
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-64: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in ornamentals

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC_{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-65: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in tobacco

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
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Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀	NOEC	EC ₅₀	NOEC	ErC ₅₀ /EyC ₅₀	NOEC	EC50
AF		470	147	55	9.54	146	800	3200
RAC (µg/L)		100	10	100	10	10	10	10
		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	88.93	18.92128	6.04966	161.69091	93.21803	6.09110	1.11163	0.2779
Step 2								
N-Europe	14.72	3.13191	1.00136	26.76364	15.42977	1.00822	0.18400	0.0460
Step 3								
D3/ditch	13.37	2.84468	0.90952	24.30909	14.01468	0.91575	0.16713	0.0418
D4/pond	1.378	0.29319	0.09374	2.50545	1.44444	0.09438	0.01723	0.0043
D4/stream	13.51	2.8744681	0.91905	24.563636	14.161426	0.92534	0.168875	0.0422
D5/pond	1.645	0.3500000	0.11190	2.990909	1.724319	0.11267	0.020563	0.0051
D5/stream	14.32	3.0468085	0.97415	26.036364	15.010482	0.98082	0.179000	0.0448
R1/pond	1.491	0.3172340	0.10143	2.710909	1.562893	0.10212	0.018638	0.0047
R1/stream	10.72	2.2808511	0.72925	19.490909	11.236897	0.73425	0.134000	0.0335
R3/stream	5.353	1.1389362	0.36415	9.732727	5.611111	0.36664	0.066913	0.0167
R4/stream	10.78	2.2936170	0.73333	19.600000	11.299790	0.73836	0.134750	0.0337
Step 4		10 metres vegetative buffe zone						

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
R3/pond	2.440	0.51915	0.16599	4.43636	2.55765	0.16712	0.03050	0.0076
Step 4		10 meteres vegetative buffe zone						
D3/ditch	1.576	0.33532	0.10721	2.86545	1.65199	0.10795	0.01970	0.0049
D4/pond	0.2950	0.06277	0.02007	0.53636	0.30922	0.02021	0.00369	0.0009
D4/stream	1.754	0.3731915	0.11932	3.189091	1.838574	0.12014	0.021925	0.0055
D5/pond	0.4194	0.0892340	0.02853	0.762545	0.439623	0.02873	0.005243	0.0013
D5/stream	1.860	0.3957447	0.12653	3.381818	1.949686	0.12740	0.023250	0.0058
R1/pond	0.2850	0.0606383	0.01939	0.518182	0.298742	0.01952	0.003563	0.0009
R1/stream	1.391	0.2959574	0.09463	2.529091	1.458071	0.09527	0.017388	0.0043
R4/stream	1.399	0.2976596	0.09517	2.543636	1.466457	0.09582	0.017488	0.0044

Table 9.5-66: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in tobacco

Group		Inverteb.
Test species		mesocosm
Endpoint (µg/L)		NOEAEC 10
AF		3
RAC (µg/L)		3.3
FOCUS Scenario	PEC _{gl-max} (µg/L)	
Step 1		
	88.93	26.9485

Group		Inverteb.
Step 2		
N-Europe	14.72	4.4606
Step 3		
D3/ditch	13.37	4.0515
D4/pond	1.378	0.4176
D4/stream	13.51	4.0939
D5/pond	1.645	0.4985
D5/stream	14.32	4.3394
R1/pond	1.491	0.4518
R1/stream	10.72	3.2485
R3/stream	5.353	1.6221
R4/stream	10.78	3.2667
Step 4	10 meteres vegetative buffe zone	
R3/pond	2.440	0.7394
Step 4	10 meteres vegetative buffe zone	
D3/ditch	1.576	0.4776
D4/pond	0.2950	0.0894
D4/stream	1.754	0.5315
D5/pond	0.4194	0.1271
D5/stream	1.860	0.5636
R1/pond	0.2850	0.0864
R1/stream	1.391	0.4215
R4/stream	1.399	0.4239

Table 9.5-67: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in tobacco

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-68: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in tobacco

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10

Group		Fish acute	Inverteb. acute	Algae
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-69: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in tobacco

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-70: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in forestry tree, Salix, Wicker

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
Test species		<i>Oncorhynchus mykiss</i>	<i>Pimephales promelas</i>	<i>Mysidopsis bahia</i>	<i>Mysidopsis bahia</i>	<i>Navicula pelliculosa</i>	<i>Chironomus riparius</i>	<i>Lemna gibba</i>
Endpoint (µg/L)		LC ₅₀ 470	NOEC 147	EC ₅₀ 55	NOEC 9.54	E _r C ₅₀ /E _y C ₅₀ 146	NOEC 800	EC50 3200
AF		100	10	100	10	10	10	10
RAC (µg/L)		4.7	14.7	0.55	0.954	14.6	80	320
FOCUS Scenario	PEC _{gl-max} (µg/L)							
Step 1								
	124.18	26.42128	8.44762	225.78182	130.16771	8.50548	1.55225	0.3881
Step 2								
N-Europe	36.77	7.82340	2.50136	66.85455	38.54298	2.51849	0.45963	0.1149
Step 3								
D3/ditch	13.37	2.84468	0.90952	24.30909	14.01468	0.91575	0.16713	0.0418
D4/pond	1.378	0.29319	0.09374	2.50545	1.44444	0.09438	0.01723	0.0043
D4/stream	13.51	2.8744681	0.91905	24.563636	14.161426	0.92534	0.168875	0.0422
D5/pond	1.645	0.3500000	0.11190	2.990909	1.724319	0.11267	0.020563	0.0051
D5/stream	14.32	3.0468085	0.97415	26.036364	15.010482	0.98082	0.179000	0.0448
R1/pond	1.491	0.3172340	0.10143	2.710909	1.562893	0.10212	0.018638	0.0047
R1/stream	10.72	2.2808511	0.72925	19.490909	11.236897	0.73425	0.134000	0.0335

Group		Fish acute	Fish prolonged	Inverteb. cute	Inverteb. long	Algae	Sed. dwell. prolonged	Aquatic Plant
R3/stream	15.17	3.2276596	1.03197	27.581818	15.901468	1.03904	0.189625	0.0474
R4/stream	10.78	2.2936170	0.73333	19.600000	11.299790	0.73836	0.134750	0.0337
Step 4		20 meters buffer zone						
D3/ditch	1.576	0.33532	0.10721	2.86545	1.65199	0.10795	0.01970	0.0049
D4/pond	0.295	0.06277	0.02007	0.53636	0.30922	0.02021	0.00369	0.0009
D4/stream	1.754	0.3731915	0.11932	3.189091	1.838574	0.12014	0.021925	0.0055
D5/pond	0.4194	0.0892340	0.02853	0.762545	0.439623	0.02873	0.005243	0.0013
D5/stream	1.86	0.3957447	0.12653	3.381818	1.949686	0.12740	0.023250	0.0058
R1/pond	0.285	0.0606383	0.01939	0.518182	0.298742	0.01952	0.003563	0.0009
R1/stream	1.391	0.2959574	0.09463	2.529091	1.458071	0.09527	0.017388	0.0043
R3/stream	1.971	0.4193617	0.13408	3.583636	2.066038	0.13500	0.024638	0.0062
R4/stream	1.399	0.2976596	0.09517	2.543636	1.466457	0.09582	0.017488	0.0044

Table 9.5-71: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for Azoxystrobin for each organism group based on FOCUS Steps 1, 2, 3 and 4 calculations for the use of CHR/F/PROTAZO in forestry tree, Salix, Wicker

Group		Inverteb.
Test species		mesocosm
Endpoint (µg/L)		NOEAEC 10
AF		3
RAC (µg/L)		3.3

Group		Inverteb.
FOCUS Scenario	PEC _{gl-max} (µg/L)	

Step 1

	124.18	37.6303
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Step 2

N-Europe	36.77	11.1424
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Step 3

D3/ditch	13.37	4.0515
D4/pond	1.378	0.4176
D4/stream	13.51	4.0939
D5/pond	1.645	0.4985
D5/stream	14.32	4.3394
R1/pond	1.491	0.4518
R1/stream	10.72	3.2485
R3/stream	15.17	4.5970
R4/stream	10.78	3.2667

Step 4	20 meters buffer zone	
D3/ditch	1.576	0.4776
D4/pond	0.295	0.0894
D4/stream	1.754	0.5315
D5/pond	0.4194	0.1271
D5/stream	1.86	0.5636
R1/pond	0.285	0.0864

Group		Inverteb.
R1/stream	1.391	0.4215
R3/stream	1.971	0.5973
R4/stream	1.399	0.4239

Table 9.5-72: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R234886 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in forestry tree, Salix, Wicker

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 150 000	EC ₅₀ 180000	E _r C ₅₀ /E _y C ₅₀ 47 000
AF		100	100	10
RAC (µg/L)		1500	1800	4700
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	58.85	0.0392	0.03	0.012521

Table 9.5-73: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R402173 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in forestry tree, Salix, Wicker

Group		Fish acute	Inverteb. acute	Algae
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Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 62 000	EC ₅₀ 100 000	E _r C ₅₀ /E _y C ₅₀ 67 000
AF		100	100	10
RAC (µg/L)		62	100	670
FOCUS Scenario	PEC _{gl-max} (µg/L)			
Step 1				
	20.76	0.3348387	0.2076	0.030985

Table 9.5-74: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for R401553 for each organism group based on FOCUS Steps 1, ~~2, 3 and 4~~ calculations for the use of CHR/F/PROTAZO in forestry tree, Salix, Wicker

Group		Fish acute	Inverteb. acute	Algae
Test species		<i>Oncorhynchus mykiss</i>	<i>Daphnia magna</i>	<i>Selenastrum capricornutum</i>
Endpoint (µg/L)		LC ₅₀ 120 000	EC ₅₀ 120000	E _r C ₅₀ /E _y C ₅₀ 120000
AF		100	100	10
RAC (µg/L)		120	120	12000
FOCUS Scenario	PEC _{gl-max} (µg/L)			

Group		Fish acute	Inverteb. acute	Algae
Step 1				
	14.77	0.1230833	0.123083	0.001231

Table 9.5-75: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for CHR/F/PROTAZO for each organism group based on Drift Calculator SWASH MODEL ver 5.3 calculations for the use of CHR/F/PROTAZO in cereals and ornamental

Intended use		
Formulation		CHR/F/PROTAZO
Application rate (g[prod]/ha)		≥ 1 X 1121 g
Entry into surface water via spraydrift (Drift calculator from SWASH)		
Buffer zone (m)	PEC _{sw} [µg prod/L]	
1	6.2917 7.2020	
Buffer zone (m)	RAC/PEC ratio	
	Daphnia magna =EC50 2 420 g/L	
	RAC=24.2 (AF=100)	
1	0.260 0.3	
Buffer zone (m)	RAC/PEC ratio	
	Pseudokirchmeirella subcapitata =ErC50 3 050g/L	
	RAC=305 (AF=10)	
1	0.0206 0.024	

Table 9.5-76: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for CHR/F/PROTAZO for each organism group based on Drift Calculator SWASH MODEL ver 5.3 calculations for the use of CHR/F/PROTAZO in forestry tree

Intended use		CHR/F/PROTAZO
Formulation		
Application rate (g[prod]/ha)		
		1 X 1121 g
Entry into surface water via spraydrift (Drift alculator from SWASH)		
Buffer zone (m)	PEC _{sw} [µg prod/L]	
20	9.7298	
Entry into surface water via spraydrift (Drift alculator from SWASH)		
Buffer zone (m)	RAC/PEC ratio Daphnia magna =EC50 2 420 g/L RAC=24.2 (AF=100)	
20	0.402	
Buffer zone (m)	RAC/PEC ratio Pseudokirchmeirella subcapitata =ErC50 3 050g/L RAC=305 (AF=10)	
20	0.0319	

Table 9.5-77: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for CHR/F/PROTAZO for each organism group based on Drift Calculator SWASH MODEL ver 5.3 calculations for the use of CHR/F/PROTAZO in soya, sunflower

SWASH MODEL FOR ENVIRONMENTAL RISK ASSESSMENT OF CHEMICALS IN SURFACE WATER	
Intended use	CHR/F/PROTAZO
Formulation	
Application rate (g[prod]/ha)	
	1 X 1121 g
Entry into surface water via spraydrift (Drift alculator from SWASH)	

Buffer zone (m)	PEC _{sw} [µg prod/L]
1	5.9546
Entry into surface water via spraydrift (Drift alculator from SWASH)	
Buffer zone (m)	RAC/PEC ratio Daphnia magna =EC50 2 420 g/L RAC=24.2 (AF=100)
1	0.246
Buffer zone (m)	RAC/PEC ratio Pseudokirchmeirella subcapitata =ErC50 3 050g/L RAC=305 (AF=10)
1	0.0195

Table 9.5-78: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for CHR/F/PROTAZO for each organism group based on Drift Calculator SWASH MODEL ver 5.3 calculations for the use of CHR/F/PROTAZO in tobacco

SWASH MODEL FOR DRIFT CALCULATIONS FOR THE USE OF CHL/PT/RS/MS IN CROPS	
Intended use	CHR/F/PROTAZO
Formulation	
Application rate (g[prod]/ha)	
	1 X 1121 g
Entry into surface water via spraydrift (Drift alculator from SWASH)	
Buffer zone (m)	PEC _{sw} [µg prod/L]
1	4.6222 5.3448
Entry into surface water via spraydrift (Drift alculator from SWASH)	

Buffer zone (m)	RAC/PEC ratio Daphnia magna =EC50 2 420 g/L RAC=24.2 (AF=100)
1	0.191 0.22
Buffer zone (m)	RAC/PEC ratio Pseudokirchmeirella subcapitata =ErC50 3 050g/L RAC=305 (AF=10)
1	0.0152 0.018

Table 9.5-79: Aquatic organisms: acceptability of risk (PEC/RAC < 1) for CHR/F/PROTAZO for each organism group based on Drift Calculator SWASH MODEL ver 5.3 calculations for the use of CHR/F/PROTAZO in oilseed rape, mustard, breadseed poppy

Intended use	CHR/F/PROTAZO 1 X 1121 g
Formulation	
Application rate (g[prod]/ha)	
Entry into surface water via spraydrift (Drift alculator from SWASH)	
Buffer zone (m)	PEC _{sw} [µg prod/L]
1	7.2020
Entry into surface water via spraydrift (Drift alculator from SWASH)	
Buffer zone (m)	RAC/PEC ratio Daphnia magna =EC50 2 420 g/L RAC=24.2 (AF=100)
1	0.298

Buffer zone (m)	RAC/PEC ratio Pseudokirchmeirella subcapitata =ErC50 3 050g/L RAC=305 (AF=10)
1	0.0236

Each of the active substances in Kajman 375 S.C. - azoxytrobin and prothioconazole are particularly toxic to the same group of aquatic organisms - invertebrates, daphnia. According to Regulation 284/2013, tests on fish should be submitted for multi-component formulations when active substances show particular toxicity to different taxonomic groups of aquatic organisms. However, Both substances are particularly toxic to the same group of aquatic organisms, so this provision does not apply to Kajman 375 SC. In addition, in order to protect the vertebrate organisms and their life cycle according to AGD point 7.5 and point 11.4, we avoid duplicating studies on vertebrate animals, so in the risk assessment we referred to the endpoints approved when introducing these active substances to Annex 1. Therefore, also below we prepared calculation of mixture toxicity on fish used only active substance's endpoints.

Review Comments:

Regulation (EC) No 1107/2009 clearly requires "the use of non-animal test methods and other RA strategies should be promoted. Animal testing for the purposes of this Regulation should be minimised and tests on vertebrates should be undertaken as a last resort". Therefore, aquatic RA alternatives to experimental testing are specifically recommended for fish.

Regarding the acute toxicity the most sensitive trophic level is not aquatic vertebrates, but invertebrates. Based on the formulation's studies for aquatic invertebrates and algae, the calculated MDR values are between 0.2 and 5, indicating that the formulation does not cause an unexpected increased toxicity compared to the active substances for these organisms. Thus, it is not necessary to submit a study on fish.

Decision scheme for mixture toxicity risk assessment for CHR/F/PROTAZO 375 SC

Step 1. Are measured toxicity data (EC_x) available for the given endpoint (typically chronic data available only for a.s.)?

Only for the a.s. ($EC_{x,a.s.}$): Go to 7

For both formulation ($EC_{x,PPP}$) and a.s. ($EC_{x,a.s.}$): Go to 2

Answer: Measured toxicity data for the formulation and the a.s. are available for daphnia, algae and macrophytes. As these are the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only for these two organism groups. → Go to 2

Measured toxicity data for fish are provided on active substances endpoints. As these are not the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only on actives substances. → Go to 1

STEP 2. Check the plausibility of the measured formulation toxicity ($EC_{x,PPP}$) against the calculated mixture toxicity $EC_{x,mix}$ -CA (assuming CA, Equation 13) for exactly the mixture composition of the a.s. in the formulation ($EC_{x,PPP}$) by means of the model deviation ratio (MDR = $EC_{x,mix}$ -CA/ $EC_{x,PPP}$).

If $MDR = 0.2 - 5$ (CA approximately holds for the mixture)

If $MDR > 5$ (mixture more toxic than CA)

If $MDR < 0.2$ (mixture less toxic than CA)

Equation 13:

$$EC_{X_{mix}-CA} = \left(\sum_{i=1}^n \frac{p_i}{EC_{X_i}} \right)^{-1}$$

Equation 15:

$$MDR = \frac{EC_{X_{mix}-CA} \text{ (calculated mixture toxicity)}}{EC_{X_{PPP}} \text{ (measured mixture toxicity)}}$$

Calculation of the acute mixture toxicity of the formulation

Table 1. Composition of CHR/F/PROTAZO 375 SC

Name/code of the product	CHR/F/PROTAZO		
Name of the active substance A	Azoxystrobin		
Name of the active substance B	Prothioconazole		
Density [g product/cm ³]	1.121		
	Nominal [g a.s./kg or L product]	Fraction considering density [%]	p_{i-mix} = Fraction of active substance i in the mixture with $\sum p_{i-mix} = 100$ [%]

Concentrations of the active substance Azoxystrobin in the product	200	17.8%	53.3%
Concentrations of the active substance Prothioconazole in the product	175	15.6%	46.7%

Table 2. Toxicity of CHR/F/PROTAZO 375 SC and active substance

Endpoint/Test species	Toxicity of the product [mg product/L]	Toxicity of the product (a.s. based) (EC_{x_PPP}) [mg a.s./L]	Toxicity of the a.s. Azoxystrobin (EC_{x_A}) [mg a.s./L]	Toxicity of the a.s. Prothioconazole (EC_{x_B}) [mg a.s./L]	Triggers (from EFSA Journal 2013;11(7):3290)
LC ₅₀ -fish	-	0.000	0.47	1.83	
EC ₅₀ -daphnids	2.42	0.810	0.055	1.3	0.01
EC ₅₀ -algae	3.05	1.020	0.146	2.18	0.1

Table 3. Calculation of toxicity exposure in CHR/F/PROTAZO 375 SC

Toxicity per fraction of the Azoxystrobin ($1/TU_A$) [mg a.s./L]	Toxicity per fraction of the Prothioconazole ($1/TU_B$) [mg a.s./L]	Calculated mixture toxicity (a.s. in product) ($EC_{x_mix_CA} = 1/\sum (TU_i)$) [mg a.s./L]	Model deviation ratio (MDR = $EC_{x_mix_CA}/EC_{x_PPP}$)	$EC_{x_mix_CA}$ (a.s. in product)/ $EC_{x_mix_CA}$ (a.s. in PEC _{mix}) (at lower exposure tier)
0.88125	3.921428571	0.720	-	1.431
0.103125	2.785714286	0.099	0.123	1.656

0.27375	4.671428571	0.259	0.253	1.626
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~~Answer: MDRs for daphnias are below 0.2 , but MDR for daphnias is between 0.2-5. Therefore , go to Step 9 and 3 respectively.~~

~~Step 9. Carefully recheck the apparent antagonism as observed in the measured mixture toxicity data (ECx PPP) regarding potential impacts of the default assumption of CA and/or heterogeneous input data used for the CA calculation. Does the apparent antagonism remain and no toxicologically plausible explanation is available (e.g. special feature of the formulation type)?~~

Yes:	Go to step 3
If measured data are not available or if the assessment in point 3 indicates that the mixtures are not similar (use modified ETR trigger values, see section 10.3.4):	Go to step 8
No:	Go to step 3

~~Answer: Yes. → Go to step 3~~

~~Step 3. Check whether the mixture composition in the formulation study giving the measured mixture toxicity (ECx PPP) in terms of the relative proportions of the individual a.s. is similar to the mixture composition at the PECmix. As a direct comparison on the basis of the relative proportions of the a.s. at the ECx PPP with the relative proportion at the PECmix is not informative as such, the comparison is done based on calculated mixture toxicity (assuming CA) for both mixture compositions. Therefore, calculate ECx mix-CA (see Equation 13) for the mixture composition of the a.s. at the PECmix and compare with the estimate calculated for the formulation (as already done in step 2 above).~~

Table 5. Results of compare ECmix-CA(a.s. in PPP) to ECmix-CA (a.s. in PECmix)

		Triggers
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Endpoint/Test species	EC _x mix-CA (a.s. in product)/EC _x mix-CA (a.s. in PEC _{mix})	0.8-1.2	<0.8 or >1.2
EC50 daphnids	1.680		Yes
ErC50 algae	0.568		Yes

Answer: Calculated factors for daphnia and algae gives results outside 0.8-1.2 Therefore, go to step 5.

STEP 5. Check whether one mixture component clearly drives the toxicity if considering the measured mixture toxicity (EC_x PPP), that is, does the largest part of the sum of toxic units (Equation 14) calculated for the formulation (≥ 90 %) comes from a single a.s. (TUi)?

Table 7. Results of toxicity driver's calculation

Endpoint/Test species	Calculated mixture toxicity (a.s. in product) (EC _x mix-CA) [mg a.s./L]	Azoxystrobin		Prothioconazole		Triggers	
		Toxicity per fraction (1/TU _i) [mg a.s./L]	Deviation from mixture toxicity = $1 - EC_{x \text{ mix-CA}} \times (1/EC_{x \text{ mix-CA}} \cdot TU_i)$ [%]	Toxicity per fraction (1/TU _i) [mg a.s./L]	Deviation from mixture toxicity = $1 - EC_{x \text{ mix-CA}} \times (1/EC_{x \text{ mix-CA}} \cdot TU_i)$ [%]	≥90% for one a.s.	≥90% for no a.s.
EC50 daphnids	0.423	0.431	98.0%	21.429	1.97%	Yes	
ErC50 algae	0.117	0.471	24.9%	0.156	75.1%	Yes	

Answer: Toxicity driver was found for daphnias and algae, therefore mixture toxicity for them is covered by active substance assessment

STEP 7. Is there evidence that synergistic interactions between mixture components might occur (e.g. based on toxicological knowledge from literature or from counter-checking measured and calculated mixture toxicity in other species) which cannot be ruled out for the given species with sufficient certainty?

Answer: No. Therefore, go to step 8.

