REGISTRATION REPORT

Part B

Section 6

Mammalian Toxicology

Detailed summary of the risk assessment

Product code: ADM.03503.F.1.A

Product name(s): see Part A

Chemical active substances:

Fluxapyroxad, 75 g/L

Prothioconazole, 150 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

(authorization)

Applicant: Country organisation / representative

as specified in Part A

Submission date: April 2022

MS Finalisation date: May 2023 (initial Core Assessment)

November 2023 (final Core Assessment)

Version history

When	What
2022/04	Version 1 Applicant
May 2023	Initial zRMS assessment The report in the dRR format has been prepared by the Applicant, therefore all comments, additional evaluations and conclusions of the zRMS are presented in grey commenting boxes. Minor changes are introduced directly in the text and highlighted in grey. Not agreed or not relevant information are ~~struck through~~ and shaded for transparency.
November 2023	Final report (Core Assessment updated following the commenting period) Additional information/assessments included by the zRMS in the report in response to comments received from the cMS and the Applicant are highlighted in yellow. Not agreed or not relevant information are ~~struck through~~ and shaded for transparency.

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DATA PROTECTION CLAIM

In order to present a dossier fully compliant with today's requirements (Reg. 284/2013), studies have been performed on ADM.03503.F.1.A. Under Article 59, Regulation 1107/2009/EC, on behalf of the Sponsor Company the applicant claims data protection for the studies conducted with ADM.03503.F.1.A. The data protection status and corresponding justification as valid for the respective country will be confirmed in the respective PART A.

STATEMENT FOR OWNERSHIP

The summaries and evaluations contained in this document may be based on unpublished proprietary data submitted for the purpose of the assessment undertaken by the regulatory authority that prepared it. Other registration authorities should not grant, amend, or renew a registration on the basis of the summaries and evaluation of unpublished proprietary data contained in this document unless they have received the data on which the summaries and evaluation are based, either –

- from the owner of the data, or
- from a second party that has obtained permission from the owner of the data for this purpose or,
- following expiry of any period of exclusive use, by offering – in certain jurisdictions – mandatory compensation, unless the period of protection of the proprietary data concerned has expired.

Reviewer comments:

This application has been submitted for the approval under Art.33 of EU Regulation 1107/2009 of the product with commercial name Avastel 225 EC (developmental code ADM.03503.F.1.A) an emulsifiable concentration (EC) formulation containing fluxapyroxad 75 g/L and prothioconazole 150g/L ADM.03503.F.1.A is a fungicide (details see GAP dRR B0) to support registration of the product in Poland and zonal registration for which PL was designated zRMS.
ADM.03503.F.1.A was not a representative formulation reviewed during the Annex I inclusion/active substance renewal and has not previously been evaluated in any EU countries according to the Uniform Principles, thus it is not possible to refer to the DRAR conclusion on fluxapyroxad and prothioconazole with regard to the formulation studies. Therefore, relevant data on the plant protection product ADM.03502.F.1.A had to be generated for authorization purposes.
For the current product registration, APPL provided an assessment of the toxicological potential based on content of relevant ingredients/calculation method (ATEmix; for details refer Part C) also *in vivo* and *in vitro* studies. ~~Regarding fact that outcome of the mentioned above assessments are in general similar zRMS PL decided , in accordance with the EC recommendations to avoid tests on animals, for the purposes of hazard classification to take into account data obtained using the calculation method. *In vivo* studies has been considered as supplementary. Some of *in vitro* studies has not been accepted by the zRMS.~~
Reflecting comments made by cMS, zRMS decided to reconsider hazard assessment and taking into account outcome of the *in vivo* tests.
Note: Considering information available in GD OECD 439 revision 14 June 2021 INITIAL CONSIDERATIONS AND LIMITATIONS Subsection 8: p.2 (..) data indicates a lack of applicability of the RhE based in vitro skin irritation test for agrochemical formulations (47). (..). See also: Kolle S.N, van Ravenzwaay B. and Landsiedel R. (2017). Regulatory accepted but out of domain: In vitro skin irritation tests for agrochemical formulations. Regul. Toxicol. Pharmacol 89, 125-130, study [redacted] 2021b (*In Vitro* Skin Irritation Test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) Using Reconstructed Human Epidermis Tissues (RhE) (OECD 439)) has not been accepted.
NDE assessment for operator, workers and B&R exposure to the fluxapyroxad, prothioconazole and PTZ-desthio considering all critical use(s) and all tasks, identify safe use of the product ADM.03503.F.1.A /Avastel 225 EC.

Based on the results of the acute toxicity and non-dietary risk assessments conducted for ADM.03503.F.1.A /Avastel 225 EC, the following personal protective equipment (PPE)/risk management measures (RMM) are recommended:
Operators, based on NDE assessment must wear adequate workwear covering arms, body and legs during M/L and also protective gloves during M/L.
Note: additional RMM based on classification & labelling:
Due to the classification of the product with ~~Skin Corr. 1B H314~~ H318: Causes serious eye damage ~~protective gloves, protective clothing and~~ eye protection/face protection should be worn when handling the product.

Worker: Worker should use adequate workwear covering arms, body and legs when entering in a treated area.

6 Mammalian Toxicology (KCP 7)

6.1 Summary

Table 6.1-1: Information on ADM.03503.F.1.A *

Product name and code	ADM.03503.F.1.A
Formulation type	Emulsifiable concentrate [Code: EC]
Active substances (incl. content)	Fluxapyroxad; 75 g/L Prothioconazole; 150 g/L
Function	Fungicide
Product already evaluated as the 'representative formulation' during the approval of the active substances	No
Product previously evaluated in another MS according to Uniform Principles	No

* Information on the detailed composition of ADM.03503.F.1.A can be found in the confidential dRR Part C.

Justified proposals for classification and labelling

According to the criteria given in Regulation (EC) No 1272/2008 of the European Parliament and of the Council of 16 December 2008, the following classification and labelling with regard to toxicological data is proposed for the preparation:

Table 6.1-2: Justified proposals for classification and labelling for ADM.03503.F.1.A according to Regulation (EC) No 1272/2008

Hazard classes, categories	Eye Dam. 1 Lact. ~~Acute Tox. 4~~ ~~Skin Corr. 1B~~
Codes for hazard pictograms	 GHS05  ~~GHS07~~
Signal word	Danger
Hazard statements	~~H302: Harmful if swallowed~~ ~~H314: Causes severe skin burns and eye damage~~ H318: Causes serious eye damage H362: May cause harm to breast-fed children.
Precautionary statements	P201 Obtain special instructions before use. P263: Avoid contact during pregnancy/while nursing. P260: Do not breathe dust/mist P280: Wear ~~protective gloves/protective clothing~~ eye protection/face protection. P305 + P351 + P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. ~~P303 + P361 + P353: IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].~~
Additional labelling phrases	To avoid risks to man and the environment, comply with the instructions for use. [EUH401]

Table 6.1-3: Summary of risk assessment for operators, workers, residents and bystanders for ADM.03503.F.1.A

	Result	PPE / Risk mitigation measures
Operators	Acceptable	Gloves during mixing/loading Additional RMM: regarding proposed product hazard ~~classyfication~~ classification Operators must wear ~~protective gloves/protective clothing~~ eye protection/face protection
Workers	Acceptable	None
Residents	Acceptable	None
Bystanders	Acceptable	None

No unacceptable risk for operators, workers, residents and bystanders was identified when the product is used as intended and provided that the PPE/ risk mitigation measures stated in Table 6.1-3 are applied.

A summary of the critical uses and the overall conclusion regarding exposure for operators, workers and residents/bystanders is presented in the following table.

Table 6.1-4 Critical uses and overall conclusion of exposure assessment

1	2	3	4	5	6	7	8	9	10			
Use-No.*	**Crops and situation (e.g. growth stage of crop)**	**F, Fn, Fpn G, Gn, Gpn or I ******	**Application**		**Application rate**		**PHI (d)**	**Remarks:** **(e.g. safener/synergist (L/ha))** **critical gap for operator, worker, resident or bystander exposure based on [Exposure model]**	**Acceptability of exposure assessment**			
			Method / Kind **(incl. application technique *****)**	**Max. number (min. interval between applications)** **a) per use** **b) per crop/ season**	**Max. application rate kg as/ha** **a) Fluxapyroxad** **b) Prothioconazole**	**Water L/ha** **min / max**			**Operator**	**Worker**	**Residents**	**Bystander**
1	Winter wheat, (BBCH 30-69)	F	Spraying, LCTM	1 ; 1	a) 0.09375 b) 0.1875	125 - 400	Not applicable	Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products; EFSA Journal 2014;12(10):3874				

* 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

*** e.g. LC: low crops, HC: high crop, TM: tractor-mounted, HH: hand-held

Explanation for column 10 "Acceptability of exposure assessment"

A	Exposure acceptable without PPE / risk mitigation measures
R	Further refinement and/or risk mitigation measures required
N	Exposure not acceptable/ Evaluation not possible

Data gaps

Noticed data gaps are:

- No data gaps were identified

6.2 Toxicological Information on Active Substances

Information regarding classification of the active substances and on EU endpoints and critical areas of concern identified during the EU review are given in Table 6.2-1.

Table 6.2-1: Information on active substances

	Active substance 1	**Active substance 2**	**Prothioconazole-desthio** (Toxicological relevant metabolite of active substance prothioconazole*)
Common Name	Fluxapyroxad	Prothioconazole	Prothioconazole-desthio
CAS-No.	907204-31-3	178928-70-6	120983-64-4
Classification and proposed labelling			
With regard to toxicological endpoints (according to the criteria in Reg. 1272/2008, as amended)	Hazard class, category: Lact. Code for hazard pictogram: None Signal word: None Hazard statement: H362	None	None

	Active substance 1	Active substance 2	**Prothioconazole-desthio** (Toxicological relevant metabolite of active substance prothioconazole*)
	Precautionary statements: P201 P260 P263 P264 P270 P308+P313		
Additional C&L proposal	None	None	None
Agreed EU endpoints			
AOEL systemic	0.04 mg/kg bw/d (corrected for 68% oral absorption)	Prothioconazole: 0.2 mg/kg bw/d Prothioconazole-desthio: 0.01 mg/kg bw/d	0.01 mg/kg bw/d (desthio-prothioconazole (JAU 6476-desthio)* (based on the rat developmental study NOAEL 1 mg/kg bw and the critical effects with increased rudimentary supernumerary ribs)
Reference	EFSA Conclusion EFSA Journal 2012;10(1):2522	EFSA Conclusion EFSA Scientific Report (2007) 106, 1-98	EFSA Conclusion EFSA Scientific Report (2007) 106, 1-98
Conditions to take into account/critical areas of concern with regard to toxicology			
EFSA Conclusion for active substance	None	The metabolite prothioconazole-desthio is more toxic than prothioconazole in the rat and rabbit developmental studies	

*as stated in DAR (2005), B.6.15.1 Operator exposure (III 7.2.1), p.327: "It has been found that JAU 6476-desthio (SXX 0665) may be formed in diluted prothioconazole formulations. This may happen on clothing, skin or certain plant surfaces during the drying process. The degradation product, JAU 6476-desthio, is known to have an embryotoxic potential in experimental animals."

6.3 Toxicological Evaluation of Plant Protection Product

A summary of the toxicological evaluation for ADM.03503.F.1.A is given in the following tables. To support the evaluation in the EU, the Acute Toxicity Estimate was calculated based on the acute toxicity profile of the ingredients in the formulation. In addition, to satisfy regulatory requirements of countries outside of the EU, *in vitro* and *in vivo* acute toxicity studies were conducted with this formulation. The results of all available data are presented in the table below. Full summaries of studies on the product that have not been previously considered within an EU peer review process are described in detail in Appendix 1. For those instances where the results of the studies with the mixture show a different result to the ATE based on the ingredients, the experimental results of the formulation will be used in accordance with Regulation (EC) No 1272/2008, para. 3.1.3.4.1.

Table 6.3-1: Summary of evaluation of the studies on acute toxicity including irritancy and skin sensitisation for ADM.03503.F.1.A

Acute toxicity endpoint	Type of test, species, model system (Guideline)	Result	Acceptability	Classification (acc. to the criteria in Reg. 1272/2008)	Reference
Acute oral toxicity	Calculation method	ATE_{mix}=978	~~Yes~~ Not considered for hazard assessment**	Acute Tox. 4 H302	Regulation (EC) No 1272/2008
	LD_{50} oral, rat (OECD 423)	> 2000 mg/kg bw	~~Supplementary~~ Yes	**None**	[redacted] 2021a *
Acute dermal toxicity	Calculation method	> 2000 mg/kg bw	~~Yes~~ Not considered for hazard	None	Regulation (EC) No 1272/2008

			assessment**		
	LD_{50} dermal, rat (OECD 402, waiver report)	> 2000 mg/kg bw	~~Supplementary~~ Yes	None	[redacted], 2021b
Acute inhalation toxicity	Calculation method	> 2000 mg/kg bw	~~Yes~~ Not considered for hazard assessment**	None	Regulation (EC) No 1272/2008
	LC_{50} inhalation, rat (OECD 436)	> 5.28 mg/L air	~~Supplementary~~ Yes	**None**	[redacted], 2021a *
Skin irritation	Calculation method	Corrosive	~~Yes~~ Not considered for hazard assessment**	Skin Corr. 1B H314	Regulation (EC) No 1272/2008
	Skin corrosion, RhE (OECD 431)	Non-corrosive	No	None	[redacted], 2021a
	Skin irritation, RhE (OECD 439)	Irritant	No	~~Skin Irrit. 2 H315~~ Not possible to classify***	Barad I. M., 2021b
	Skin irritation, rabbit (OECD 404)	Non-irritant	~~Supplementary~~ Yes	**None**	[redacted], 2021c *
Eye irritation	Calculation method	Irritant	~~Yes~~ Not considered for hazard assessment**	Eye Dam. 1 H318	Regulation (EC) No 1272/2008
	Eye irritation, BCOP (OECD 437)	Inconclusive	No	Inconclusive	[redacted], 2021b
	Eye irritation, rabbit (OECD 405)	Irritant	~~Supplementary~~ Yes	**Eye Dam. 1 H318**	[redacted], 2021d *
Skin sensitisation	Calculation method	Non-sensitising	~~Yes~~ Not considered for hazard assessment**	None	Regulation (EC) No 1272/2008
	Skin sensitisation, DPRA (OECD 442C)	Inconclusive	No	Not possible to classify using 2 out of 3 defined approach	[redacted] 2021c
	Skin sensitisation, KeratinoSens (OECD 442D)	Positive	No	Not possible to classify using 2 out of 3 defined approach	[redacted], 2021d
	Skin sensitisation, mouse (OECD 429, LLNA)	Non-sensitising	~~Supplementary~~ Yes	**None**	[redacted], 2021c *
	Supplementary studies for combinations of plant protection products	No data – not required	--		

* These in vivo studies were not performed with intention for use within the EU, but to satisfy the regulatory requirements of countries outside of the EU. The results do not indicate that a more severe classification is required than has been determined based on the ingredients of ADM.03503.F.1.A. However, they are submitted with this application as relevant information.

**Reflecting cMS comments zRMS decided to reconsider hazard assessment and taking into account outcome of the *in vivo* tests (recommendation provided in Regulation (EC) No. 1272/2008 to give animal data preference)

- Considering that the Acute Oral Toxicity Study "[redacted], 2021" is valid from a scientific point of view, study resulted in no classification in line with Regulation (EC) No 1272/2008
- Considering that the Acute Dermal Toxicity Study "[redacted], 2021" is valid from a scientific point of view, study resulted in no classification in line with Regulation (EC) No 1272/2008
- Considering that the Acute Inhalation Toxicity Study "[redacted], 2021" is valid from a scientific point of view, study resulted in no classification in line with Regulation (EC) No 1272/2008
- Considering that the Skin Irritation Study "[redacted], 2021" is valid from a scientific point of view, study resulted in no classification in line with Regulation (EC) No 1272/2008

- Considering that the Eye Irritation Study "[redacted], 2021" is valid from a scientific point of view, **study resulted in H318 classification** in line with Regulation (EC) No 1272/2008
- Considering that the Skin Sensitization Study "[redacted], 2021" is valid from a scientific point of view, study resulted in no classification in line with Regulation (EC) No 1272/2008

***Conclusion is only possible if the study data is used in combination with the data from the other *in vitro* study

Table 6.3-2: Additional toxicological information relevant for classification/labelling of ADM.03503.F.1.A

	Substance (concentration in product, % w/w)	Classification of the substance (acc. to the criteria in Reg. 1272/2008)	Reference	Classification of product (acc. to the criteria in Reg. 1272/2008)
Toxicological properties of active substances (relevant for classification of product)	Fluxapyroxad (7.5%)	Lact. H362 (criteria ≥0.3%)	Reg. 1272/2008	Lact. H362
Toxicological properties of non-active substances (relevant for classification of product)	Confidential information submitted in Part C			Acute Tox. 4 H302 Skin Corr. 1B H314 Eye Dam. 1 H318
Further toxicological information	No data – not required			

* Please use concentration range or concentration limit (e.g. 1-10% or > 1%) as provided in MSDS.
** Material safety data sheet by the applicant

6.4 Toxicological Evaluation of Groundwater Metabolites

All metabolite concentrations for prothioconazole are predicted to stay below 0.1 µg/L – no groundwater assessment is required.

The following data on metabolites of fluxapyroxad with the potential to reach the groundwater in concentrations above 0.1 µg/L and requiring relevance assessment were submitted. Note that the relevance assessment of the metabolites is reported in Part B.10; the submitted toxicological studies are summarised in this document.

6.4.1 M700F001

An overview of the results of the accepted toxicological studies for groundwater metabolite M700F001 is given in the following table.