STEP 8. Conduct a mixture RA based on calculated mixture toxicity

Table 8. Results of exposure of mixture toxicity's calculation to aquatic species for cereals, oilseed rape, ornamental <50 cm, breadseed poppy, mustard, tobacco

Exposure		(lower exposure tier)	(higher exposure tier)											
Exposure tier (FOCUS step)	Azoxystrobin	Step 2	Step 3 (D3 ditch)	Step 3 (D4 pond)	Step 3 (D4 stream)	Step 3 (D5 pond)	Step 3 (D5 stream)	Step 4 (R1 pond, 10m vbf and 5 nsbz)	Step 4 (R1 stream, 10m vbf and 5 nsbz)	Step 4 (R3 stream, 10m vbf and 5 nsbz)	Step 4 (R4 stream, 10m vbf and 5 nsbz)			
PEC _{sw} [mg a.s./L]		0.014720	0.001108	0.000856	0.000902	0.000272	0.000972	0.000172	0.001690	0.001849	0.001907			
Exposure tier (FOCUS step)	Prothioconazole	Step 2	Step 3 (D3)	Step 3 (D4)	Step 3 (D4s)	Step 3 (D5)	Step 3 (D5)	Step 3 (R1)	Step 3 (R1)	Step 3 (R3)	Step 3 (R4 stream)			

step)			ditch)	pond)	trea m)	pon d)	strea m)	pon d)	strea m)	strea m))			
PEC _{sw} [mg a.s./L]		0.001 420	0.000 059	0.00 0061	0.00 0188	0.00 002 6	0.00 007 0	0.00 013 9	0.00 081 4	0.00 079 1	0.0016 04			
Total exposure concentr ation of the mixture (a.s. based) (PEC _{mi x}) [mg/L]	-	0.016 140	0.001 167	0.00 0918	0.00 1090	0.00 029 8	0.00 104 2	0.00 031 1	0.00 250 4	0.00 264 0	0.0035 11			
	-													
Endpoint /Test species	-			Calculated mixture toxicity (a.s. in PEC _{mix}) ($EC_{x-mix-CA} = \sum (p_i \cdot PEC_i / EC_{x-i})$) [mg a.s./L]										
LC50 fish	-	0.503	0.488	0.49 5	0.50 2	0.49 5	0.70 3	0.70 3	0.62 0	0.60 5	0.712			
Endpoint /Test species	-	$ETR_{mix} = PEC_{mix} / EC_{x-PPP}$												Tri age rs
ErC50 algae	-	0.032	0.002	0.00 2	0.00 2	0.00 1	0.00 1	0.00 0	0.00 4	0.00 4	0.005			0.0 1

Table 9. Results of exposure of mixture toxicity's calculation to aquatic species for salix, wicker, forestry tree, ornamental > 50 cm

Exposure		(lower exposure tier)	(higher exposure tier)											
Exposure tier (FOCUS step)	Azoxystrobin	Step 2	Step 4 (D3 ditch, 20m bz)	Step 4 (D4 pond, 20m bz)	Step 4 (D4s trea m, 20m bz)	Step 4 (D5 pond, 20m bz)	Step 4 (D5 strea m, 20m bz)	Step 4 (R1 pond, 20m bz)	Step 4 (R1 strea m, 20m bz)	Step 4 (R3 strea m, 20m bz)	Step 4 (R4 strea m, 20m bz)			
PEC _{sw} [mg a.s./L]		0.124 180	0.001 576	0.00 0295	0.00 1754	0.00 041 9	0.00 186 0	0.00 028 5	0.00 139 1	0.00 197 1	0.00 139 9			
Exposure tier (FOCUS step)	Prothioconazole	Step 2	Step 4 (D3 ditch, 10m bz)	Step 3 (D4 pond)	Step 4 (D4s trea m, 10m bz)	Step 3 (D5 pond)	Step 4 (D5 strea m, 10m bz)	Step 3 (R1 pond)	Step 3 (R1 strea m)	Step 4 (R3 strea m, 10m bz)	Step 3 (R4 strea m)			
PEC _{sw} [mg a.s./L]		0.034 430	0.005 316	0.00 0720	0.00 6920	0.00 074 4	0.00 627 4	0.00 073 2	0.00 936 2	0.00 664 3	0.00 941 7			
Total exposure concentration of the mixture (a.s. based)		0.158 610	0.006 892	0.00 1015	0.00 8674	0.00 116 3	0.00 813 4	0.00 101 7	0.01 075 3	0.00 861 4	0.01 081 6			

(PEC _{mix}) [mg/L]														
Endpoint / Test species				Calculated mixture toxicity (a.s. in PEC _{mix}) $(EC_{x-mix, CA} = \sum (p_i \cdot PEC_i / EC_{x,i}))$ [mg a.s./L]										
LC50 fish		0.560	1.101	0.99 4	0.89 6	1.10 1	1.01 1	1.01 1	1.33 2	1.10 1	1.33 2			
Endpoint / Test species		$ETR_{mix} = PEC_{mix} / EC_{x, PPP}$												Trigger
ErC50 algae		0.283	0.006	0.00 1	0.01 0	0.00 1	0.00 8	0.00 1	0.00 8	0.00 8	0.00 8			0.0 1

Answer: ETR_{mix} for higher exposure tier are below the triggers. Therefore, CHR/F/PROTAZO no poses unacceptable mixture toxicity to aquatic species.

Decision scheme for mixture toxicity risk assessment for CHR/F/PROTAZO 375 for cereals, oilseed rape, mustard, breadseed poppy, soya, sunflower, ornamental and tobacco

Step 1. Are measured toxicity data (EC_x) available for the given endpoint (typically chronic data available only for a.s.)?

Only for the a.s. (EC_{x,a.s.}): Go to 7

For both formulation (EC_{x,PPP}) and a.s. (EC_{x,a.s.}): Go to 2

Answer: Measured toxicity data for the formulation and the a.s. are available for daphnia, algae and macrophytes. As these are the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only for these two organism groups. ☐ Go to 2

Measured toxicity data for fish are provided on active substances endpoints. As these are not the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only on actives substances. ☐ Go to 1

STEP 2. Check the plausibility of the measured formulation toxicity (EC_xPPP) against the calculated mixture toxicity EC_xmix-CA (assuming CA, Equation 13) for exactly the mixture composition of the a.s. in the formulation (EC_xPPP) by means of the model deviation ratio (MDR = EC_xmix-CA/EC_xPPP).

If MDR = 0.2–5 (CA approximately holds for the mixture)

If MDR > 5 (mixture more toxic than CA)

If MDR < 0.2 (mixture less toxic than CA)

Equation 13:

$$EC_{x_{mix-CA}} = \left(\sum_{i=1}^n \frac{p_i}{EC_{x_i}} \right)^{-1}$$

Equation 15:

$$MDR = \frac{EC_{x_{mix-CA}} \text{ (calculated mixture toxicity)}}{EC_{x_{PPP}} \text{ (measured mixture toxicity)}}$$

Calculation of the acute mixture toxicity of the formulation

Table 1. Composition of CHR/F/PROTAZO

Name/code of the product	CHR/F/PROTAZO	
Name of the active substance A	Azoxystrobin	
	Prothioconazole	
Name of the active substance B		
Density [g product/cm ³]	1.121	

	Nominal [g a.s./kg or L product]	Fraction considering density [%]	$p_{i \text{ mix}} = \text{Fraction of active substance } i \text{ in the mixture with } \sum p_{i \text{ mix}} = 100 \text{ [%]}$
Concentrations of the active substance Azoxystrobin in the product	200	17.8%	53.3%
Concentrations of the active substance Prothioconazole in the product	175	15.6%	46.7%

Endpoint/Test species	Toxicity of the product [mg product/L]	Toxicity of the product (a.s. based) ($EC_{x \text{ PPP}}$) [mg a.s./L]	Toxicity of the a.s. Azoxystrobin ($EC_{x \text{ A}}$) [mg a.s./L]	Toxicity of the a.s. Prothioconazole ($EC_{x \text{ B}}$) [mg a.s./L]	Triggers (from EFSA Journal 2013;11(7):3290)
LC50 fish	-	0.000	0.47	1.83	0.01
EC50 daphnids	2.42	0.810	0.23	1.3	0.01
ErC50 algae (Pseudokirchenella)	3.05	1.020	0.36	2.18	0.1

Table 3. Calculation of toxicity exposure in CHR/F/PROTAZO

Toxicity per fraction of the Azoxystrobin (1/TU _A) [mg a.s./L]	Toxicity per fraction of the Prothioconazole (1/TU _B) [mg a.s./L]	Calculated mixture toxicity (a.s. in product) (EC _{x mix-CA} = 1/Σ (TU _i)) [mg a.s./L]	Model deviation ratio (MDR = EC _{x mix-CA} /EC _{x PPP})	EC _{x mix-CA} (a.s. in product)/EC _{x mix-CA} (a.s. in PEC _{mix}) (at lower exposure tier)
0.88125	3.921428571	0.720	-	1.431
0.43125	2.785714286	0.373	0.461	1.506

$$\text{Equation 13: } EC_{x \text{ mix-CA}} = \left(\sum_{i=1}^n \frac{p_i}{EC_{x_i}} \right)^{-1}$$

where:

- n: number of mixture components
- i: index from 1...n mixture components
- p_i: the ith component as a relative fraction of the mixture composition (note: Σ p_i must be 1)
- EC_{x_i}: concentration of component i provoking x % effect (pragmatically, NOEC_i may be inserted, too).

0.675	4.671428571	0.590	0.578	1.518
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Answer: MDRs for algae and daphnias are between 0.2 -5. Therefore , go to Step 3.

Step 3. Check whether the mixture composition in the formulation study giving the measured mixture toxicity (EC_x PPP) in terms of the relative proportions of the individual a.s. is similar to the mixture composition at the PEC_{mix}. As a direct comparison on the basis of the relative proportions of the a.s. at the EC_x PPP with the relative proportion at the PEC_{mix} is not informative as such, the comparison is done based on calculated mixture toxicity (assuming CA) for both mixture compositions. Therefore, calculate EC_{x mix-CA} (see Equation 13) for the mixture composition of the a.s. at the PEC_{mix} and compare with the estimate calculated for the formulation (as already done in step 2 above).

Table 4. Results of compare ECmix-CA(a.s. in PPP) to ECmix-CA (a.s. in PECmix)

Endpoint/Test species	ECx mix-CA (a.s. in product)/ECx mix-CA (a.s. in PECmix)	Triggers	
		0.8-1.2	<0.8 or >1.2
EC50 daphnids	1.506		Yes
ErC50 algae	1.518		Yes

Answer: Calculated factors for daphnia and algae gives results outside 0.8-1.2 Therefore, go to step 5.

STEP 5. Check whether one mixture component clearly drives the toxicity if considering the measured mixture toxicity (ECx PPP), that is, does the largest part of the sum of toxic units (Equation 14) calculated for the formulation ($\geq 90\%$) comes from a single a.s. (TU_i)?

Equation 14:

$$\sum_{i=1}^n TU_i = \sum_{i=1}^n \frac{c_i}{EC_{X_i}}$$

Table 5. Results of toxicity driver's calculation

Endpoint/Test species	Calculated mixture toxicity (a.s. in product) (EC _{x mix-CA}) [mg a.s./L]	Azoxystrobin		Prothioconazole		Triggers	
		Toxicity per fraction (1/TU _i) [mg a.s./L]	Deviation from mixture toxicity = 1-EC _{x mix-CA} x (1/EC _{x mix-CA} -TU _i) [%]	Toxicity per fraction (1/TU _i) [mg a.s./L]	Deviation from mixture toxicity = 1-EC _{x mix-CA} x (1/EC _{x mix-CA} -TU _i) [%]	$\geq 90\%$ for one a.s.	$\geq 90\%$ for no a.s.

EC50 daphnids	0.373	0.431	86.6%	2.786	13.41%		no
ErC50 algae	0.590	0.675	87.4%	4.671	12.6%		no

Answer: No toxicity drivers found for daphnias and algae. Go to step 8.

STEP 7. Is there evidence that synergistic interactions between mixture components might occur (e.g. based on toxicological knowledge from literature or from counter-checking measured and calculated mixture toxicity in other species) which cannot be ruled out for the given species with sufficient certainty?

Answer: No. Therefore, go to step 8

STEP 8. Conduct a mixture RA based on calculated mixture toxicity

Table 7. Results of exposure of mixture toxicity's calculation to aquatic species

Table 9. Results of exposure of mixture toxicity's calculation to aquatic species for salix, wicker, forestry tree, ornamental > 50 cm

Exposure		(lower exposure tier)	(higher exposure tier)												
Exposure tier (FOCUS step)	Azoxystrobin	Step 2	Step 3 (D3 ditch)	Step 3 (D4 pond)	Step 3 (D4stream)	Step 3 (D5 pond)	Step 3 (D5 stream)	Step 4 (R1 pond, 10m vbf and 5 nsbz)	Step 4 (R1 stream, 10m vbf and 5 nsbz)	Step 4 (R3 stream, 10m vbf and 5 nsbz)	Step 4 (R4 stream, 10m vbf and 5 nsbz)				
PEC _{sw} [mg a.s./L]		0.014720	0.001108	0.000856	0.000902	0.000272	0.000972	0.000172	0.001690	0.001849	0.001907				
Exposure tier (FOCUS step)	Prothioconazole	Step 2	Step 3 (D3 ditch)	Step 3 (D4 pond)	Step 3 (D4stream)	Step 3 (D5 pond)	Step 3 (D5 stream)	Step 3 (R1 pond)	Step 3 (R1 stream)	Step 3 (R3 stream)	Step 3 (R4 stream)				

PECsw [mg a.s./L]		0.001420	0.000059	0.000061	0.000188	0.000026	0.000070	0.000139	0.000081	0.000079	0.001604			
Total exposure concentration of the mixture (a.s. based) (PECmix) [mg/L]		0.016140	0.001167	0.000917	0.001090	0.000298	0.001042	0.000311	0.002504	0.002640	0.003511			
Endpoint/Test species		Calculated mixture toxicity (a.s. in PEC _{mix}) (EC _{1,mix,CA} = $\sum (p_{i,PEC}/EC_{i,PEC})$) [mg a.s./L]												
LC50 fish		0.503	0.488	0.494	0.503	0.495	0.704	0.704	0.620	0.605	0.712			
EC50 daphnids		0.248	0.240	0.243	0.268	0.248	0.243	0.364	0.314	0.305	0.369			
ErC50 algae (Pseudokirchnerella)		0.389	0.376	0.381	0.421	0.388	0.381	0.574	0.494	0.480	0.582			
Endpoint/Test species		ETR _{mix} = PEC _{mix} /EC _{1,ref}												
LC50 fish		0.032	0.002	0.002	0.002	0.001	0.001	0.000	0.004	0.004	0.005			Trigger s 0.01
EC50 daphnids		0.065	0.005	0.004	0.004	0.001	0.004	0.001	0.008	0.009	0.010			0.01
ErC50 algae (Pseudokirchnerella)		0.042	0.003	0.002	0.003	0.001	0.003	0.001	0.005	0.005	0.006			0.1

Answer: ETR_{mix} for higher exposure tier are below the triggers. Therefore, CHR/F/PROTAZO no poses unacceptable mixture toxicity to aquatic species with 10meters vegetative and 5 meters no spray buffer zone for cereals, oilseed rapes, mustard, breadseed poppy, soya, sunflower, ornamental and tobacco.

Decision scheme for mixture toxicity risk assessment for CHR/F/PROTAZO 375 for forestry tree, ornamental >50 cm, tobacco

Step 1. Are measured toxicity data (ECx) available for the given endpoint (typically chronic data available only for a.s.)?

Only for the a.s. (ECx a.s.): Go to 7

For both formulation (EC_{xPPP}) and a.s. (EC_{xa.s.}): Go to 2

Answer: Measured toxicity data for the formulation and the a.s. are available for daphnia, algae and macrophytes. As these are the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only for these two organism groups. □ Go to 2

Measured toxicity data for fish are provided on active substances endpoints. As these are not the most sensitive aquatic organisms, it is justified to conduct the mixture toxicity risk assessment only on actives substances. □ Go to 1

STEP 2. Check the plausibility of the measured formulation toxicity (EC_{xPPP}) against the calculated mixture toxicity EC_{xmix-CA} (assuming CA, Equation 13) for exactly the mixture composition of the a.s. in the formulation (EC_{xPPP}) by means of the model deviation ratio (MDR = EC_{xmix-CA}/EC_{xPPP}).

If MDR = 0.2–5 (CA approximately holds for the mixture)

If MDR > 5 (mixture more toxic than CA)

If MDR < 0.2 (mixture less toxic than CA)

Equation 13:

$$EC_{x_{mix-CA}} = \left(\sum_{i=1}^n \frac{p_i}{EC_{x_i}} \right)^{-1}$$

Equation 15:

$$MDR = \frac{EC_{x_{mix-CA}} \text{ (calculated mixture toxicity)}}{EC_{x_{PPP}} \text{ (measured mixture toxicity)}}$$

Calculation of the acute mixture toxicity of the formulation

Table 1. Composition of CHR/F/PROTAZO

Name/code of the product	CHR/F/PROTAZO
Name of the active substance A	Azoxystrobin

Name of the active substance B	Prothioconazole		
Density [g product/cm ³]	1.121		
	Nominal [g a.s./kg or L product]	Fraction considering density [%]	$p_{i\text{ mix}} = \text{Fraction of active substance } i \text{ in the mixture with } \sum p_{i\text{ mix}} = 100 [\%]$
Concentrations of the active substance Azoxystrobin in the product	200	17.8%	53.3%
Concentrations of the active substance Prothioconazole in the product	175	15.6%	46.7%

Endpoint/Test species	Toxicity of the product [mg product/L]	Toxicity of the product (a.s. based) (EC _{x PPP}) [mg a.s./L]	Toxicity of the a.s. Azoxystrobin (EC _{x A}) [mg a.s./L]	Toxicity of the a.s. Prothioconazole (EC _{x B}) [mg a.s./L]	Triggers (from EFSA Journal 2013;11(7):3290)
LC50 fish		0.000	0.47	1.83	0.01
EC50 daphnids	2.42	0.810	0.23	1.3	0.01
ErC50 algae (Pseudokirichenella)	3.05	1.020	0.36	2.18	0.1

Table 3. Calculation of toxicity exposure in CHR/F/PROTAZO

Toxicity per fraction of the Azoxystrobin (1/TU _A) [mg a.s./L]	Toxicity per fraction of the Prothioconazole (1/TU _B) [mg a.s./L]	Calculated mixture toxicity (a.s. in product) (EC _{x mix-CA} = 1/Σ (TU _i)) [mg a.s./L]	Model deviation ratio (MDR = EC _{x mix-CA} /EC _{x PPP})	EC _{x mix-CA} (a.s. in product)/EC _{x mix-CA} (a.s. in PEC _{mix}) (at lower exposure tier)
0.88125	3.921428571	0.720	-	1.284

$$\text{Equation 13: } EC_{x \text{ mix-CA}} = \left(\sum_{i=1}^n \frac{p_i}{ECx_i} \right)^{-1}$$

where:

- n: number of mixture components
- i: index from 1...n mixture components
- p_i: the ith component as a relative fraction of the mixture composition (note: Σ p_i must be 1)
- ECx_i: concentration of component i provoking x % effect (pragmatically, NOEC_i may be inserted, too).

0.43125	2.785714286	0.373	0.461	1.334
0.675	4.671428571	0.590	0.578	1.341

Answer: MDRs for algae and daphnias are between 0.2 -5. Therefore , go to Step 3.

Step 3. Check whether the mixture composition in the formulation study giving the measured mixture toxicity (EC_x PPP) in terms of the relative proportions of the individual a.s. is similar to the mixture composition at the PEC_{mix}. As a direct comparison on the basis of the relative proportions of the a.s. at the EC_x PPP with the relative proportion at the PEC_{mix} is not informative as such, the comparison is done based on calculated mixture toxicity (assuming CA) for both mixture compositions. Therefore, calculate EC_x mix-CA (see Equation 13) for the mixture composition of the a.s. at the PEC_{mix} and compare with the estimate calculated for the formulation (as already done in step 2 above).

Table 4. Results of compare ECmix-CA(a.s. in PPP) to ECmix-CA (a.s. in PECmix)

Endpoint/Test species	ECx mix-CA (a.s. in product)/ECx mix-CA (a.s. in PECmix)	Triggers	
		0.8-1.2	<0.8 or >1.2
EC50 daphnids	1.334		Yes
ErC50 algae	1.341		Yes

Answer: Calculated factors for daphnia and algae gives results outside 0.8-1.2 Therefore, go to step 5.

STEP 5. Check whether one mixture component clearly drives the toxicity if considering the measured mixture toxicity (ECx PPP), that is, does the largest part of the sum of toxic units (Equation 14) calculated for the formulation ($\geq 90\%$) comes from a single a.s. (TUi)?

Equation 14:

$$\sum_{i=1}^n TU_i = \sum_{i=1}^n \frac{c_i}{EC_{X_i}}$$

Table 5. Results of toxicity driver's calculation

Azoxystrobin	Prothioconazole	Triggers
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Endpoint/Test species	Calculated mixture toxicity (a.s. in product) ($EC_{x \text{ mix-CA}}$) [mg a.s./L]	Toxicity per fraction ($1/TU_i$) [mg a.s./L]	Deviation from mixture toxicity = $1-EC_{x \text{ mix-CA}} \times (1/EC_{x \text{ mix-CA}}-TU_i)$ [%]	Toxicity per fraction ($1/TU_i$) [mg a.s./L]	Deviation from mixture toxicity = $1-EC_{x \text{ mix-CA}} \times (1/EC_{x \text{ mix-CA}}-TU_i)$ [%]	>=90% for one a.s.	>=90% for no a.s.
EC50 daphnids	0.373	0.431	86.6%	2.786	13.41%		no
ErC50 algae	0.590	0.675	87.4%	4.671	12.6%		no

Answer: No toxicity drivers found for daphnias and algae. Go to step 8.

STEP 7. Is there evidence that synergistic interactions between mixture components might occur (e.g. based on toxicological knowledge from literature or from counter-checking measured and calculated mixture toxicity in other species) which cannot be ruled out for the given species with sufficient certainty?

Answer: No. Therefore, go to step 8

STEP 8. Conduct a mixture RA based on calculated mixture toxicity

Table 7. Results of exposure of mixture toxicity's calculation to aquatic species

Table 9. Results of exposure of mixture toxicity's calculation to aquatic species for salix, wicker, forestry tree, ornamental > 50 cm

Exposure		(lower exposure tier)	(higher exposure tier)											
Exposure tier (FOCUS step)	Azoxystrobin	Step 2	Step 4 (D3 ditch, 20m bz)	Step 4 (D4 pond, 20m bz)	Step 4 (D4stream, 20m bz)	Step 4 (D5 pond, 20m bz)	Step 4 (D5 stream, 20m bz)	Step 4 (R1 pond, 20m bz)	Step 4 (R1 stream, 20m bz)	Step 4 (R3 stream, 20m bz)	Step 4 (R4 stream, 20m bz)			
PEC _{sw} [mg a.s./L]		0.124180	0.001576	0.000295	0.001754	0.000419	0.001860	0.000285	0.001391	0.001971	0.001399			

Exposure tier (FOCUS step)	Prothioconazole	Step 2	Step 4 (D3 ditch, 20m bz)	Step 3 (D4 pond)	Step 4 (D4stream, 20m bz)	Step 3 (D5 pond)	Step 4 (D5 stream, 10m bz)	Step 3 (R1 pond)	Step 3 (R1 stream)	Step 4 (R3 stream, 10m bz)	Step 3 (R4 stream)			
PEC _{sw} [mg a.s./L]		0.034430	0.001376	0.000720	0.001532	0.000744	0.001624	0.000732	0.001215	0.001719	0.001222			
Total exposure concentration of the mixture (a.s. based) (PEC _{mix}) [mg/L]		0.158610	0.002952	0.001015	0.003286	0.001163	0.003484	0.001017	0.002606	0.003690	0.002621			
Endpoint/Test species		Calculated mixture toxicity (a.s. in PEC _{mix}) ($EC_{50mixture} = \sum (p_{iPEC}/EC_{50i})$) [mg a.s./L]												
LC50 fish		0.560	0.719	0.994	0.896	0.719	1.011	1.011	0.719	0.719	0.719			
EC50 daphnids		0.280	0.373	0.553	0.373	0.486	0.373	0.564	0.373	0.373	0.373			
ErC50 algae (Pseudokirchnerella)		0.440	0.589	0.883	0.589	0.772	0.589	0.902	0.589	0.589	0.589			
Endpoint/Test species		ETR _{mix} = PEC _{mix} /EC _{50m}												
LC50 fish		0.283	0.004	0.001	0.004	0.002	0.003	0.001	0.004	0.005	0.004			Trigger s 0.01
EC50 daphnids		0.566	0.008	0.002	0.009	0.002	0.009	0.002	0.007	0.010	0.007			0.01
ErC50 algae (Pseudokirchnerella)		0.361	0.005	0.001	0.006	0.002	0.006	0.001	0.004	0.006	0.004			0.1

Answer: ETR_{mix} for higher exposure tier are below the triggers. Therefore, CHR/F/PROTAZO no poses unacceptable mixture toxicity to aquatic species with 20 meters vegetative and no-spray buffer zone for forestry tree, Salix and Wicker.

9.5.3 Overall conclusions

Based on the predicted rates of CHR/F/PROTAZO 375 SC, the TER values describing the risk for aquatic species following exposure to CHR/F/PROTAZO 375 SC according to the GAP of the formulation CHR/F/PROTAZO 375 SC achieve the acceptability criteria with **applying appropriate mitigation measures**

- ~~10 m vegetative and 5 m buffer zone for cereals, oilseed rape, breadseed poppy, mustard, ornamental <50 cm~~
- ~~20 meters vegetative and 5 meters buffer zone for ornamental <50cm for MS where R4 stream is relevant scenario~~
- ~~20m vegetive and 20m nospray buffer zone for forestry tree, tobacco > 50 cm~~

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 initial risk assessment was based on the worst case PEC values and the results of laboratory toxicity testing. The PEC_{sw} Step 1-2 and Step 3 and 4 were used.

The CHR/F/PROTAZO 375 SC applications close to surface water pose acceptable risk to aquatic organisms with appropriate mitigation measures.

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 Prothioconazole, azoxystrobin and their relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on bees of CHR/F/PROTAZO were not evaluated as part of the EU assessment of Prothioconazole and azoxystrobin. New data submitted with this application are listed in Table 9.6.-1 and summarised in Appendix 2.

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

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

Species	Substance	Exposure System	Results	Reference
Apis mellifera	Prothioconazole	Oral	LD ₅₀ > 71 µg/bee	EFSA Scientific Report (2007) 106
Apis mellifera	Prothioconazole	Contact	LD ₅₀ > 200 µg/bee	EFSA Scientific Report (2007) 106
Apis mellifera	Azoxystrobin	Oral	LD ₅₀ > 25 µg/bee	EFSA Journal 2010; 8(4):1542
Apis mellifera	Azoxystrobin	Contact	LD ₅₀ > 200 µg/bee	EFSA Journal 2010;

Species	Substance	Exposure System	Results	Reference
				8(4):1542
Apis mellifera	CHR/F/PROTAZO	Oral	LD ₅₀ > 200 µg/bee	M. Grzesica, 2019 Study code: B/42/19
Apis mellifera	CHR/F/PROTAZO	Contact	LD ₅₀ > 200 µg/bee	M. Grzesica, 2019 Study code: B/43/19
Apis mellifera	CHR/F/PROTAZO	Chronic Oral	LC50= 1837.4578 [mg of prod/kg od food] LDD50= 41.0230 [µg of prod/bee/day]	U. Orzechowska, Study code: 0038/0018/E
Apis Mellifera	CHR/F/PROTAZO	Larval	LC50= 1214.545 [mg of prod/kg od food] LDD50= 186.854 [µg of prod/larva]	U. Orzechowska, Study code: 0038/0016/E
Higher-tier studies (tunnel test, field studies)				

9.6.2 Risk assessment

The evaluation of the risk for bees was performed in accordance with the recommendations of the “Guidance Document on Terrestrial Ecotoxicology”, as provided by the Commission Services (SANCO/10329/2002 rev.2 (final), October 17, 2002). To achieve a concise risk assessment, the risk envelope approach is applied.

9.6.2.1 Hazard quotients for bees

Table 9.6-2: First-tier assessment of the risk for bees due to the use of CHR/F/PROTAZO in cereals (worst case use from GAP table)

Intended use			
Active substance		Prothioconazole	
Application rate (g/ha)		2 × 175	
Test design	LD ₅₀ (lab.) (µg/bee)	Single application rate (g/ha)	Q _{HO} , Q _{HC} criterion: Q _H ≤ 50
Oral toxicity	71	175	2.46
Contact toxicity	200		0.875
Product		Azoxystrobin	
Application rate (g/ha)		2 × 200	
Test design	LD ₅₀ (lab.) (µg/bee)	Single application rate (g/ha)	Q _{HO} , Q _{HC} criterion: Q _H ≤ 50
Oral toxicity	25	200	8
Contact toxicity	200		1
Product		CHR/F/PROTAZO	

Application rate (g/ha)		2×1121	
Test design	LD ₅₀ (lab.) (µg/bee)	Single application rate (g/ha)	Q _{HO} , Q _{HC} criterion: Q _H ≤ 50
Oral toxicity	200	1121	5.6
Contact toxicity	200		5.6

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

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

Not relevant.

9.6.3 Effects on bumble bees

Not required.

9.6.4 Effects on solitary bees

Not required.

9.6.5 Overall conclusions

All hazard quotients (HQ) are considerably less than 50, indicating that CHR/F/PROTAZO 375 SC applied at the maximum use rate poses low risk to bees.

Review Comments:

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

The submitted risk assessment, based on laboratory studies, has been accepted. It can therefore be concluded that there will be negligible risk associated with the exposure of bees to CHR/F/PROTAZO 375 SC.

The applicant fulfilled the data requirements according to Commission regulation No. 284/2013 and submitted chronic tests on bees with formulated product.

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 Prothioconazole, azoxystrobin and their relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents.

Effects on non-target arthropods of CHR/F/PROTAZO were not evaluated as part of the EU assessment of Prothioconazole and azoxystrobin. New data submitted with this application are listed in Appendix I

and summarised in Appendix 2.

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

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

Species	Substance	Exposure System	Results	Reference
<i>Typhlodromus pyri</i> (protonymphs)	CHR/F/PROTAZO	Laboratory test glass plates (2D)	LR ₅₀ = 1.7 L/ha, which is equivalent to 1905.7 g prod/ha	M. Grzesica, 2020, Study code: B/40/19
<i>Aphidius rhopalosiphi</i> (adults)	CHR/F/PROTAZO	Laboratory test glass plates (2D)	LR ₅₀ = 1.7 L/ha, which is equivalent to 1905.7 g prod/ha	M, Grzesica 2020, Study code: B/41/19
Field or semi-field tests				

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.

Table 9.7-2: First- and higher-tier assessment of the in-field risk for non-target arthropods due to the use of CHR/F/PROTAZO in cereals (worse case use from GAP table)

Intended use			
Active substance/product		CHR/F/PROTAZO	
Application rate (g/ha)		2 x 1121	
MAF		1.7	
Test species	LR₅₀ (lab.) (g/ha)	PER_{in-field} (g/ha)	HQ_{in-field} criterion: HQ ≤ 2
Tier I			
<i>Typhlodromus pyri</i>	1905.7 (1.7L/ha)	1905.7 (1.7L/ha)	1
<i>Aphidius rhopalosiphi</i>	1905.7 (1.7L/ha)		1

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.

9.7.2.2 Risk assessment for off-field exposure

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

Table 9.7-3: First- and higher-tier assessment of the off-field risk for non-target arthropods due to the use of CHR/F/PROTAZO in cereals (worse case use from GAP table)

Intended use		Field crops			
Active substance/product		CHR/F/PROTAZO			
Application rate (g/ha)		2 × 1121			
MAF		1.7			
vdf		10 (Tier 1)			
Test species Tier I	LR ₅₀ (lab.) (g/ha)	Drift rate	PER _{off-field} (g/ha)	CF	HQ _{off-field} criterion: HQ ≤ 2
<i>Typhlodromus pyri</i>	1905.7	0.0238	4.54	10	0.0238
<i>Aphidius rhopalosiphi</i>	1905.7				0.0238
Intended use		Ornamentals > 50 cm			
Active substance/product		CHR/F/PROTAZO			
Application rate (g/ha)		2 × 1121			
MAF		1.7			
vdf		10 (Tier 1)			
Test species Tier I	LR ₅₀ (lab.) (g/ha)	Drift rate	PER _{off-field} (g/ha)	CF	HQ _{off-field} criterion: HQ ≤ 2
<i>Typhlodromus pyri</i>	1905.7	0.0732	13.95	10	0.0732
<i>Aphidius rhopalosiphi</i>	1905.7				0.0732
Intended use		Salix and Wicker			
Active substance/product		CHR/F/PROTAZO			
Application rate (g/ha)		2 × 1121			
MAF		1.7			
vdf		10 (Tier 1)			
Test species Tier I	LR ₅₀ (lab.) (g/ha)	Drift rate	PER _{off-field} (g/ha)	CF	HQ _{off-field} criterion: HQ ≤ 2
<i>Typhlodromus pyri</i>	1905.7	0.2553	48.65	10	0.255
<i>Aphidius rhopalosiphi</i>	1905.7				0.255

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.

~~Calculations for cereals covered calculation for oilseed rape, mustard, soya, sunflower, breadseeds poppy, tobacco and ornamentals. Calculation for forestry tree covered calculation for Salix and Wicker~~

9.7.2.3 Additional higher-tier risk assessment

Not relevant.

9.7.2.4 Risk mitigation measures

No risk mitigation needed.

9.7.3 Overall conclusions

All hazard quotients (HQ) are considerably less than 2, indicating that CHR/F/PROTAZO 375 SC applied at the maximum use rate poses no risk to non-target arthropods. No risk mitigation needed.

Review Comments:

Based on Tier 1 risk assessment it can be concluded that low risk for non-target arthropods is expected from the use of CHR/F/PROTAZO 375 SC according to the proposed use pattern. No unacceptable effects on non-target arthropods are expected in in-field and off-field habitats.

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 Prothioconazole, azoxystrobin and their relevant metabolites. Full details of these studies are provided in the respective EU DAR and related documents. Effects on earthworms and other non-target soil organisms (meso- and macrofauna) of CHR/F/PROTAZO were not evaluated as part of the EU assessment of prothioconazole and azoxystrobin. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

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

Table 9.8-1: 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>	Prothioconazole-dethio	long term	NOEC 1 mg p.m./kg dw	EFSA Scientific Report (2007) 106
<i>Eisenia fetida</i>	Prothioconazole-S-methyl	long-term	NOEC= 100 mg p.m./kg dw	EFSA Scientific Report (2007) 106
<i>Eisenia fetida</i>	Azoxystrobin	long-term	NOEC= 20 mg a.s./kg d.w soil	EFSA Journal 2010; 8(4):1542
<i>Eisenia andrei</i>	CHR/F/PROTAZO	long term	NOEC \geq 1000 mg test item/kg dw	A. Wróblel, 2019, Study code: G/34/19
<i>Folsomia candida</i>	Prothioconazole	long term	NOEC= 64 mg a.s/kg dw	EFSA Scientific Report (2007) 106
<i>Folsomia candida</i>	Prothioconazole-dethio	long term	NOEC= 62.5 mg p.m./kg dw	EFSA Scientific Report (2007) 106
<i>Folsomia candida</i>	Prothioconazole-S-methyl	long term	NOEC= 31.6 mg p.m./kg bw	EFSA Scientific Report (2007) 106
<i>Folsomia candida</i>	Azoxystrobin	long term	NOEC = 25 50 mg a.s./kg dw soil	EFSA Journal 2010; 8(4):1542

Species	Substance	Exposure System	Results	Reference
<i>Folsomia candida</i>	CHR/F/PROTAZO	long term	NOEC= 180 mg test item/kg dw	P. Holewik, 2019, Study code: G/35/19
<i>Hypoaspis aculeifer</i>	Prothioconazole	long term	NOEC= 100 mg a.s./kg dw	EFSA Scientific Report (2007) 106
<i>Hypoaspis aculeifer</i>	CHR/F/PROTAZO	long term	NOECrep.= 1000 mg test item/kg dw EC10rep. = 923.541 mg tet item/kg dw	P. Holewik, 2019, Study code: G/36/19
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.

9.8.2 Risk assessment

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

9.8.2.1 First-tier risk assessment

The relevant PEC_{soil} for risk assessments covering the proposed use pattern are taken from Section 8 (Environmental Fate), Chapter 8.7.2, Table 8.7-3. According to the assessment of environmental-fate data, multi-annual accumulation in soil is need to be considered for prothioconazole and azoxystrobin. To achieve a concise risk assessment, the risk envelope approach is applied.

Table 9.8-2: First-tier assessment of the acute and chronic risk for earthworms and other non-target soil organisms (meso- and macrofauna) due to the use of CHR/F/PROTAZO in cereals (worst case use from GAP table)

Intended use					
Acute effects on earthworms					
Product/active substance	LC ₅₀ (mg/kg dw)	PEC _{soil} (mg/kg dw)		TER _a (criterion TER ≥ 10)	
Not required					
Chronic effects on earthworms					
Product/active substance	NOEC (mg/kg dw)*	PEC _{soil} (mg/kg dw)		TER _{lt} (criterion TER ≥ 5)	
Prothioconazole	-	-		-	
Prothioconazole-desthio	0.5	0.0873	0.0944	5.72	5.3

Prothioconazole-S-methyl	50	0.0208	0.0214	2.403	2336.4
Azoxystrobin	10	0.4189	0.6765	23	14.8
CHR/F/PROTAZO	500	2.3915	1.1957	209	418.2
Chronic effects on other soil macro- and mesofauna <i>Folsomia candida</i>					
Product/active substance	NOEC* (mg/kg dw)	PEC_{soil} (mg/kg dw)		TER_{lt} (criterion TER ≥ 5)	
Prothioconazole	32	0.3382	0.3397	94	94.2
Prothioconazole-desthio	31.25	0.0873	0.0944	357	331.0
Prothioconazole-S-methyl	15.8	0.0208	0.0214	759	738.3
Azoxystrobin	12.5 25	0.4189	0.6765	30	37.0
CHR/F/PROTAZO	90	2.3915	1.1957	37	75.3
Chronic effects on other soil macro- and mesofauna <i>Hypoaspis aculeifer</i>					
Product/active substance	NOEC* (mg/kg dw)	PEC_{soil} (mg/kg dw)		TER_{lt} (criterion TER ≥ 5)	
Prothioconazole	50	0.3382	0.3397	147	147.2
CHR/F/PROTAZO	500 461.77	2.3915	1.1957	209	386.2

TER values shown in bold fall below the relevant trigger.

* NOEC correct by factor 2 (log Pow > 2)

9.8.2.2 Higher-tier risk assessment

Not relevant.

9.8.3 Overall conclusions

The long term risk to earthworms and other non-target soil organisms (meso- and macrofauna) was assessed as low for CHR/F/PROTAZO 375 SC in a first-tier risk assessment.

Review Comments:

The long-term risks of CHR/F/PROTAZO 375 SC to soil meso- and macro-organisms were assessed from toxicity exposure ratios between toxicity endpoints and maximum PEC_{soil}. The relevant predicted environmental concentrations in soil (PEC_{soil}) for risk assessments covering the proposed use pattern are taken from Part B Section 8 (Environmental Fate).

Safe use of CHR/F/PROTAZO 375 SC was confirmed based on TER_{LT} calculations for active substances, their metabolites and for formulation.

9.9 Effects on soil microbial activity (KCP 10.5)

9.9.1 Toxicity data

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

Effects on soil microorganisms of CHR/F/PROTAZO were not evaluated as part of the EU assessment of prothioconazole and azoxystrobin. New data submitted with this application are listed in Appendix 1 and summarised in Appendix 2.

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

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

Endpoint	Substance	Exposure System	Results	Reference
N-mineralisation	Prothioconazole	28 d, aerobic soil type	2.0 kg a.s./ha (≥ 2.71 mg/kg soil dw)	EFSA Scientific Report (2007) 106,
N-mineralisation	Prothioconazole-desthio	28 d, aerobic soil type	0.2 kg p.m./ha (≥ 0.274 mg/kg soil dw)	EFSA Scientific Report (2007) 106,
N-mineralisation	Prothioconazole-desthio	28 d, aerobic soil type	1.0 kg p.m./ha (≥ 1.37 mg/kg soil dw)	EFSA Scientific Report (2007) 106,
N-mineralisation	Prothioconazole-S-methyl	28 d, aerobic type soil	2.0 kg p.m/ha (≥ 2.71 mg/kg soil dw)	EFSA Scientific Report (2007) 106,
N-mineralisation	R 234886	28 d, aerobic type soil	No effect at 1 and 10 mg/kg soil dry weight	EFSA Journal 2010; 8(4):1542
N-mineralisation	R401553	28 d, aerobic type soil	No effect at 0.528 and 2.643 mg test item /kg dry wt soil	EFSA Journal 2010; 8(4):1542
N-mineralisation	R401173	28 d, aerobic type soil	No effect at 0.826 and 4.131 mg test item/kg dry soil	EFSA Journal 2010; 8(4):1542
N-mineralisation	CHR/F/PROTAZO	28 d, aerobic type soil	11.96 mg of the test item / kg dry weight of soil (1.94 mg of the prothioconazole/kg dry weight of soil and 2.19 mg of the azoxystrobin/kg dry weight of soil) and 5 x PEC: 59.78 mg of the test item / kg dry weight of soil (9.70 mg of the prothioconazole/kg dry weight of soil and 10.95 mg of the azoxystrobin/kg dry weight of soil) did not exceed 25% on 28 day of analysis.	P. Holewik, 2019, study code: G/37/19

9.9.2 Risk assessment

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

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

Table 9.9-2: Assessment of the risk for effects on soil micro-organisms due to the use of CHR/F/PROTAZO in cereals (worse case use from GAP table)

Intended use				
N-mineralisation				
Product/active substance	Max. conc. with effects $\leq 25\%$ (mg/kg dw)	PEC_{soil} (mg/kg dw)		Risk acceptable?
Prothioconazole	2.71	0.3382	0.3397	yes
Prothioconazole-desthio	1.37	0.0873	0.0944	yes
Prothioconazole-S-methyl	2.69	0.0208	0.0214	yes
R 234886	No effect at 1 and 10 mg/kg soil dry weight	0.0266	0.0564	yes
R401553	No effect at 0.528 and 2.643 mg test item /kg dry wt soil	0.0003	0.0004	yes
R402173	No effect at 0.826 and 4.131 mg test item/kg dry soil	0.0016	0.0026	yes
CHR/F/PROTAZO	11.96 mg of the test item / kg dry weight of soil (1.94 mg of the prothioconazole/kg dry weight of soil and 2.19 mg of the azoxystrobin/kg dry weight of soil) and 5 x PEC: 59.78 mg of the test item / kg dry weight of soil (9.70 mg of the prothioconazole/kg dry weight of soil and 10.95 mg of the azoxystrobin/kg dry weight of soil) did not exceed 25% on 28 day of analysis.	2.3915	1.1957	yes
C-mineralisation				

Product/active substance	Max. conc. with effects $\leq 25\%$ (mg/kg dw)	PEC _{soil} (mg/kg dw)	Risk acceptable?
Not required			

9.9.3 Overall conclusions

The Predicted Environmental Concentrations of the formulation CHR/F/PROTAZO 375 SC and its active substance prothioconazole in soil are below the concentrations at which no unacceptable effects ($< 25\%$) regarding the soil microbial activity were observed after 28 days or more of exposure, indicating that the proposed use of CHR/F/PROTAZO 375 SC poses an acceptable risk to soil microorganisms.

Review Comments:

CHR/F/PROTAZO 375 SC had no significant effect on soil micro-organisms at 59.78 mg form./kg dry soil. Based on it, can be concluded that CHR/F/PROTAZO 375 SC under field conditions, use at the proposed rates poses no unacceptable risk to non-target soil micro-organisms.

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

9.10.1 Toxicity data

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

Effects on non-target terrestrial plants of CHR/F/PROTAZO were not evaluated as part of the EU assessment of prothioconazole and azoxystrobin. New data submitted with this application are listed in Appendix 1 summarised in Appendix 2.

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

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

Species	Substance	Exposure System	Results (plant without roots)	Reference
Helianthus annuus	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/39/19
Brassica oleracea var. capitata	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/39/19
Pisum sativum	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g	P. Holewik, 2019, Study code: G/39/19

Species	Substance	Exposure System	Results (plant without roots)	Reference
			test item/ha	
Daucus carota	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/39/19
Lolium perenne	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/39/19
Avena sativa	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/39/19
Helianthus annuus	CHR/F/PROTAZO	21 d Vegetative vigor	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha ER50 > 666.7 ml test item/ha, which is equivalent to 1626.08 g test item/ha	P. Holewik, 2019, Study code: G/38/19
Brassica oleracea var. capitata	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 747 g test item/ha	P. Holewik, 2019, Study code: G/38/19
Pisum sativum	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/38/19
Daucus carota	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/38/19
Lolium perenne	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/38/19
Avena sativa	CHR/F/PROTAZO	21 d Seedling emergence	ER50 > 2000 ml test item/ha, which is equivalent to 2242 g test item/ha	P. Holewik, 2019, Study code: G/38/19

m: monocotyledonous; d: dicotyledonous

9.10.2 Risk assessment

9.10.2.1 Tier-1 risk assessment (based screening data)

Not relevant.

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

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

Table 9.10-2: Assessment of the risk for non-target plants due to the use of CHR/F/PROTAZO in cereals (drift rate: 2.77%) – risk envelope cover: oilseed rape, mustard, breadseed poppy, ornamentals, tobacco, soya, sunflower

Intended use		CHR/F/PROTAZO		
Active substance/product		2 × 1121		
Application rate (g/ha)		1.7		
MAF				
Test species	ER ₅₀ (g/ha)	Drift rate	PER _{off-field} (g/ha)	TER criterion: TER ≥ 5
Helianthus annuus	2242	0.0238	45.36	49.43
Brassica oleracea var. capitata	2242	0.0238	45.36	49.43
Pisum sativum	2242	0.0238	45.36	49.43
Daucus carota	2242	0.0238	45.36	49.43
Lolium perenne	2242	0.0238	45.36	49.43
Avena sativa	2242	0.0238	45.36	49.43
Helianthus annuus	747	0.0238	45.36	16.47
Brassica oleracea var. capitata	2242	0.0238	45.36	49.43
Pisum sativum	2242	0.0238	45.36	49.43
Daucus carota	2242	0.0238	45.36	49.43
Lolium perenne	2242	0.0238	45.36	49.43
Avena sativa	2242	0.0238	45.36	49.43
Intended use		Worst case drift rate for single application rate (covers all intended uses)		

Active substance/product		CHR/F/PROTAZO		
Application rate (mL/ha)		1000		
Test species	ER₅₀ (mL/ha)	Drift rate	PER_{off-field} (mL/ha)	TER criterion: TER ≥ 5
All species	2000	0.292	292	6.8

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

~~Calculations for cereals covered calculation for oilseed rape, mustard, soya, sunflower, breadseeds poppy, tobacco and ornamentals. Calculation for forestry tree covered calculation for Salix and Wicker.~~

~~Table 9.10-3: Assessment of the risk for non-target plants due to the use of
CHR/F/PROTAZO in cereals (drift rate: 2.77%)—risk envelope cover: oilseed
rape, mustard, breadseed poppy, ornamentals, tobacco, soya, sunflower~~

Intended use				
Active substance/product		CHR/F/PROTAZO		
Application rate (g/ha)		2 × 1121		
MAF		1.7		
Test species	ER₅₀ (g/ha)	Drift rate	PER_{off-field} (g/ha)	TER criterion: TER ≥ 5
Helianthus annuus	2242	0.0723	137.78	16.27
Brassica oleracea var. capitata	2242	0.0723	137.78	16.27
Pisum sativum	2242	0.0723	137.78	16.27
Daucus carota	2242	0.0723	137.78	16.27
Lolium perenne	2242	0.0723	137.78	16.27
Avena sativa	2242	0.0723	137.78	16.27
Helianthus annuus	747	0.0723	137.78	5.42
Brassica oleracea var. capitata	2242	0.0723	137.78	16.27
Pisum sativum	2242	0.0723	137.78	16.27
Daucus carota	2242	0.0723	137.78	16.27
Lolium perenne	2242	0.0723	137.78	16.27
Avena sativa	2242	0.0723	137.78	16.27

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

9.10.2.3 Higher-tier risk assessment

Not relevant.

9.10.2.4 Risk mitigation measures

No risk mitigation is required.

9.10.3 Overall conclusions

Based on the predicted rates of CHR/F/PROTAZO 375 SC in off-field areas, the TER values describing the risk for non-target plants following exposure to CHR/F/PROTAZO 375 SC according to the GAP of the formulation CHR/F/PROTAZO 375 SC achieve the acceptability criteria without buffer zone and risk mitigation.

Review Comments:

The risk assessment is based on the “Guidance Document on Terrestrial Ecotoxicology”, (SANCO/10329/2002 rev.2 final, 2002).

Based on the risk assessment it can be concluded that the proposed use of CHR/F/PROTAZO 375 SC poses no unacceptable risk to non-target plants, if applied according to the recommended use pattern. Particular precautions to reduce the environmental concentrations resulting from CHR/F/PROTAZO 375 SC applications are not required for the protection of terrestrial non-target plants.

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

Not relevant

9.12 Monitoring data (KCP 10.8)

Not available

9.13 Classification and Labelling


Based on experimental data with the formulation, CHR/F/PROTAZO 375 SC is estimated to not be very toxic to aquatic life. (Please refer to study reports in Section B9 for details).

Prothioconazol is classified as Aquatic Chronic Category 2.

Azoxystrobin is classified as Aquatic Acute Category 1 and Aquatic Chronic Category 1; M factor=10.