Table 6.4-1: Summary of the results of toxicity studies for M700F001

Type of test, species (Guideline)	Result	Acceptability	Reference*
Rat oral LD50	> 2000 mg/kg bw	Yes	EFSA Journal 2012;10(1):2522
Ames, in vitro chromosome aberration, in vitro mammalian gene cell mutation, in vivo micronucleus tests).	Not genotoxic	Yes	EFSA Journal 2012;10(1):2522
90-day dietary rat study	NOAEL 1000 mg/kg bw/day (highest dose level tested).	Yes	EFSA Journal 2012;10(1):2522
Rabbit developmental toxicity study	NOAEL for maternal and developmental toxicity 250 mg/kg bw/day (highest dose level tested; severe maternal severe	Yes	EFSA Journal 2012;10(1):2522

Type of test, species (Guideline)	Result	Acceptability	Reference*
	maternal toxicity at ≥500 mg/kg/day in range-finding studies		

* indicates that a study was reviewed at EU level

6.4.2 M700F002

An overview of the results of the accepted toxicological studies for groundwater metabolite M700F002 is given in the following table.

Table 6.4-2: Summary of the results of toxicity studies for M700F002

Type of test, species (Guideline)	Result	Acceptability	Reference*
Rat oral LD50	> 2000 mg/kg bw	Yes	EFSA Journal 2012;10(1):2522
Ames, in vitro chromosome aberration, in vitro mammalian gene cell mutation, in vivo micronucleus tests).	Not genotoxic	Yes	EFSA Journal 2012;10(1):2522
90-day dietary rat study	NOAEL 1000 mg/kg bw/day (highest dose level tested).	Yes	EFSA Journal 2012;10(1):2522
Rabbit developmental toxicity study	NOAELs: 300 mg/kg bw/day for maternal (based on reduction of body weight gain and food consumption) and 1000 mg/kg bw/day developmental toxicity (the highest dose tested).	Yes	EFSA Journal 2012;10(1):2522

* indicates that a study was reviewed at EU level

6.5 Dermal Absorption (KCP 7.3)

A summary of the dermal absorption rates for the active substances in ADM.03503.F.1.A are presented in the following table.

Table 6.5-1: Dermal absorption rates for active substances in ADM.03503.F.1.A

	Fluxapyroxad		Prothioconazole		Prothioconazole-desthio	
	Value	Reference	Value	Reference	Value	Reference
Concentrate	~~5.0%~~ 5.2%	New study reported in Appendix 1	25%	Default values in 2017 EFSA Guidance on dermal absorp-tion	-	New study reported in Appendix 1
Dilution 1	~~9.0%~~ 9.5% (1:320, 0.234 g/L)		70%		12% (1:290, 0.469 g/L)	
Dilution 2	~~12%~~ 13% (1:400, 0.188 g/L)		70%		13% (1:363, 0.375 g/L)	

6.5.1 Justification for proposed values - Fluxapyroxad

Proposed dermal absorption rates for fluxapyroxad are based on dermal absorption studies using ADM.03503.F.1.A. The study results are summarised in the following table. Full summaries of studies on the dermal absorption of fluxapyroxad that have not previously been evaluated within an EU peer review process are described in detail in Appendix 1.

Table 6.5-2: Summary of the results of submitted dermal absorption studies for fluxapyroxad

Test	Concentrate	Spray dilution	Formulation in study	Acceptability of study	Justification provided on representativity of study formulation for current product	Acceptability of justification	Reference*
In vitro (human)	~~5.0%~~ 5.2%	~~9.0% (1:320, 0.234 g/L)~~ ~~12.0% (1:400, 0.188 g/L)~~ 9.5% (1:320, 0.234 g/L) 13.0% (1:400, 0.188 g/L)	ADM.03503.F.1.A	Yes	Not required	Justification accepted. Endpoint can be used for current product	[redacted], 2021a

* indicates that a study was reviewed at EU level

6.5.2 Justification for proposed values - prothioconazole

No data on dermal absorption for prothioconazole in ADM.03503.F.1.A is available. Justifications for default values according to Guidance on Dermal Absorption (EFSA Journal 2017; 15(6):4873) are presented in the following table.

Table 6.5-3: Default dermal absorption rates for prothioconazole

	Value	Justification for value	Acceptability of justification
Concentrate	25%	ADM.03503.F.1.A is an EC formulation. Prothioconazole is present at a concentration of >50 g/L.	Justification accepted. Endpoint can be used for current product
Dilution	70%	ADM.03503.F.1.A is an EC formulation. Prothioconazole is present at a concentration of <50 g/L.	Justification accepted. Endpoint can be used for current product

6.5.3 Justification for proposed values – Prothioconazole-desthio

Proposed dermal absorption rates for prothioconazole-desthio are based on dermal absorption studies using ADM.03503.F.1.A. The study results are summarised in the following table. Full summaries of studies on the dermal absorption of prothioconazole-desthio that have not previously been evaluated within an EU peer review process are described in detail in Appendix 1.

Table 6.5-4: Summary of the results of submitted dermal absorption studies for prothioconazole-destio

Test	Concentrate	Spray dilution	Formulation in study	Acceptability of study	Justification provided on representativity of study formulation for current product	Acceptability of justification	Reference*
In vitro (human)	-	12% (1:290, 0.469 g/L) 13% (1:363, 0.375 g/L)	ADM.03503.F.1.A	Yes	Not required	Justification accepted. Endpoint can be used for current product	[redacted], 2021b

* indicates that a study was reviewed at EU level

6.6 Exposure Assessment of Plant Protection Product (KCP 7.2)

Reviewer comment:

The NDE calculations performed by the applicant using EFSA Operator Model (75th quantile regression) are acceptable and zRMS agrees to the conclusions.
The risk assessment/calculated exposure for operators, workers and B&R are acceptable under conditions of intended uses.

Table 6.6-1: Product information and toxicological reference values used for exposure assessment

Product name and code	ADM.03503.F.1.A	
Formulation type	EC	
Category	Fungicide	
Container size(s), short description	Please refer to Section 4.1 for further information on containers	
Active substance (incl. content)	**Fluxapyroxad** 75 g/L	
AOEL systemic	0.04 mg/kg bw/d	
Inhalation absorption	100%	
Oral absorption	68%	
Dermal absorption	~~Concentrate: 5.0%~~ ~~Dilutions:~~ ~~9.0% (0.234 g a.s./L)~~ ~~12.0% (0.188 g a.s./L)~~ Concentrate: 5.2% Dilutions: 9.5% (0.234 g a.s./L) 13.0% (0.188 g a.s./L) Based on product (formulation), Finlayson, Z. (2021a) reported in Appendix 1	
Vapour pressure	8.1×10^{-9} Pa at 25°C (1), *i.e.* low volatile substances having a vapor pressure of $<5 \times 10^{-3}$ at 25°C	
	Active substance **Prothioconazole** 150 g/L	Toxicological relevant metabolite of active substance* **Prothioconazole-desthio**
AOEL systemic	0.2 mg/kg bw/d (2)	0.01 mg/kg bw/d (2)
Inhalation absorption	100%	100%
Oral absorption	100%	100%
Dermal absorption	Concentrate: 25% Dilution: 70% (Default values based on EFSA 2017 guidance (3))	Concentrate: ~~Not applicable~~ 12% (measured value for dilution) Dilutions: 12% (0.469 g a.s./L) 13% (0.375 g a.s./L) (Based on product (formulation), Finlayson, Z. (2021b) reported in Appendix 1) 23% (0.213 g a.s./L) (*pro-rata e*xtrapolated value)
Vapour pressure	$<<4 \times 10^{-7}$ Pa at 20°C (1), $<<4 \times 10^{-7}$ Pa at 25°C (1), *i.e.* low volatile substances having a vapor pressure of $<5 \times 10^{-3}$ at 25°C	Parent value

(1) European Food Safety Authority; Conclusion on the peer review of the pesticide risk assessment of the active substance fluxapyroxad (BAS 700 F). EFSA Journal 2012;10(1):2522. [90 pp.] doi:10.2903/j.efsa.2012.2522.
(2) European Food Safety Authority (2007). Conclusion regarding the peer review of the pesticide risk assessment of the active substance prothioconazole. EFSA Journal 2007; 5(8): RN-106, 1-98. doi:10.2903/j.efsa.2007.106r.
(3) European Food Safety Authority (2017). Guidance on dermal absorption. EFSA Journal 2017;15(6):4873. doi: 10.2903/j.efsa.2017.4873
* In addition to the risk assessment of the active substance prothioconazole, the risk of the metabolite prothioconazole-desthio is assessed (based on the EFSA conclusion for prothioconazole).

Overall considerations in the exposure assessment of ADM.03503.F.1.A

According to the DAR (2005)[1], diluted prothioconazole can degrade to the metabolite prothioconazole-desthio:

"It has been found that JAU 6476-desthio (SXX 0665) may be formed in diluted prothioconazole formulations. This may happen on clothing, skin or certain plant surfaces during the drying process. The degradation product, JAU 6476-desthio, is known to have an embryotoxic potential in experimental animals."

According to the EFSA conclusion of prothioconazole[2], the degradation product prothioconazole-desthio is more toxic than the parent compound and is therefore considered in the risk assessments of all relevant population groups. The content of prothioconazole-desthio in the concentrate is however assumed to be very low. This assumption is based on the Commission Implementing Regulation (EU) No 540/2011 (amending Regulation (EU) No 1107/2009), in which it is declared that the amount of prothioconazole-desthio may not exceed 0.5 g/kg, i.e. 0.05 % (*w/w*) in the technical material.[3]

However, as a highly conservative approach exposure to prothioconazole-desthio has been considered during mixing and loading the concentrated product since a very low level of conversion may take place when the concentrate is exposed to humid air or sweat on the skin of the operator. Since storage stability data have shown that ADM.03503.F.1.A is stable as a concentrate 100% conversion upon exposure to low levels of moisture is not considered realistic and therefore represents a theoretical worst case.

In a conservative approach, three exposure assessments are conducted for each relevant exposure group. One assessment reflects exposure to 100 % prothioconazole. A further scenario has also been considered in which both substances are present at 50%. It is considered that for mixing and loading even 50% conversion is an over estimation and provides a highly conservative exposure estimate for prothioconazole-desthio.

~~To reflect this non-availability of prothioconazole-desthio when handling the concentrate, *i.e.* during mixing and loading, the value for the dermal absorption of prothioconazole-desthio in the concentrate was set to 0 %.~~

~~In a conservative approach, two exposure assessments are conducted for each relevant exposure group. One assessment reflects exposure to 100 % prothioconazole when handling the concentrate, the dilution or the dried formulation, while the other assessment reflects exposure to 100 % prothioconazole-desthio when handling the dilution or the dried formulation.~~

For the estimation of the amount of prothioconazole-desthio that may be formed when handling the product, a conversion factor is calculated based of the molecular weights of prothioconazole-desthio and prothioconazole (Equation 1):

$$\frac{M_{prothioconazole-desthio}}{M_{prothioconazole}} = \frac{312.2\ \text{g/mol}}{344.26\ \text{g/mol}} = 0.907 \qquad \text{Equation 1}$$

- $M_{prothioconazole}$ = Molecular weight of prothioconazole (EFSA, 2007)

[1] DAR (2005), B.6.15.1 Operator exposure (III 7.2.1), p. 327
[2] Conclusion regarding the peer review of the pesticide risk assessment of the active substance prothioconazole. EFSA Journal 2007; 5(8): RN-106, 1-98. doi:10.2903/j.efsa.2007.106r
[3] dRR Part B Section 1: Identity, Section 2: Physical and chemical properties, Section 4: Further information, 2020

- $M_{prothioconazole-desthio}$ = Molecular weight of prothioconazole-desthio (EFSA, 2007)

Taking the above calculated conversion factor of 0.907 into account, 0.17 kg prothioconazole-desthio/ha is to be considered for an application rate of 1.25 L prod./ha, containing 0.1875 kg prothioconazole/ha.

Reviewer comment regarding conversion factor PTZ to PTZ-desthio: Some experimental data implies that a conversion rate of 50% is closer to reality (observed conversion rates in exposure studies were between 1 and 70%; 50% conversion was the 90th percentile from the whole dataset). However, in the absence of new experimental data, which would allow a recalculation, zRMS as a precautionary approach propose to use of a conversion rate of 100% as worst case. However, the reviewer is aware that some experts from cMS may prefer a conversion factor of 50%.

6.6.1 Selection of critical use(s) and justification

The critical GAP used for the exposure assessment of the plant protection product is shown in Table 6.1-4. A list of all intended uses within the zone is given in Part B, Section 0.

Justification

ADM.03503.F.1.A is a fungicide applied as spray in cereals. All applications are done via tractor-mounted downward spraying. The highest application rate is 1.25 L product/ha.

The product is applied in a water volume of 125 L/ha to 400 L/ha in cereals. For fluxapyroxad a dermal absorption value for the spray dilution of ~~9.0~~ 9.5% has been assumed for the exposure assessment. This value has been derived for a spray concentration of 0.234 g/L which corresponds to the maximum application rate applied in the maximum water volume. For prothioconazole-desthio a dermal absorption value of 13% has been assumed. This value corresponds to a spray concentration of 0.375 g prothioconazole-desthio/L, which is a slightly lower spray concentration than is obtained from applying 1.25 L product/ha in 400 L/ha. These dermal absorption values provide a worst case for the intended use on cereals. For completeness the assessment for residents considers applications made using both the minimum and maximum water volumes for which different dermal absorption values apply.

As default dermal absorption values are used for prothioconazole, the exposure assessment considers applications are made in the minimum water volume, as this provides a worst-case risk assessment for bystanders and residents exposure from spray drift.

6.6.2 Operator exposure (KCP 7.2.1)

6.6.2.1 Estimation of operator exposure

A summary of the exposure models used for estimation of operator exposure to the active substances during application of ADM.03503.F.1.A according to the critical use is presented in Table 6.6-2. The outcome of the estimation is presented in
Table 6.6-3 (longer term exposure). Detailed calculations are in Appendix 2.

Table 6.6-2: Exposure models for intended uses

Critical use	Winter wheat (max. 1.25 L product/ha)
Model	Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products; EFSA Journal 2014;12(10):3874 calculator version: 30/03/2015

Table 6.6-3: Estimated operator exposure (longer term exposure)

		Fluxapyroxad	
Model data	**Level of PPE**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to cereals			
Application rate		0.09375 kg a.s./ha	
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) M/L and A	~~0.0148~~ 0.0154	~~36.99~~ 38.50
	Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A	~~0.0016~~ 0.0017	~~3.95~~ 4.14

		Prothioconazole		Prothioconazole-desthio*	
Model data	**Level of PPE**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to cereals					
Application rate: 1.25 L product/ha (0.1875 kg prothioconazole/ha or 0.17 kg prothioconazole-desthio/ha in a worst-case approach)					
~~**Spray application** (AOEM; 75th percentile) Body weight: 60 kg~~	~~Work wear (arms, body and legs covered) M/L and A~~	~~0.1332~~	~~66.58~~	~~0.0029~~	~~28.96~~
	~~Mixing/Loading~~	~~0.1162~~	~~58.11~~	~~-~~	~~-~~
	~~Application~~	~~0.0170~~	~~8.48~~	~~0.0029~~	~~28.96~~
~~**Spray application** (AOEM; 75th percentile) Body weight: 60 kg~~	~~Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A~~	~~0.0204~~	~~10.22~~	~~0.0029~~	~~28.96~~
	~~Mixing/Loading~~	~~0.0035~~	~~1.74~~	~~-~~	~~-~~
	~~Application~~	~~0.0170~~	~~8.48~~	~~0.0029~~	~~28.96~~
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) M/L and A	0.1332	66.58	0.0547	547
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A	0.0204	10.22	0.0045	44.89

*First tier estimates of exposure assume as a theoretical worst case that there is 100% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. For this conversion 1 kg prothioconazole yields 0.907 kg prothioconazole-desthio. This conversion can occur during the drying process on clothing and skin so for spray operators the exposure assessment only considers exposure from application of the spray solution

Dermal absorption:
Fluxapyroxad - Concentrate: 5.2%, Dilution: 9.5% (0.234 g a.s./L)
Prothioconazole - Concentrate: 25%, Dilution: 70% (default values)
Prothioconazole-desthio - Concentrate: 12%, Dilution: 13% (0.375 g a.s./L)

An assessment is also presented which takes into account a conversion factor of 50% of prothioconazole to prothioconazole-desthio which may be considered more realistic. New calculations on operator exposure estimates are presented below:

Applying a conversion rate of 50% of prothioconazole to its desthio-metabolite, 0.094 kg prothioconazole /ha is to be considered for an application rate of 1.25 L prod./ha and for prothioconazole-desthio an amount of 0.085 kg prothioconazole-desthio/ha by the following equation:

Application rate prothioconazole-desthio = application rate of prothioconazole x MW prothioconazole-desthio / MW prothioconazole x conversion rate (%).

Thus, the calculated application rate of prothioconazole-desthio in 1.25 L prod./ha is 0.09375 kg a.s./ha x 312.2 g/mol / 344.3 g/mol x 1 = 0.085 kg prothioconazole-desthio/ha.

With a 50% conversion rate the concentration of prothioconazole-desthio in the spray dilution when applying 1.25 L product/ha in 400 L/ha would be 0.213 g/L. This value is less concentrated than the concentrations tested in the dermal absorption study and therefore a dermal absorption value has been extrapolated from the lowest concentration tested (13% for 0.375 g/L). For prothioconazole-desthio a dermal absorption value of 23% has therefore been assumed

Table 6.6-4: Estimated operator exposure (longer term exposure) – 50% conversion rate

		Fluxapyroxad	
Model data	**Level of PPE**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to cereals			
Application rate		0.09375 kg a.s./ha	
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) M/L and A	0.0154	38.50
	Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A	0.0017	4.14

		50% Prothioconazole		50% Prothioconazole-desthio*	
Model data	**Level of PPE**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**	**Total absorbed dose (mg/kg/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to cereals					
Application rate: 1.25 L product/ha (50 % conversion to prothioconazole-desthio)					
		0.094 kg a.s./ha Prothioconazole		0.085 kg a.s./ha Prothioconazole-desthio	
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) M/L and A	0.0766	38.3	0.0329	329
Spray application (AOEM; 75th percentile) Body weight: 60 kg	Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A	0.0104	5.2	0.0034	34.4

*Estimates of exposure assume that there is 50% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. For this conversion 1 kg prothioconazole yields 0.907 kg prothioconazole-desthio.
Dermal absorption:
Fluxapyroxad - Concentrate: 5.2%, Dilution: 9.5% (0.234 g a.s./L)
Prothioconazole - Concentrate: 25%, Dilution: 70% (default values)
Prothioconazole-desthio - Concentrate: 12%, Dilution: 23% (0.213 g a.s./L *pro rata* extrapolated)

6.6.2.2 Measurement of operator exposure

Since the operator exposure estimations carried out indicated that the acceptable operator exposure levels (AOELs) will not be exceeded under conditions of intended uses and considering above mentioned personal protective equipment (PPE), a study to provide measurements of operator exposure was not necessary and was therefore not performed.

6.6.3 Worker exposure (KCP 7.2.3)

6.6.3.1 Estimation of worker exposure

Table 6.6-5 shows the exposure model used for estimation of worker exposure after entry into a previously treated area or handling a crop treated with ADM.03503.F.1.A according to the critical use. Outcome of the estimation is presented in

Table 6.6-6 (longer term exposure). Detailed calculations are in Appendix 2.

Table 6.6-5: Exposure models for intended uses

Critical use	Winter wheat (max. 1.25 L product/ha)
Model	Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products; EFSA Journal 2014;12(10):3874 calculator version: 30/03/2015

Table 6.6-6: Estimated worker exposure (longer term exposure)

		Fluxapyroxad			
Model data	**Level of PPE**	**Total absorbed dose (mg/kg bw/day)**		**% of systemic AOEL**	
Crop inspection in cereals Outdoor Work rate: 2 hours/day, DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable					
Number of applications and application rate		1 x 0.09375 kg a.s./ha			
Body weight: 60 kg	Work wear (arms, body and legs covered) TC: 1400 cm²/person/h	0.0012		~~2.95~~ 3.12	
		Prothioconazole		**Prothioconazole-desthio**	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.1875 kg a.s./ha		1 x 0.17 kg a.s./ha in a worst-case approach	
Body weight: 60 kg	Work wear (arms, body and legs covered) TC: 1400 cm²/person/h	0.0184	9.19	0.0031	30.94

Dermal absorption:
Fluxapyroxad - Concentrate: 5.2%, Dilution: 9.5% (0.234 g a.s./L)
Prothioconazole - Concentrate: 25%, Dilution: 70% (default values)
Prothioconazole-desthio - Concentrate: 12%, Dilution: 13% (0.375 g a.s./L)

Additional estimation of worker exposure taking into account a conversion factor of 50% of prothioconazole to prothioconazole -desthio are presented below:

Table 6.6-7: Estimated worker exposure (longer term exposure) – 50% conversion rate

		Fluxapyroxad			
Model data	**Level of PPE**	**Total absorbed dose (mg/kg bw/day)**		**% of systemic AOEL**	
Crop inspection in cereals Outdoor Work rate: 2 hours/day, DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable					
Number of applications and application rate		1 x 0.09375 kg a.s./ha			
Body weight: 60 kg	Work wear (arms, body and legs covered) TC: 1400 cm²/person/h	0.0012		3.12	
		Prothioconazole		**Prothioconazole-desthio**	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.094 kg a.s./ha		1 x 0.085 kg a.s./ha	
Body weight: 60 kg	Work wear (arms, body and legs covered) TC: 1400 cm²/person/h	0.0092	4.6	0.0027	27.4

Dermal absorption:
Fluxapyroxad - Concentrate: 5.2%, Dilution: 9.5% (0.234 g a.s./L)
Prothioconazole - Concentrate: 25%, Dilution: 70% (default values)
Prothioconazole-desthio - Concentrate: 12%, Dilution: 23% (0.213 g a.s./L *pro rata* extrapolated)

6.6.3.2 Refinement of generic DFR value (KCP 7.2)

Since the worker exposure estimations carried out indicated that the acceptable operator exposure level (AOEL) will not be exceeded under conditions of intended uses and considering work wear, a study to provide measurements of dislodgeable foliar residues for fluxapyroxad, prothioconazole and prothioconazole-desthio was not necessary and was therefore not performed.

6.6.3.3 Measurement of worker exposure

Since the worker exposure estimations carried out indicated that the acceptable operator exposure levels (AOELs) will not be exceeded under conditions of intended uses and considering work wear, a study to provide measurements of worker exposure was not necessary and was therefore not performed.

6.6.4 Resident and bystander exposure (KCP 7.2.2)

6.6.4.1 Estimation of resident and bystander exposure

No bystander risk assessment is required for PPPs that do not have significant acute toxicity or the potential to exert toxic effects after a single exposure. Exposure in this case will be determined by average exposure over a longer duration, and higher exposures on one day will tend to be offset by lower exposures on other days. Therefore, exposure assessment for residents also covers bystander exposure.

Table 6.6-8 shows the exposure model used for estimation of resident and bystander exposure to fluxapyroxad, prothioconazole and prothioconazole-desthio. The outcome of the estimation is presented in Table 6.6-9 (longer term resident exposure). Detailed calculations are in Appendix 2.

Table 6.6-8: Exposure models for intended uses

Critical use	Winter wheat (max. 1 x 1.25 L product/ha)
Model	Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products; EFSA Journal 2014;12(10):3874 calculator version: 30/03/2015

Table 6.6-9: Estimated resident exposure (longer term exposure)

		Fluxapyroxad	
Model data		**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to winter wheat – maximum water volume 400 L/ha Buffer zone: 2-3 (m) Drift reduction technology: No DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable Dermal absorption values: Fluxapyroxad 9.5%, Prothioconazole 70%, Prothioconazole-desthio 13%			
Number of applications and application rate		1 x 0.09375 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	~~0.0006~~ 0.0006	~~1.43~~ 1.51
	Vapour (75th perc.)	~~0.0011~~ 0.0011	~~2.68~~ 2.68
	Deposits (75th perc.)	~~0.0002~~ 0.0002	~~0.44~~ 0.45
	Entry (75th perc.)	~~0.0014~~ 0.0015	~~3.56~~ 3.76
	Sum (mean)	~~0.0026~~ 0.0027	~~6.62~~ 6.83
Resident adult Body weight: 60 kg	Drift (75th perc.)	~~0.0001~~ 0.0001	~~0.34~~ 0.36
	Vapour (75th perc.)	~~0.0002~~ 0.0002	~~0.58~~ 0.58
	Deposits (75th perc.)	~~0.0001~~ 0.0001	~~0.14~~ 0.15
	Entry (75th perc.)	~~0.0008~~ 0.0008	~~1.98~~ 2.09
	Sum (mean)	~~0.0010~~ 0.0010	~~2.42~~ 2.52

		Prothioconazole		**Prothioconazole-desthio**	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.1875 kg a.s./ha		1 x 0.17 kg a.s./ha in a worst-case approach	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0088	4.40	0.0015	14.91
	Vapour (75th perc.)	0.0011	0.54	0.0011	10.70
	Deposits (75th perc.)	0.0021	1.03	0.0005	4.60
	Entry (75th perc.)	0.0221	11.07	0.0037	37.29
	Sum (mean)	0.0251	12.55	0.0052	52.03
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0021	1.05	0.0004	3.56
	Vapour (75th perc.)	0.0002	0.12	0.0002	2.30
	Deposits (75th perc.)	0.0009	0.45	0.0002	1.51
	Entry (75th perc.)	0.0123	6.15	0.0021	20.72
	Sum (mean)	0.0117	5.85	0.0022	21.61

		Fluxapyroxad	
Model data		**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to winter wheat – minimum water volume 125 L/ha Buffer zone: 2-3 (m) Drift reduction technology: No DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable Dermal absorption values: Fluxapyroxad 9.5%, Prothioconazole 70%, Prothioconazole-desthio 12%			
Number of applications and application rate		1 x 0.09375 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	~~0.0018~~ 0.0019	~~4.57~~ 4.82
	Vapour (75th perc.)	~~0.0011~~ 0.0011	~~2.68~~ 2.68
	Deposits (75th perc.)	~~0.0002~~ 0.0002	~~0.44~~ 0.45
	Entry (75th perc.)	~~0.0014~~ 0.0015	~~3.56~~ 3.76
	Sum (mean)	~~0.0033~~ 0.0035	~~8.36~~ 8.66
Resident adult Body weight: 60 kg	Drift (75th perc.)	~~0.0004~~ 0.0005	~~1.09~~ 1.15
	Vapour (75th perc.)	~~0.0002~~ 0.0002	~~0.58~~ 0.58
	Deposits (75th perc.)	~~0.0001~~ 0.0001	~~0.14~~ 0.15
	Entry (75th perc.)	~~0.0008~~ 0.0008	~~1.98~~ 2.09
	Sum (mean)	~~0.0011~~ 0.0012	~~2.77~~ 2.90

		Prothioconazole		Prothioconazole-desthio	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.1875 kg a.s./ha		1 x 0.17 kg a.s./ha in a worst-case approach	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0282	14.09	0.0044	44.06
	Vapour (75th perc.)	0.0011	0.54	0.0011	10.70
	Deposits (75th perc.)	0.0021	1.03	0.0004	4.35
	Entry (75th perc.)	0.0221	11.07	0.0034	34.43
	Sum (mean)	0.0358	17.88	0.0066	65.65
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0067	3.37	0.0011	10.51
	Vapour (75th perc.)	0.0002	0.12	0.0002	2.30
	Deposits (75th perc.)	0.0009	0.45	0.0001	1.39
	Entry (75th perc.)	0.0123	6.15	0.0019	19.13
	Sum (mean)	0.0139	6.95	0.0024	23.56

Additional estimation of resident and bystander exposure taking into account a conversion factor of 50% of prothioconazole to prothioconazole-desthio are presented below.

With a 50% conversion rate the concentration of prothioconazole-desthio in the spray dilution when applying 1.25 L product/ha in 400 L/ha would be 0.213 g/L. This value is less concentrated than the concentrations tested in the dermal absorption study and therefore a dermal absorption value has been extrapolated from the lowest concentration tested (13% for 0.375 g/L). For the maximum water volume a dermal absorption value of 23% has therefore been extrapolated *pro-rata*. For 1.25 L product/ha in 125 L/ha wa-

ter the concentration of prothioconazole-desthio is 0.68 g/L and therefore a value of 12% from the dermal absorption study is applicable.

Table 6.6-10: Estimated resident exposure (longer term exposure) – 50% conversion rate

		Fluxapyroxad	
Model data		**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to winter wheat – maximum water volume 400 L/ha Buffer zone: 2-3 (m) Drift reduction technology: No DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable Dermal absorption values: Fluxapyroxad 9.5%, Prothioconazole 70%, Prothioconazole-desthio 23%			
Number of applications and application rate		1 x 0.09375 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0006	1.51
	Vapour (75th perc.)	0.0011	2.68
	Deposits (75th perc.)	0.0002	0.45
	Entry (75th perc.)	0.0015	3.76
	Sum (mean)	0.0027	6.83
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0001	0.36
	Vapour (75th perc.)	0.0002	0.58
	Deposits (75th perc.)	0.0001	0.15
	Entry (75th perc.)	0.0008	2.09
	Sum (mean)	0.0010	2.52

		Prothioconazole		**Prothioconazole-desthio**	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.094 kg a.s./ha		1 x 0.085 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0044	2.21	0.0013	13.2
	Vapour (75th perc.)	0.0011	0.54	0.0011	10.7
	Deposits (75th perc.)	0.0010	0.52	0.0004	3.54
	Entry (75th perc.)	0.0111	5.55	0.0033	33.0
	Sum (mean)	0.0131	6.56	0.0047	46.8
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0011	0.53	0.0003	3.14
	Vapour (75th perc.)	0.0002	0.12	0.0002	2.30
	Deposits (75th perc.)	0.0004	0.22	0.0001	1.33
	Entry (75th perc.)	0.0062	3.08	0.0018	18.3
	Sum (mean)	0.0060	2.99	0.0019	19.4

		Fluxapyroxad	
Model data		**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Tractor mounted boom spray application outdoors to winter wheat – minimum water volume 125 L/ha			

Buffer zone: 2-3 (m) Drift reduction technology: No DT_{50}: 30 days DFR: 3 µg/cm²/kg a.s./ha Interval between treatments: Not applicable Dermal absorption values: Fluxapyroxad 9.5%, Prothioconazole 70%, Prothioconazole-desthio 12%			
Number of applications and application rate		1 x 0.09375 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0019	4.82
	Vapour (75th perc.)	0.0011	2.68
	Deposits (75th perc.)	0.0002	0.45
	Entry (75th perc.)	0.0015	3.76
	Sum (mean)	0.0035	8.66
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0005	1.15
	Vapour (75th perc.)	0.0002	0.58
	Deposits (75th perc.)	0.0001	0.15
	Entry (75th perc.)	0.0008	2.09
	Sum (mean)	0.0012	2.90

		Prothioconazole		**Prothioconazole-desthio**	
		Total absorbed dose (mg/kg bw/day)	**% of systemic AOEL**	**Total absorbed dose (mg/kg bw/day)**	**% of systemic AOEL**
Number of applications and application rate		1 x 0.094 kg a.s./ha		1 x 0.085 kg a.s./ha	
Resident child Body weight: 10 kg	Drift (75th perc.)	0.0141	7.07	0.0022	22.0
	Vapour (75th perc.)	0.0011	0.54	0.0011	10.7
	Deposits (75th perc.)	0.0010	0.52	0.0002	2.18
	Entry (75th perc.)	0.0111	5.55	0.0017	17.2
	Sum (mean)	0.0185	9.23	0.0038	38.2
Resident adult Body weight: 60 kg	Drift (75th perc.)	0.0034	1.69	0.0005	5.25
	Vapour (75th perc.)	0.0002	0.12	0.0002	2.30
	Deposits (75th perc.)	0.0004	0.22	0.0001	0.69
	Entry (75th perc.)	0.0062	3.08	0.0010	9.56
	Sum (mean)	0.0071	3.54	0.0013	12.9

6.6.4.2 Measurement of resident and/or bystander exposure

Since the resident and/or bystander exposure estimations carried out indicated that the acceptable operator exposure level (AOEL) for fluxapyroxad, prothioconazole and prothioconazole-desthio will not be exceeded under conditions of intended uses and considering above mentioned risk mitigation measures, a study to provide measurements of resident/bystander exposure was not necessary and was therefore not performed.

6.6.5 Combined exposure

The product is a mixture of two active substances. The metabolite prothioconazole-desthio is also included in the risk assessment for combined exposure.

6.6.5.1 Exposure assessment of fluxapyroxad, prothiconazole and the metabolite prothioconazole-desthio in ADM.03503.F.1.A

Note: The combined toxicological effect of these active substances has not been investigated with regard to repeated dose toxicity.
At the first tier, combined exposure is calculated as the sum of the component exposures without regard to the mode of action or mechanism/target of toxicity. First tier estimates of exposure to ADM.03503.F.1.A assume as a theoretical worst case that there is 100% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. For this conversion 1 kg prothioconazole yields 0.907 kg prothioconazole-desthio. This conversion can occur during the drying process on clothing and skin so for spray operators the exposure assessment only considers exposure to prothioconazole-desthio from application of the spray solution. Parent prothioconazole is only considered for combined risk assessment for operators mixing and loading. For workers and resident exposure, the combined risk assessment considers exposure to fluxapyroxad and the metabolite prothioconazole-desthio as this combination gives the highest estimates for combined exposure.
Exposure estimates to ADM.03503.F.1.A have also been provided assuming that there is 50% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. This scenario is considered more realistic than 100% conversion. The combined exposure assessment considers exposure to fluxapyroxad and both parent prothioconazole and to the metabolite prothioconazole-desthio.
Initially, the individual Hazard Quotients (HQ) are calculated for all active substances in the PPP by assessing the exposure according to appropriate models and dividing the individual exposure levels by the respective systemic AOEL. This is equivalent to the predicted exposure as % of systemic AOEL from Table 6.6-3 converted to decimal. The Hazard Index (HI) is the sum of the individual HQs.

Table 6.6-11: Risk assessment from combined exposure 100% conversion of parent prothioconazole to prothioconazole-desthio

Application scenario	Active ingredient	Estimated exposure / AOEL (HQ)
Operators – Field crop (boom) sprayer application in cereals	Fluxapyroxad	0.040
	Prothioconazole (mixing and loading)	~~0.017~~ 0.449
	Prothioconazole-desthio (application)	0.290
	Cumulative risk operators (HI)	~~0.347~~ **0.489**
Workers – Crop Inspection	Fluxapyroxad	0.030
	Prothioconazole-desthio	0.309
	Cumulative risk workers (HI)	**0.339**
Resident – child (application in minimum water volume (125 L/ha) provides the worst case	**Fluxapyroxad**	
	Drift	~~0.046~~ 0.048
	Vapour	~~0.027~~ 0.027
	Deposits	~~0.004~~ 0.004
	Entry	~~0.036~~ 0.038
	Sum of all pathways	~~0.084~~ 0.087
	Prothioconazole-desthio	
	Drift	0.441
	Vapour	0.107
	Deposits	0.044
	Entry	0.344
	Sum of all pathways	0.657
	Cumulative risk resident – child (HI)	
	Drift	~~0.486~~ 0.489

Application scenario	Active ingredient	Estimated exposure / AOEL (HQ)
	Vapour	~~0.134~~ 0.134
	Deposits	~~0.048~~ 0.048
	Entry	~~0.380~~ 0.382
	Sum of all pathways	~~**0.740**~~ **0.744**
Resident - adult	**Fluxapyroxad**	
	Drift	~~0.011~~ 0.012
	Vapour	~~0.006~~ 0.006
	Deposits	~~0.001~~ 0.002
	Entry	~~0.020~~ 0.021
	Sum of all pathways	~~0.028~~ 0.029
	Prothioconazole-desthio	
	Drift	0.105
	Vapour	0.023
	Deposits	0.014
	Entry	0.191
	Sum of all pathways	0.236
	Cumulative risk resident – adult (HI)	
	Drift	~~0.116~~ 0.117
	Vapour	~~0.029~~ 0.029
	Deposits	~~0.015~~ 0.016
	Entry	~~0.211~~ 0.212
	Sum of all pathways	~~**0.263**~~ 0.265

Table 6.6-9: Risk assessment from combined exposure assuming 50% conversion of parent prothioconazole to prothioconazole-desthio

Application scenario	Active ingredient	Estimated exposure / AOEL (HQ)
Operators – Field crop (boom) sprayer application in cereals Work wear (arms, body and legs covered) + gloves M/L and work wear (arms, body and legs covered) A	Fluxapyroxad	0.040
	Prothioconazole (mixing/loading and application)	0.052
	Prothioconazole-desthio	0.340
	Cumulative risk operators (HI)	**0.432**
Workers – Crop Inspection	Fluxapyroxad	0.030
	Prothioconazole	0.046
	Prothioconazole-desthio	0.274
	Cumulative risk workers (HI)	**0.350**
Resident – child (application in minimum water volume (125 L/ha) provides the worst case	**Fluxapyroxad**	
	Drift	0.048
	Vapour	0.027
	Deposits	0.005
	Entry	0.038
	Sum of all pathways	0.087
	Prothioconazole	

Application scenario	Active ingredient	Estimated exposure / AOEL (HQ)
	Drift	0.071
	Vapour	0.005
	Deposits	0.005
	Entry	0.056
	Sum of all pathways	0.092
	Prothioconazole-desthio	
	Drift	0.220
	Vapour	0.107
	Deposits	0.022
	Entry	0.172
	Sum of all pathways	0.382
	Cumulative risk resident – child (HI)	
	Drift	0.339
	Vapour	0.139
	Deposits	0.032
	Entry	0.266
	Sum of all pathways	0.561
Resident - adult	**Fluxapyroxad**	
	Drift	0.012
	Vapour	0.006
	Deposits	0.002
	Entry	0.021
	Sum of all pathways	0.029
	Prothioconazole	
	Drift	0.017
	Vapour	0.001
	Deposits	0.002
	Entry	0.031
	Sum of all pathways	0.035
	Prothioconazole-desthio	
	Drift	0.053
	Vapour	0.023
	Deposits	0.007
	Entry	0.096
	Sum of all pathways	0.129
	Cumulative risk resident – adult (HI)	
	Drift	0.082
	Vapour	0.03
	Deposits	0.011
	Entry	0.148
	Sum of all pathways	0.193

The Hazard Index is < 1 for all scenarios. Thus, combined exposure to all active substances in ADM.03503.F.1.A is not expected to present a risk for operators, workers, residents and bystanders. No further refinement of the assessment is required.