Based on the classification of the active substances the product is to be classified as follows:

Hazard class(es), categories:	H410 - Very toxic to aquatic life with long lasting effects
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Hazard pictograms:	 GHS09
Signal word:	Warning
Hazard statement(s):	H410 - very toxic to aquatic life with long lasting effects
Precautionary statement(s):	P501 Dispose of contents/container in accordance with local regulation P391 Collect spillage

Appendix 1 Lists of data considered in support of the evaluation

List of data submitted by the applicant and relied on

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 10.1.1	K. Florynski	2020	CHR/F/PROTAZO - TER Calculations for Terrestrial Vertebrates no GLP Unpublished	N	Chemrol
KCP 10.1.2	K. Florynski	2020	CHR/F/PROTAZO - TER Calculations for Terrestrial Vertebrates no GLP Unpublished	N	Chemrol
KCP 10.2/01	D. Jenota	2019	Prothioconazole + Azoxystrobin (175 + 200 SC) [CHR/F/PROTAZO 375 SC] Daphnia magna, Acute immobilisation test Study code: W/43/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
KCP 10.2/02	D. Jenota	2019	Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] Raphidocelis subcapitata SAG 61.81 (formerly Pseudokirchneriella subcapitata) Growth inhibition test Study code: W/44/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
KCP 10.3/01	M. Grzesica	2019	Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] Honeybees (Apis mellifera L.), Acute Oral Toxicity Test Study code: B/42/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department	N	Chemrol

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
			of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished		
KCP 10.3/02	M. Grzesica	2019	Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] Honeybees (Apis mellifera L.), Acute Contact Toxicity Test Study code: B/43/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemiroł
10.3/03	M. Grzesica	2020	A laboratory test for evaluating the effects of Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] on the predatory mite, Typhlodromus pyri (Sch.) Study code: B/40/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemiroł
10.3/04	M. Grzesica	2020	A laboratory test for evaluating the effects of Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] on the parasitic wasp, Aphidius rhopalosiphi (De Stefani-Perez) Study code: B/41/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemiroł
KCP 10.3/05	U. Orzechowska	2020	Honey Bee, Chronic Oral Toxicity Test according to OECD 245 Study code: 0038/0018/E SORBOLAB Research Laboratory LLC, Zaniemska 11 Street, 61-029 Poznań GLP Unpublished	N	Chemiroł
KCP	U. Orzechowska	2020	Chronic Toxicity Test for Honey Bee Larvae according to OECD GD 239	N	Chemiroł

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
10.3/06			Study code: 0038/0016/E SORBOLAB Research Laboratory LLC, Zaniemyska 11 Street, 61-029 Poznań GLP Unpublished		
10.4/01	A. Wróbel	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO 375 SC) Earthworm Reproduction Test (Eisenia andrei) Study code: G/34/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
10.4/02	P. Holewik	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Collembolan (Folsomia candida) Reproduction Test Study code: G/35/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
10.4/03	P. Holewik	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Predatory mite (Hypoaspis (Geolaelaps) aculeifer) reproduction test in soil Study code: G/36/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
10.5	P. Holewik	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Soil Microorganisms: Nitrogen Transformation Test Study code: G/37/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland	N	Chemrol

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		
10.6/01	P. Holewik	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test Study code: G/39/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol
10.6/02	P. Holewik	2019	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Terrestrial Plant Test: Vegetative Vigour Test Study code: G/38/19 Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna Department of Ecotoxicological Studies, Doświadczalna 27, 43-200 Pszczyna, Poland GLP Unpublished	N	Chemrol

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

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 10.1/01	xxxxxxxxxxx	1999	JAU 6476 techn.ai: Acute oral toxicity for bobwhite quail (Colinus virginianus) Report No.: BAR/LD028 xxxxxxxxxxxxx GLP	Y	BAY

[illegible]

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
			xxxxxxxxxx GLP Unpublished		
KCP 10.1/08	xxxxxxxxxx	2002	JAU6476-desthio techn. Ai.: Effects of a subchronic dietary exposure to the northern bobwhite quail including effects on reproduction and behaviour Report No.: BAR/REP 006 xxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.1/09	xxxxxxxxxx	2001	Desthio JAU-6476: A reproduction study with the mallard (Anas platyrhynchos) Report No.: 110617 xxxxxxxxxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/01	xxxxxxxxxx	1999	JAU 6476 – Acute toxicity (96 haours) to Rainbow trout (Oncorhynchus mykiss) in a static test Report No.: DOM99076 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/02	xxxxxxxxxx	1999	JAU 6476 – Acute toxicity (96 hours) to bluegill (Lepomis macrochirus) in a static test Report No.: DOM 99090 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/03	xxxxxxxxxx	2000	JAU 6476 – Acute toxicity (96 hours) to common carp (cyprinus carpio) in a static test Report No.: DOM 20010 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY

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 10.2/04	xxxxxxxxxx	1990	SXX 0665 techn. Acute toxicity to rainbow trout in a static test Report No.: FF-298 xxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/05	xxxxxxxxxx	1991	SXX 0665: Acute toxicity to golden orfe in a static test Report No.: FO-1253 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/06	xxxxxxxxxx	2001	JAU 6476-S-methyl – Acute toxicity (96 hours) to raumbow trout (Oncorhynchus mykiss) in a semi-static test Report No.: DOM 21047 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/07	xxxxxxxxxx	1983	Report on the test for acute toxicity of CGA 98032 to rainbow trout Report No.: 821418 xxxxxxxxxxxxxxxxxx non GLP Unpublished	Y	Bay
KCP 10.2/08	xxxxxxxxxx	2002	1,2,4-Triazole-Juvenile growth test, fish (Oncorhynchus mykiss) Report No.: DOM 21060 xxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/09	xxxxxxxxxx	2001	JAU 6476 – Early life-stage toxicity test with rainbow trout (Oncorhynchus mykiss) under flow-through conditions Report No.: DOM 20028 xxxxxxxxxxxxxxxxxx	Y	BAY

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP Unpublished		
KCP 10.2/10	xxxxxxxxxxx	2002	JAU 6476-desthio: Early life-stage toxicity test with rainbow trout (Oncorhynchus mykiss) under flow-through conditions Report No.: 1022.013.321 xxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/11	xxxxxxxxxxx	2001	(14C)-JAU 6476 – Bioconcentration and biotransformation in bluegill (Lepomis macrochirus) under flow-through conditions Report No.: DOM 21003 xxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/12	xxxxxxxxxxx	2001	[14]-JAU 6476-desthio- Bioconcentration and biotransformation in bluegill (Lepomis macrochirus) under flow-through conditions Report No.: DOM 20006 xxxxxxxxxxx GLP Unpublished	Y	BAY
KCP 10.2/13	Schneider, J.	2002	Estimation of Partition Coefficient in Octanol-Water of JAU 6476-S-methyl Report No.: MO-02-002532 Bayer AG non GLP Unpublished	N	BAY
KCP 10.2/14	Heimbach, F.	1999	Acute toxicity of JAU 6476 (tech.) to water fleas (Daphnia magna) Report No.: HBF/DM 212 Bayer AG GLP Unpublished	N	BAY

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 10.2/15	Heimbach, F.	1990	Acute toxicity of SXX 0665 (techn.) to waterfleas (Daphnia magna) Report No.: HBF/DM 95 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/16	Dorgerloh, M. Sommer, H.	2001	Acute toxicity of JAU 6476-S-methyl to waterfleas (Daphnia magna) Report No.: DOM 21055 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/17	Rufli, H.	1983	Report on the test for acute toxicity of CGA 98032 to Daphnia magna Report No.: 821416 Ciba-Gergy Limited, Basel, Switzerland Bayer AG non GLP Unpublished	N	BAY
KCP 10.2/18	Hendel, B. Sommer, H.	2001	Influence of JAU 6476 (tech) on the reproduction rate of water fleas Report No.: HDB/RDM 67 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/19	Dorgerloh, M. Sommer, H.	2001	Influence of JAU 6476-desthio on the reproduction rate of water fleas in a static renewal laboratory test system Report No.: DOM 21036 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/20	Dorgerloh, M.	2000	JAU 6476 – Influence on the growth of the green alga, Selenastrum capricornutum Report No.: DOM 99107 Bayer AG	N	BAY

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP Unpublished		
KCP 10.2/21	Heimbach, F.	1990	Growth inhibition of green algae (<i>Scenedesmus subspicatus</i>) by SXX 0665 (tech.) Report No.: HBF/AL 78 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/22	Dorgerloh, M. Sommer, H.	2001	JAU 6476-S-methyl – Influence on the growth of the green alga, <i>Selenastrum capricornutum</i> Report No.: DOM 21028 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/23	Palmer, S.J. Kendall, T.Z. Krueger, H.O.	2001	1,2,4-Triazole: A 96-hour toxicity test with the freshwater alga (<i>Selenastrum capricornutum</i>) Report No.: 528A-101 Wildlife International Ltd., Easton, MD, USA Bayer AG GLP Unpublished	N	BAY
KCP 10.2/24	Hendel, B.	2000	Influence of JAU 6476 (tech.) on development and emergence of larvae of <i>Chironomus riparius</i> in a water-sediment system Report No.: HDB/CH 42 Bayer AG GLP Unpublished	N	BAY
KCP 10.2/25	Hendel, B.	2000	Influence of SXX 0665 (tech.) on development and emergence of larvae of <i>Chironomus riparius</i> in a water-sediment system Report No.: HDB/CH 43 Bayer AG GLP Unpublished	N	BAY

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 10.3/01	Wilhelmy, H.	1999	JAU 6476 a.i. – Acute effects on the honeybee <i>Apis mellifera</i> Report No.: IBA64051 Noack Laboratorium, Sarstedt, Germany Bayer AG GLP Unpublished	N	BAY
KCP 10.3/02	Bruhnke, C.	2001	JAU 6476 EC 250 – Acute effects on <i>Typhlodromus pyri</i> (Acari: Phytoseiidae) in coffin-cells Report No.: IRC71732 Bayer AG GLP Unpublished	n	BAY
KCP 10.3/03	Gossmann, A.	2001	Effects of JAU 6476 375 SC on the predatory mite <i>Typhlodromus pyri</i> – extended laboratory study (dose response test) Report No.: 10193062 Ibacon GmbH, Rossdorf, Germany Bayer AG GLP Unpublished	N	BAY
KCP 10.3/04	Gossmann, A.	2001	Effects of JAU 6476 375 SC on the predatory mite <i>Typhlodromus pyri</i> under extended laboratory conditions (aged residue test) Report No.: 10194062 Ibacon GmbH, Rossdorf, Germany Bayer AG GLP Unpublished	N	BAY
KCP 10.3/05	Dechert, G.	2000	JAU 6476 EC 250 – Laboratory test on <i>Aphidius rhopalosiphii</i> Report No.: IWA73572 Noack Laboratorium, Sarstedt, Germany Bayer AG GLP	N	BAY

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
			Unpublished		
KCP 10.3/06	Maus, C.	2000	Effects of JAU 6476 EC 250 on the ladybird beetle (<i>Coccinella septempunctata</i>) under laboratory conditions Report No.: MAUS/CS Bayer AG GLP Unpublished	N	BAY
KCP 10.3/07	Drexler, A.	2001	Effects of JAU 6476 EC 250 on the lacewing <i>Chrysoperla carnea</i> Steph. (Neuroptera, Chrysopidae) in the laboratory – multi dose test Report No.: 10192046 Ibacon GmbH, Rossdorf, Germany Bayer AG GLP Unpublished	N	BAY
KCP 10.4/01	Meisner, P.	2000	Influence of JAU 6476 EC 250 on the reproduction of earthworms (<i>Eisenia fetida</i>) Report No.: MPE/RG 235 Bayer AG GLP Unpublished	N	BAY
KCP 10.4/02	Meisner, P.	2000	Influence of JAU 6476-desthio on the reproduction of earthworms (<i>Eisenia fetida</i>) Report No.: MPE/RG 332/00 Bayer AG GLP Unpublished	N	BAY
KCP 10.4/03	Heimbach, F.	2000	Influence of JAU 6476-S-Methyl on the reproduction of earthworms (<i>Eisenia fetida</i>) Report No.: HBF/RG 317 Bayer AG GLP Unpublished	N	BAY

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 10.5/01	Anderson, J. P. E.	1999	Influence of JAU 6476 technical ingredient on glucose simulated respiration in soils Report No.: AJO/203099 Bayer AG GLP Unpublished	N	BAY
KCP 10.5/02	Anderson, J. P. E.	1999	Influence of JAU 6476 technical ingredient on the microbial mineralization of nitrogen in soils Report No.: AJO/203199 Bayer AG GLP Unpublished	N	BAY
KCP 10.5/03	Anderson, J. P. E.	2000	Influence of the metabolite JAU-6476-desthio on the microbial mineralization of nitrogen in soils Report No.: AJO/209400 Bayer AG GLP Unpublished	N	BAY
KCP 10.5/04	Anderson, J. P. E.	2001	Influence of the metabolite JAU 6476-desthio on the microbial mineralization of nitrogen in soils Report No.: AJO/219101 Bayer Ag GLP Unpublished	N	BAY
KCP 10.5/05	Anderson, J. P. E.	1999	Influence of the metabolite JAU 6476-S-methyl on glucose stimulated respiration in soils Reoirt No.: AJO/203399 Bayer AG GLP Unpublished	N	BAY
KCP 10.5/06	Anderson, J. P. E.	1999	Influence of the metabolite Jau 6476-S-methyl on the microbial mineralization of nitrogen in soils Report No.: AJO/203399 Bayer AG GLP Unpublished	N	BAY

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 10.4/04	Nienstedt, K. M.	2002	Reproduction toxicity test exposing Folsomia candida (collembola) to JAU 6476 technical Report No.: 1022.028.641 Springborn Laboratories AG, Horn, Switzerland Bayer AG GLP Unpublished	N	BAY
KCP 10.4/05	Hoogendoorn, G. M.	2000	An extended laboratory study to evaluate the effects of JAU 6476 on the predaceous mite Hypoaspis aculeifer canestrini (acari: Laelapidae) Report No.: B060HAE MITOX Stichting Bevordering Duurzame Plaagbestrijding, Amsterdam, Netherlands Bayer AG GLP Unpublished	N	BAY
KCP 10.4/06	Moser, T. Roembke, J.	2001	Acute and reproduction toxicity of JAU 6476-Desthio to the collembolan species Folsomia candida according to the ISO Guideline 11267 Report No.: P1CR ECT GmbH, Floersheim, Germany Bayer AG GLP Unpublished	N	BAY
KCP 10.4/07	Nienstedt, K. M. Novent, O.	2001	Reproduction toxicity test exposing Folsomia candida (Collembola) to JAU 6476-desthio Report No.: 1022.020.641 Springborn Laboratories AG, Horn, Switzerland Bayer AG GLP Unpublished	N	BAY
KCP 10.4/08	Moser, T. Scheffczyk, A.	2001	Acute and reproduction toxicity of JAU 6476-S-methyl to the collembolan species Folsomia candida Report No.: P35CR ECT GmbH, Floersheim, Germany Bayer AG	N	BAY

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP Unpublished		
KCP 10.5/07	Mueller, G.	1999	Investigation of the cological properties of JAU 6476 Report No.: 839 N/99 Bayer AG GLP Unpublished	N	BAY
Azoxystrobin					
KCP 10.1/10	xxxxxxxxxxxxx	2005	Avian Acute Oral Toxicity Test for ICI-A-5504 SC RL143164-AVO-B, Syngenta File No ICI5504/2949 xxxxxxxxxxxxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	SYN
KCP 10.1/11	Murfiitt, R.	2008	Review of effect of lower bodyweight upon wild mammal population dynamics TMJ5073, Syngenta File No N/1184 Syngenta- Jealott's Hill International, Bracknell, Berkshire, United Kingdom no GLP Unpublished	Y	SYN
KCP 10.2/26	xxxxxxxxxxxxx	2002	R401553 (Azoxystrobin metabolite): Acute toxicity to rainbow trout (Oncorhynchus mykiss) 7252/B, 2013675, Syngenta File No SYN501657/0002 xxxxxxxxxxxxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	Syn
KCP 10.2/27	xxxxxxxxxxxxx	2002	R402173 (Azoxystrobin metabolite): Acute toxicity to rainbow trout (Oncorhynchus mykiss) 7338/B, 2013671, Syngenta File No SYN511114/0001 xxxxxxxxxxxxxxxxxxxxxxxxxxxxx GLP Unpublished	Y	SYN
KCP	xxxxxxxxxxxxx	2002	R401553 (Azoxystrobin metabolite): Acute toxicity to Daphnia magna	N	SYN

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
10.2/28			BL7253/B, 2013672, Syngenta File No SYN501657/0003 XXXXXXXXXXXXXXXXXXXX GLP Unpublished		
KCP 10.2/29	Wallace, S.J.	2002	R402173 (Azoxystrobin metabolite): Acute toxicity to Daphnia magna BL7339/B, 2013670 , Syngenta File No SYN511114/0002 Syngenta Crop Protection AG, Basel, Switzerland Brixham Environmental Laboratory, Brixham, United Kingdom, GLP Unpublished	N	Syn
KCP 10.2/30	Smyth DV, Sankey SA, Kent SJ, Stanley RD	1994	ICIA5504: Toxicity to the Freshwater Diatom Navicula pelliculosa, BL5087/B Syngenta File No ICI5504/0965 Zeneca Agrochemicals, Jealott's Hill, United Kingdom GLP, Unpublished	N	SYN
KCP 10.2/31	Smyth DV, Kent SJ, Sankey SA, Shearing JM	1994	ICIA5504: Toxicity to the Blue-Green Alga Anabaena flos-aquae BL5054/B Syngenta File No ICI5504/0967 Zeneca Agrochemicals, Jealott's Hill, United Kingdom , GLP, Unpublished	N	Syn
KCP 10.2/32	Bowles A J, Wallace S J	2002	R401553 (Azoxystrobin metabolite): Toxicity to the green alga Selenastrum capricornutum BL7254/B, 2013669 Syngenta File No SYN501657/0004 Syngenta Crop Protection AG, Basel, Switzerland Brixham Environmental Laboratory, Brixham, United Kingdom, GLP Unpublished	N	Syn
KCP 10.2/33	Wallace SJ, Woodyer JM	2002	R402173 (Azoxystrobin metabolite): Toxicity to the green alga Selenastrum capricornutum BL7340/B, 2013668 Syngenta File No SYN511114/0003 Syngenta Crop Protection AG, Basel, Switzerland Brixham Environmental Laboratory, Brixham, United Kingdom,	N	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
			GLP, Unpublished		
KCP 10.2/34	Smyth DV, Sankey SA, Kent SJ, Stanley RD	1994	ICIA5504: Toxicity to the Duckweed Lemma gibba BL5000/B, Syngenta File No ICI5504/0963 Zeneca Agrochemicals, Jealott's Hill, United Kingdom GLP Unpublished	N	SYN
KCP 10.2/35	Kent SJ, Sankey SA, Grinell AJ	1993	ICIA5504: Acute Toxicity to Mysid Shrimp (Mysidopsis bahia) BL4785/B, Syngenta File No ICI5504/0925 Zeneca Agrochemicals, Jealott's Hill, United Kingdom GLP, Unpublished	N	SYN
KCP 10.2/36	Kent SJ, Sankey SA, Caunter JE, Grinell AJ	1994	ICIA5504: Acute Toxicity to Larvae of the Pacific Oyster (Crassostrea gigas) BL4842/B, Syngenta File No ICI5504/0927 Zeneca Agrochemicals, Jealott's Hill, United Kingdom GLP Unpublished	N	SYN
KCP 10.2/37	Boeri RL, Magazu JP, Ward TJ	1997	Chronic Toxicity of Azoxystrobin to the Mysid Mysidopsis bahia 1350-ZE, Syngenta File No ICI5504/0952 Zeneca Agrochemicals, Jealott's Hill, United Kingdom GLP Unpublished	N	SYN
KCP 10.2/38	Smyth DV, Kent SJ, Sankey SA, Johnson PA		ICIA5504: Toxicity to the Marine Alga Skeletonema costatum Zeneca Agrochemicals, Jealott's Hill, United Kingdom BL5053/B, Syngenta File No ICI5504/0966 GLP Unpublished	N	SYN
KCP 10.2/39	Cole J F H, Everett C J, Gentle W, Ashwell	2000	Azoxystrobin: An Outdoor Pond Microcosm Study Report No.: RJ2857B	N	SYN

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
	J A and Goggin U		Syngentam Jealott,S Hill Research Station, UK GLP Unpublished		
KCP 10.4/09	Friedrich S.	2002	Acute toxicity of R234886 to the earthworm Eisenia fetida 2013645, 01 10 48 076 , Syngenta File No R234886/0001 Syngenta Crop Protection AG, Basel, Switzerland BioChem agrar, Gerichshain, Germany GLP, Unpublished	N	SYN
KCP 10.4/10	Friedrich S.	2008	SYN501657 - Acute toxicity to the earthworm Eisenia fetida 071048052S T003940-07, Syngenta File No SYN501657/0006 Syngenta Crop Protection AG, Basel, Switzerland BioChem agrar, Gerichshain, Germany GLP Unpublished	N	SYN
KCP 10.4/11	Friedrich S.	2008	SYN501114 - Acute toxicity to the earthworm Eisenia Foetida 071048051S T003941-07 , Syngenta File No SYN501114/0001 Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom BioChem agrar, Gerichshain, Germany, GLP Unpublished	N	SYN
KCP 10.5/08	Lemnitzer B	2002	Effects of R234886 (metabolite of Azoxystrobin) on the activity of soil microflora 02 10 35 1001, 2023502 , Syngenta File No R234886/0002 Syngenta Crop Protection AG, Basel, Switzerland BioChem agrar, Gerichshain, Germany GLP Unpublished	N	SYN
KCP 10.5/09	Schulz L.	2008	SYN501657 - Effects on the activity of soil microflora 071048046C/N T003946-07, Syngenta File No SYN501657/0007 Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom	N	SYN

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
			BioChem agrar, Gerichshain, Germany, GLP Unpublished		
KCP 10.5/10	Schulz L.	2008	SYN501114 - Effects on the activity of soil microflora 071048045C/N T003947-07, Syngenta File No SYN501114/0002 Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom BioChem agrar, Gerichshain, Germany, GLP, Unpublished	N	SYN

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

No new studies was performed.

A 2.2 KCP 10.2 Effects on aquatic organisms

A 2.2.1.1.1 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 202 and according to the principles of GLP. No deviations to the guideline were noted.
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Reference: KCP 10.2/01

Report Prothioconazole + Azoxystrobin (175 + 200 SC) [CHR/F/PROTAZO 375 SC] *Daphnia magna*, Acute immobilisation test, D. Jenota, 2019, Study code: W/43/19

Guideline(s): the OECD Guideline No. 202 (2004)/ EU method C.2.

Deviations: No

GLP: Yes

Acceptability: Yes

Duplication No
(if vertebrate study)

Materials and methods

Test item: Prothioconazole + Azoxystrobin (175 + 200 SC) [CHR/F/PROTAZO 375 SC]; batch no.: 03; content of azoxystrobin 205.1 g/L and content of prothioconazole: 181.5 g/L; production date: June 13, 2019; expiry date: June 31, 2021.

Test organism: *Daphnia magna* Straus (< 24 h old at exposure initiation); not first brood progeny; neonates collected from a laboratory culture cultivated at the Łukasiewicz Research Network - Institute of Industrial Organic Chemistry, Branch Pszczyna..

Test design: Static test (48 h of exposure); 4 replicates per each test item concentration and the control; 5 *Daphnia magna* in each replicate.

Nominal test item concentrations: 10, 5, 2.5, 1.25, 0.625, 0.313 mg/L plus the control.

Test conditions: Temperature: 19.8 – 21.1°C; pH of the control: 7.39 – 7.60; dissolved oxygen concentration in the control: 8.0 – 8.1 mg/L; daily cycle 16 h light : 8 h dark; fluorescent light source; no feeding; no aeration; medium: Elendt M7.

Chemical determinations: The concentrations of azoxystrobin and prothioconazole were chemically determined with a validated liquid chromatographic method with DAD detection.

Statistical analysis: Probit method, Step-down Cochran-Armitage Test Procedure.

Endpoint values: EC50/48 h, NOEC/48 h, LOEC/48 h.

Results and discussions

The effect of the test item on immobilisation of *Daphnia magna* was assessed. The test item concentration used in the definitive test was determined on the basis of the preliminary test results. The *Daphnia magna* were considered immobile if they showed no ability to swim within 15 seconds after gentle swirling of the test vessel..

Preliminary test (non-GLP)

In the test, the recorded temperature during exposure was in a range of 20.9 – 21.6°C. The measured pH values were in the ranges of 6.84 – 7.04 at exposure initiation and in the range of 6.82 – 6.93 at exposure termination. The measured dissolved oxygen concentrations were 8.4 mg/L at exposure initiation and in the range of 8.2 – 8.4 mg/L at exposure termination.

In the preliminary test, in the control and the test item concentrations of 0.1, no immobilisation of *Daphnia magna* was observed during the exposure. At exposure termination, in the test item concentrations of 1.0 mg/L the immobilisation was 65% and in the test item concentrations of 10, 100 mg/L the immobilisation of *Daphnia magna* was 100%, respectively.

Table 5. Immobilisation of *Daphnia magna*, preliminary test (non-GLP)

Nominal test item concentration [mg/L]	Number of <i>Daphnia magna</i>	Number of immobilised <i>Daphnia magna</i>								Total of immobilised <i>Daphnia magna</i> [%]	
		24 h				48 h					
		Replicates									
		A	B	C	D	A	B	C	D	24 h	48 h
Control	20	0	0	0	0	0	0	0	0	0	0
0.1	20	0	0	0	0	0	0	0	0	0	0
1.0	20	0	1	0	0	3	3	4	3	5	65
10	20	5	5	5	5	5	5	5	5	100	100
100	20	5	5	5	5	5	5	5	5	100	100

Time of exposure: 14.08.2019 – 16.08.2019

Results of chemical determinations

In the preliminary test, the concentrations of prothioconazole and azoxystrobin were not chemically determined.

Definitive test

In the test, the recorded temperature during exposure was in the range of 19.8 – 21.1°C and constant within 1.3 °C (Figure 15). The measured pH values at exposure initiation were in the range of 7.30 – 7.39 in the control and the test item concentrations. The measured pH values at exposure termination were in the range of 7.41 – 7.60 in the control and the test item concentrations. The measured dissolved oxygen concentrations at exposure initiation were in the range of 8.0 – 8.2 in the control and the test item concentrations. The measured dissolved oxygen concentrations at exposure termination were in the range of 7.9 – 8.0 in the control and the test item concentrations.