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 7.1.1/01	[redacted]	2021a	Acute oral toxicity study of ADM.03503.F.1.A (Fluxapyroxad 75 prothioconazole 150 g/L EC) in rats (acute toxic class method) [redacted] GLP Unpublished	Y	ADM
KCP 7.1.2/01	[redacted]	2021b	Acute dermal toxicity study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rats (fixed dose preocedure) [redacted] GLP Unpublished	N	ADM
KCP 7.1.3/01	[redacted]	2021a	Acute inhalation toxicity study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rats (acute toxic class method) [redacted] GLP Unpublished	Y	ADM
KCP 7.1.4/01	[redacted]	2021a	In vitro skin corrosion test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) using reconstructed human epidermis tissues (RhE) [redacted] GLP Unpublished	N	ADM
KCP 7.1.4/02	[redacted]	2021b	In vitro skin irritation test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) using reconstructed human epidermis tissues (RhE) [redacted] GLP Unpublished	N	ADM
KCP 7.1.4/03	[redacted]	2021c	Acute dermal irritation study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rabbits [redacted]	Y	ADM

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 7.1.5/01	[redacted]	2021b	In vitro eye irritation test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) using bovine corneal opacity and permeability test [redacted] GLP Unpublished	N	ADM
KCP 7.1.5/02	[redacted]	2021d	Acute eye irritation study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rabbits [redacted] GLP Unpublished	Y	ADM
KCP 7.1.6/01	[redacted]	2021c	In vitro skin sensitisation study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC): keratinocyte based ARE-NRF2 luciferase reporter gene test [redacted] GLP Unpublished	N	ADM
KCP 7.1.6/02	[redacted]	2021d	In chemico skin sensitisation of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC): Direct peptide reactivity assay (DPRA) [redacted] GLP Unpublished	N	ADM
KCP 7.1.6/03	[redacted]	2021c	Skin sensitisation study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) by local lymph node assay in mice [redacted] GLP Unpublished	Y	ADM
KCP 7.3/01	[redacted]	2021a	The In Vitro Percutaneous Absorption of Radiolabelled Fluxapyroxad in a Concentrate and Two In-Use Dilutions of the Fluxapyroxad 75 g/L + Prothioconazole 150 g/L EC Formulation (ADM.03503.F.1.A) Through Human Split-Thickness Skin	N	ADM

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**
			[redacted] GLP Unpublished		
KCP 7.3/02	[redacted]	2021b	The In Vitro Percutaneous Absorption of Radiolabelled Prothioconazole-desthio in Two In-Use Dilutions of the Fluxapyroxad 75 g/L + Prothioconazole 150 g/L EC Formulation (ADM.03503.F.1.A) Through Human Split-Thickness Skin [redacted] GLP Unpublished	N	ADM
KCP 7.4/01	Anonymous	2022	Safety Data Sheet – ADM.03503.F.1.A ADAMA Makhteshim Ltd Not GLP / GEP Unpublished	N	N/A

ADM = Property of ADAMA Agricultural Solutions and all affiliates.
Under Article 59 of Regulation 1107/2009/EC, the Sponsor Company claims data protection for all ADM studies.

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**
-	-	-	-	-	-

List of data submitted by the applicant and not relied on

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

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

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

Appendix 1 Detailed evaluation of the studies relied upon

A 1.1 Statement on bridging possibilities

Comments of zRMS:	Bridging is not applicable. Hazard classification via the application of bridging principles is not possible since data on a similar mixture are not available.

Bridging is not necessary since the studies submitted have been conducted using ADM 03503.F.1.A.

For the calculation method further details are provided in Part C, Confidential information.

A 1.2 Acute oral toxicity (KCP 7.1.1)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. Thus, in a first approach, classification based on the complete composition of the plant protection product is taken into account for classification purpose.
In addition, a vertebrate study is available for acute toxicity of ADM.03503.F.1.A via the oral route. This study was not performed with intention for use within the EU, but it was performed to satisfy the regulatory requirements of countries outside of the EU. For transparency reasons, the study is provided with this dossier and summarised hereafter.
Although, using the calculation method based on the ingredients of the formulation a classification of acute tox 4 is required, this classification is not required based on the acute oral toxicity study in the rat (A 1.2.2). Taking all available information into consideration no classification is required according to Regulation (EC) No. 1272/2008.

A 1.2.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Acute oral toxicity has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.1.3.6. Since the composition of the plant protection product is confidential, this approach is presented in the confidential part C of this dossier.

Using the calculation method, the acute toxicity estimate of the mixture is 978.44 mg/kg bw. Thus, classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as Acute Tox. 4 H302.

A 1.2.2 Study 2

Comments of zRMS:	~~From the scientific point of view study Acute Oral Toxicity Study Disha Chande, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection has not been taken into account (please refer ZRMS consideration in the preface to this dRR reflecting cMS comments). Study ([redacted]) has been reviewed for compliance with the current guidelines resulting from scientific progress (OECD 423 rev 2022). Study ([redacted]) implements 3R rules minimizing the number of animals required to estimate the acute oral toxicity of a chemical. No deviation has been noted. Results of the study and conclusions are adequate for risk assessment and classification purpose.

Reference KCP 7.1.1/01

Report author:	[redacted]
Report year ;	2021
Report title:	Acute Oral Toxicity Study Of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 G/L EC) in Rats
Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 423 (2001)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: In an acute oral toxicity study, a set of fasted rat [RccHan:WIST], 09-11 weeks old) (3 females) were given a single oral dose of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) using water as vehicle at a dose of 2000 mg/kg body weight (for set I and set II) and observed for 14 days.

There was no mortality, clinical signs (lethargy along with a decrease in body weight) were observed in one of the six rats treated.

A decrease in body weight was observed in one of the six rats treated at 2000 mg/kg body weight.

The acute oral median lethal dose of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rat was found to be greater than 2000 mg/kg body weight.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Unclassified**

Under the experimental conditions, the oral LD_{50} of ADM.03503.F.1.A is higher than 2000 mg/kg bw in rats. Thus, **no classification is required according to Regulation (EC) No. 1272/2008.**

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid
	Lot/Batch #:	1162-230719-011
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)

	Date of Expiry	05 September 2021

2. Vehicle and/or Positive Control: The test item was liquid, and a suitable vehicle was selected following a solubility assessment. The test item formed homogeneous suspension in RO water. Therefore a 200 mg/mL dose formulation of the test item was prepared by mixing the required quantity of the test item in RO water.

3.	**Test Animals:**		
	Species:	Rat	
	Strain:	RccHan:WIST	
	Age/Weight at Dosing:	9 to 11 weeks, Weight (g) Minimum: 179.3, Maximum: 198.5 g	
	Source:	Animal Breeding Facility, Jai Research Foundation	
	Housing:	Three rats/cage	
	Feed:	Teklad certified Global High Fiber Rat and Mice Feed manufactured by Harlan, U.S.A. *ad libitum* with the exception of overnight fasting, prior to dosing, until three hours post-dosing.	
	Water:	UV sterilized water filtered through Kent Reverse Osmosis water filtration system was provided *ad libitum*	
	Environmental Conditions:	**Temperature:** **Humidity:** **Air Changes:** **Photoperiod:**	19 to 23 °C 56 to 66% Minimum 15 air changes/h 12 h dark/ 12 h light
	Acclimation Period:	6 to 10 days	

B. STUDY DESIGN AND METHODS

1. In-life Dates –
Start: 02 April 2021 End: 26 April 2021

2. Animal Assignment and Treatment - Following overnight fasting, prior to dosing, rat were given a single dose of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) by gavage, then observed daily and weighed on day 1, 3, 7 and on 14 days. All the rats were sacrificed and a necropsy was performed in all animals. A total of six rats was used.

3. Statistics – : Not Applicable

II. RESULTS AND DISCUSSION

A. Mortality – Mortality data are presented in Text Table 1.

Text Table 1. Dose, Mortality/Animals Treated

Dose (mg/kg body weight)	Female Rat (mortality/total)
2000	**0/6**

No mortality was observed in female rat treated with 2000 mg ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)/kg body weight.

B. Clinical Observations – clinical signs (lethargy) was observed in one of the six rats treated at the dose level of 2000 mg/kg body weight.

C. Body Weight - Decrease in body weight was observed in one of the six rats treated at 2000 mg/kg body weight with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC).

D. Necropsy

External

External examination of terminally sacrificed female rat did not reveal any abnormality of pathological significance.

Internal
Visceral examination of female rat sacrificed at termination did not reveal any abnormal lesions.

E. Conclusion:

The acute oral median lethal dose of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in female rat was found to be greater than 2000 mg/kg body weight, according to OECD 423.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Unclassified**

Under the experimental conditions, the oral LD_{50} of ADM.03503.F.1.A is higher than 2000 mg/kg bw in rats. Thus, **no classification is required according to Regulation (EC) No. 1272/2008.**

A 1.3 Acute percutaneous (dermal) toxicity (KCP 7.1.2)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. Thus, in a first approach, classification based on the complete composition of the plant protection product is taken into account for classification purpose.
In addition, a waiver report is available for acute toxicity of ADM.03503.F.1.A via the dermal route. This study was not performed with intention for use within the EU, but it was performed to satisfy the regulatory requirements of countries outside of the EU. For transparency reasons, the waiver report is provided with this dossier and summarised hereafter.
Based on both the calculation method and the waiver report for the acute dermal toxicity study no classification is required according to Regulation (EC) No. 1272/2008.

A 1.3.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Acute dermal toxicity has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.1.3.6.

No ingredients in ADM.03503.F.1.A are classified for acute dermal toxicity. Thus, no classification is required according to Regulation (EC) No. 1272/2008.

A 1.3.2 Study 2

Comments of zRMS:	~~From the scientific point of view study Disha Chande, 2021 Acute dermal toxicity study of ADM.03503.F.1.A is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary~~ zRMS agree with applicant regarding acute dermal study waiver. The test item, ADM.03503.F.1.A was found to have an acute oral median lethal dose in Wistar rats of greater than 2000 mg/kg body weight therefore meets the waiver criteria (Guidance Document on Waiving or Bridging of Mammalian Acute Toxicity Tests (OECD, 2016), point number 16 states that "a dermal toxicity study may be waived if the test item has shown no adverse effects in an acute oral toxicity test up to 2000 mg/kg body weight). In accordance

	with the mentioned Guidance and on the basis of weight of evidence, it is scientifically justify that the use of animals in the Acute Dermal Toxicity Study of ADM.03503.F.1.A in Rats is not required and can to be waived in this instance. Conclusions are adequate for risk assessment and classification purpose.

Reference KCP 7.1.2/01

Report author:	[redacted]
Report year ;	2021
Report title:	Acute dermal toxicity study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in rats (fixed dose procedure),
Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 402 (2017)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	No

Summary

The acute dermal toxicity of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was investigated in a study compliant with OECD 402 (2017) test guideline criteria. OECD 402 (2017) requires the application of a stepwise process, namely the start of the dermal study only upon the results from an oral acute toxicity study. The approach in OECD 402 (2017) is based on a re-evaluation of the additional information on toxic properties that can be gained from an acute dermal toxicity study, when acute oral toxicity data is already available, with respect to animal welfare considerations.

According to the Guidance Document on Waiving or Bridging of Mammalian Acute Toxicity Tests (OECD, 2016), point number 16 states that "a dermal toxicity study may be waived if the test item has shown no adverse effects in an acute oral toxicity test up to 2000 mg/kg body weight. Reviews comparing the classification of oral and dermal hazards indicate that it is rare for the dermal test to yield a more severe classification (Thomas and Dewhurst, 2007; Creton *et al.*, 2010; Seidle *et al.*, 2011, Moore *et al.*, 2013)". Novel data support this assessment (van der Kamp and Elliot, 2021) and this approach is supported by the US Environmental Protection Agency (EPA 705-G-2020-3722, 2020).

The test item, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was found to have an acute oral median lethal dose in Wistar rats of greater than 2000 mg/kg body weight (ADAMA Reference: 000107565) and so meets the waiver criteria detailed above. Therefore, in accordance with the criterion and on the basis of weight of evidence, it is scientifically mandated that the use of animals in the Acute Dermal Toxicity Study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in Rats is not required and can to be waived in this instance.

The acute dermal study waiver proposal is further supported by the weight of evidence available for the active substances present in the test material: The conclusion of the EFSA Journal, 2012, of the pesticide risk assessment of the active substance Fluxapyroxad concludes low acute toxicity has been observed when fluxapyroxad was administered by the oral, dermal or inhalation routes. The active substance fluxapyroxad did not produce skin or eye irritation, and did not have a potential for skin sensitisation.

The conclusion of the EFSA Journal, 2007, of the pesticide risk assessment of the active substance Prothioconazole concludes low acute toxicity *via* oral (LD_{50} > 6200 mg/kg body weight), dermal (LD_{50} > 2000 mg/kg body weight) or inhalation (LC_{50} > 4990 mg/m^3) routes. It does not show any eye and skin irritation or sensitizing potential.

According to OECD 402 (2017) and based on the considerations above, the acute dermal lethal median dose (LD_{50}) of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is greater than 2000 mg/kg body weight.

Conclusion

Under the experimental conditions, the dermal LD_{50} of ADM.03503.F.1.A is higher than 2000 mg/kg bw. Thus, **no classification is required according to Regulation (EC) No. 1272/2008**.

A 1.4 Acute inhalation toxicity (KCP 7.1.3)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. Thus, in a first approach, classification based on the complete composition of the plant protection product is taken into account for classification purpose.
In addition, a vertebrate study is available for acute toxicity of ADM.03503.F.1.A via the inhalation route. This study was not performed with intention for use within the EU, but it was performed to satisfy the regulatory requirements of countries outside of the EU. For transparency reasons, the study is provided with this dossier and summarised hereafter.
Based on both the calculation method and the acute inhalation study in the rat no classification is required according to Regulation (EC) No. 1272/2008.

A 1.4.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Acute inhalation toxicity has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.1.3.6.

No ingredients in ADM.03503.F.1.A are classified for acute inhalation toxicity. Thus, no classification is required according to Regulation (EC) No. 1272/2008.

A 1.4.2 Study 2

Comments of zRMS:	~~From the scientific point of view study Ramesh Verma, 2021 Acute Inhalation Toxicity Study Of ADM.03503.F.1.A is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary.~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection has not been taken into account (please refer ZRMS consideration in the preface to this dRR reflecting cMS comments). Results of the study and conclusions are adequate for risk assessment and classification purpose. Study accepted.

Reference KCP 7.1.3/01

Report author:	[redacted]
Report year ;	2021
Report title:	Acute Inhalation Toxicity Study Of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 G/L EC) in Rats
Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 436 (2009)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: In an acute inhalation toxicity study, a group of 3 male and 3 female Wistar rats (9 to 11 weeks old) were exposed to the aerosol of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in a dynamic, flow-past, nose-only inhalation exposure system. The test item aerosol was generated using a nebuliser, and the actual concentration was measured gravimetrically from the breathing zone. Rats were exposed for 4 h followed by a 14 day post-exposure observation period.

The test atmosphere was generated at an acceptable concentration (5.28 mg/L, measured gravimetrically) and the particle size (the mass median aerodynamic diameter (MMAD) was 2.96 µm with an average geometric standard deviation (GSD) of 1.63).

One female and one male rat were found dead on day 1 and day 2 post-exposure, respectively, exposed to 5.28 mg ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)/L air.

Clinical signs of lethargy, abdominal breathing and gasping were observed in rats exposed to 5.28 mg ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)/L air.

A decrease in the body weight was observed on day 1, 3 and 7, which recovered by day 14 in two surviving rats (one male and one female rat) when compared with their day 0 body weight. In the case of the other two surviving rats (one male and one female rat), a decrease in the body weight was observed on day 1, 3, 7, and 14, when compared with day 0 body weight, but recovery was observed on day 7 and 14 when compared to their day 3 body weight.

An external examination of the found-dead and terminally sacrificed rats did not reveal any abnormality. An internal examination of the found-dead rats revealed lungs: reddish discolouration, whereas the other found dead and terminally sacrificed rats did not reveal any abnormality.

The results are summarised below:

Nominal conc. (mg/L)	Actual Conc. (mg/L)	MMAD µm	GSD	Mortality (number dead / total)		
				Males	Females	Combined
9.22	5.28	2.96	1.63	1/3	1/3	2/6

The 4 h acute inhalation median lethal concentration of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), in male and female Wistar rats, was greater than the actual (gravimetric) concentration of 5.28 mg/L air, and the LC_{50} cut-off value was 12.5 mg/L air.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Category 5**

Under the experimental conditions, the inhalation LC_{50} of ADM.03503.F.1.A is higher than 5.28 mg/L air in rats. Thus, **no classification is required according to Regulation (EC) No. 1272/2008**.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid
	Lot/Batch #:	1162-230719-011
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
	Expiry date	05 September 2021

2. Vehicle and/or Positive Control: The test item was a liquid end-use product and was tested undiluted

(at a constant concentration).

3.	**Test Animals:**		
	Species:	Rat	
	Strain:	RccHan:WIST	
	Age at Dosing:	9 to 11 weeks,	
	Weight at Dosing (g):	Male: Minimum: 275.3, Maximum: 279.4 Female: Minimum: 197.4, Maximum: 204.2	
	Source:	Animal Breeding Facility, Jai Research Foundation	
	Housing:	three rats/cage	
	Feed:	Teklad certified Global High Fiber Rat and Mice Feed manufactured by Harlan, U.S.A. *ad libitum* with the exception of overnight fasting, prior to dosing, until three hours post-dosing.	
	Water:	UV sterilized water filtered through Kent Reverse Osmosis water filtration system was provided *ad libitum*	
	Environmental Conditions:	**Temperature:** **Humidity:** **Air Changes:** **Photoperiod:**	19 to 23 °C 56 to 65% Minimum 15 air changes/h 12 h dark/ 12 h light
	Acclimation Period:	7 days	

B. STUDY DESIGN AND METHODS

1. In-life Dates –

Start: 06 April 2021 End: 27 April 2021

2. Exposure conditions:

Generation of the test atmosphere/chamber description: The inhalation chamber has 2 main parts namely outer plenum and inner plenum. Inner plenum volume is 862 cm^3 and outer plenum volume is 1,200 cm^3 so the total volume = 2062 cm^3 x 2 = 4.124 litres. The exposure unit (outer plenum) is made of stainless steel with 12 portholes to accommodate transparent perspex rat exposure tubes.

The inhalation equipment is designed to sustain a dynamic airflow that ensures an adequate air exchange of at least 2-3 times the respiratory minute volume of rats (i.e., between 0.5 to 1.0 L/min per exposure port), an adequate oxygen content of at least 19% and exposure atmosphere CO_2 levels of less than 1%. The exposure chamber was operated at a slight positive pressure so that the inlet air flow rate was slightly more than the exhaust flow rate. Aerosol not breathed and expired air passed into the outlet unit, connected to a suction pump, and left the chamber via an impinger containing 1.0% sodium hydroxide solutions and moisture traps.

Test atmosphere concentration: The acute toxic class method uses serial steps of fixed concentrations (0.05, 0.5, 1 or 5 mg/L) to provide a ranking of test article toxicity. Absence or presence of test item-related mortality of the animals dosed at one step will determine the next step, as per Annex 3d, OECD 436. The initial target exposure concentration was selected by the Sponsor as 5 mg/L air (or the maximum attainable concentration) based on data from similar formulations of the test material.

Particle size determination: Particle size analysis was carried out to determine the consistency and stability of particle size distribution twice during the 4-hour exposure period using Intox Seven Stage cascade impactor. Samples were drawn through the cascade impactor at a flow rate of 0.74 L/min for 1 minute. The stages were weighed pre and then post sampling to determine the

total mass on each stage. The increase in the weight of each collection plate is the mass of particles in the size range of that impact stage. The total mass of particles and the percent mass of particles in each size range is calculated. The Mass Median Aerodynamic Diameter (MMAD) and Geometric Standard Deviation (GSD) was calculated from the % particle size distribution (Figure 3).

Animal Assignment and Treatment - The rats were exposed for 4 h (nose only) followed by a 14-day post exposure observation period during which body weight and clinical observations were recorded. Survivors were sacrificed and a necropsy was performed in all animals.

Animals were observed daily, and body weights were recorded prior to exposure on day 0 and on days 1, 3, 7 and 14 after exposure. Survivors were sacrificed and a necropsy was performed in all animals.

3. Statistics – For the Inhalation LC_{50}: Not Applicable as limit study

II. RESULTS AND DISCUSSION

A. Atmosphere Analysis

The actual concentration (measured gravimetrically) of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in the exposure chamber was monitored at intervals throughout the 4- hour exposure. The overall mean achieved concentration was 5.28 mg/L air.

Group	Nominal Concentration (mg/L air)	Achieved Gravimetric Concentration (mg/L air)	Mass Median Aerodynamic Diameter MMAD (µM)	Geometric Standard Deviation (GSD)
1	9.22	5.28	2.96	1.63

Chamber conditions during the exposure were as follows:

Group	Chamber temperature (ºC)	Relative Humidity (%)	O_2 Content (%)	CO_2 content (%)
1	20.8 to 21.9	58.3 to 63.1	20.6 to 20.9	0.03

B. Mortality – Mortality data are presented in Text Table 1.

Text Table 1. Dose, Mortality/Animals Treated

Estimated Gravimetric Breathing Zone Concentration (mg/L air)	Total mortality
5.28	**2/6**

C. Clinical Observations Clinical signs of lethargy, abdominal breathing and gasping were observed in rats exposed to 5.28 mg ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)/L air.

D. Body Weight - A decrease in the body weight was observed on day 1, 3 and 7, which recovered by day 14 in two surviving rats (one male and one female rat) when compared with their day 0 body weight. In the case of the other two surviving rats (one male and one female rat), a decrease in the body weight was observed on day 1, 3, 7, and 14, when compared with day 0 body weight, but recovery was observed on day 7 and 14 when compared to their day 3 body weight.

E. Necropsy

External

An external examination of the found-dead and terminally sacrificed rats did not reveal any abnormality.

Internal

An internal examination of the found-dead rats revealed lungs: reddish discolouration (Rat N° 6), whereas the other found dead and terminally sacrificed rats did not reveal any abnormality.

F. Conclusion:

The 4 h acute inhalation median lethal concentration of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), in male and female Wistar rats, was greater than the actual (gravimetric) concentration of 5.28 mg/L air, and the LC_{50} cut-off value was 12.5 mg/L air.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Category 5**

Under the experimental conditions, the inhalation LC_{50} of ADM.03503.F.1.A is higher than 5.28 mg/L air in rats. **Thus, no classification is required according to Regulation (EC) No. 1272/2008.**

A 1.5 Skin irritation (KCP 7.1.4)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. In consideration of the content and the classification of the individual components in the formulation a classification H314 would be required. Since the composition of the plant protection product is confidential, this approach is presented in the confidential part C of this dossier.
Two *in vitro* studies are available, one study investigated *in vitro* skin corrosion (OECD 431). ADM.03503.F.1.A was predicted to be not corrosive in this study. In the *in vitro* skin irritation test (OECD 439) ADM.03503.F.1.A was predicted to be a skin irritant.
However, an *in vivo* skin irritation study according to OECD guideline 404 (2015) was conducted but not performed with intention for use within the EU, but to satisfy the regulatory requirements of countries outside of the EU. Considering the results of the animal study the product does not need to be classified as skin irritant according to CLP Regulation (EC) 1272/2008.
In a weight of evidence approach based on the *in vitro* and *in vivo* data, ADM.03503.F.1.A requires no classification as skin irritant according to Regulation (EC) No. 1272/2008 and subsequent regulations.

A 1.5.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Skin irritation has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.2.3.3.

An ingredient in ADM.03503.F.1.A is classified for Skin Corr. 1B H314. This ingredient is present in the formulation at a concentration >5%.

Thus, classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as Skin Corr. 1B H314.

A 1.5.2 Study 2

Comments of zRMS:	~~From the scientific point of view study In Vitro Skin Corrosion Test of ADM.03503.F.1.A Indrajitsinh M. Barad, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has not been taken into the consideration.~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection or *in vitro* studies has not been taken into account (please refer zRMS consideration in the preface to this dRR reflecting cMS comments).

Reference KCP 7.1.4/01

Report author:	[redacted]
Report year:	2021
Report title:	*In Vitro* Skin Corrosion Test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) using Reconstructed Human Epidermis Tissues (RhE)
Report no:	[redacted]
ADAMA reference number:	[redacted]
Guidelines followed in study:	OECD 431 (2019) and EC Method B40 (2008)
Deviations from current test guideline:	None
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: This study was performed to evaluate the corrosive potential of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), using reconstructed human epidermis (RhE) tissue, as per the United Nations Globally Harmonized System of Classification and Labelling of Chemicals (UN GHS).

Tissues were exposed to ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) (test item) and sterile distilled water (negative control) for 3 minutes at room temperature and 60 minutes at 37 ± 1 °C in 5 ± 1% CO_2 using three replicates/time point. Positive control tissues were exposed for 60 minutes at 37 ± 1 °C in 5 ± 1% CO_2. Killed negative control (3 minutes and 60 minutes), test item (3 minutes and 60 minutes), and positive control (60 minutes) treated tissues were also exposed in the same manner using two replicates to correct the direct MTT reduction obtained by the test item and positive control during the pre-test.

No significant reduction in the percent cell viability was observed after 3-minute and 60-minute exposure in the treated tissues, when compared with that of the concurrent negative control. The Non-specific MTT reduction calculation (NSMTT) for the positive control killed tissues was < 50%. Therefore, the true MTT Metabolic conversion of treated tissue was not determined. The difference between the viability of the treated tissues was less than 6%, i.e., % CV.

Treatment	Viability	
	3 Minutes Exposure	60 Minutes Exposure
Negative control (Sterile distilled water)	100%	100%
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	88.77%	91.95%
Positive control (8N KOH)	-	2.38%

All-Optical Density (OD) values (corrected OD) for negative control replicates were between 1.101 and 1.129, against a guideline requirement of ≥ 0.8 and ≤ 3.0 (the acceptance criteria for SkinEthicTM RhE model). The positive control showed 2.38 % cell viability, against a guideline requirement of <15%, when compared with that of the concurrent negative control, demonstrating the efficiency of the SkinEthicTM RhE model.

All criteria for a valid study were met. From the results of this study, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is concluded to be non-corrosive in the in vitro skin corrosion test, using reconstructed human epidermis (RhE) tissues.

COMPLIANCE: Signed and dated GLP, Quality Assurance and data confidentiality statements are provided.

I. MATERIALS AND METHODS

Experimental start: 27 April 2021, Experimental completion 04 May 2021

A. MATERIALS:

1. Test Item : ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
   Description : Yellowish to brownish, transparent liquid
   Lot/Batch # : 1162-230719-011
   Purity : Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6)
   Prothioconazole: 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
   Storage : Room temperature
   Expiry Date : 05 September 2021

2. **Vehicle and/or Positive Control:**
   Negative control : Sterile distilled water.
   Positive control : Potassium hydroxide (8N KOH)
   Concentration : 8N KOH
   Suggested Retest Date: January 21, 2022
3. **Test System:**
   Reconstructed human epidermis (SkinEthic™ RhE) tissue
   Source: SkinEthic Laboratories, Episkin 4, Rue Alexander Fleming, 69366 Lyon Cedex 07, France

B. STUDY DESIGN AND METHODS

1. Pre-Tests

Prior to the main experiment, preliminary tests, i.e., a colour interference test, direct MTT reduction test, and mesh compatibility were performed to select appropriate adapted controls and use of nylon mesh on the apical surface of each tissue.

1. **Color Interference**: Glass test tube with 0.3 mL Isopropanol+ 40 µL of test item and 40 µL of Isopropanol as negative control was incubated at 37 ± 1 ºC and 5 ± 1% CO_2 in 95% humidified atmosphere for 60 minutes. Absorbance was measured.

2. **MTT Reduction Test** : 6 Well Plate containing 1 mL MTT Solution+ 40 µL of test item and 40 µL of distilled water in 1Ml MTT Solution serve as negative control was incubated at 37 ± 1 ºC and 5 ± 1% CO_2 in 95% humidified atmosphere in dark

3. **Mesh Compatibility Test (Only for Liquid Test item)**: Distilled water or Test item on slide with Mesh and Incubated at Room Temperature

Main Study

Methods:

Well Plate and Media used during Treatment : 24-Well Plate with 0.3 mL maintenance media

Standard condition during treatment: 3-Minutes: at Room Temperature
60-Minutes: at 37 ± 1 ºC and 5 ± 1% CO2 in 95%

	humidified atmosphere. Nylon Mesh applied on liquid materials for uniform spread of material
Rinsing	Rinsed 20 times with constant soft stream of 1 mL DPBS at a 5-8 cm distance from the insert to remove all test item
MTT test and Incubation	24-Well Plate with 0.3 mL MTT Solution at 37 ± 1 ºC and 5 ± 1% CO_2 in 95% humidified atmosphere
Formazan Extraction and Incubation	24-Well Plate with 1.5 mL Isopropanol and incubated at Room temperature overnight in dark
OD Measurement at 570 nm	200 µL/well and three replicates/tissue in 96-well plate

Statistics:

Data from individual tissue replicates i.e., OD values and corrected OD values obtained from software were used and processed further to calculate mean, standard deviation and percentage cell viability, in addition to the evaluation of classification. Data of percent coefficient of variation between tissue replicates were determined for each exposure time.

II. RESULTS AND DISCUSSION

Pre-Tests

1. Colour Interference Test

No significant difference in the absorbance was observed between the negative control (isopropanol) and the test item. Therefore, the results of the colour interference test show that the interference was not observed due to the test item. The results of the colour interference test are provided in the below-mentioned table:

Treatment	Optical Density (nm)	Interaction
Negative Control (isopropanol)	0.044	No
	0.060	
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	0.050	No
	0.053	

Key: No = Colour was not observed.

2. Direct MTT Reduction Test

The test item did produce a colour change when compared with that of the concurrent negative control (distilled water). Since there was evidence of direct MTT reduction (in the absence of tissues), additional adapted controls were included in the main study. Results of the direct MTT reduction test are summarised below:

Treatment	Colour Observed	Interaction
Negative Control (Distilled water)	Yellow	No
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	Blue	Yes

3. Mesh Compatibility Test

No interaction of mesh with the test item or the negative control (distilled water) were observed when examined under a light microscope and the un-aided eye.

Main Study

1. Negative Control

The optical density (OD) values (corrected ODs) of the negative controls in all tissues were between 1.101 and 1.129 (TABLE 1) and met the acceptance range for the prediction model Skin-

Ethic™ RHE (≥ 0.8 and ≤ 3.0), as per the OECD test guideline 431. The results of the negative control are summarised below:

Exposure time	Mean OD (3 tissues)	Mean % viability	SD	%CV
3 minutes	1.119	100	0.00	0.00
60 minutes	1.105	100	0.00	0.00

Note: For Negative control of 3 minutes and 60 minutes, SD and CV were calculated using corrected OD at 570 nm.

As per the certificate of analysis for the batch of tissues used in the study (Batch# 21-RHE-061), the average optical density was 1.3 in QC testing at SkinEthic Laboratories

Therefore, the observed results were within the acceptance/historical range of SkinEthic Laboratories/OECD TG acceptance criteria. Based on these observations, the results of the negative control are considered acceptable for addition into the laboratory's historical control database.

2. Positive Control

Exposure to the positive control for 1 hour induced a decrease in the relative absorbance (OD) and cell viability as compared to the negative control. The mean percent viability of the positive control was 2.38% which met OECD 431 acceptance criteria, i.e., <15% viability.

Exposure time	Mean OD (3 tissues)	Mean % viability	SD	%CV
60 minutes	0.026	2.38	0.14	5.88

Note: For the positive control, SD and % CV of % viability was calculated using % viability/tissue.

For adapted positive control-treated tissues, the NSMTT was < 0% relative to that of the negative control, therefore the true MTT metabolic conversion of positive control treated tissue was not determined.

3. Observation of Tissues after MTT Test

MTT reduction was observed in the negative control and ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated live and killed tissues for the exposure periods of 3 minutes and 60 minutes. MTT reduction was not observed in the positive control treated live and killed tissues exposed for 60 minutes.

4. Test Item

No significant reduction in the percent cell viability was observed after 3-minute and 60-minute exposure in treated tissues when compared with that of the concurrent negative control. The difference between viability of treated tissues (% CV) was less than 1% which meets the assay acceptance criteria (< 30%).

The results of the ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated tissues are summarised below:

Exposure time	Mean OD (3 tissues)	Mean % viability	SD	%CV
3 minutes	0.993	88.77	0.63	0.71
60 minutes	1.016	91.95	0.46	0.50

For the test item, SD and % CV of % viability was calculated using % viability/tissue.

For test item treated adapted controls (killed tissues) exposed for period of 3 and 60 minutes, NSMTT was < 50% relative to the negative control, therefore a true MTT metabolic conversion of treated tissue was not determined.

5. Interpretation of Results

The test item did directly reduce MTT in the pre-test, and so adapted test item controls were included in the main study, alongside additional adapted positive and negative control tissues (freeze killed tissues). Since NSMTT was < 50% relative to the negative control, therefore a true MTT metabolic conversion of treated and positive control tissue was not determined.

The negative and positive control optical density values were within the historical control range and met the assay acceptance criteria, as described in the OECD 431 guideline. The efficiency of the test system was demonstrated and all criteria for a valid study were met.

The mean percent viability of the positive control, after 60 minutes exposure, was less than 15%, compared to the negative control. This correctly predicts 8N KOH as corrosive to skin.

Following the exposure to the test item, there was no reduction in tissue viability compared to the negative control. Therefore, viability exceeded the cut-off values of 50% after 3 minutes exposure and 15% after 60 minutes exposure. Therefore, based on the results of this study, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is classified as non-corrosive as per the "United Nations Globally Harmonized System of Classification and Labelling of Chemicals".