Table 6. pH values and dissolved oxygen concentrations, definitive test

Nominal test item concentration [mg/L]	Measured at exposure initiation [#]		Measured at exposure termination [*]	
	pH value	Dissolved oxygen concentration [mg/L]	pH value	Dissolved oxygen concentration [mg/L]
Control	7.39	8.1	7.60	8.0
0.313	7.35	8.1	7.48	8.0
0.625	7.30	8.1	7.43	8.0
1.25	7.33	8.1	7.41	8.0
2.5	7.35	8.2	7.42	8.0
5.0	7.35	8.2	7.43	7.9
10	7.32	8.0	7.42	7.9

[#]- pH values and dissolved oxygen concentrations measured in samples before split up into replicates

^{*}- pH values and dissolved oxygen concentrations measured in samples of pooled replicates

No immobilisation of *Daphnia magna* was observed during the period of exposure in the control and in the test item concentrations of 0.313, 0.625, 1.25 mg/L. At exposure termination, in the test item concentration of 2.5 mg/L, the immobilization at exposure termination was 60%, whereas in the test item concentrations of 5 and 10 mg/L, the immobilization was 100%. No abnormal behavior of *Daphnia magna* was observed during exposure. 7.

Table 7. Immobilisation of *Daphnia magna*, definitive test

Nominal test item concentration [mg/L]	Number of <i>Daphnia magna</i>	Number of immobilised <i>Daphnia magna</i>								Total of immobilised <i>Daphnia magna</i> [%]	
		24 h				48 h					
		Replicates									
		A	B	C	D	A	B	C	D	24 h	48 h
Control	20	0	0	0	0	0	0	0	0	0	0
0.313	20	0	0	0	0	0	0	0	0	0	0
0.625	20	0	0	0	0	0	0	0	0	0	0
1.25	20	0	0	0	0	0	0	0	0	0	0
2.5	20	3	2	2	3	3	4	2	3	50	60
5.0	20	5	4	4	4	5	5	5	5	85	100
10	20	5	5	5	5	5	5	5	5	100	100

Time of exposure: 15.10.2019 – 17.10.2019

Results of chemical determinations

The concentrations of prothioconazole and azoxystrobin were chemically determined using a validated liquid chromatographic method [SOP/C/505]. Samples of each treatment were collected at exposure initiation and at exposure termination. At exposure initiation the determined concentration of prothioconazole in the test item concentrations of 0.625, 12.5, 2.5, 5.0, 10 mg/L were in the range of 97.8 – 102.8% of nominal concentration. In the test item concentration of 0.313 mg/L the determined nominal concentration of prothioconazole was below the limit of quantification. The determined concentrations of azoxystrobin in all test item concentrations were in the range of 101.7 – 103.6% of nominal concentration. The results confirm that the test item concentrations were prepared correctly. At exposure termination the determined concentration of prothioconazole in the test item concentrations of 0.625, 12.5, 2.5, 5.0, 10 were in the range of 92.9 – 102.3% of the nominal concentration. In the test item concentration of 0.313 mg/L the determined nominal concentration of prothioconazole was below the limit of quantification. The determined concentration of azoxystrobin in all test item concentrations were in the range of 97.9 – 104.2%. Therefore, the test item concentrations were stable under test conditions.

Table 8. Concentration and stability of Prothioconazole, definitive test

Nominal test item concentration [mg/L]	Nominal concentration of Prothioconazole [mg/L]	Average determined concentration of Prothioconazole (n=3) in samples collected [mg/L]			
		at exposure initiation	% of nominal concentration	at exposure termination	% of nominal concentration
Control	---	<LoD	---	<LoD	---
0.313	0.051*	0.049*	96.7	0.046*	90.8
0.625	0.101	0.099	97.8	0.094	92.9
1.25	0.202	0.203	100.3	0.207	102.3
2.5	0.405	0.411	101.5	0.401	99.1
5.0	0.810	0.827	102.2	0.815	100.7
10	1.619	1.665	102.8	1.638	101.2

*- values below LoQ
 LoQ = 0.1 mg/L
 LoD = 0.03 mg/L
 --- no value

Table 9. Concentration and stability of Azoxystrobin, definitive test

Nominal test item concentration [mg/L]	Nominal concentration of Azoxystrobin [mg/L]	Average determined concentration of Azoxystrobin (n=3) in samples collected [mg/L]			
		at exposure initiation	% of nominal concentration	at exposure termination	% of nominal concentration
Control	---	<LoD	---	<LoD	---
0.313	0.0573	0.0584	102.0	0.0567	99.0
0.625	0.1144	0.117	102.3	0.1137	99.4
1.25	0.229	0.237	103.6	0.224	97.9
2.5	0.458	0.469	102.5	0.455	99.5
5.0	0.915	0.931	101.7	0.914	99.9
10	1.830	1.863	101.8	1.906	104.2

LoQ = 0.0005 mg/L
 LoD = 0.00015 mg/L
 --- no value

Endpoint values

The endpoint values were determined based on the nominal test item concentrations. The endpoint values were calculated with a probit method. To make calculations and to conduct statistical analyzes, the ToxRat Professional commercial software was used.

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

Endpoint value [mg/L]	Time of exposure	
	24 h	48 h
EC ₅₀	2.82 (2.32 – 3.43)	2.42 (n.d.)
EC ₂₀	1.93 (1.42 – 2.35)	2.19 (n.d.)
EC ₁₀	1.59 (1.06 – 1.98)	2.08 (n.d.)
LOEC	2.5	2.5
NOEC	1.25	1.25

Calculations were made according to [7]. [SOP/W/68].

(–) – 95% confidence interval

n.d. – not determined

The median concentration causing 50% immobilisation of *Daphnia magna* after 24 h of exposure, i.e. the EC₅₀/24 h value is 2.82 mg/L (95% confidence interval: 2.32 – 3.43). The EC₂₀/24 h value is 1.93 mg/L (95% confidence interval: 1.42 – 2.35). The EC₁₀/24 h value is 1.59 mg/L (95% confidence interval: 1.06 – 1.98).

The median concentration causing 50% immobilisation of *Daphnia magna* after 48 h of exposure, i.e. the EC₅₀/48 h value is 2.42 mg/L (95% confidence interval: not determined). The EC₂₀/48 h value is 2.19 mg/L (95% confidence interval: not determined). The EC₁₀/48 h value is 2.08 mg/L (95% confidence interval: not determined).

The data on immobilisation of the *Daphnia magna* at exposure termination were analysed using Step-down Cochran-Armitage Test Procedure, which showed a significant difference between nominal test item concentrations of 2.50, 5.0 and 10 mg/L and the control. Therefore, the LOEC/48 h value is 2.5 mg/L and the NOEC/48 h value 1.25 mg/L..

TEST VALIDITY CRITERIA

In the definitive test the validity criteria were met according to the OECD Guideline No. 202 (2004):

- the percentage of immobilisation of *Daphnia magna* in the control was 0% (criterion: not more than 10%),
- the dissolved oxygen concentrations in the test vessels were within the range of 7.9 – 8.2 mg/L (criterion: not less than 3 mg/L)

A 2.2.1.1.2 Study 2

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.
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Reference: KCP 10.2/02

Report Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] *Raphidocelis subcapitata* SAG 61.81 (formerly *Pseudokirchneriella subcapitata*) Growth inhibition test, D. Jenota, 2019, Study code: W/44/19

Guideline(s): the OECD Guideline No. 201 (2006)/EU method C.3.

Deviations: No

GLP: Yes

Acceptability: Yes
Duplication No
(if vertebrate study)

Materials and methods

Test item: Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]; batch no. – 03, content of azoxystrobin: 205.1 g/L and the content of prothioconazole is 181.5 g/L; production date: 13.06.2019, expiry date: 13.06.2021.

Test organism: The unicellular freshwater green algae, *Raphidocelis subcapitata* (formerly *Pseudokirchneriella subcapitata* (Korshikov) Hindák, *Selenastrum capricornutum* Prinz) SAG 61.81 cultivated at the Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicological Studies, Laboratory of Aquatic Toxicology. The algae were obtained from the Culture Collection of Algae at Göttingen, University, Germany

Test design: 72 hours of exposure; three replicates per each test item concentration; six replicates per the control; a background for each treatment; initial algal cell density: 1 x 10⁴ cells/mL.

Nominal test item concentrations: 10, 3.3, 1.1, 0.37, 0.12 mg/L plus the control.

Test conditions: 21.4 – 22.7°C; pH of the control: 7.50 – 7.86; mean light intensity: 6420 – 6540 lux; constant illumination and shaking; medium: AAP.

Chemical determinations: The concentrations of azoxystrobin and prothioconazole were determined with a validated liquid chromatographic method with DAD detection.

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

Endpoint values: ErC₅₀/72 h, EyC₅₀/72 h, NOEC/72 h, LOEC/72 h.

Results and discussions

The effect of the test item on the green algal growth was assessed. The range of the test item concentrations used in the definitive test were determined on the basis of the preliminary test results. The growth inhibition was estimated on the basis of the density of the algae cells determined in the definitive test

Preliminary test (non-GLP)

In the preliminary test, the recorded temperature was in the range of 22.3 – 22.6°C, the mean light intensity was in the range of 8023 – 8078 lux. The pH values were in the ranges of 6.86 – 7.17 at exposure initiation and 7.02 – 7.31 at exposure termination.

The average transmittance values were in the range of 99.5 – 100.0% at exposure initiation and 100.0% at exposure termination when compared with the control. Hence, the indirect method was adequate to determine the number of algal cells. The growth rate inhibition after 72 h of exposure was 0.2% in the test item concentration of 0.01 mg/L, 2.4% in the test item concentration of 0.1 mg/L, 12.3% in the test item concentration of 1.0 mg/L and 101.4% in the test item concentration of 10 mg/L. The yield inhibition after 72 hours of exposure was 1.1% in the test item concentration of 0.01 mg/L, 10.4% in the test item concentration of 0.1 mg/L, 42.9% in the test item concentration of 1.0 mg/L and 100.0% in the test item concentration of 10 mg/L.

Table 5. Inhibition of growth rate and yield, preliminary test (non-GLP)

Nominal test item concentration [mg/L]	% inhibition after 72 h of exposure (growth rate)	% inhibition after 72 h of exposure (yield)
Control	0.0	0.0
0.01	0.2	1.1
0.1	2.4	10.4
1.0	12.3	42.9
10	101.4*	100.0

* inhibition of growth rate and yield are higher than 100% means that the algae cell density at observation during exposure was lower than in the initial algae cell density

Definitive test

The definitive test was performed using the test item concentrations of 10, 3.3, 1.1, 0.37, 0.12 mg/L plus the control. The recorded temperature was in the range of 21.4 – 22.7 °C with a variation of up to 1.3°C . The mean light intensity was in the range of 6420 – 6540 lux. The pH values measured at exposure initiation were in the range of 7.04 – 7.50 and at exposure termination were in the range of 7.86 – 7.15. In all the test item concentrations no differences in shape, size and colour of algal cells were reported as compared to the algae cells in the control. The average transmittance values were in the range of 99.8 – 100.0% at exposure initiation and 100.0% at exposure termination when compared with the control. Hence, the indirect method was adequate to determine the number of algal cells.

Table 11. Growth rate and yield inhibition, definitive test

Nominal test item concentration [mg/L]	% inhibition after 72 h of exposure (growth rate)	% inhibition after 72 h of exposure (yield)
Control	0.0	0.0
0.12	0.6	2.7
0.37	5.7	25.0
1.1	20.3	64.2
3.3	56.8	94.8
10	77.1	98.5

*calculated inhibition values are lower than 0%, what means that the algal cell density at exposure termination was higher than the algal cell density in the control

Results of the chemical determinations

The concentrations of prothioconazole and azoxystrobin were chemically determined using a validated liquid chromatographic method [SOP/C/505]. Samples of each treatment were collected at exposure initiation and at exposure termination.

Table 12. Concentration and stability of azoxystrobin, definitive test

Nominal test item concentration [mg/L]	Nominal concentration of azoxystrobin [mg/L]	Average determined concentration of azoxystrobin (n=3) in samples collected			
		at exposure initiation [mg/L]	% of nominal concentration	at exposure termination [mg/L]	% of nominal concentration
Control	---	< LoD	---	< LoD	---
0.12	0.0220	0.0227	103.4	0.0210	95.6
0.37	0.0677	0.0690	101.9	0.0677	100.0
1.1	0.2010	0.2090	103.8	0.1970	97.9
3.3	0.604	0.6470	107.1	0.6270	103.8
10	1.830	1.9300	105.5	1.911	104.4

LoQ = 0.0005 mg/L

LoD = 0.00015 mg/L

--- no value

Table 13. Concentration and stability of prothioconazole, definitive test

Nominal test item concentration [mg/L]	Nominal concentration of prothioconazole [mg/L]	Average determined concentration of prothioconazole (n=3) in samples collected				
		at exposure initiation [mg/L]	% of nominal concentration	at exposure termination [mg/L]	% of nominal concentration	Geometric mean of determined concentration of prothioconazole [mg/L]
Control	---	< LoD	---	< LoD	---	---
0.12	0.019	< LoD	---	< LoD	---	---
0.37	0.060	0.061	101.8	< LoQ (0.047)	---	0,05
1.1	0.178	0.180	101.1	0.146	82.0	0,16
3.3	0.534	0.550	102.9	0.533	99.8	5,41
10	1.619	1.670	103.2	1.613	99.6	1,64

* The geometric mean of the determined test item concentrations was calculated according to the formula given in the OECD Series on Testing and Assessment No. 23, Annex 2, page 50

LoQ = 0.1 mg/L

LoD = 0.03 mg/L

--- no value

At exposure initiation, the determined concentrations of azoxystrobin were in the range of 101.9 – 107.1% of the nominal concentration. The determined concentration of **prothioconazole** ~~azoxystrobin~~ were in the range of 101.1% - 103.2% of the nominal concentration. The results confirm that the test item concentrations were prepared correctly. At exposure termination, the determined concentrations of azoxystrobin were in the range of 95.6 – 104.4% of the nominal concentration. The determined concentration of prothioconazole were in the range of 82.0% - 99.8% of the nominal concentration. Therefore, the concentrations of azoxystrobin and prothioconazole were stable under test conditions..

Endpoint values

The endpoint values were determined on the basis of the nominal test item concentrations. The ECx values were calculated with a probit method. The lowest observed effect concentration (LOEC) and the no observed effect concentration (NOEC) were estimated on the basis of statistical analyses. To conduct statistical analyses, the ToxRat Professional commercial software was used.

Table 14. Growth rate endpoint values based on the nominal test item concentrations, definitive test

Endpoint value [mg/L]	Time of exposure:		
	24 h	48 h	72 h
ErC₅₀	14.16 (7.06 – 74.77)	3.54 (3.13 – 4.03)	3.05 (2.71 – 3.45)
ErC₂₀	1.29 (0.38 – 2.36)	0.98 (0.79 – 1.17)	0.96 (0.78 – 1.13)
ErC₁₀	0.37 (0.04 – 0.89)	0.50 (0.37 – 0.64)	0.52 (0.39 – 0.66)
LOEC	1.10	0.37	0.37
NOEC	0.37	0.12	0.12

(-) – 95% confidence interval

Calculations were made according to [8], [SOP/W/68]

n.d. – not determined

Table 15. Yield endpoint values based on the nominal test item concentrations, definitive test

Endpoint value [mg/L]	Time of exposure:		
	24 h	48 h	72 h
E_yC₅₀	3.04 (1.84 – 5.74)	0.95 (0.80 – 1.12)	0.74 (0.67 – 0.82)
E_yC₂₀	0.53 (0.16 – 0.98)	0.36 (0.26 – 0.45)	0.31 (0.26 – 0.36)
E_yC₁₀	0.21 (0.03 – 0.48)	0.21 (0.14 – 0.29)	0.20 (0.16 – 0.24)
LOEC	1.10	0.37	0.37
NOEC	0.37	0.12	0.12

(-) – 95% confidence interval

Calculations were made according to [8], [SOP/W/68]

n.d. – not determined

The median test item concentration causing 50% inhibition of the average specific growth rate of *Raphidocelis subcapitata*, i.e. the ErC₅₀/72 h value is 3.05 mg/L (95% confidence interval: 2.71 – 3.45). The ErC₂₀/72 h value is 0.96 mg/L (95% confidence interval: 0.78 – 1.13) and the ErC₁₀/72 h value is 0.52 mg/L (95% confidence interval: 0.39 – 0.66). Statistical tests based on the growth rate data were the Shapiro-Wilk's Test on Normal Distribution which confirmed normal distribution of the data, the Levene's Test on Variance Homogeneity (with . Residuals) which showed that the variances were homogeneous and Williams Multiple Sequential t-test Procedure which showed a significant difference between test item concentration of 0.37, 1.1, 3.3 and 10 mg/L and the control. Therefore, the lowest observed effect concentration, i.e. the LOEC/72 h is 0.37 mg/L and the no observed effect concentration, i.e. the NOEC/72 h is 0.12 mg/L. The median test item concentration causing 50% yield inhibition of *Raphidocelis*

subcapitata, i.e. the EyC50/72 h value is 0.74 mg/L (95% confidence interval: 0.67 – 0.82). The EyC20/72 value is 0.31 mg/L (95% confidence interval: 0.26 – 0.36) and EyC10/72 h value is 0.20 mg/L (95% confidence interval: 0.16 – 0.24). Statistical tests based on the yield data were the Shapiro-Wilk's Test on Normal Distribution which confirmed normal distribution of the data, the Levene's Test on Variance Homogeneity (with Residuals) which showed that the variances were homogenous and Williams Multiple Sequential t-test Procedure which showed a significant difference between test item concentration of 0.37, 1.1, 3.3, 10 mg/L and the control. Therefore, the lowest observed effect concentration, i.e. the LOEC/72 h is 0.37 mg/L and the no observed effect concentration, i.e. the NOEC/72 h is 0.12 mg/L.

TEST VALIDITY CRITERIA

In the definitive test, the following validity criteria specified in OECD Guideline No. 201 (2006) and EU Method C.3 were met:

- the biomass in the control increased by a factor of 149.3 within the 72-hour test period (criterion: at least a 16-fold growth),
- the coefficient of variation of the mean specific growth rate after the 72-hour test period (exposure initiation – exposure termination) in the control culture was 1.2% (criterion: it must not exceed 7%).
- the mean coefficient of variation for the section-by-section growth rate in the control culture was 18.8% (criterion: it must not exceed 35%).

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 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 213 and according to the principles of GLP. No deviations to the guideline were noted.
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Reference:	KCP 10.3/02
Report	Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] Honeybees (<i>Apis mellifera</i> L.), Acute Oral Toxicity Test, M. Grzesica, 2019, Study code: B/42/19
Guideline(s):	the OECD Guideline for the Testing of Chemicals No. 213 (1998) and the EU Method C.16. (2008)
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication	No

(if vertebrate study)

Materials and methods

Test item:

name: Prothioconazole + Azoxystrobin
(175 + 200) SC
[CHR/F/PROTAZO 375 SC]
active substance: 181.5 g/L of
prothioconazole
205 g/L of azoxystrobin
batch number: 03
manufacturing date: 13.06.2019
expiry date: 13.06.2021

Biological test system:

the honeybee. *Apis mellifera* L.
strain: carnica
source: an apiary at the Łukasiewicz
Research Network – Institute of Industrial
Organic Chemistry, Branch Pszczyna,
Department of Ecotoxicological Studies
age: approximately 3 weeks

Test design:

test item:
exposure time: 48 hours
number of doses: 5 doses and a control
number of replicates: 3 replicates
number of bees: 10 bees/replicate
- reference item:
exposure time: 24 hours
number of doses: 3 doses
number of replicates: 3 replicates
number of bees: 10 bees/replicate

Test item doses:

12.5, 25.0, 50.0, 100.0 and 200.0 µg
test item/bee and a control (0.0
µg/bee)

Reference item doses:

0.1, 0.2 and 0.4 µg a.i./bee

Test conditions:

temperature: 24 – 26°C, relative air
humidity: 57 – 64%
place: a dark room

Endpoints:

- honeybee mortality after 24 and 48
hours of exposure
- LD50/24h and LD50/48h of the
test item
- LD50/24h of the reference item
(dimethoate)

Statistical method:

regression analysis using the log-
probit method

Results and discussions

Preliminary test

After 48 hours there was one dead bee in the control group. Mortality corrected by Abbott's formula, after 48 hours in the doses 8.0, 40.0 and 200.0 µg/bee were 0.0, 0.0 and 20.0%, respectively.

Table 1. Honeybee mortality after 24 hours of exposure – preliminary test

Dose [µg /bee]	Number of tested bees [no.]	Mortality	
		Number of dead bees [no.]	Total
		replicates	
		I	[%]
0.0 (Control)	10	0	0.0
8.0	10	0	0.0
40.0	10	0	0.0
200.0	10	1	10.0

Table 2. Honeybee mortality after 48 hours of exposure – preliminary test

Dose [µg/bee]	Number of tested bees [no.]	Mortality	
		Number of dead bees [no.]	Total
		replicates	
		I	[%]
0.0 (Control)	10	0	0.0
8.0	10	0	0.0
40.0	10	0	0.0
200.0	10	2	20.0

The preliminary test was performed between 16 – 18.07.2019

Definitive test

The results refer to all doses, time points and replicates, and they are presented in the form of raw data collected during the observations. After 48 hours of exposure, mortality of the control group was 0.0% and for the treated groups mortality percentages at the doses 12.5, 25.0, 50.0, 100.0 and 200.0 µg/bee, were 0.0, 0.0, 3.3, 0.0 and 0.0%, respectively. The median lethal doses (LD50/24 h and LD50/48 h oral) are higher than the highest dose used in the test, i.e. 200.0 µg/honeybee..

Table 3. Honeybee mortality after 4 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality				
		Number of dead bees [no.]			Total	
		replicates				
		I	II	III	[no.]	[%]
0.0 (Control)	30	0	0	0	0	0.0
12.5	30	0	0	0	0	0.0
25.0	30	0	0	0	0	0.0
50.0	30	0	0	0	0	0.0
100.0	30	0	0	0	0	0.0
200.0	30	0	0	0	0	0.0

Table 4. Honeybee mortality and the LD₅₀ after 24 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality					LD ₅₀ [µg/bee]
		Number of dead bees [no.]			Total		
		replicates					
		I	II	III	[no.]	[%]	
0.0 (Control)	30	0	0	0	0	0.0	> 200
12.5	30	0	0	0	0	0.0	
25.0	30	0	0	0	0	0.0	
50.0	30	1	0	0	1	3.3	
100.0	30	0	0	0	0	0.0	
200.0	30	0	0	0	0	0.0	

Table 5. Honeybee mortality and the LD₅₀ after 48 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality					LD ₅₀ [µg/bee]
		Number of dead bees [no.]			Total		
		I	II	III	[no.]	[%]	
0.0 (Control)	30	0	0	0	0	0.0	> 200
12.5	30	0	0	0	0	0.0	
25.0	30	0	0	0	0	0.0	
50.0	30	1	0	0	1	3.3	
100.0	30	0	0	0	0	0.0	
200.0	30	0	0	0	0	0.0	

The definitive test was performed between 24 – 26.09.2019

Reduction [%] in food consumption calculated using the formula of Abbott (1925) presented below:

$$C = \left(1 - \frac{nT}{nK} \right) \times 100$$

C: reduction of food consumption [%].

nT : the amount of sucrose solution consumed by bees exposed to the test item.

nK : the amount of sucrose solution consumed by control bees.

Mortality of the bees receiving the reference item registered after 4 and 24 hours and the LD50 values after 24 hours are presented below. The median lethal dose of dimethoate (LD50 oral) after 24 hours determined with the log-probit method. is 0.26 µg a.i./bee (confidence limits: 0.21 – 0.33).

Table 10. Honeybee mortality after 4 hours of exposure on the reference item Bi 58 Top 400 EC (dimethoate) – definitive test

Dose [µg a.i./bee]	Number of tested bees [no.]	Mortality				
		Number of dead bees [no.]			Total	
		replicates				
		I	II	III	[no.]	[%]
0.0 (control)	30	0	0	0	0	0.0
0.1	30	0	0	1	1	3.3
0.2	30	1	0	1	2	6.7
0.4	30	2	1	1	4	13.3

Table 11. Honeybee mortality and the LD₅₀ after 24 hours of exposure to Bi 58 Top 400 EC (dimethoate) – definitive test

Dose [µg a.i./bee]	Number of tested bees [no.]	Mortality					LD ₅₀ [µg a.i./bee]
		Number of dead bees [no.]			Total		
		replicates					
		I	II	III	[no.]	[%]	
0.0 (control)	30	0	0	0	0		0.26* (0.21 – 0.33)
0.1	30	1	1	1	3	10.0	
0.2	30	3	3	3	9	30.0	
0.4	30	8	7	8	23	76.7	

*: the LD₅₀ (with 95% confidence limits) was calculated with the log-probit method (ToxRat Professional 3.3.0. software [SOP/B/67]).

a.i.: active ingredient

DEFINITIONS OF THE ENDPOINTS

The LD50 (median lethal dose) oral is a statistically derived single dose of a test or reference item that can cause death in 50 per cent of biological test systems when administered by the oral route. The LD50 is expressed in µg of the test item per bee or in µg of the active ingredient contained in the reference item per bee. It was calculated with the log-probit method using ToxRat Professional software, Version 3.3.0. Mortality: a honeybee is considered dead if it is completely immobile..

7. VALIDITY OF THE STUDY

The following validity criteria were met during the test:

- the average mortality for the control was 0.0% at the end of the experiment (criterion: it must not exceed 10%).
- the LD50/24 h of the reference item (dimethoate) was 0.26 µg a.i./bee (criterion: 0.10 – 0.35 µg a.i./bee).

A 2.3.1.1.3 KCP 10.3.1.1.2 Acute contact toxicity to bees

A 2.3.1.1.4 Study 1

Comments of zRMS:	The study was conducted to OECD guideline 214 and according to the principles of GLP. No deviations to the guideline were noted.
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Reference:	KCP 10.3/01
Report	Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] Honeybees (<i>Apis mellifera</i> L.), Acute Contact Toxicity Test, M. Grzesica, 2019, Study code: B/43/19
Guideline(s):	the OECD Guideline for the Testing of Chemicals No. 214 (1998) and the EU Method C.17. (2008)
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:	name: Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] active substance: 181.5 g/L of prothioconazole 205 g/L of azoxystrobin batch number: 03 manufacturing date: 13.06.2019 expiry date: 13.06.2021
Biological test system:	the honeybee, <i>Apis mellifera</i> L., strain: carnica source: an apiary at the Institute of Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicological Studies age: approximately 3 weeks
Test design:	- the test item: exposure duration: 48 hours number of doses: 5 doses and a control number of replicates: 3 replicates number of bees: 10 bees/replicate - the reference item: exposure duration: 24 hours number of doses: 3 doses number of replicates: 3 replicates number of bees: 10 bees/replicate
Test item doses:	12.5, 25.0, 50.0, 100.0 and 200.0 µg test item/bee and a control (0.0 µg/bee)
Reference item doses:	0.1, 0.2 and 0.4 µg a.i./bee

Test conditions: temperature: 24 – 26°C, relative air
humidity: 58 – 63%
place: a dark room

Endpoints:

- ☐ honeybee mortality after 24 and 48 hours of exposure
- ☐ LD₅₀/24h and LD₅₀/48h of the test item
- ☐ LD₅₀/24h of the reference item (dimethoate)

Statistical method: regression analysis using the log-probit method

Results and discussions

Preliminary test

Mortality results obtained in the preliminary experiment are presented in Tables 1 and 2. Mortality of the control group after 48 hours of exposure was 0.0%. The percentages of mortality of the bees treated with the test item at the doses of 8.0, 40.0 and 200.0 µg/honeybee were 0.0, 0.0 and 0.0%, respectively. No abnormal behavioural effects were observed during the test.

Table 1. Honeybee mortality after 24 hours of exposure – preliminary test

Dose [µg/bee]	Number of tested bees [no.]	Mortality	
		Number of dead bees [no.]	Total
		replicate	
		I	
0.0 (control)	10	0	0.0
8.0	10	0	0.0
40.0	10	0	0.0
200.0	10	0	0.0

Table 2. Honeybee mortality after 48 hours of exposure – preliminary test

Dose [µg/bee]	Number of tested bees [no.]	Mortality	
		Number of dead bees [no.]	Total
		replicate	
		I	
0.0 (control)	10	0	0.0
8.0	10	0	0.0
40.0	10	0	0.0
200.0	10	0	0.0

The preliminary test was conducted between 18 – 20.07.2019.

Definitive test

Mortality of the control group after 48 hours of exposure was 0.0%. The mortality percentages of the bees treated groups 12.5, 25.0, 50.0, 100 and 200.0 µg/honeybee after 48 hours of exposure were 0.0, 0.0, 0.0, 0.0 and 3.3% respectively. The median lethal doses (LD50/24 h and LD50/48 h contact) are higher than the highest dose used in the test, i.e. 200.0 µg/honeybee.

Table 3. Honeybee mortality after 4 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality				
		Number of dead bees [no.]			Total	
		replicates				
		I	II	III	[no.]	[%]
0.0 (control)	30	0	0	0	0	0.0
12.5	30	0	0	0	0	0.0
25.0	30	0	0	0	0	0.0
50.0	30	0	0	0	0	0.0
100.0	30	0	0	0	0	0.0
200.0	30	0	0	0	0	0.0

Table 4. Honeybee mortality and the LD₅₀ after 24 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality					LD ₅₀ [µg/bee]
		Number of dead bees [no.] replicates			Total		
		I	II	III	[no.]	[%]	
0.0 (control)	30	0	0	0	0	0.0	>200
12.5	30	0	0	0	0	0.0	
25.0	30	0	0	0	0	0.0	
50.0	30	0	0	0	0	0.0	
100.0	30	0	0	0	0	0.0	
200.0	30	0	0	0	0	0.0	

Table 5. Honeybee mortality and the LD₅₀ after 48 hours of exposure – definitive test

Dose [µg/bee]	Number of tested bees [no.]	Mortality					LD ₅₀ [µg/bee]
		Number of dead bees [no.]			Total		
		I	II	III	[no.]	[%]	
0.0 (control)	30	0	0	0	0	0.0	>200
12.5	30	0	0	0	0	0.0	
25.0	30	0	0	0	0	0.0	
50.0	30	0	0	0	0	0.0	
100.0	30	0	0	0	0	0.0	
200.0	30	1	0	0	1	3.3	

The definitive test was conducted between 24 – 26.09.2019.

DEFINITIONS OF THE ENDPOINTS

The LD50 (median lethal dose) contact, is a statistically derived single dose of a substance that can cause death in 50 per cent of animals when administered by contact route. The LD50 is expressed in µg test item/bee or µg a.i./bee. It was calculated with the log-probit method. Mortality: a honeybee is dead if it is completely immobile..

VALIDITY OF THE STUDY

The following validity criteria were met during the test:

- the average mortality for the control was 0.0% after 48 h (criterion: it must not exceed 10%),
- the LD50/24 h of the reference item (dimethoate) was 0.24 µg a.i./bee (criterion: 0.10–0.30 µg a.i./bee).).

A 2.3.1.2 KCP 10.3.1.2. Chronic toxicity to bees

Comments of zRMS:	<p>The study was conducted to OECD guideline 245 and according to the principles of GLP. The following deviations to the guideline were noted:</p> <ul style="list-style-type: none"> - During the range-finding test, periodic, humidity fluctuations of short duration were recorded as an effect of feedings and observations record, which did not affect the condition of research system. - During the definitive test, slight increase of temperature above 35°C was noted (required: 33±2°C). The average air humidity was 75.1% (required: 50-70%). The changes were minor, which did not affect the condition of the research system. - For analytical analysis were submitted test item solutions only at the beginning of storage period, due to the conducting of stability test, which confirmed test item stability in solutions at the end of storage period (4 days). <p>Above deviations had no effect on the course of the test, the study met validity criteria.</p>
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Reference:	KCP 10.3/05
Report	Honey Bee, Chronic Oral Toxicity Test according to OECD 245, U. Orzechowska, Study code: 0038/0018/E, 2020
Guideline(s):	OECD 245
Deviations:	No Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test design	<p>stability test: tested concentrations and control in one replicate range-finding, definitive and reference test: tested concentrations and control in three replicates, 10 bees per replicate</p>
Test cages	<p>stability test: storage conditions: plastic containers of 100 mL volume test conditions: plastic syringe of 2 mL volume range-finding, definitive and reference test: cages 20x20x20 cm</p>

Duration time

stability test:
storage conditions: 72 hours
test conditions: 24 hours
range-finding, definitive and reference test:
10 days

Tested concentrations

stability test:
control; 2.5 mg/kg; 2500 mg/kg of food
range-finding test:
control; 2.5 mg/kg; 25 mg/kg; 250 mg/kg;
2500 mg/kg of food

definitive test:

control

156.25 mg/kg of food (test item intake dose: 3.345 µg/bee/day)
312.5 mg/kg of food (test item intake dose: 7.315 µg/bee/day)
625 mg/kg of food (test item intake dose: 15.890 µg/bee/day)
1250 mg/kg of food (test item intake dose: 24.591 µg/bee/day)
2500 mg/kg of food (test item intake dose: 58.655 µg/bee/day)

reference test:

control; dimethoate: 0.5 g/kg of
food

Study conditions

stability test:
storage conditions: average temperature
5.201°C (minimal temperature 4.7°C; maximal
temperature 6.5°C); darkness
test conditions: average temperature 34.596°C
(minimal temperature 34.2°C; maximal
temperature 34.8°C); average humidity:
79.600% (minimal humidity 78.8%; maximal
humidity 80.4%); darkness
range-finding test:
average temperature 34.399°C (minimal
temperature 33.4°C; maximal temperature
34.5°C); average humidity: 70.292% (minimal
humidity 32.7%; maximal humidity 78.2%);
darkness

definitive test and reference test:

average temperature 34.4°C (minimal temperature 32.5°C; maximal temperature 35.6°C); average
humidity: 75.1% (minimal humidity 54.7%; maximal humidity 85.9%); darkness

Chronic oral toxicity test of the test item CHR/F/PROTAZO 375 SC on honey bee (*Apis mellifera* L.) was conducted. The aim of the test was to determine the concentration causing 50% mortality of population (LC50 value), the dose causing mortality of 50% of population after 10 days (LDD50 value) and determine NOEC and NOEDD values. The test was conducted according to OECD 245 Guideline.

Final results

In the course of the study, test item has shown apitoxic effect in honey bee mortality after 10 days of the test. On the basis of data analysis, value LC10 = 576.9500 mg of test item/kg of food; LC20 = 858.6873 mg of test item/kg of food; LC50 = 1837.4578 mg of test item/kg of food and LDD10 = 13.1047 µg of test item/bee/day; LDD20 = 19.3891 µg µg of test item/bee/day; LDD50 = 41.0230 µg of test item/bee/day for mortality. Values LOEC = 1250 mg of test item/kg of food; NOEC = 625 mg of test item/kg of food; LOEDD = 24.591 µg µg of test item/bee/day and NOEDD = 15.89 µg µg of test item/bee/day were also determined.

Final results calculated using ToxRat Professional software			
Parameter	Concentration [mg of test item/kg of food]	Parameter	Dose [µg of test item/bee/day]
LC ₁₀	576.9500 (354.8453 – 769.0704)*	LDD ₁₀	13.1047 (8.1579 – 17.3566)*
LC ₂₀	858.6873 (614.1198 – 1098.2848)*	LDD ₂₀	19.3891 (13.9930 – 24.6935)*
LC ₅₀	1837.4578 (1426.8865 – 2668.9917)*	LDD ₅₀	41.0230 (31.9393 – 59.6197)*
NOEC	625	NOEDD	15.890
LOEC	1250	LOEDD	24.591

LC₁₀ test item concentration causing 10% reduction

LC₂₀ test item concentration causing 20% reduction

LC₅₀ test item concentration causing 50% reduction

NOEC the highest test item concentration not causing statistically significant differences in comparison to the control

LOEC the lowest test item concentration causing statistically significant differences in comparison to the control

LDD₁₀ daily dietary test item dose causing 10% reduction

LDD₂₀ daily dietary test item dose causing 20% reduction

LDD₅₀ daily dietary test item dose causing 50% reduction

NOEDD the highest daily dietary test item dose not causing statistically significant differences in comparison to the control

LOEDD the lowest daily dietary test item dose causing statistically significant differences in comparison to the control

*) upper and lower confidence limit (95%)

Validity criteria

The test met the validity criteria of the experiment:

- bee mortality in control after 10 days was 3.3% (required: ≤15%).
- bee mortality in the reference test after 10 days was 80.0% (required: ≥50%)

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

Comments of zRMS:	<p>The study was conducted to OECD guideline 239 and according to the principles of GLP. The following deviations to the guideline were noted:</p> <ul style="list-style-type: none"> - On day 8 of the test, desiccator with larvae was transferred to test room (temperature 34-35°C, humidity 60-80%), what poses a deviation from Standard Experimental Procedure and Study Plan. The alteration was introduced due to observable improvement of larvae development outside incubator in conducted breeding tests. - In course of the range-finding and definitive test, periodic decreases of temperature and humidity occurred. It resulted from daily feedings and observations. These drops were short-termed, did not affect the condition of the test system. - For analytical analysis were submitted test item solutions only at the beginning of storage period, due to the conducting of stability test, which confirmed test item stability in solutions at the end of validity period. <p>The deviations had no effect of the test results. The test met the validity criteria.</p>
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Reference:	KCP 10.3/06
Report	Chronic Toxicity Test for Honey Bee Larvae according to OECD GD 239, U. Orzechowska, 2020, Study code: 0038/0016/E
Guideline(s):	OECD GD 239
Deviations:	No Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test design	stability test: tested concentrations and control in one replicate range-finding, definitive, reference test: tested concentrations and control in one replicate; 36 larvae per replicate
Test cages	stability test: volumetric flasks of 250 mL volume range-finding, definitive, reference test: 48-well breeding plates with queen-cell cups placed in the dissector and placed in incubator; from day 8 dissectors placed in test room; from day 15 of the test – transparent plastic boxes placed in test room
Exposition time	4 days (from day 3 to day 6)
Duration of the test	stability test: 72 hours range-finding, definitive, reference test: 22 days
Tested concentrations	stability test: control 0.05 g/L, corresponding to 0.65 mg/kg of food 50 g/L, corresponding to 650 mg/kg of food range-finding test: control 0.65 mg/kg of food, corresponding to 0.1 µg/larva 6.5 mg/kg of food, corresponding to 1 µg/larva 65 mg/kg of food, corresponding to 10 µg/larva 650 mg/kg of food, corresponding to 100 µg/larva definitive test: control 16.64 mg/kg of food, corresponding to 2.56 µg/larva 41.6 mg/kg of food, corresponding to 6.4 µg/larva 104 mg/kg of food, corresponding to 16 µg/larva 260 mg/kg of food, corresponding to 40

Test conditions

µg/larva
650 mg/kg of food, corresponding to 100
µg/larva
reference test:
control
fenoxycarb 0.32 mg/kg of food, corresponding
to 49.28 µg/larva
stability test:
average temperature 5.092°C (minimal
temperature 4.8°C; maximal temperature
6.3°C); darkness
range-finding test:
average temperature 33.986°C (minimal
temperature 32.2°C; maximal temperature
35.2°C); average humidity: 81.527% (minimal
humidity 45.0%; maximal humidity 98.8%);
darkness
definitive test and reference test:
average temperature 34.387°C (minimal
temperature 23.9°C; maximal temperature
35.5°C); average humidity: 81.751% (minimal
humidity 41.1%; maximal humidity 99.9%);
darkness

The assessment test of the test item CHR/F/PROTAZO 375 SC effect on Honey Bee larvae (*Apis mellifera carnica*) was conducted. Study was performed in accordance with document ENV/JM/MONO(2016)34 OECD GD 239 Guidance Document Draft on Honey Bee (*Apis mellifera* L.) Larval Toxicity Test, Repeated Exposure, 15 July 2016. The aim of the study was determination of the concentration causing 50% mortality of population (LC50 value) and the dose causing mortality of 50% of the population after 22 days (LD50 value). Values NOEC and NOED were calculated for the emerged bees on day 22. In course of the experiment, the test item has shown no apitoxic effect in mortality of following developmental stages of bees after 22 days of the test. At the end of the study, the concentration and the dose causing 50% mortality of the population in the test (LC50 and LD50 values) were determined, as well as LC10, LC20, LD10, LD20, NOEC and NOED values were determined at 22 day.

Parameter	Concentration [mg of test item/kg of food]	Parameter	Dose [µg of test item/larva]
LC ₁₀	304.522 (161.163 – 438.730)*	LD ₁₀	46.849 (24.794 – 67.497)*
LC ₂₀	489.620 (335.304 – 851.233)*	LD ₂₀	75.326 (51.585 – 130.959)*
LC ₅₀	1214.545 (739.708 – 5569.216)*	LD ₅₀	186.854 (113.801 – 856.827)*
NOEC	≥650	NOED	≥100

* upper and lower confidence limits (95%) given in the brackets

LC₁₀ test item concentration causing reduction by 10%

LC₂₀ test item concentration causing reduction by 20%

LC₅₀ test item concentration causing reduction by 50%

NOEC the highest test item concentration not causing statistically significant differences in relations to the control

LD₁₀ test item dose causing reduction by 10%

LD₂₀ test item dose causing reduction by 20%

LD₅₀ test item dose causing reduction by 50%

NOED the highest test item dose not causing statistically significant differences in relations to the control

Validity criteria

The test met the validity criteria of the experiment listed in OECD GD 239 Guideline:

- cumulative larval mortality in control on days 3-8 was 0.0% (required: ≤15%),
- the adults emergence rate in control on day 22 was ~~72.2~~ 75% (required: ≥70%),
- larval mortality in control on day 8 for dimethoate was ~~66.7~~ 11.1% (required: ≥50%), for fenoxycarb, the adults emergence rate on day 22 was 5.6% (required: ≤20%)

A 2.3.1.4 KCP 10.3.1.4 Sub-lethal effects

A 2.3.1.4.1 Study 1

Comments of zRMS:	<p>The study follows the guideline and according to the principles of GLP.</p> <p>The following deviations to the guideline were noted:</p> <ul style="list-style-type: none"> - In the preliminary test, temperature was higher than recommended in the Guideline range of 25 ± 2°C %. This short term deviation did not have an impact on the results of the preliminary test. - According to the guideline developed by the IOBC, BART, EPPO Joint Initiative, as a food source only pollen is used. However, in the experiment additional food in the form of the two-spotted spider mite (<i>T. urticae</i>) eggs, was used. Another food source prevents the mites from escaping from discs. <p>The deviations had no effect of the test results.</p> <p>The study is considered to be valid and suitable for the risk assessment.</p>
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Report	A laboratory test for evaluating the effects of Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] on the predatory mite, <i>Typhlodromus pyri</i> (Sch.)E. Kulec-Płoszczyca, 2019, Study code: B/40/19
Guideline(s):	to the ESCORT 1 (Barrett K. L. et al., 1994) and the ESCORT 2 (Candolfi M. P. et al., 2001) guidance documents and the guidelines developed by the IOBC, BART, and EPPO Joint Initiative (Blümel S. et al., 2000)
Deviations:	<input checked="" type="checkbox"/> No <input type="checkbox"/> Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:	name: Prothioconazole + Azoxystrobin (175 + 200) SC producent code: [CHR/F/PROTAZO 375 SC] active substance: 181.5 g/L of prothioconazole 205.1 g/L of azoxystrobin batch number: 03 production date: 13.06.2019 expiry date: 13.06.2021
Biological test system:	the predatory mite, <i>Typhlodromus pyri</i> (Sch.) (Acari: <i>Phytoseiidae</i>)
– age:	24-hour-old protonymphs
– source:	a laboratory culture at the Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna; the culture was augmented by a commercial breeder
Experimental design:	5 study groups: 5 study groups: – a control group <input type="checkbox"/> Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 0.425 L/ha <input type="checkbox"/> Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 0.85 L/ha <input type="checkbox"/> Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 1.7 L/ha– Bi 58 Top 400 EC at the rate of 9.0 mL/ha

number of replicates: 3; number of mites in each replicate: 20

Test conditions:

– temperature:	23 – 26°C
– relative air humidity:	69 – 88%
– photoperiod:	16 h light : 8 h dark
– light intensity:	1097 lux
Statistical analysis:	Probit analysis using max. likelihood regression, Step-down Cochran-Armitage test procedure, Chi ² 2x2 Table Test with Bonferroni Correction

Endpoints:

– mite mortality after 7 days of the treatment

– LR₅₀ and NOER_{mortality}

RESULTS

Mortality and escape of *Typhlodromus pyri*

In the preliminary test, mortality of the control group after 7 days of exposure was 7.5%. After 7 days of exposure to Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rates of 0.11, 0.43 and 1.7 L/ha, the *T. pyri*, corrected [1] mortality percentages were equal to 5.4, 24.3 and 24.3%, respectively.

Table 1. Mortality of *T. pyri* after 7 days of exposure – preliminary non GLP test

Study group [rate] [L/ha]	Number of tested mites [no.]	Mortality				
		Number of dead & escaped mites [no.]		Total dead & escaped		
		I	II	[no.]	[%]	[%] ^c
Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]						
Control	60	3	0	3	7.5	–
0.11	60	3	2	5	12.5	5.4
0.43	60	5	7	12	30.0	24.3
1.7	60	5	7	12	30.0	24.3

c: mortality corrected according to the Abbott's formula [1]

The preliminary test was performed between 20.09 – 27.09.2019

In the definitive test, mortality of the control group after 7 days of exposure was 5.0%. After 7 days of exposure to Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rates of 0.425, 0.85 and 1.7 L/ha, the *T. pyri*, mortality percentages, corrected by Abbott's formula, were equal to 12.3, 17.5 and 29.8%, respectively. There were statistically significant differences in mortality between groups treated with the test item at rates of 0.425, 0.85 and 1.7 L/ha in comparison to the control group (Step-down Cochran-Armitage test procedure, $p(\text{tend}) > \alpha$).

Table 2. Mortality of *T. pyri* after 7 days of exposure – definitive test

Study group [rate] [L/ha]	Number of tested mites [no.]	Mortality					
		Number of dead & escape mites [no.]			Total dead & escape		
		Replicates					
		I	II	III	[no.]	[%]	[%] ^c
Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]							
Control	60	1	0	2	3	5.0	–
0.425 ⁺	60	2	3	3+(2) ^e	10	16.7	12.3
0.85 ⁺	60	2	5	6	13	21.0	17.5
1.7 ⁺	60	7	6	7	20	33.3	29.8
LR ₅₀		> 1.7 [L/ha]					
NOER _{mortality}		< 0.425 [L/ha]					
[mL/ha]	Bi 58 Top 400 EC						
9.0	60	17	20	17	54	90.0	89.5

c: mortality corrected according to the Abbott's formula [1]

+: statistically significant differences

e: number of escaped mites

The definitive test was performed between 21.11 – 28.11.2019.

On the basis of the obtained results the LR50 value could not be estimated. It could be assumed that LR50 value is higher than 1.7 L/ha. The NOER_{mortality} value is below to 0.425 L/ha. After 7 days of exposure to Bi 58 Top 400 EC at the rate of 9.0 mL/ha, the mortality was 89.5%. Therefore, the validity criterion specified in the Method description was met. The results obtained in the reference item group showed that the test organisms were sensitive to dimethoate. Based on the results it can be stated that Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]/ha has an adverse effect on mortality of the tested organisms at the rates of 0.425, 0.85 and 1.7L/ha..

TEST VALIDITY CRITERIA

The following validity criteria were met during the study:

- ☐ mortality of the control group was 5.0% on day 7 of exposure (criterion: a maximum of 20%),
- ☐ corrected mortality of the mites exposed to the reference item at the rate of 9.0 mL/ha was 89.5% on day 7 of exposure (criterion: from 50 to 100%),

A 2.3.1.4.2 Study 2

Comments of zRMS:	The study follows the guideline and according to the principles of GLP. The study is considered to be valid and suitable for the risk assessment.
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Reference: KCP 10.3/04

Report A laboratory test for evaluating the effects of Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] on the parasitic wasp, *Aphidius rhopalosiphii* (De Stefani-Perez), M. Grzesica, 2020, Study code: B/41/19

Guideline(s): the ESCORT 1 (Barrett K.L. et al., 1994) and the ESCORT 2 (Candolfi M.P.

et al., 2001) guidance documents and the guidelines developed by the IOBC, BART, and EPPO Joint Initiative (Mead-Briggs M.A. et al., 2000)

Deviations: No
GLP: Yes
Acceptability: Yes
Duplication (if vertebrate study) No

Materials and methods

Test item: name: Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]
active substances: 181.5 g/L of prothioconazole
205.1 g/L of azoxystrobin
batch number: 03
manufacturing date: 13.06.2019
expiry date: 13.06.2021

Biological test system: the parasitic wasp, *Aphidius rhopalosiphi* (De Stefani-Perez); Hymenoptera: Braconidae, Aphidinae

– **age:** adult wasps (24 - 48 hours after emerging from mummies)

– **source:** a laboratory culture at the Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicological Studies; the culture was obtained from commercial supplier.

Experimental design: 5 study groups:
– a control group (0.0 L/ha)
– Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 0.425 L/ha,
– Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 0.85 L/ha,
– Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rate of 1.7 L/ha,
– Bi 58 Top 400 EC at the rate of 0.12 mL/ha (0.048 g a.i./ha)
number of replicates: 4 replicates/group
number of wasps: 10 wasps/replicate

Test conditions:

– **temperature:** 18 – 20°C

– **relative air humidity:** 67 – 77%

– **photoperiod:** 16 hours light: 8 hours dark

– **light intensity:** mortality assessment: 1408 lx

Statistical analyses: Chi2 2x2 Table Test with Bonferroni Correction.

Endpoints:

– wasp mortality after 48 hours of exposure
– determination of the LR₅₀ and the NOER_{mortality}

RESULTS

Mortality of *A. rhopalosiphi*

In the preliminary test mortality of the control wasps after 48 hours was 10.0%. The percentages of mortality corrected using the formula of Abbott [after 48 hours of exposure to Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rates of 0.11, 0.43 and 1.7 L/ha were 0.0, 5.6 and (-5.6)%, respectively. Negative value indicates that the mortality in the treated group was lower than in the control group.

Table 3. Mortality of *A. rhopalosiphi* after 2 hours – definitive test.