D. Conclusion:

From the results of this study, it is concluded that ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is predicted to be non-corrosive to the skin as indicated in OECD 431, under the specified conditions of this study using reconstructed human epidermis (RhE) tissues.

A 1.5.3 Study 3

Comments of zRMS:	Considering information available in GD OECD 439 revision 14 June 2021 INITIAL CONSIDERATIONS AND LIMITATIONS Subsection 8: p.2 (..) data indicates a lack of applicability of the RhE based *in vitro* skin irritation test for agrochemical formulations (47). (..) See also: Kolle S.N, van Ravenzwaay B. and Landsiedel R. (2017). *Regulatory accepted but out of domain: In vitro skin irritation tests for agrochemical formulations*. Regul. Toxicol. Pharmacol 89, 125-130. Thus, taking into account mentioned above information zRMS decided to conclude assessment in this hazard category for the ADM.03503.F.1.A based on *in vivo* study. Study outcome ([redacted] 2021 *In Vitro Skin Irritation Test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) Using Reconstructed Human Epidermis Tissues (RhE)*, has not been considered for hazard assessment. ~~Based on the skin irritation of the individual components classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as Skin Corr. 1B H314. Composition and calculation details are provided in dRR Part C is relevant and sufficient for hazard evaluation.~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Reference KCP 7.1.4/02

Report author:	[redacted]
Report year:	2021
Report title:	*In Vitro* Skin Irritation Test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) Using Reconstructed Human Epidermis Tissues (RhE)
Report no:	[redacted]
ADAMA reference number:	[redacted]
Guidelines followed in study:	OECD 439 (June 2020), EC method B46 (2012)
Deviations from current test guideline:	None
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: This study was performed to evaluate the skin irritation potential of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), using a reconstructed human epidermis (RhE) tissue, as per the United Nations Globally Harmonized System of Classification and Labelling of Chemicals (UN GHS).

Tissues were exposed to the negative control (Dulbecco's Phosphate Buffered Saline (DPBS)), positive control (sodium dodecyl sulfate, 5% aqueous (SDS)) and ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in triplicate for 42 minutes, at room temperature. After exposure, the tissues were rinsed, dried, and incubated in a growth medium for 42 hours. The tissues were then incubated with MTT (3-(4, 5-Dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide) solution for 180 minutes, and the blue formazan salt was extracted, and the optical density (OD) measured. The OD values obtained with the test item were used to calculate the mean percentage tissue viability (after exposure and post-treatment incubation) and provide the *in vivo* prediction.

The mean percent cell viability in tissues treated with the test item is shown below. Significant reduction in the percent cell viability was observed in ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated tissues when compared with that of the concurrent negative control.

Treatment	**Mean Percent Viability**
	(42 Minutes Exposure)
Negative control (Dulbecco's Phosphate Buffered Saline (DPBS))	100
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	6.8
Positive control (Sodium dodecyl sulfate (5% aq.))	3.9

The negative and positive controls met the acceptance range for the OECD guideline and the efficiency of the test system was demonstrated. All criteria for a valid study were met as described in the OECD 439 guideline.

Based on the results of this study, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is predicted to be a skin irritant.

COMPLIANCE: Signed and dated GLP, Confidentiality and Quality Assurance Statements are provided.

I. MATERIALS AND METHODS

Experimental start date: 27 April 2021, Experimental completion date: 05 May 2021

A. MATERIALS:

1. Test Item: ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
   Description: Yellowish to brownish, transparent liquid
   Lot/Batch #: 1162-230719-011
   Purity: Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6)
   Prothioconazole: 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
   Storage: Room temperature
   Expiry Date: 05 September 2021

2. **Vehicle and/or Positive Control:**
   Negative control tissues were treated with sterile Dulbecco's Phosphate Buffered Saline (DPBS, Lot N°: RNBH6544)
   Positive control tissues were treated with sterile sodium dodecyl sulfate (5% aq.):

3. **Test System:**
   Reconstructed human epidermis (SkinEthic™ RhE)
   Source: SkinEthic Laboratories, Episkin 4, Rue Alexander Fleming, 69366 Lyon Cedex 07, France

B. STUDY DESIGN AND METHODS

Pre-Tests

Prior to the main experiment, preliminary tests such as colour interference test, direct MTT reduction test and mesh compatibility test were performed to select the appropriate adapted controls.

Well Plate and Media Used for Pre-Test and Incubation

1. **Color Interference:** Glass test tube with 0.3 mL Isopropanol at 37 ± 1 ºC and 5 ± 1% CO_2 in 95% humidified atmosphere for 60 minutes. Absorbance was measured.

2. **MTT Reduction Test :** 24 Well Plate containing MTT Solution and test item at room temperature for 180 minutes. Visual examination was performed to check the MTT reduction.

3. **Mesh Compatibility Test (Only for Liquid Test item):** Distilled water or Test item on slide with Mesh and Incubated at Room Temperature for 42 minutes. Interaction was checked using a light microscope and the un-aided eye.

Main Study

Methods:

Well Plate and Media used during Treatment	24-Well Plate with 0.3 mL maintenance media
Standard condition during treatment	42-Minutes at Room Temperature. Nylon Mesh applied on liquid materials for uniform spread of material.
Rinsing	25 times with constant soft stream of 1 mL PBS at a 5-8 cm distance from the insert to remove all test item
MTT test and Incubation	24-Well Plate with 0.3 mL MTT Solution at 37 ± 1 ºC and 5 ± 1% CO2 in 95% humidified atmosphere
Formazan Extraction and Incubation	24-Well Plate with 1.5 mL Isopropanol and extracted at room temperature with gentle shaking (150 rpm) for 2 hours
OD Measurement at 570 nm	200 µL/well and three replicates/tissue in 96-well plate

Statistics:

For controls and test item, data from individual tissue replicates e.g., OD values and corrected OD values obtained from software were used and were processed further to calculate percentage cell viability for test item, including classification were reported in tabular form. In addition, mean and ranges of viability, standard deviation and CVs between tissue replicates for controls and test item were reported.

II. RESULTS AND DISCUSSION

Pre-Tests

1. Colour Interference Test

Any difference in the absorbance due to the colour interference was not observed between the negative control (Isopropanol) and the test item, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC). Therefore, the results of the colour interference test did not show any interference in the optical density due to the test item. Results of the colour interference test are summarised in the table below:

Treatment	Optical Density (nm)	Interaction
Negative Control (Isopropanol)	0.044	No
	0.060	
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	0.045	No
	0.045	

2. Direct MTT Reduction Test

ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) produced a colour change when compared with that of the concurrent negative control (Distilled water), indicating direct MTT reduction and therefore adapted controls were included in the main study. Results of the direct MTT reduction test are summarised in the table mentioned below:

Treatment	Colour Observed	Interaction
Negative Control (Distilled water)	Yellow	No
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	Blue	Yes

3. Mesh Compatibility Test

The result shows that there was no interaction of the test item and negative control with mesh (distilled water) when observed under a light microscope and the un-aided eyes.

Main Study

1. Negative Control

The OD values (corrected ODs) of negative controls in all tissues was between 0.899 and 0.922. Therefore, the results of the negative control met the acceptance range of the OECD test guideline 439 for the prediction model SkinEthic™ RHE. Test guideline requirement of optical density is ≥ 0.8 and ≤ 3.0 (the acceptance criteria for SkinEthic™ RHE model as per the OECD TG 439). The standard deviation value of negative controls in all tissues met the acceptance range of OECD test guideline 439 for the prediction model SkinEthic™ RHE.

The results of the negative control are summarised below:

Mean OD (3 tissues)	Mean % viability	SD	%CV
0.912	100	0.006	0.66

Note: For Negative control, SD and CV were calculated using corrected OD at 570 nm.

As per the certificate of analysis for the batch of tissues used in the study (Batch# 21-RHE-061), the average optical density observed was 1.3 in QC testing at SkinEthic Laboratories. Based on these observations, results of the negative control are considered acceptable for addition into the laboratory's historical control database.

2. Positive Control

The mean percent viability of the positive control was 3.9%, which met the acceptance criteria of the OECD test guideline 439, i.e., <40% viability. The Standard Deviation value for all tissues met the acceptance criteria of not more than 18%.

The results of the positive control are summarised below:

Mean OD (3 tissues)	Mean % viability	SD	%CV
0.035	3.9	0.208	5.33

Note: For positive control, SD and CV of % viability was calculated using % viability/tissue.

3. Test Item

The mean percent viability of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated tissues, expressed as % of the negative control, was 6.8%. The standard deviation of each intra-batch mean was 2.623 and met the acceptance criteria of not more than 18%.

The results of the test item treated tissues summarised below:

Mean OD (3 tissues)	Mean % viability	SD	%CV
0.062	6.8	2.623	38.57

For the test item, SD and CV of % viability were calculated using % viability/tissue.

Since the Non-Specific MTT reduction (NSMTT) of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated killed tissues was < 0% relative to the negative control, a true MTT metabolic conversion of treated tissue was not determined.

4. Interpretation of Results

Since direct MTT reduction was observed in the pre-tests, adapted controls were included in the main study. However, the Non-Specific MTT reduction calculation (NSMTT) showed no difference relative to the negative control (<0%), and so the test item was not interacting with MTT in viable tissues.

As the negative and positive control optical density values met the assay acceptance criteria, as described in the OECD 439 guideline, they were considered acceptable for addition in the historical control data. The efficiency of the test system was demonstrated, and all criteria for a valid study were met.

The mean percent viability of the positive control, after exposure and post-treatment incubation, was less than 50%. This correctly predicts SDS (5% aq) as an irritant to the skin (UN GHS Category 1 or 2).

Since the mean percent viability of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated tissues, after exposure and post-treatment incubation, was less than 50%, the *in vitro* prediction for the test item is an irritant to skin (UN GHS Category 2 "Irritant"), as per the "United Nations Globally Harmonized System of Classification and Labelling of Chemicals".

D. Conclusion:

Based on the results of this study, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is predicted to be a skin irritant.

A 1.5.4 Study 4

Comments of zRMS:	~~From the scientific point of view study Acute Dermal Irritation Study Of ADM.03503.F.1.A Disha Chande, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary.~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection or *in vitro* studies has not been taken into account (please refer zRMS consideration in the preface to this dRR reflecting cMS comments). Test product was applied to the skin of an experimental animal; untreated skin areas of the test animal serve as the control. The degree of irritation/corrosion was read and scored at specified intervals in order to provide a complete evaluation of the effects. The duration of the study was sufficient to evaluate the reversibility or irreversibility of the effects observed. There was no deviation from studies protocol. Results of the study and conclusions are adequate for risk assessment and classification purpose. Study accepted.

Reference KCP 7.1.4/03

Report author:	[redacted]
Report year ;	2021
Report title:	Acute Dermal Irritation Study Of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 G/L EC) in Rabbits
Report no:	[redacted]

Reference number:	[redacted]
Guidelines followed in study:	OECD 404 (2015)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: In this acute dermal irritation study, three adult female New Zealand White rabbits were dermally exposed to 0.5 mL ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC). Initially, one rabbit was tested with a single patch applied evenly over the intact skin for a period of 4 hours. Based on the observations at 24-h post-patch removal, two additional rabbits were tested simultaneously to confirm the irritation response.

The test item was applied over an approximately 6 cm^2 area of skin. The control skin site of each rabbit was untreated. The treated and control sites were covered with a semi-occlusive dressing (a gauze patch, secured at margins by a non-irritating tape) for a period of 4 hours. At the end of the 4-h exposure period, any residual test item was removed with a piece of cotton wool soaked in distilled water. All test sites were evaluated for skin irritation and corrosion after patch removal. Skin reactions were recorded at 1, 24, 48, and 72 hours after removing the patches for all rabbits.

ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) caused a minimal dermal reaction in all rabbits, which was fully reversible by 24-h post-patch removal. The mean dermal irritation score for each rabbit, for erythema and oedema, observed at 24, 48, and 72-hours post-patch removal was 0.00 in all cases.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019):
Non-irritating to rabbit skin

Under the experimental conditions, ADM.03503.F.1.A is not a skin irritant. Thus, **no classification is required according to Regulation (EC) No. 1272/2008**.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid
	Lot/Batch #:	1162-230719-011
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
	Expiry date:	05 September 2021

2. Vehicle and/or Positive Control: The test item was a liquid end-use product and was tested undiluted.

3.	**Test Animals:**	
	Species:	Rabbit
	Strain:	New Zealand White
	Age at Dosing:	14 to 15 weeks,
	Weight at Dosing:	Minimum: 1.824 kg, Maximum: 1.933 kg
	Source:	Animal Breeding Facility, Jai Research Foundation
	Housing:	Individual rabbit.
	Feed:	Teklad certified Global High Fibre Rabbit Feed manufactured by Envigo, USA.
	Water:	UV sterilised water filtered through Reverse Osmosis water filtration system.

	Environmental Conditions:	**Temperature:** **Humidity:** **Air Changes:** **Photoperiod:**	20 to 23 °C 64 to 66% Minimum 15 air changes/h 12 h dark/ 12 h light
	Acclimation Period:	6 to 9 days	

B. STUDY DESIGN AND METHODS

1. In-life Dates –

Start: 06 May 2021 End: 18 May 2021

2. Animal Assignment and Treatment - Approximately 24 hours prior to the treatment, hair from the dorsal region at two contralateral sites of each rabbit was closely clipped using an electric animal clipper. Care was taken to ensure that the skin was not abraded while clipping. An area greater than 6 cm^2 was clipped at both the sites. Initially one rabbit was tested with a single patch applied evenly to the intact skin for a period of 4 h. Based on the observations at 24-h post-patch removal, two additional rabbits were tested simultaneously to confirm the irritation response. An amount of 0.5 ml of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was applied evenly to one of the clipped sites of each rabbit. The other clipped site was untreated and served as the control site. The treated and the control sites were covered with gauze patches of approximately 6 cm^2 which were not more than 8-ply and were secured at the margins by non-irritating tape to prevent evaporation of the test item and to ensure that the rabbits did not ingest it. The entire trunk of each animal was wrapped with non-irritating semi-occlusive adhesive tape to avoid dislocation of the patch(es). At the end of the 4-h exposure period (day 0), the residual test item was removed with cotton wool, soaked in distilled water.
The rabbits were observed for morbidity and mortality twice a day and detailed clinical signs were recorded at least once a day.
Skin reactions were observed at 1, 24, 48 and 72 h post-patch removal. The site of application was visually assessed and scored according to the numerical scoring system listed in OECD 404. Observation for signs of irritation (such as hyperplasia, scaling, discolouration, fissures and scabs) were also recorded.
Mean irritation scores were calculated across three scoring times (24, 48 and 72 hours after patch removal) for each animal for erythema & eschar formation grades and for oedema grades, separately.

3. Statistics – : Not Applicable

II. RESULTS AND DISCUSSION

A. Skin Reactions

At 1-hour post-patch removal, the treated skin site revealed very slight erythema (barely perceptible) (score of 1) and very slight oedema (barely perceptible) (score of 1) in all rabbits. There was no skin reaction observed at 24-hour post-patch removal. All rabbits recovered completely and appeared normal till the end of the experimental period.

The mean score, calculated from the treated site of all rabbits at 24, 48, and 72-hours observation time-points, was 0.00 for erythema and 0.00 for oedema.

B. Clinical Observations –

No clinical sign related to the treatment was observed in any rabbit throughout the experimental period, and there was no adverse effect on the body weight.

C. Conclusion:

Based on the results of this study, an indication of the classification forADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019):
Non-irritating to rabbit skin.

Under the experimental conditions, ADM.03503.F.1.A is not a skin irritant. Thus, **no classification is required according to Regulation (EC) No. 1272/2008.**

A 1.6 Eye irritation (KCP 7.1.5)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. In consideration of the content and the classification of the individual components in the formulation, a classification of ADM.03503.F.1.A with H318 would be required. Since the composition of the plant protection product is confidential, this approach is presented in the confidential part C of this dossier.

An *in vitro* eye irritation study according to OED guideline 437 (2020) was conducted. The results of this study showed that no stand-alone prediction could be made. An *in vivo* eye irritation study according to OECD guideline 405 (2017) was conducted but not performed with intention for use within the EU, but to satisfy the regulatory requirements of countries outside of the EU to clarify the results of the *in vitro* study. Based on the available information, ADM.03503.F.1.A is a severe eye irritant. Thus, classification as Eye Damage Category 1 H318 is required according to Regulation (EC) No. 1272/2008.

A 1.6.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity end-points *in vivo* studies results has been used as basis to assessment.

Eye irritation has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.3.3.3.

Various ingredients in ADM.03503.F.1.A are classified for Eye Dam. 1 H318 and Eye Irrit. 2 H319. The ingredient classified as Eye Dam. 1 H318 is present in the formulation at a concentration >3%.

Thus, classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as Eye Dam. 1 H318.

A 1.6.2 Study 2

Comments of zRMS:	~~From the scientific point of view study *In Vitro* Eye Irritation Test of ADM.03503.F.1.A Ramesh Verma, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has not been taken into the consideration.~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection or *in vitro* studies has not been taken into account (please refer zRMS consideration in the preface to this dRR reflecting cMS comments).

Reference KCP 7.1.5/01

Report author:	[redacted]
Report year ;	2021
Report title:	*In Vitro* Eye Irritation Test of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) Using Bovine Corneal Opacity

	and Permeability Test
Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 437 (2020)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: This study was conducted to evaluate the ocular irritancy of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), as measured by the potential to induce opacity and increase permeability in isolated bovine cornea. The BCOP test can identify test item classified as UN GHS Category 1 (severe eye irritants) or UN GHS no category.

Isolated bovine corneas (three corneas per set) were treated with normal saline (Set 1 - control), undiluted dimethylformamide (Set 2 - positive control) and ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) (Set 3 – test group). After a 10 minute application, corneas were washed and incubated for approximately 2 h at 32 ± 1 ºC. At the end of incubation, opacity readings were recorded, and the corneas were applied with 1 mL of fluorescein sodium solution (4 mg/mL) on to the anterior surface of the cornea followed by incubation for approximately 90 minutes at 32 ± 1 ºC. At the end of incubation, the Optical Density (OD) was measured at 490 nm from fluid collected from the posterior chamber. The *in vitro* irritancy score for each treatment group was calculated from the mean opacity and permeability values.