Study group [application rate]	Tested wasps [no.]	Mortality (dead + moribund)					
		Dead wasps [no.]				Total	
		Replicates					
		I	II	III	IV	[no.]	[%]
Control [0.0]	40	0	0	0	0	0	0.0
Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]							
0.425	40	0	0	0	0	0	0.0
0.85	40	0	0	0	0	0	0.0
1.7	40	0	0	0	0	0	0.0
[mL/ha]	Reference item						
0.12	40	0	1	1	1	3	7.5

Table 4. Mortality of *A. rhopalosiphi* after 24 hours – definitive test.

Table 1: Mortality of <i>A. mellonella</i> after 24 hours – definitive test.							
Study group [application rate]	Tested wasps [no.]	Mortality (dead + moribund)					
		Dead wasps [no.]				Total	
		I	II	III	IV	[no.]	[%]
Control [0.0]	40	0	0	0	0	0	0.0
Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]							
0.425	40	0	0	0	0	0	0.0
0.85	40	0	0	0	0	0	0.0
1.7	40	1	1	0	0	2	5.0
[mL/ha]	Reference item						
0.12	40	9	8	8	9	34	85.0

Table 5. Mortality of *A. rhopalosiphi* after 48 hours – definitive test.

Study group/rate [L/ha]	Tested wasps [no.]	Mortality (dead + moribund)					
		Dead wasps [no.]				Total	
		Replicates					
		I	II	III	IV	[no.]	[%]
Control [0.0]	40	0	0	0	0	0	0.0
Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC]							
0.425	40	0	0	0	0	0	0.0
0.85	40	0	0	0	0	0	0.0
1.7 ⁺	40	1	1	0	1	3	7.5
LR ₅₀	Test item [L/ha] ≥1.7			Active ingredients [g/ha] (348.7 ^a + 308.6 ^b)			
NOER _{mortality}	Test item [L/ha] 0.85			Active ingredients [g/ha] (174.3 ^a + 154.3 ^b)			

a: azoxystrobin

b: prothioconazole

⁺: statistically significant difference

The definitive test was performed between 05 – 07.11.2019.

In the definitive test, after 48 hours mortality of the control wasps was 0.0%. After 48 hours of exposure to Prothioconazole + Azoxystrobin (175 + 200) SC [CHR/F/PROTAZO 375 SC] at the rates of 0.425, 0.85 and 1.7 L/ha the percentages of mortality were 0.0, 0.0 and 7.5%, respectively. The LR₅₀ value is higher than 1.7 L/ha and the NOER_{mortality} is equal to 0.85 L/ha. Mortality of the wasps exposed to Bi 58 Top 400 EC at the rate of 0.12 mL/ha, was 85.0% after 24 hours. Therefore, the validity criterion specified in the Method description was met. The results showed that the test organisms were sensitive to dimethoate

TEST VALIDITY CRITERIA

The following validity criteria were met during the study [4]:

- after 48 hours mortality of the control group was 0.0% (criterion: a maximum of 13.0%),
- after 24 hours mortality of the group treated with the reference item at the rate of 0.12 mL/ha was 85.0% (criterion: from 75 to 100%).

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 2.4 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 2.4.1.1.1 Study 2

Comments of zRMS:	<p>The study was conducted to OECD guideline 222 and according to the principles of GLP.</p> <p>The following deviation to the guideline was noted:</p> <ul style="list-style-type: none"> - The range of temperature registered in the definitive test was 17.1 – 22.4°C. Short-term deviation from the recommended range of temperature did not have impact on the generated results. <p>The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference:	KCP 10.4/01
Report	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO 375 SC) Earthworm Reproduction Test (<i>Eisenia andrei</i>), A. Wróbel, 2019, Study code: G/34/19
Guideline(s):	OECD Guideline No. 222 (2016)
Deviations:	No Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO 375 SC) batch no.: 03
Active substance:	prothioconazole – 181.5 g/L azoxystrobin – 205.1 g/L
Artificial soil:	10% sphagnum peat, 20% kaolin clay, 70% airdried quartz sand
Test organism:	the earthworm, <i>Eisenia andrei</i> obtained from a standard laboratory culture cultivated at the Łukasiewicz Research Network Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicological Studies, Laboratory of Soil Toxicology
Test design:	test duration: 8 weeks; number of replicates: 4 replicates/concentration + 8 replicates/control; number of earthworms: 10 earthworms/replicate
Concentrations of the test item:	control, 3.2, 5.6, 10.0, 18.0, 32.0, 56.0, 100.0, 180.0, 320.0, 560.0, and 1000.0 mg/kg dry weight of the artificial soil
Test conditions:	temperature: 17.1 – 22.4°C; pH at the beginning of the experiment: 5.51 – 5.58; pH at the end of the experiment: 5.50 – 5.60; soil moisture content at the beginning of the experiment: 24.4 – 25.2% (48.1 – 49.7% of the maximum water holding capacity); soil moisture content at the end of the experiment: 22.2 – 24.1% (43.9 – 47.5% of the maximum water holding capacity)

	capacity); light-dark cycle: 16h : 8h; light intensity at the beginning of the experiment: 586 – 670 lux light intensity at the end of the experiment: 586– 692 lux
Statistical analysis:	EC ₁₀ , EC ₂₀ , EC ₅₀ , LC ₅₀ – probit analysis using linear max. likelihood regression, NOEC: reproduction – Shapiro-Wilk's Test on Normal Distribution, Bartlett's Test Procedure on Variance Homogeneity, Williams Multiple Sequential t-test Procedure survival – Fisher's Exact Binomial Test with Bonferroni Correction LOEC: a value suggested by the ToxRat Professional 2.10 statistical computer software
Endpoint:	EC ₁₀ , EC ₂₀ , EC ₅₀ , NOEC, LOEC LC ₅₀ , NOEC, LOEC

RESULTS

Mortality of the adult earthworms

. On the basis of the results, it was concluded that at concentrations ranging from 3.2 to 1000.0 mg of the test item/kg dry weight of artificial soil, after 4 weeks of exposure to the test item, mortality of the adult earthworms was ranging from 0.0 to 2.5%. As for the control groups, it was equal to 0.0%. The concentration of the test item causing 50% mortality of the adult earthworms (LC₅₀) is above 1000 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil)

Observations of the earthworms

After 4 weeks of the experiment, the treated living earthworms did not exhibit any changes in appearance and behaviour..

Table 6. Results of the observations of the adult earthworms (*Eisenia andrei*) for changes in behaviour and in morphology.

Concentration [mg/kg dry weight of the artificial soil]	Replicate	Number of tested earthworms [no.]	Changes in behaviour and in morphology
0.0 (control)	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
	5	10	10 nc
	6	10	10 nc
	7	10	10 nc
	8	10	10 nc
3.2	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
5.6	1	10	10 nc
	2	10	9 nc 1 d
	3	10	10 nc
	4	10	10 nc
10.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
18.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
32.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
56.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
100.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
180.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
320.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc
560.0	1	10	10 d
	2	10	10 d
	3	10	9 nc 1 d
	4	10	10 d
1000.0	1	10	10 nc
	2	10	10 nc
	3	10	10 nc
	4	10	10 nc

nc – no changes;
d – dead

Body weights of the living adult earthworms

After the application of the test item at the concentrations ranging from 3.2 to 1000.0 mg/kg dry weight of artificial soil, the body weight increase was between 8.3 and 19.3%. As for the control group, it was equal to 5.0%..

4.4. Impact of the test item on reproduction of the earthworms

Table 11. Endpoint values determined during the earthworm reproduction test (*Eisenia andrei*).

Parameter	Value [mg test item/kg dry weight of artificial soil]	Value [mg of prothioconazole/kg dry weight of artificial soil]	Value [mg of azoxystrobin/kg dry weight of artificial soil]
EC ₁₀	> 1000.0	> 161.9	> 183.0
EC ₂₀	> 1000.0	> 161.9	> 183.0
EC ₅₀	> 1000.0	> 161.9	> 183.0
NOEC (reproduction)	≥ 1000.0	≥ 161.9	≥ 183.0
LOEC (reproduction)	> 1000.0	> 161.9	> 183.0
LC ₅₀	> 1000.0	> 161.9	> 183.0
NOEC (survival)	≥ 1000.0	≥ 161.9	≥ 183.0
LOEC (survival)	> 1000.0	> 161.9	> 183.0

After the application of the test item at the concentrations ranging from 3.2 to 1000.0 mg/kg dry weight of the artificial soil, the mean number of juveniles was between 53 and 71 per replicate. The mean number of juveniles in the control group was equal to 65 per replicate.

After 8 weeks of the experiment, it was concluded that Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO 375 SC) had no statistically significant impact on reproduction of the earthworms at the concentrations ranging from 3.2 to 1000.0 mg/kg dry weight of the artificial soil.

The concentration of the test item causing a 10% reduction in the number of juveniles produced within the exposure period (EC₁₀) is above 1000.0 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil).

The concentration of the test item causing a 20% reduction in the number of juveniles produced within the exposure period (EC₂₀) is above 1000.0 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil).

The concentration of the test item causing a 50% reduction in the number of juveniles produced within the exposure period (EC₅₀) is above 1000.0 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil).

The highest concentration at which the test item is observed to have no statistically significant effects on reproduction (NOEC) is higher than or equal to 1000.0 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil).

The lowest concentration at which the test item is observed to have a statistically significant effect on reproduction (LOEC) is higher than 1000.0 mg/kg dry weight of artificial soil (161.9 mg of prothioconazole + 183.0 mg of azoxystrobin/kg dry weight of artificial soil).

4.5. Observations of the juveniles of earthworms

After 8 weeks of the experiment, the juveniles of earthworms did not exhibit any changes in appearance and behaviour.

4.6. Results of the reference test

Table 12. Reference substance – carbendazim. Number of juvenile earthworms (*Eisenia andrei*).

Concentration [mg/kg dry soil]		Replicate	Number of juveniles [no.]	Mean ±SD	Comparison to the control [%]	CV* [%]
0.0 (control with acetone)		1	67	66 ± 17	-	26.0
		2	60			
		3	41			
		4	77			
		5	45			
		6	76			
		7	71			
		8	93			
0.0 (control)		1	59	71 ± 11	106.4	15.0
		2	66			
		3	57			
		4	82			
		5	68			
		6	71			
		7	73			
		8	88			
1.0		1	66	70 ± 32	104.9	45.6
		2	75			
		3	107			
		4	30			
1.5		1	44	53* ± 7	79.6	12.8
		2	59			
		3	57			
		4	51			
2.25		1	69	52*± 20	78.5	38.5
		2	28			
		3	68			
		4	43			
3.37		1	25	28*± 11	41.5	41.0
		2	44			
		3	19			
		4	22			
5.0		1	9	5*± 4	6.8	86.1
		2	6			
		3	0			
		4	3			
NOEC	mg/kg dry weight of the artificial soil	1.0				
LOEC		1.5				

*statistically significant differences between the control and the treatment groups (Williams Multiple Sequential t- test Procedure, alpha = 0.05, one-sided smaller).

According to the OECD Guideline No. 222, the LOEC should be between 1 – 5 mg/kg dry weight of the artificial soil; hence, it may be concluded that the sensitivity of the test organisms was proper.

VALIDITY CRITERIA

The results are considered valid because the following criteria were satisfied in the controls:

- each replicate produced 65 juveniles (mean) at the end of the experiment - (criterion: ≥ 30 juveniles by the end of the experiment),
- the coefficient of variation of reproduction was 18.8% (criterion: ≤ 30%),
- adult mortality over the initial 4 weeks of the experiment was 0.0% (criterion: ≤ 10%).

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 2.4.2.1.1 Study 2

Comments of zRMS:	<p>The study was conducted to OECD guideline 232 and according to the principles of GLP.</p> <p>The following deviations to the guideline was noted:</p> <ul style="list-style-type: none"> - culturing of collembolans takes place in plastic containers containing an artificial substrate consisting of plaster and charcoal in ratio 9:1 and not 10:1 or 8:1 as is mentioned in OECD Guideline No. 232 (2016), - at the end of the test the soil moisture content was determined by drying small sample of the artificial soil in 105°C instead of weighing the test vessels as it is mentioned in OECD Guideline No. 232 (2016), - physiological or pathological symptoms or distinct changes in behaviour were not described. <p>The deviation did not affect the results of the study.</p> <p>The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference: KCP 10.4/02

Report Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO)
 Collembolan (*Folsomia candida*) Reproduction Test, P. Holewik, 2019,
 Study code: G/35/19

Guideline(s): OECD Guideline No. 232 (2016)

Deviations: ~~No~~ Yes

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Materials and methods

Test item:

Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO)
 batch no.: 03

Active substance:

prothioconazole : 181.5 g/L

azoxystrobin: 205.1 g/L

Artificial soil:

5% sphagnum peat, 20% kaolin clay, and 75% air-dried industrial sand

Test organism:

the collembolan, *Folsomia candida* obtained from a standard laboratory culture at the Łukasiewicz Research Network – Institute of Industrial Organic Chemistry, Branch Pszczyna, Laboratory of Soil Toxicology. The collembolans used in the study were 9 – 12 days old.

Test design:

test duration: 28 days

number of replicates: 4 replicates / concentration + 8 replicates / control; number of collembolans: 10 / replicate

Concentrations of the test item:

a control, 18.0, 32.0; 56.0; 100.0; 180.0; 320.0; 560.0; 1000.0 mg of the test item/kg of dry weight of the artificial soil

Test conditions:

temperature: 18.5 – 20.0°C;

pH at the beginning of the test: 6.04 – 6.25;

pH at the end of the test: 5.89 – 6.01;

soil moisture content at the beginning of the test: 12.8 – 13.2% (44.4– 45.7% of the maximum water holding capacity);

soil moisture content at the end of the test: 11.9 – 12.8% (41.2 – 44.4% of the maximum water holding capacity);

lighting: 16 h light and 8h dark;

light intensity at the beginning of the experiment: 436 – 468 lux

light intensity at the end of the experiment: 439 – 463 lux

Statistical analysis:

EC10, EC20, EC50, LC10, LC20 and LC50 – a 4-parameter logistic analysis or probit analysis

NOEC (number of juveniles):

- Shapiro-Wilk's Test on Normal Distribution,
- Levene's Test on Variance Homogeneity (with Residuals),
- Williams Multiple Sequential t-test Procedure,

NOEC (survival):

- Fisher's Exact Binomial Test with Bonferroni Correction

Endpoints:

EC10, EC20, EC50, NOEC

LC10, LC20, LC50, NOEC

RESULTS

Mortality

Table 6. Endpoint values - the impact of the test item on the mortality of adult collembolans (*Folsomia candida*).

Endpoint	Value [mg test item /kg dry weight of the artificial soil]	Value [mg of prothioconazole /kg dry weight of the artificial soil]	Value [mg of azoxystrobin /kg dry weight of the artificial soil]
LC ₁₀	> 1000.0	> 161.9	> 183.0
LC ₂₀	> 1000.0	> 161.9	> 183.0
LC ₅₀	> 1000.0	> 161.9	> 183.0
NOEC	≥ 1000.0	≥ 161.9	≥ 183.0

After the application of the test item at the concentrations ranging from 18.0 to 1000.0 mg/kg dry weight of the artificial soil, mortality was between 2.5 to 20.0%. As for the control group, it was equal to 6.3%. The concentration of the test item causing a 50% mortality of adults within the exposure period (LC₅₀) is above 1000.0 mg/kg dry weight of the artificial soil (161.9 mg of prothioconazole /kg dry weight of the artificial soil and 183.0 mg of azoxystrobin /kg dry weight of the artificial soil).

Impact on reproduction

Table 8. Endpoint values - the impact of the test item on reproduction of collembolans (*Folsomia candida*).

Endpoint	Value [mg test item /kg dry weight of the artificial soil]	Value [mg of prothioconazole /kg dry weight of the artificial soil]	Value [mg of azoxystrobin /kg dry weight of the artificial soil]
EC ₁₀	189.4	30.7	34.7
EC ₂₀	222.2	36.0	40.7
EC ₅₀	291.7	47.2	53.4
NOEC	180.0	29.1	32.9

After the application of the test item at the concentrations ranging from 18.0 to 1000.0 mg/kg dry weight of the artificial soil, the mean number of juveniles was between 3.0 – 530.8 per replicate. As for the control group, the number of juveniles was equal to 431.4 per replicate.

The obtained results led to the following conclusions:

* The concentration of Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) causing a 10% reduction in the number of juveniles produced within the exposure period (EC₁₀) is equal to 189.4 mg/kg dry weight of the artificial soil (30.7 mg of prothioconazole /kg dry weight of the artificial soil and 34.7 mg of azoxystrobin /kg dry weight of the artificial soil).

* The concentration of Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) causing a

20% reduction in the number of juveniles produced within the exposure period (EC20) is equal to 222.2 mg/kg dry weight of the artificial soil (36.0 mg of prothioconazole /kg dry weight of the artificial soil and 40.7 mg of azoxystrobin /kg dry weight of the artificial soil).

* The concentration of Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) causing a 50% reduction in the number of juveniles produced within the exposure period (EC50) is equal to 291.7 mg/kg dry weight of the artificial soil (47.2 mg of prothioconazole /kg dry weight of the artificial soil and 53.4 mg of azoxystrobin /kg dry weight of the artificial soil).

* The highest concentration at which the test item is observed to have no statistically significant effects on collembolan reproduction (NOEC) is equal to 180.0 mg/kg dry weight of the artificial soil (29.1 mg of prothioconazole /kg dry weight of the artificial soil and 32.9 mg of azoxystrobin /kg dry weight of the artificial soil).

Results of the reference test

The concentration of boric acid causing a 50% reduction in the number of juveniles produced within the exposure period (EC50) is 100.3 mg/kg dry weight of the artificial soil.

According to the OECD Guideline No. 232, the EC50 should be about 100 mg/kg dry weight of the artificial soil; hence, it may be concluded that the sensitivity of the test organisms was proper.

The test was conducted 25.02.2019 – 29.03.2019.

VALIDITY CRITERIA

The results are considered valid because the following criteria were satisfied in the controls:

- * mean adult mortality: 6.3% (criterion: $\leq 20\%$),
- * the mean number of juveniles per vessel at the end of the test: 431.4 (criterion: ≥ 100 juveniles at the end of the test),
- * the coefficient of variation calculated for the number of juveniles: 13.2 (criterion: $\leq 30\%$).

A 2.4.2.1.2 Study 2

Comments of zRMS:	<p>The study was conducted to OECD guideline 226 and according to the principles of GLP.</p> <p>The following deviations to the guideline was noted:</p> <ul style="list-style-type: none"> - According to the OECD Guideline No. 226 (2016) the water content of the soil substrate should be maintained throughout the test by weighing and if needed re-watering the vessels periodically. In the study to maintain proper moisture content, a small sample of soil was drying at 105°C and re-weighing at the beginning, after 7 days of the test and at the end of the test. - Due to the use of the temperature extraction method, there was no need for euthanasia of the extracted organisms since the mites are fixed in a 70% ethanol solution. - Due to the use of the temperature extraction method, there was no impossible to record the symptoms with behavioural and morphology changes of the extracted predatory mites <p>The deviation did not affect the results of the study.</p> <p>The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference: KCP 10.4/03

Report Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO)
 Predatory mite (Hypoaspis (Geolaelaps) aculeifer) reproduction test in soil,
 P. Holewik, 2019, Study code: G/36/19

Guideline(s): the OECD Guideline No. 226 (2016)

Deviations: ~~No~~ Yes
GLP: Yes
Acceptability: Yes
Duplication (if vertebrate study) No

Materials and methods

Test item:

Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) batch number: 03

Active substance:

prothioconazole: 181.5 g/L

azoxystrobin: 205.1 g/L

Artificial soil:

5% sphagnum peat, 20% kaolin clay, and 75% air-dried industrial sand

Test organism:

the predatory mites, *Hypoaspis* (*Geolaelaps*) *aculeifer* (adult female mites from a synchronized culture) obtained from a standard laboratory culture at the Łukasiewicz Research Network - Institute of Industrial Organic Chemistry, Branch Pszczyna, Department of Ecotoxicological Studies, Laboratory of Soil Toxicology. The mites were introduced 7 – 14 days after becoming adult.

Test design:

test duration: 14 days

number of replicates: 4 replicates / concentration + 8 replicates / control; number of mites: 10 mites / replicate

Concentrations of the test item:

a control, 5.6, 10, 18, 32, 56, 100, 180, 320, 560 and 1000 mg test item/kg dry weight of the artificial soil.

Test conditions:

temperature: 18.5 – 21.5C

pH at the beginning of the test: 5.64 – 5.70

pH at the end of the test: 5.51 – 5.74

soil moisture content at the beginning of the test: 13.2 – 14.9% (41.9 – 47.3% of the maximum water holding capacity)

soil moisture content in the middle of the test: 13.6 – 15.5% (43.1 – 49.2% of the maximum water holding capacity)

soil moisture content at the end of the test: 14.2 – 15.2% (45.0 – 48.2% of the maximum water holding capacity)

light-dark cycle: 16 h light and 8 h dark

light intensity at the beginning of the test: 501 – 524 lux

light intensity at end of the test: 508 – 533 lux

Statistical analysis:

EC10, EC20, EC50 – a logit analysis using linear max. likelihood regression

LC10, LC20, LC50 – a probit analysis using linear max. likelihood regression

NOEC:

- offspring number – Shapiro-Wilk's Test on Normal Distribution, Bartlett's Test Procedure on Variance Homogeneity, Williams Multiple Sequential t-test Procedure

- survival – Fisher's Exact Binomial Test with Bonferroni Correction

Endpoints:
EC10, EC20, EC50, NOEC
LC10, LC20, LC50, NOEC

RESULTS

Mortality of adult females

Table 7. Endpoint values – the impact of the test item on survival of adult females

(Hypoaspis aculeifer).

Endpoint	Value [mg/kg dry weight of the artificial soil]	Value [mg of prothioconazole / kg dry weight of the artificial soil]	Value [mg of azoxystrobin / kg dry weight of the artificial soil]
LC ₁₀	> 1000.0	> 161.9	> 183.0
LC ₂₀	> 1000.0	> 161.9	> 183.0
LC ₅₀	> 1000.0	> 161.9	> 183.0
NOEC (survival)	≥ 1000.0	≥ 161.9	≥ 183.0

Mortality of the predatory mites exposed to the test item at the concentrations ranging from 5.6 to 1000 mg/kg dry weight of the artificial soil was between 0.0% and 5.0%. Mortality of the control group was equal to 1.3%. The concentration of the test item causing a 50% mortality of adults within the exposure period (LC₅₀) is above 1000 mg/kg dry weight of the artificial soil (above 161.9 mg of prothioconazole / kg dry weight of the artificial soil and 183.0 mg of azoxystrobin / kg dry weight of the artificial soil).

Impact on reproduction

Table 9. Endpoint values - the impact of the test item on reproduction of the predatory mites

(Hypoaspis aculeifer).

Endpoint	Value [mg/kg dry weight of the artificial soil]	Value [mg of prothioconazole / kg dry weight of the artificial soil]	Value [mg of azoxystrobin / kg dry weight of the artificial soil]
EC ₁₀	923.541 (822.9 – 951.3)	149.5 (133.2 – 154.0)	169.0 (150.6 – 174.1)
EC ₂₀	> 1000.0	> 161.9	> 183.0
EC ₅₀	> 1000.0	> 161.9	> 183.0
NOEC (reproduction)	≥ 1000.0	≥ 161.9	≥ 183.0

After the application of the test item at the concentrations ranging from 5.6 to 1000 mg/kg dry weight of the artificial soil, the mean number of juveniles was between 100.8 – 144.8 per replicate. The mean number of juveniles in the control group was equal to 121.5 per replicate.

The obtained results led to the following conclusions:

* The concentration of the test item causing a 10% reduction in the number of mites produced within the exposure period (EC10) is equal to 923.5 mg/kg dry weight of the artificial soil (equal to 149.5 mg of prothioconazole / kg dry weight of the artificial soil and 169.0 mg of azoxystrobin / kg dry weight of the artificial soil).

* The concentration of the test item causing a 20% reduction in the number of mites produced within the exposure period (EC20) is above 1000 mg/kg dry weight of the artificial soil (above 161.9 mg of prothioconazole / kg dry weight of the artificial soil and 183.0 mg of azoxystrobin / kg dry weight of the artificial soil).

* The concentration of the test item causing a 50% reduction in the number of mites produced within the exposure period (EC50) is above 1000 mg/kg dry weight of the artificial soil (above 161.9 mg of prothioconazole / kg dry weight of the artificial soil and 183.0 mg of azoxystrobin / kg dry weight of the artificial soil).

* The highest concentration at which the test item is observed to have no statistically significant effects on mite reproduction (NOEC) is above or equal to 1000 mg/kg dry weight of the artificial soil (above or equal to 161.9 mg of prothioconazole / kg dry weight of the artificial soil and 183.0 mg of azoxystrobin / kg dry weight of the artificial soil).

Results of the reference test

concentration of boric acid causing a 50% reduction in the number of juveniles produced within the exposure period (EC50) is 240.155 mg/kg dry weight of the artificial soil. According to the OECD Guideline No. 226, the EC50 should be between 100 and 500 mg/kg dry weight of the artificial soil; hence, it may be concluded that the sensitivity of the test organisms was proper.