Treatment	***In Vitro* Irritancy Score (IVIS)**
Negative control (Saline)	0.46
Positive control (DMF)	84.41
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)	15.40

The negative and positive controls met the acceptance criteria, as described in the study plan, and confirmed the reliability of the test procedure.

The mean IVIS score for the corneas treated with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was found to be 15.40. This value falls between the cut-off values for predicting serious eye damage or no classification according to the UN GHS.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals: No stand-alone prediction can be made

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid
	Lot/Batch #:	1162-230719-011
	Expiration date:	05 September 2021
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)

2. Vehicle and/or Positive Control: Normal saline was used as negative control and N,N-dimethylformamide was used as positive control in the study.

3. Details of Test System:

Test System	Isolated cornea from the eyes of freshly slaughtered cattle
Source	Deonar Abattoir slaughterhouse, Mumbai, Maharashtra
Bovine Age	Between 1 to 5 years (the age of the animals was determined based on the teeth count and horn ring count at the site of collection. In addition to the Horizontal Diameter of corneas and central corneal thickness were used to determine the age of the animal by the test facility)
Transportation Condition	Transported (in a sealed plastic container) under cold condition in Hanks' Balanced Salt Solution containing antibiotics [penicillin at 100 IU/mL and streptomycin at 100 µg/mL

B. STUDY DESIGN AND METHODS

1. In-life Dates –

Start: 29 May 2021 End: 29 May 2021

2. Treatment

Corneas having an opacity value < 7 opacity units for the opacitometer were used in the study. Three corneas were tested in each group

Application procedure:

Negative control, positive control and test item were used undiluted, for each isolated eye 750 µL was introduced into the anterior chamber. Corneas were exposed for approximately 10 mins at 32 ºC.

At the end of the exposure period, corneal epithelium was washed, after which the corneas were incubated for approximately 2 hours at 32 ºC. After which, opacity of each cornea was recorded using an opacitometer. After the opacity measurement, the permeability was determined using fluorescein solution.

3. Statistics

The change in opacity for each cornea (including the negative control corneas) was calculated by subtracting the initial opacity reading from the final opacity reading. These values were corrected by subtracting from each the average change in opacity observed for the negative control corneas. The mean opacity value for each treatment was calculated by averaging the corrected opacity values of each cornea for a given treatment.

Permeability was calculated by calculating the corneas having an opacity value < 7 opacity units for the opacitometer were used in the study. Three corneas were tested in each group mean OD_{490} for the blank wells. The mean blank OD_{490} was subtracted from the OD_{490} of each well (corrected OD_{490}).

Final corrected OD_{490} = (OD_{490} – mean blank OD_{490}) – Average corrected negative control OD_{490}

The mean OD_{490} for each treatment group was calculated by averaging the final corrected OD_{490} values of the treated corneas for that treatment condition.

An *in vitro* irritation score was calculated for each individual treatment with the following formula:

In vitro score = Opacity value + (15 x permeability OD_{490} value)

II. RESULTS AND DISCUSSION

A. Corneal Opacity – The mean final opacity values for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated eyes showed an observable increase in comparison to the control group. An observable marked increase in the final mean opacity was observed in corneas treated with the positive control, *N,N*-dimethylformamide in comparison to the control group, as shown below:

Treatment:	Negative Control (Saline)	Test Item	Positive Control (DMF)
Mean Opacity value	0.33	8.83	72.59

B. Corneal Permeability- The mean final corneal permeability values for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated eyes showed an observable increase in comparison to the control group. An observable marked increase in the mean final corneal permeability was observed in corneas treated with the positive control, *N,N*-dimethylformamide in comparison to the control group, as shown below:

Treatment:	Negative Control (Saline)	Test Item	Positive Control (DMF)
Mean permeability value	0.009	0.438	0.788

***C. In vitro* Irritancy Score (IVIS) -** The mean *In Vitro* Irritancy Score (IVIS) of normal saline (control), *N,N*-dimethylformamide (positive control) and test item treated corneas are shown below:

Treatment:	Negative Control (Saline)	Test Item	Positive Control (DMF)
Mean IVIS score	0.46	15.40	84.41

The mean IVIS score of the positive control (DMF) was greater than 55 and predicts serious eye damage (UN GHS Category 1).

The mean IVIS score for the corneas treated with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was found to be 15.40. This value falls between the cut-off values for predicting serious eye damage or no classification according to the UN GHS.

D. Validity of the Test-

The mean *In Vitro* Irritancy Score (IVIS) of dimethylformamide (positive control) treated corneas (84.41) was within the range of two standard deviation of the mean of the historical control data , confirming the reliability of the test procedure.

The opacity and permeability values of the negative control (saline) treated corneas (0.33 and 0.009, respectively) were less than the established upper limits for background values, confirming the reliability of the test procedure.

E. Conclusion:

From the results of this *in vitro* assay in isolated bovine cornea, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is No stand-alone prediction can be made.

A 1.6.3 Study 3

Comments of zRMS:	~~From the scientific point of view study Acute Eye Irritation Study Of ADM.03503.F.1.A Disha Chande, 2021 Ramesh Verma, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection or *in vitro* studies has not been taken into account (please refer zRMS consideration in the preface to this dRR reflecting cMS comments). Study (Disha Chande, 2021) has been reviewed for compliance with the current guidelines resulting from scientific progress (OECD 405 rev 2017). The update from 2012 mainly focused on the use of analgesics and anesthetics without impacting the basic concept and structure of the TG. ICCVAM (*Interagency Coordinating Committee on the Validation of Alternative Methods*) reviewed the usefulness and limitations of routinely using topical anesthetics, systemic analgesics, during *in vivo* ocular irritation safety testing. The review concluded that the use of topical anesthetics and systemic analgesics could avoid most or all pain and distress without affecting the outcome of the test, and recommended that these substances should always be used. In the discussed study (Disha Chande, 2021) topical anesthetics has been used. In the Reviewer opinion study implements 3R rules and humane endpoints minimizing pain and distress of animals.

	In the mentioned study degree of eye irritation/serious eye damage were evaluated by scoring lesions of conjunctiva, cornea, and iris, at specific intervals. Duration of the study was sufficient to evaluate the reversibility or irreversibility of the effects. Results of the study and conclusions are adequate for risk assessment and classification purpose. Study accepted.

Reference KCP 7.1.5/02

Report author:	[redacted]
Report year ;	2021
Report title:	Acute Eye Irritation Study Of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 G/L EC) in Rabbits
Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 405 (2017)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: In an acute eye irritation study, three adult male New Zealand White rabbits were given a single ocular application of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC).

Initially, one rabbit was tested. Based on observations at 24-hours post-test item application, the irritation response was confirmed by testing two additional rabbits sequentially.

A volume of 0.1 mL of test item was applied in their right eyes, while their left eyes were kept untreated and served as the control. Observations were made at an interval of 1, 24, 48, and 72-hours and on day 7 and 14 for all the rabbits and on day 21 in rabbit N° 2 and 3 post-test item application. General health status was also checked.

Corneal opacity (score of 1 to 2) was evident at day 7 in rabbit N° 2, and on day 14 and 21 in N° 2 and 3 rabbits. Conjunctival redness (score of 1) was evident at 1, 24, 48, and 72 h, and on day 7 in one rabbit which resolved by day 14 and conjunctival redness (score of 1 to 2) was evident at 1, 24, 48, and 72 h, and on day 7, 14, and 21 rabbit N° 2 and 3. Conjunctival chemosis (score 1) was evident at 1, 24, 48, and 72 h in rabbit N° 1, which resolved by day 7 whereas conjunctival chemosis (score 1) was evident at 1, 24, 48, and 72 h, and on day 7 and 14 in two rabbits, which resolved by day 21.

An examination with fluorescein dye and cobalt blue filter revealed no corneal epithelium damage at 24, 48, and 72-hours and on day 7 and 14 post-test item application for all the rabbits and in rabbit N° 2 and 3 on day 21.

The control eye did not show any abnormal reaction, during the study. Moreover, there was no sign of systemic toxicity in any rabbit observed.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Category 1**

Under the experimental conditions, ADM.03503.F.1.A is a severe eye irritant/irreversible effects. Thus, classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as **Eye Dam. 1 H318.**

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid
	Lot/Batch #:	1162-230719-011
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
	Expiry date	05 September 2021

2. Vehicle and/or Positive Control: The test item was a liquid end-use product and was tested undiluted.

3.	**Test Animals:**		
	Species:	Rabbit	
	Strain:	New Zealand White	
	Age at Dosing:	4 to 5 months	
	Weight at Dosing:	Minimum: 2.587 kg, Maximum: 2.816 kg	
	Source:	Vab Bio Sciences, Hyderabad, India	
	Housing:	Individual rabbit.	
	Feed:	Teklad certified Global High Fibre Rabbit Feed manufactured by Envigo, USA.	
	Water:	UV sterilised water filtered through Reverse Osmosis water filtration system.	
	Environmental Conditions:	**Temperature:** **Humidity:** **Air Changes:** **Photoperiod:**	20 to 23 °C 65 to 66% Minimum 15 air changes/h 12 h dark/ 12 h light
	Acclimation Period:	6 to 11 days	

B. STUDY DESIGN AND METHODS

1. In-life Dates –

Start: 29 May 2021 End: 30 June 2021

2. Animal Assignment and Treatment –

Prior to the treatment, both eyes of each rabbit were examined for ocular lesions to confirm no pre-existing eye disease, corneal damage or any other defects. The eyes of all rabbits were found to be normal and the rabbits were accepted for the study.

Anaesthetic Regime: Appropriate anaesthetic and systemic analgesia were applied.

Initially, a single rabbit was tested. A single amount of 0.1 ml of the test item was placed into the conjunctival sac of the right eye. Since severe effects were not observed in the first treated rabbit, two additional rabbits were sequentially treated in an identical manner.

Rabbits were observed twice daily for mortality and morbidity. Clinical signs were recorded once a day.

Eye reactions: Both eyes of each rabbit were observed for signs of ocular irritation at an interval of 1, 24, 48, and 72 h and on day 7, 14 and/or after test item application on day 0. The irritant responses were scored following the Draize numerical evaluation. For each observation occasion, scoring for ocular lesions was undertaken for the corneal opacity, the iris and the conjunctiva (redness and chemosis, including lids and/or nictitating membranes). Any other ocular lesions were also recorded, if present. Fluorescein staining was used to assess any corneal damage at 24, 48, and 72 hours and on day 7, 14 for rabbit N° 1 and 21 post TIA application (Swinyard, 1990).

3. Statistics – : Not Applicable

II. RESULTS AND DISCUSSION

A. Mean Eye Irritation Scores

The rabbit mean eye irritation score, observed for corneal opacity (0.00), iritis (0.00), conjunctival redness (1.00 to 2.00) and conjunctival chemosis (1.00) following grading at 24, 48, and 72-h post-test item application.

B. Narrative Description of Eye Irritation

At 1 h post-TIA, treated eyes revealed conjunctival redness [some blood vessels definitely hyperaemic (injected) (score of 1) in rabbit N° 1 and 3 to diffuse, crimson colour, individual vessels not easily discernible (score of 2) in rabbit N° 2] and conjunctival chemosis [some swelling above normal (includes nictitating membranes) (score of 1) in all the rabbits].

At 24, 48, 72 h post-TIA, treated eyes revealed conjunctival redness [some blood vessels definitely hyperaemic (injected) (score of 1) in rabbit N° 1 to diffuse, crimson colour, individual vessels not easily discernible (score of 2) in rabbit N° 2 and 3] and conjunctival chemosis [some swelling above normal (includes nictitating membranes) (score of 1) in all the rabbits].

On day 7 post-TIA, treated eyes revealed corneal opacity [scattered /diffuse areas of opacity (other than slight dulling of normal lusture), details of iris clearly visible in rabbit N° 2 (score of 1)], conjunctival redness [some blood vessels definitely hyperaemic (injected) in rabbit N° 1 (score of 1) to diffuse, crimson colour, individual vessel not easily discernible in rabbit N° 2 and 3 (score 2)] and conjunctival chemosis [some swelling above normal (includes nictitating membranes) in rabbit N° 2 and 3 (score of 1)].

On day 14 post-TIA, treated eyes revealed corneal opacity [easily discernible translucent area, details of iris slightly obscured in rabbit N° 2 and 3 (score of 2)], conjunctival redness [some blood vessels definitely hyperaemic (injected) in rabbit N° 2 (score of 1) to diffuse, crimson colour, individual vessel not easily discernible in rabbit N° 3 (score 2)] and conjunctival chemosis [some swelling above normal (includes nictitating membranes) in rabbit N° 2 and 3 (score of 1)]. Treated eye of rabbit N° 1 recovered completely and appeared to be normal.

On day 21 post-TIA of rabbits revealed corneal opacity [scattered /diffuse areas of opacity (other than slight dulling of normal lusture), details of iris clearly visible in rabbit N° 2 and 3 (score of 1)] and conjunctival redness [some blood vessels definitely hyperaemic (injected) in rabbit N° 2 and 3 (score of 1)].
An examination with fluorescein dye and cobalt blue filter revealed no corneal epithelium damage at 24, 48, and 72-hours and on day 7 and 14 post-TIA for all the rabbits and on day 21 for rabbit N° 2 and 3.

Iritis was not observed in any rabbit, throughout the experimental period.
Since one rabbit had shown complete recovery, the same was terminated after the 14 day examination while other two rabbits were observed till the end of the examination period of 21 day post-TIA.

Score at timepoint	Cornea Max Score:4	Iris Max Score: 2	Conjunctivae Max Score 3	Chemosis Max score 4
1h	0/0/0	0/0/0	1/2/1	1/1/1
24h	0/0/0	0/0/0	1/2/2	1/1/1
48h	0/0/0	0/0/0	1/2/2	1/1/1
72h	0/0/0	0/0/0	1/2/2	1/1/1
Day 7	0/1/0	0/0/0	1/2/2	0/1/1
Day 14	0/2/2	0/0/0	0/1/2	0/1/1
Day 21	NA/1/1	NA/0/0	NA/1/1	NA/0/0
Average 24, 48, 72h	**0/0/0**	**0/0/0**	**1/2/2**	**1/1/1**

NA = not applicable

C. <u>Clinical Observations</u> –

No sign of systemic toxicity, including clinical observation and body weight, was observed in any rabbit, throughout the experimental period.

D. Conclusion:

Based on results of this study, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is considered to be severe eye irritant/irreversible effects on the rabbit eye.
Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Category 1.**
Under the experimental conditions, ADM.03503.F.1.A is a severe eye irritant/irreversible effects. Thus, classification is required according to Regulation (EC) No. 1272/2008. ADM.03503.F.1.A is classified as **Eye Dam. 1 H318**.

A 1.7 Skin sensitisation (KCP 7.1.6)

Classification via the application of bridging principles is not possible since data on a similar mixture are not available. Thus, in a first approach, classification based on the complete composition of the plant protection product is taken into account for classification purpose. Since the composition of the plant protection product is confidential, this approach is presented in the confidential part C of this dossier.
In addition, two *in vitro* skin sensitization studies were conducted (OECD 442C and OECD 442D) based on these in vitro studies it was not possible to determine a classification for the formulation.
In addition, a vertebrate study is available for skin sensitisation of ADM.03503.F.1.A. This study was not performed with intention for use within the EU, but it was performed to satisfy the regulatory requirements of countries outside of the EU. The study is provided with the dossier and summarized hereafter.
Based on all the available information, ADM.03503.F.1.A is considered not a skin sensitizer. Thus, no classification is required according to Regulation (EC) No. 1272/2008.

A 1.7.1 Study 1 – calculation method

Comments of zRMS:	~~Acute oral toxicity assessment based on product composition is relevant and sufficient for hazard evaluation. Calculation has been accepted (for details see Part C).~~ Reflecting cMS comments and considering Regulation (EC) No. 1272/2008 which give animal data preference for hazard assessment, zRMS revised their approach and do not take into account toxicity assessment based on product composition. For toxicity endpoints *in vivo* studies results has been used as basis to assessment.

Skin sensitisation has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.4.3.3.

No ingredients in ADM.03503.F.1.A are classified for skin sensitisation. Thus, no classification is required according to Regulation (EC) No. 1272/2008.

A 1.7.2 Study 2

Comments of zRMS:	Based on outcome of the study In Vitro Skin Sensitization of ADM.03503. F.1.A Indrajitsinh M. Barad, 2021 it was not possible to classify using 2 out of 3 defined approach therefore study has not been taken into the final consideration. ~~Skin sensitization has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.4.3.3.~~

Reference KCP 7.1.6/01

Report author:	[redacted]
Report year:	2021
Report title:	*In Vitro* Skin Sensitization of ADM.03503. F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) Keratinocyte-Based ARE-Nrf2 Luciferase Reporter Gene Test
Report no:	[redacted]
ADAMA reference number:	[redacted]
Guidelines followed in study:	OECD 442D (2018), EC B60 (2017)
Deviations from current test guideline:	None
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: The skin sensitisation potential of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was assessed *in vitro* using the Keratinocyte-Based ARE-Nrf2 Luciferase Reporter Gene method. This assay examines the second key event (keratinocyte activation) in the skin sensitisation adverse outcome pathway.

Following the solubility assessment, dimethyl sulfoxide (DMSO) was selected as the vehicle, and the test item was tested in three independent experiments, alongside positive and negative controls, with three replicates/experiment. KeratinoSens™ (HaCaT) cells were exposed to ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) at 12 concentrations ranging from 0.20 µg/mL to 400 µg/mL, to the positive control (*trans*-cinnamaldehyde) at concentrations of 4 to 64 µM, and the negative control (DMSO) and were incubated for 48 ± 2 hours in 5 ± 1% CO_2 at 37 ± 1 °C. After incubation, induction of the luciferase gene was assessed by measuring the luminescence in the cells. Cell viability of the concurrently treated cells was also evaluated using the thiazolyl blue tetrazolium bromide (MTT) test with a separate set of plates. In experiment 2, although >1.5 fold induction was obtained for the test item, due to the lack of clear dose-response, experiment 2 was considered inconclusive. Therefore, Experiment 3 was performed as the confirmatory experiment. Finally, Experiment 1 and 3 were considered valid for final evaluation.