VALIDITY CRITERIA

The results are considered valid because the following criteria were satisfied in the control:

- * mean adult mortality: 1.3% (criterion: $\leq 20\%$),
- * the mean number of juveniles per vessel at the end of the test: 121.5 (criterion: ≥ 50 juveniles at the end of the test,
- * the coefficient of variation for the number of juveniles: 17.8 (criterion: $\leq 30\%$).

A 2.5 KCP 10.5 Effects on soil nitrogen transformation

A 2.5.1.1.1 Study 1

Comments of zRMS:	<p>The study was conducted to OECD guideline 216 and according to the principles of GLP.</p> <p>The following deviations to the guideline was noted:</p> <ul style="list-style-type: none"> - According the Guideline, the soil extraction should be conducted at 150 rpm for 60 min. However, in this study, the extraction was performed at 90 rpm for 24 hours. The modification resulted from the optimization of the nitrate extraction which showed that the extraction was more effective when the shaking rate was lower and the extraction lasted longer. - The predicted environmental concentration (PEC) was calculated assuming 1 cm of the soil depth according to the German conditions for the substances with the mobility in soil $K_{Foc} > 500$ mL/g. Thus, the applied soil depth is a deviation from OECD Guideline No. 216 (2000) and EU Method C.21 where the PEC is calculated by using 5 cm of the soil depth. <p>The deviation did not affect the results of the study.</p> <p>The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference:	KCP 10.5/01
Report	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Soil Microorganisms: Nitrogen Transformation Test), P. Holewik, 2019, Study code: G/37/19
Guideline(s):	the OECD Guideline No. 216 (2000)/EU Method C.21.
Deviations:	No Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test material:

Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO)
batch no.: 03

Active substance:

prothioconazole: 181.5 g/L
azoxystrobin: 205.1 g/L

Soil:

Agricultural soil collected from a place belonging to the Łukasiewicz Research Network - Institute of Industrial Organic Chemistry, Branch Pszczyna.

Test design:

Three portions of soil (3 x 1500 g), i.e. one control group and two treated groups. Every portion was divided into three replicates (3 x 500 g). The soil was enriched with the organic substrate, i.e. lucerne at dose of 5 g/kg dry weight of soil. Test duration: 28 days.

Concentrations of the test item:

control, PEC: 11.96 mg of the test item / kg dry weight of soil (1.94 mg of the prothioconazole/kg dry weight of soil and 2.19 mg of the azoxystrobin/kg dry weight of soil), 5 x PEC: 59.78 mg of the test item / kg dry weight of soil (9.70 mg of the prothioconazole/kg dry weight of soil and 10.95 mg of the azoxystrobin/kg dry weight of soil).

Test conditions:

temperature: 20.0 – 22.0°C,
soil moisture: 49.0% – 53.9% of the maximum water holding capacity, incubation in darkness

Endpoints:

The concentration of nitrate [mg/kg dry soil] after 0, 7, 14 and 28 days of incubation.

The nitrate formation rate [mg/kg dry weight of soil/day] for selected time intervals of soil incubation, i.e. 0 – 7, 0 – 14, 0 – 28 days.

Percent deviation from the control in nitrate formation rate calculated for selected time intervals i.e. 0 – 7, 0 – 14, 0 – 28 days.

Statistical analysis:

- Shapiro-Wilk's test on Normal Distribution
- Levene's Test on Variance Homogeneity (with Residuals)
- Williams Multiple Sequential t-test Procedure

RESULTS

Table 5. Concentration of the nitrate ions on day 0 of incubation.

Concentration	Control			PEC			5 x PEC		
Replicate	I	II	III	I	II	III	I	II	III
Reading* [mg/L]	9.951	10.581	9.951	15.501	14.561	15.241	15.501	15.761	16.981
Nitrate ion concentration [mg/kg of dry soil]	49.76	52.91	49.76	77.51	72.81	76.21	77.51	78.81	84.91
Mean nitrate ion concentration [mg/kg of dry soil] \pm SD	50.81 \pm 1.82			75.51* \pm 2.43			80.41* \pm 3.95		
CV	3.6			3.2			4.9		

* - values adjusted for the value of the blank sample

+ - statistically significant difference between the control and the treatment group (Williams Multiple Sequential t-test Procedure, significance level = 0.05, two sided)

Table 6. Concentration of the nitrate ions on day 7 of incubation.

Concentration	Control			PEC			5 x PEC		
Replicate	I	II	III	I	II	III	I	II	III
Reading* [mg/L]	38.745	41.335	44.235	43.945	42.895	45.825	43.655	51.565	53.575
Nitrate ion concentration [mg/kg dry of soil]	193.73	206.68	221.18	219.73	214.48	229.13	218.28	257.83	267.88
Mean nitrate ion concentration [mg/kg of dry soil] \pm SD	207.19 \pm 13.73			221.11 \pm 7.42			247.99* \pm 26.22		
CV	6.6			3.4			10.6		

* - values adjusted for the value of the blank sample

+ - statistically significant difference between the control and the treatment group (Williams Multiple Sequential t-test Procedure, significance level = 0.05, two sided)

Table 7. Concentration of the nitrate ions on day 14 of incubation.

Concentration	Control			PEC			5 x PEC		
Replicate	I	II	III	I	II	III	I	II	III
Reading* [mg/L]	70.899	69.439	69.299	79.509	74.289	80.019	77.209	91.289	82.789
Nitrate ion concentration [mg/kg of dry soil]	354.50	347.20	346.50	397.55	371.45	400.10	386.05	456.45	413.95
Mean nitrate ion concentration [mg/kg of dry soil] \pm SD	349.40 \pm 4.43			389.70 \pm 15.86			418.81* \pm 35.45		
CV	1.3			4.1			8.5		

* - values adjusted for the value of the blank sample

* - statistically significant difference between the control and the treatment group (Williams Multiple Sequential t-test Procedure, significance level = 0.05, two sided)

Table 8. Concentration of the nitrate ions on day 28 of incubation.

Concentration	Control			PEC			5 x PEC		
Replicate	I	II	III	I	II	III	I	II	III
Reading* [mg/L]	85.603	80.753	87.333	93.753	90.063	93.043	99.253	98.503	102.363
Nitrate ion concentration [mg/kg of dry soil]	428.02	403.77	436.67	468.77	450.32	465.22	496.27	492.52	511.82
Mean nitrate ion concentration [mg/kg of dry soil] \pm SD	422.82 \pm 17.06			461.43* \pm 9.79			500.20* \pm 10.23		
CV	4.0			2.1			2.0		

* - values adjusted for the value of the blank sample

* - statistically significant difference between the control and the treatment group (Williams Multiple Sequential t-test Procedure, significance level = 0.05, two sided)

There were no statistically significant differences between the control and the group treated with the test item at the concentration corresponding to the PEC in nitrate concentration after 7 and 14 days of incubation.

There were statistically significant differences between the control and the group treated with the test item at the concentration corresponding to the 5xPEC in nitrate concentration after 7 and 14 days of incubation.

There were statistically significant differences between the control and the group treated with the test item at both concentrations i.e PEC and 5xPEC in nitrate concentration after 0 and 28 days of incubation

Table 9. Nitrate formation rate* [mg nitrate/kg dry weight of soil/day] for selected time intervals.

Time interval [d]	Control				PEC				5 x PEC			
	Replicate			Mean ± SD	Replicate			Mean ± SD	Replicate			Mean ± SD
	I	II	III		I	II	III		I	II	III	
0 – 7	20.417	22.267	24.339	22.341 ± 1.96	20.603	19.853	21.946	20.80 ± 1.06	19.696	25.346	26.781	23.941 ± 3.75
0 – 14	21.692	21.171	21.121	21.328 ± 0.32	23.003	21.139	23.185	22.44 ± 1.13	21.831	26.860	23.824	24.17 ± 2.53
0 – 28	13.472	12.606	13.781	13.286 ± 0.61	14.045	13.386	13.918	13.783 ± 0.35	14.852	14.718	15.408	14.993* ± 0.37

* - Rate of nitrate ions formation per a day = [(mg nitrate / kg of soil dry weight on sampling day 'a') - (mg nitrate / kg of soil dry weight on day 0)]/ 'a' day; 'a' = 7, 14 and 28 day

* - statistically significant difference between the control and the treatment group (Williams Multiple Sequential t-test Procedure, significance level = 0.05, two sided)

At the time interval: 0 – 7 and 0 - 14 there were no statistically significant difference between the control and the group treated with test item at both concentrations i.e PEC and 5xPEC in nitrate formation rate

At the time interval: 0 – 28 there were no statistically significant differences between the control and the groups treated with test item at the concentrations corresponding to the PEC in nitrate formation rate. For the concentration of the test item corresponding to the 5xPEC the statistically significant difference in nitrate formation rate in comparison to the control group was observed.

The difference in the nitrate formation rate between the control soil and the one treated with the test item at the concentration corresponding to the PEC: 11.96 mg of the test item / kg dry weight of soil (1.94 mg of the prothioconazole/kg dry weight of soil and 2.19 mg of the azoxystrobin/kg dry weight of soil) and 5 x PEC: 59.78 mg of the test item / kg dry weight of soil (9.70 mg of the prothioconazole/kg dry weight of soil and 10.95 mg of the azoxystrobin/kg dry weight of soil) did not exceed 25% on 28 day of analysis.

When the difference in the nitrates formation rate between the lower treatment (PEC) and a control is equal to or less than 25% at any sampling day after day 28, the product can be evaluated as having no long-term influence on nitrogen transformation in soil.

On the basis of the results, it was concluded that Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) at the concentration corresponding to the PEC: 11.96 mg of the test item / kg dry weight of soil (1.94 mg of the prothioconazole/kg dry weight of soil and 2.19 mg of the azoxystrobin/kg dry weight of soil) and 5 x PEC: 59.78 mg of the test item / kg dry weight of soil (9.70 mg of the prothioconazole/kg dry weight of soil and 10.95 mg of the azoxystrobin/kg dry weight of soil) did not have any long-term adverse effects on the process of nitrogen transformation in aerobic surface soils.

VALIDITY CRITERION

The coefficients of variation (CV) in the control group were 3.6, 6.6, 1.3 and 4.0%, after 0, 7, 14 and 28 days of incubation. The validity criterion was met, because the variation between replicate control samples is less than ± 15%.

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

A 2.6.1 KCP 10.6.1 Summary of screening data

A 2.6.2 KCP 10.6.2 Testing on non-target plants

Comments of zRMS:	The study was conducted to OECD guideline 227 and according to the principles of GLP.
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	<p>The following deviations to the guideline was noted:</p> <ul style="list-style-type: none"> - According to OECD Guideline No. 227 (2006), the light intensity should be $350 \pm 50 \mu\text{E}/\text{m}^2/\text{s}$. However, these values are recommended for tests conducted in greenhouses. The experiment was conducted in a test room, where only artificial lighting was used. The light intensity was between $87.3 - 149.3 \mu\text{E}/\text{m}^2/\text{s}$. Good control plant vigour was observed. Therefore, it was concluded that the light intensity was suitable for plant growing. <p>The deviation did not affect the results of the study. The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference: KCP 10.6/02

Report: Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Terrestrial Plant Test: Vegetative Vigour Test), P. Holewik, 2019, Study code: G/38/19

Guideline(s): the OECD Guideline No. 227 (2006)

Deviations: ☒ No ☐ Yes

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) No

Materials and methods

Test item:
 Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) batch no: 03

active substance:
 prothioconazole: 181.5 g/L
 azoxystrobin: 205.1 g/L

Test species:
 sunflower (*Helianthus annuus*), cabbage (*Brassica oleracea* var. *capitata*), pea (*Pisum sativum*), carrot (*Daucus carota*), perennial ryegrass (*Lolium perenne*), oats (*Avena sativa*).

Soil:
 sandy loam

Study design:
 number of rates: 5 application rates + control; number of replicates: 4 replicates/rate for carrot, oats, perennial ryegrass and 7 replicates/rate for cabbage, pea, sunflower . The total number of plants per application rate – 20 for carrot, oats, perennial ryegrass and 21 for sunflower, pea, cabbage.
 Test termination: 21 days after the spraying.

Application rates:
 - 24.7, 74.1, 222.2, 666.7 and 2000.0 ml test item/ha (i.e. 9.5, 28.6, 85.9, 257.7 and 773.2 g a.s./ha) - sunflower, cabbage, pea, carrot, perennial ryegrass and oats.
 volume of deionised water used to prepare the highest rate: 300 L water/ha

Test conditions:
 temperature: $15.5 - 24.6^\circ\text{C}$, humidity: 46.6 – 93.3%, controlled light – dark cycles (16h:8h), light intensity: $87.3 - 149.3 \mu\text{E}/\text{m}^2/\text{s}$, carbon dioxide concentration: 315 – 346 ppm.

Statistical analysis:

ER₂₅, ER₅₀ – probit analysis.

NOER (shoot length) - Shapiro-Wilk's Test on Normal Distribution, Levene's Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure.

NOER (shoot dry weight) - Shapiro-Wilk's Test on Normal Distribution, Levene's Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure.

Endpoints:

ER₂₅, ER₅₀, NOER.

RESULTS

Table 30. ER₂₅, ER₅₀ and NOER values (mL of the test item/ha).

	Sunflower <i>Helianthus annuus</i>	Cabbage <i>Brassica oleracea</i> <i>var. capitata</i>	Pea <i>Pisum sativum</i>	Carrot <i>Daucus carota</i>	Perennial ryegrass <i>Lolium perenne</i>	Oats <i>Avena sativa</i>
Plant number at the end of the experiment						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0
Shoot length (plants without roots)						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 666.7	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0
Plant dry weight (plants without roots)						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 666.7	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0

The ER₂₅, ER₅₀, and NOER values were calculated using the ToxRat Professional 2.10 computer software.

Table 31. ER₂₅, ER₅₀ and NOER values (g of active substance /ha).

	Sunflower <i>Helianthus annuus</i>	Cabbage <i>Brassica oleracea</i> <i>var. capitata</i>	Pea <i>Pisum sativum</i>	Carrot <i>Daucus carota</i>	Perennial ryegrass <i>Lolium perenne</i>	Oats <i>Avena sativa</i>
Plant number at the end of the experiment						
ER₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2
Shoot length (plants without roots)						
ER₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 257.7	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2
Plant dry weight (plants without roots)						
ER₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 257.7	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2

The ER₂₅, ER₅₀ and NOER values were calculated using the ToxRat Professional 2.10 computer software.

Sunflower (*Helianthus annuus*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the sunflower shoot length was between 83.6 and 99.6% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the sunflower shoot dry weight was between 85.0 and 100.5% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage were not observed.

Cabbage (*Brassica oleracea* var. *capitata*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the cabbage shoot length was between 93.5 and 104.7% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the cabbage shoot dry weight was between 87.3 and 102.7% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Pea (*Pisum sativum*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the pea shoot length was between 96.5 and 107.4% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the pea shoot dry weight was between 102.4 and 120.4% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the

plant damage were observed.

Carrot (*Daucus carota*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the carrot shoot length was between 90.1 and 103.5% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the carrot shoot dry weight was between 88.5 and 116.9% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage were not observed.

Perennial ryegrass (*Lolium perenne*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed .

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the perennial ryegrass shoot length was between 101.6 and 104.4% of the control shoot length .

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the perennial ryegrass shoot dry weight was between 96.0 and 127.3% of the control shoot weight (Table 24).

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Oats (*Avena sativa*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, plants mortality were not observed.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the oats shoot length was between 102.6 and 106.8% of the control shoot length .

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the oats shoot dry weight was between 97.2 and 115.1% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

CONCLUSIONS

1. The test item, i.e. Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) applied at rates ranging from 24.7 to 2000.0 mL of the test item/ha had no impact on the process of growth on vegetative vigour of all analyzed species.
2. The test item did not cause mortality of all analyzed species at rates ranging from 24.7 to 2000.0 mL of the test item/ha.
3. On the basis of NOER, ER25 and ER50 values determined from the shoot length it was proved that the test item did not inhibit the process of cabbage, pea, carrot, perennial ryegrass and oats. The test item slightly impacted the growth of sunflower.
4. On the basis of NOER, ER25 and ER50 values determined from the shoot dry weight, it was proved that the test item did not inhibit the process of growth of cabbage, pea, carrot, perennial ryegrass and oats. The test item slightly impacted the growth of sunflower.
5. Some phytotoxic symptoms were not observed of all analyzed species after 21 days of the exposure.

VALIDITY CRITERIA

The following validity criteria were met:

- the seedling emergence (validity criterion: at least 70%) was as follows:

78.6 – 97.6% – sunflower,

92.9 – 100.0% – cabbage,

81.0 – 90.5% – pea,

85.0 – 92.5% – carrot,

85.0 – 92.5% – perennial ryegrass,

75.0 – 92.5% – oats,

- the mean survival of the emerged control seedlings was 100% in sunflower, cabbage, pea, carrot, perennial ryegrass and oats,
- the control seedlings did not exhibit any visible phytotoxic symptoms,
- environmental conditions for all plants belonging to the same species were identical.

Comments of zRMS:	<p>The study was conducted to OECD guideline 208 and according to the principles of GLP.</p> <p>The following deviations to the guideline was noted:</p> <ul style="list-style-type: none"> - According to OECD Guideline No. 208 (2006), the light intensity should be $350 \pm 50 \mu\text{E}/\text{m}^2/\text{s}$. However, these values are recommended for tests conducted in greenhouses. The experiment was conducted in a test room, where only artificial lighting was used. The light intensity was between 64.8-169.0 $\mu\text{E}/\text{m}^2/\text{s}$. Good control plant vigour was observed. Therefore, it was concluded that the light intensity was suitable for plant growing. <p>The deviation did not affect the results of the study.</p> <p>The study is considered to be reliable and suitable for the risk assessment.</p>
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Reference:	KCP 10.6/01
Report	Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test), P. Holewik, 2019, Study code: G/39/19
Guideline(s):	the OECD Guideline No. 208 (2006).
Deviations:	No Yes
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	No

Materials and methods

Test item:

Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO)

batch no: 03

active substance:

prothioconazole: 181.5 g/L

azoxystrobin: 205.1 g/L (Appendix No. 1)

Test species:

sunflower (*Helianthus annuus*), cabbage (*Brassica oleracea* var. *capitata*), pea (*Pisum sativum*), carrot (*Daucus carota*), perennial ryegrass (*Lolium perenne*), oats (*Avena sativa*).

Soil:

Sandy loam

Study design:

number of rates: 5 application rates for sunflower, cabbage, pea, carrot, perennial ryegrass and oats + control; number of replicates: 4 replicates/rate

for carrot, perennial ryegrass, oats; 7 replicates/rate for cabbage, sunflower and pea. The total number of plants per application rate – 20 for carrot, oats, perennial ryegrass and 21 for cabbage, sunflower and pea

test termination: 14 days after the emergence of 50% of the control seedlings

Test concentrations:

- 24.7, 74.1, 222.2, 666.7 and 2000.0 ml test item/ha (i.e. 9.5, 28.6, 85.9, 257.7 and 773.2 g a.s./ha) - sunflower, cabbage, pea, carrot, perennial ryegrass and oats.

volume of deionised water used to prepare the highest rate: 300 L water/ha

Test conditions:

temperature: 18.5 – 24.7 °C, humidity: 54.3 – 89.0 %, lighting: 16 h light : 8 h dark; light intensity: 64.8 – 169.0 µE/m²/s; carbon dioxide concentration: 309 – 328 ppm

Statistical analysis:

ER₂₅, ER₅₀ : probit analysis,

NOER:

– emergence – Fisher’s Exact Binomial Test with Bonferroni Correction

– shoot length – Shapiro - Wilk’s Test on Normal Distribution, Levene’s Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure, Welch-t test for Inhomogeneous Variances with Bonferroni-Holm Adjustment.

– shoot dry weight - Shapiro - Wilk’s Test on Normal Distribution, Levene’s Test on Variance Homogeneity (with Residuals), Williams Multiple Sequential t-test Procedure, Welch-t test for Inhomogeneous Variances with Bonferroni-Holm Adjustment.

Endpoints:

ER₂₅, ER₅₀, NOER

RESULTS

Table 36. ER₂₅, ER₅₀ and NOER values (mL of the test item/ha).

Endpoint	Sunflower <i>Helianthus annuus</i>	Cabbage <i>Brassica oleracea</i> var. <i>capitata</i>	Pea <i>Pisum sativum</i>	Carrot <i>Daucus carota</i>	Perennial ryegrass <i>Lolium perenne</i>	Oats <i>Avena sativa</i>
Plant number at the end of the experiment						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0
Shoot length (plants without roots)						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0
Plant dry weight (plants without roots)						
ER₂₅	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
ER₅₀	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0	> 2000.0
NOER	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0	≥ 2000.0

The ER₂₅, ER₅₀ and NOER values were calculated using the ToxRat Professional 2.10 computer software.

Table 37. ER₂₅, ER₅₀ and NOER values (g of active substance / ha).

Endpoint	Sunflower <i>Helianthus annuus</i>	Cabbage <i>Brassica oleracea</i> var. <i>capitata</i>	Pea <i>Pisum sativum</i>	Carrot <i>Daucus carota</i>	Perennial ryegrass <i>Lolium perenne</i>	Oats <i>Avena sativa</i>
Plant number at the end of the experiment						
ER ₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER ₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2
Shoot length (plants without roots)						
ER ₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER ₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2
Plant dry weight (plants without roots)						
ER ₂₅	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
ER ₅₀	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2	> 773.2
NOER	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2	≥ 773.2

The ER₂₅, ER₅₀, and NOER values were calculated using the ToxRat Professional 2.10 computer software

Sunflower (*Helianthus annuus*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of sunflower was not delayed when compared with the control. The death of sunflower plants were not observed. At the control group, 90.5% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha, total number of plants at the end of the experiment ranged from 100.0 to 110.5% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the sunflower shoot length was between 101.3 and 120.1% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the sunflower shoot weight was between 91.1 and 100.7% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Cabbage (*Brassica oleracea* var. *capitata*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of cabbage was not delayed when compared with the control. The death of cabbage plants were not observed. At the control group, 100.0% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha total number of plants at the end of the experiment was equal to 100.0% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the cabbage shoot length was between 90.9 and 103.3% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the cabbage shoot weight was between 93.6 and 118.4% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Pea (*Pisum sativum*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of pea was not delayed when compared with the control. The death of plants were not

observed (Table 16). At the control group, 100.0% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha total number of plants at the end of the experiment was ranging from 81.0 to 95.2% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the pea shoot length was between 97.1 and 103.1% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the pea shoot weight was between 81.6 – 99.7% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Carrot (*Daucus carota*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of carrot was not delayed when compared with the control. The death of plans were not observed. At the control group, 100.0% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha total number of plants at the end of the experiment ranged from 75.0 to 95.0% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the carrot shoot length was between 89.4 and 107.4% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the carrot shoot weight was between 78.0 and 154.0% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Perennial ryegrass (*Lolium perenne*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of perennial ryegrass was not delayed when compared with the control. At the rates ranging from 24.7 to 2000.0 mL of the test item/ha the death of perennial ryegrass plants were not observed. At the control group, 100.0% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha total number of plants at the end of the experiment ranged from 95.0 to 100.0% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the perennial ryegrass shoot length was between 85.2 and 103.1% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the perennial ryegrass shoot weight was between 77.9 to 118.5% of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

Oats (*Avena sativa*)

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, seedling emergence of oats was not delayed when compared with the control. At the control group the death of oats plants were not observed. At the control group, 85.0% of plants emerged. At rates ranging from 24.7 to 2000.0 mL of the test item/ha, total number of plants at the end of the experiment ranged from 88.2 to 111.8% in comparison to the control group.

After the application of the test item at the rates between 24.7 to 2000.0 mL of the test item/ha, the oats shoot length was between 95.5 and 102.1% of the control shoot length.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the oats shoot weight was between 92.4 and 106.0 of the control shoot weight.

After the application of the test item at the rates ranging from 24.7 to 2000.0 mL of the test item/ha, the plant damage was not observed.

CONCLUSIONS

1. The test item, i.e. Prothioconazole + Azoxystrobin (175 + 200) SC (CHR/F/PROTAZO) applied at rates ranging from 24.7 to 2000.0 mL of the test item/ha had no impact on the growth and seedling emergence of all analyzed species.

- ## VALIDITY CRITERIA

- the seedling emergence in the control (validity criterion: at least 70%) was as follows:

100.0% - cabbage,

100.0% - pea,

100.0% – carrot.

100.0% – perennial ryegrass,

85.0% – oats,

- the mean survival of the emerged control seedlings (validity criterion: at least 90%) was as follows:

100.0% - sunflower,

100.0% - cabbage,

100.0 % - pea,

100.0% – carrot,

100.0% – perennial ryegrass,

100.0% – oats,

- the control seedlings did not exhibit any visible phytotoxic effects;

- environmental conditions for all plants of the same species were identical

A 2.6.3	KCP 10.6.3	Extended laboratory studies on non-target plants
<p>1. Test species</p> <p>2. Test conditions</p> <p>3. Test results</p>	<p>1. Test species</p> <p>2. Test conditions</p> <p>3. Test results</p>	<p>1. Test species</p> <p>2. Test conditions</p> <p>3. Test results</p>

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

A 2.8 KCP 10.8 Monitoring data