The value of I_{max} and $EC_{1.5}$ was calculated based on luciferase activity, i.e., luminescence measured (reading of three plates), while the value of IC_{50} and IC_{30} was calculated based on the results of cytotoxicity (OD values, reading of one plate). The results and KeratinoSens™ prediction were:

Test Item Name	**Luciferase gene induction**		**Cellular viability**		**KeratinoSens™ Prediction**
	I_{max}	**$EC_{1.5}$**	**IC_{50}**	**IC_{30}**	
ADM.03503.F.1.A (Fluxapyroxad75 Prothioconazole150 g/L EC))	3.81	7.87 µg/mL	77.59 µg/mL	67.62 µg/mL	Positive
Positive Control (*Trans*-Cinnamaldehyde)	18.14	10.57 µM	-	-	Positive

ADM.03503. F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) met all the evaluation criteria to be predicted as a sensitiser (positive) in an *in vitro* KeratinoSens assay. Negative and positive controls met the acceptance criteria for the controls and were correctly identified as negative and positive, respectively. All criteria for a valid study were met, as described in the study plan.

From the results of this KeratinoSens assay, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was concluded as "positive" for induction of ARE-dependent gene expressions.

COMPLIANCE: Signed and dated GLP, Confidentiality and Quality Assurance statements are provided.

I. MATERIALS AND METHODS

Experimental start date: 27 March 2021, Experimental completion 06 May 2021

A. MATERIALS:

Test Item Name	:	ADM.03503. F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
Batch/Lot Number	:	1162-230719-011
Analytical Concentration	:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
Physical State	:	Yellowish to brownish, transparent liquid
Date of Expiry	:	05 September 2021
Storage Temperature	:	Room Temperature

Negative and/or Positive Control:

Negative Control:
Dimethyl sulfoxide (DMSO) was used as the concurrent negative control for each set of experiments.

Positive Controls:
Trans-cinnamaldehyde was used as a positive control. Stock concentrations from 0.4 to 6.4 mM were prepared in DMSO, which were further diluted to achieve final concentrations of positive control ranging from 4 to 64 µM.

4. **Test System**:

The genetically modified HaCaT cell line (KeratinoSens™) obtained from Givaudan Schweiz AG, was used in this study. The KeratinoSens™ cell line contains a stable insertion of the luciferase gene under the transcriptional control of a constitutive SV40 promoter fused with an ARE of the human aldoketoreductase (AKR)1C2 gene.

The KeratinoSens™ culture was maintained in a frozen state in liquid nitrogen. The batch of the cell line was tested for mycoplasma contamination before using the cells in the experiment. Cultures free from any contamination were used in the study. For the experiments cell line passage numbers of 23-25 were used.

B. STUDY DESIGN AND METHODS

For each test item and positive control, one experiment consisting of at least two independent repetitions, each containing three replicates of each concentration, was performed. Each independent repetition was performed on a different day with fresh stock solutions of chemicals and independently harvested cells.

Total three experiments were performed for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC). Experiment 1 and 3 were considered valid for final evaluation.

Methods:

1. Solubility Test

The solubility of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was assessed in dimethyl sulfoxide (DMSO), at a concentration of 40 mg /mL and formed a solution. Hence, dimethyl sulfoxide was selected as the vehicle for this study.

2. Preparation of Cell Culture for Treatment (Day -1)

One day prior to testing, cells were harvested and distributed into 96-well plates at a cell density of 10,000 cells/well. Plates were incubated for 24 hours in 5 ± 1% CO_2 at 37 ± 1 °C.

3. Preparation of Control and Test Items

The test item and control substances were prepared on the day of testing (day 2).

Control Preparation
Positive control: *Trans*-cinnamaldehyde was dissolved in DMSO to achieve a concentration of 200 mM. This solution was further diluted to final concentration of 6.4 mM.

Negative control: DMSO was used as negative (solvent) control.

Test Item Preparation

As the test item was a formulation and had no defined molecular weight, based on guideline recommendation, the highest tested concentration was 400 µg/mL.

Serial dilutions were made using DMSO to obtain 12 stock concentrations (100 fold in DMSO) (0.020 mg/mL to 40 mg/mL).

Application of Test and Control Items (Day 2)

Stock solutions of ADM.03503. F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) were prepared 0 to 40 mg/ml in DMSO.

Test Item Exposure Procedure (Day 2)

After 24 ± 2 hours of incubation, medium from all the 4 plates (three plates for luciferase assay and one for cytotoxicity, MTT assay) was aspirated and discarded. It was replaced with 150 µL of DMEM containing 1% FBS without geneticin. The test item stock dilutions were further diluted (25-fold in culture medium).
50 µL of this stock solutions were added to three white assay plates and one cytotoxicity plate already containing 150 µL of culture medium. Final test concentrations used for the exposure were 0.20, 0.39, 0.78, 1.56, 3.13, 6.25, 12.50, 25.00, 50.00, 100.00, 200.00 and 400.00 µg/mL for the three experiments. Plates were then covered with petri-seal and incubated for 48 hours in 5% CO_2 at 37 °C.

Luminescence Measurement (Day 4)

Luciferase Activity

After 48 ± 2 hours of incubation, the medium from the 96-well white assay plates was aspirated and discarded. Cells were washed once with DPBS. After washing, 20 µL of passive lysis buffer was added into each well. Cells were then incubated for 20 minutes at room temperature. After incubation, the plates with cell lysate were placed in a luminometer. 50 µL of 1× luciferase substrate was added, and luciferase activity was recorded for 2 seconds.
Note: Readings of four wells were taken at a time.

Cytotoxicity Evaluation (Day 4)

After 48 ± 2 hours of incubation, cytotoxicity was determined using the MTT assay.

The plate was covered with petri seal and incubated at 5 ± 1 % CO_2 at 37 ± 1 °C for 4 hours. After incubation, the medium was removed and 50 µL of isopropanol solution was added to each well. The plate was sealed and covered with a petri seal and placed on shaker. After shaking for 30 minutes, the absorbance was measured at 570 nm using a luminometer for each well.

Cell viability of the concurrently treated cells was also evaluated using the MTT test with a separate set of plates.

4. Statistics:

For each concentration showing >1.5-fold gene-induction, statistical significance ($p < 0.05$) was calculated (e.g., by a two-tailed Student's t-test), comparing the luminescence values for the three replicate samples with the luminescence values in the solvent (negative) control wells. The EC1.5 value represents the lowest concentration with >1.5-fold gene induction (i.e. 50% enhanced luciferase activity). It was checked in each case whether this value was below the IC50/IC30 value, indicating that there was less than 30% reduction in cellular viability at the EC1.5 determining concentration.

II. RESULTS AND DISCUSSION

For ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) a total of three experiments were performed. In experiment 2, although >1.5 fold induction was obtained for the test item, due to the lack of clear dose-response, experiment 2 was considered inconclusive. Therefore, Experiment 3 was performed as the confirmatory experiment. Finally, Experiment 1 and 3 were considered valid for final evaluation. Results of valid experiments are presented in this report.

A. Controls

1. Negative (Solvent) Control

The coefficient of variation observed for the negative control (DMSO) during experiments 1 and 3 for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was 9.04% and 8.62%, respectively, which was below 20%. Since the variation amongst the replicates was less than 20%, the results of these runs were considered valid.

2. Positive Control

Luciferase activity induction obtained with the positive control, trans-cinnamaldehyde, was found to be >1.5 (the threshold value) at concentrations of 8, 16, 32 and 64 µM in experiments 1; 16, 32 and 64 µM in experiment 2. These values were significantly higher statistically than the vehicle control values.

The EC1.5 for the positive control was 7.66 µM in experiment 1 and 14.59 µM in experiment 3; both values are within two standard deviations of the laboratory historical control data. All the requirements of the positive control were met, and the experiments were considered valid.

The average gene induction for the positive control at a concentration of 64 µM was 30.69 in experiment 1 and 5.59 in Experiment 3. In experiment 1, the average gene induction for positive control at a concentration of 64 µM was not within the guideline acceptance limit of 2 to 8. However, a clear dose-response, with an increasing gene induction at increasing dose, was observed for trans-cinnamaldehyde in both experiments, so these results were considered valid.

3. Luciferase Activity

The maximal average fold induction of luciferase activity (Imax) following treatment with the test item was 3.81, which is greater than the 1.5-fold increase, indicate the positive result. The maximum average fold induction of luciferase activity (Imax) values for the test item exceeded a 1.5-fold increase at concentrations 6.25, 12.5, 25, and 50 µg/mL in experiment 1; at 25 and 50 µg/mL in experiment 3.

The EC1.5 mean value, representing the concentration where induction of luciferase activity is above the 1.5-fold threshold (i.e. 50% enhanced luciferase activity), was 7.87 µg/mL. There was a clear dose-dependent increase in the induction value.

B. Cytotoxicity Assessment (MTT assay)

Cytotoxicity observed was 7.69%, 9.23%, and 4.27% at the tested concentrations 100, 200 and 400 in experiment 1 and 2.00%, 2.60%, and 3.21% at the tested concentrations 100, 200 and 400 in experiment 3. Cellular viability was greater than 70% up to the tested concentration of 50 µg/mL in both experiments.

C. Interpretation

For ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) both valid experiments (Experiment-1 and Experiment-3) were considered for the final evaluation. The mean results are summarised below:

TABLE 2: The mean results are summarised below:

Test Item Name		Luciferase gene induction		Cellular viability		KeratinoSens™ Prediction
		I_{max}	$EC_{1.5}$	IC_{50}	IC_{30}	
Test Item	**Test Item**	3.81	7.87 µg/mL	77.59 µg/mL	67.62 µg/mL	Positive
Positive Control	**Positive Control (*Trans*-Cinnamaldehyde)**	18.14	10.57 µM	-	-	Positive

t item : ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)

Results of this KeratinoSensTM assay indicate that ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) met all the evaluation criteria to conclude that it is positive in this test.

The solvent and positive controls met the acceptance criteria and were correctly identified as negative and positive, respectively, confirming the suitability of the test system and procedures used.

Table 3: Summary of Mean Luciferase Gene Induction for Negative Control, Positive Control and Test Item ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)

Test Item Concentration (µg/mL)	Expt. 1	Expt. 3	Controls		Expt. 1	Expt. 3
0.20	1.47	0.82	**Blank Solvent (DMSO)**		0.96	1.08
0.39	1.45	0.84			1.05	0.99
0.78	1.52	0.96			1.00	1.08
1.56	1.33	0.94			0.99	1.03
3.13	1.47	1.00			0.98	0.94
6.25	1.61↑↑	1.20			1.02	0.88
12.5	1.71↑	1.29	**Positive Control (µM)**	**4**	1.03	0.95
25.00	2.37↑↑	1.93↑		**8**	1.54↑	1.30
50.00	4.78↑↑	2.84↑↑		**16**	1.70↑↑	1.54↑↑
100.00	0.00	0.00		**32**	3.89↑↑	2.33↑↑
200.00	0.00	0.00		**64**	30.69↑	5.59↑
400.00	0.00	0.00	**No Cell Blank**		0.0	0.0

Key: Test Item = ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), DMSO = Dimethyl sulfoxide, Positive control = *trans*-cinnamaldehyde
Note: The fold induction above the 1.5-fold threshold of luciferase activity was also statistically significant for the test item which were compared to the vehicle controls.
↑ = Significantly higher than vehicle control ($p \leq 0.05$),
↑↑ = Significantly higher than vehicle control ($p \leq 0.01$),

Table 4: Summary of Mean Cell Viability Percentage

Test Item Concentration (µg/mL)	Expt. 1	Expt. 3	Controls	Expt. 1	Expt. 3
0.20	107.80	103.57	**Blank Solvent (DMSO)**	106.78	101.77
0.39	103.36	90.15		93.45	96.36
0.78	110.54	99.77		97.21	97.76
1.56	91.91	93.76		105.41	98.56

3.13	97.04	99.17			106.61	98.16
6.25	99.09	100.17			90.55	107.38
12.5	107.46	94.16	**Positive Control (µM)**	**4**	93.62	107.38
25.00	104.90	103.17		**8**	92.77	111.59
50.00	101.65	109.58		**16**	93.11	105.58
100.00	7.69	2.00		**32**	100.46	105.78
200.00	9.23	2.60		**64**	91.23	113.39
400.00	4.27	3.21	**No Cell Blank**		0.00	0.00

Key: Test Item = ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), DMSO = Dimethyl sulfoxide, Positive control = *trans*-cinnamaldehyde

D. Conclusion:
From the results of this KeratinoSens™ assay, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) concluded as "positive" for induction of ARE-dependent gene expression.

A 1.7.3 Study 3

Comments of zRMS:	Study *In Chemico* Skin Sensitisation of ADM.03503.F.1.A Indrajitsinh M. Barad, 2021 is inconclusive also not possible to classify using 2 out of 3 defined approach, therefore study has not been taken into the final consideration. ~~Skin sensitization has been calculated using the method described in Regulation (EC) No 1272/2008 section 3.4.3.3..~~

Reference KCP 7.1.6/02

Report author:	[redacted]
Report year:	2021
Report title:	*In Chemico* Skin Sensitisation of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC): Direct Peptide Reactivity Assay (DPRA)
Report no:	[redacted]
ADAMA reference number:	[redacted]
Guidelines followed in study:	OECD 442C (2020)
Deviations from current test guideline:	None
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: This study was conducted to evaluate the skin sensitisation potential of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), using synthetic heptapeptides containing either Lysine (Ac-RFAAKAA-COOH) or Cysteine (Ac-RFAACAA-COOH). ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) was soluble in acetonitrile at a concentration of 100 mM. Therefore, acetonitrile was selected as the vehicle for this study.

Synthetic heptapeptides containing either Lysine (Ac-RFAAKAA-COOH) or Cysteine (Ac-RFAACAA-COOH), were used as the test system in this assay. Cysteine and Lysine containing peptides were incubated with the positive control and the test item (100 mM and neat chemical) for 24 ± 2 hours at 22.5-30 ºC (in the dark), separately. The relative peptide concentration was measured using the high-performance liquid chromatography (HPLC) with gradient elution and UV detection at 220 nm. Cysteine and Lysine peptide percent depletion values were calculated and used in a prediction model, which allows

assigning the test item to one of four reactivity classes used to support the discrimination between sensitisers and non-sensitisers (OECD, 2020).

All acceptance criteria for a valid assay were met, and the maximum standard deviation of the test item (100 mM and neat chemical) replicates met the test guideline requirements for a valid result. The mean percent peptide depletion values of the positive control (100 mM Cinnamaldehyde in acetonitrile) met the acceptance criteria. Following the incubation with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) (100 mM and neat chemical), the percent peptide depletion obtained for Cysteine and Lysine is as tabulated below. Precipitation was not observed in Cysteine peptide. Precipitation was observed in Lysine Peptide after incubation only with neat chemical. Therefore, samples were centrifuged at 400 x g for 10 minutes at 25 ºC to remove the precipitation before analysis. Co-elution of the test item did not occur either with Cysteine or Lysine peptides.

Test Item Name	Actual % Depletion (Cysteine)	Actual % Depletion (Lysine)	% Mean Depletion (Cysteine & Lysine)	DPRA Prediction
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) **100 mM**	0	0	0	No Reactivity (Negative)
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC **Neat**	29	73	51	High Reactivity (Positive)

From the Cysteine 1:10/lysine 1:50 prediction model, the Reactivity class is “No Reactivity” for 100mM of the test item. In contrast, the Reactivity class is “High Reactivity” for the Neat test item. The Non-concordant results between 100 mM and Neat Test item leads to a prediction that the test item is inconclusive for skin sensitisation.

From the results of this study, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) (100mM) is predicted by DPRA to be inconclusive.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided.

MATERIALS AND METHODS

A. MATERIALS:

5. Test Item : ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
   Description : Yellowish to brownish, transparent liquid
   Lot/Batch # : 1162-230719-011
   Purity : Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6)
   Prothioconazole: 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
   Storage : Room temperature
   Expiry Date : 05 September 2021
   Solvent Used : Acetonitrile
6. **Vehicle and/or Positive Control:**
   The vehicle control of acetonitrile was selected based on a solubility assessment with the test item.
   Positive Controls:
   Cinnamaldehyde was used as positive control (PC) at a concentration of 100 mM in acetonitrile.

7. **Test System:**
   Synthetic heptapeptides containing either Lysine (Ac-RFAAKAA-COOH) or Cysteine (Ac-RFAACAA-COOH) are used as the test system for the direct peptide reactivity assay.

B. STUDY DESIGN AND METHODS

1. ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is a mixture of compounds, it has no defined molecular weight. Therefore, as per guideline recommendation, apparent molecular weight was considered to prepare 100 mM solution. The test item was found to be soluble in acetonitrile at 100 mM and precipitation was not observed. Hence, acetonitrile was selected as the vehicle for the experiment.
2. Stock solutions (0.667 mM) of Cysteine (Ac-RFAACAA-COOH) and Lysine (Ac-RFAAKAA-COOH) were freshly prepared just before their incubation with the test item.

3. Test item was prepared as a 100 mM solution in acetonitrile based on the apparent molecular weight, immediately prior to use. two aliquots of test item were received in two separate vials for Cysteine and Lysine peptide. As the test item is mixture of compounds, it was also tested as neat chemical. For testing of liquid test item as a neat chemical, test item was exposed as such by incubating it at 1:10 and 1:50 ratio with the cysteine and lysine peptides, respectively.

4. Some batches of acetonitrile have a negative impact on Cysteine peptide stability. Hence, stability was assessed by injecting the Cysteine Reference Control A at 0, 24, and 48 h in three replicates. Peak areas were compared and %RCV was calculated.
5. HPLC conditions:
   Instrumental Parameters

Column	Column: ZorbaX SB-C18 (2.1 mm x 100 mm x 3.5 micron) with Guard Column Phenomenex Security Guard C18 (4 mm x 2 mm)			
Column Temperature	30 ± 2 °C			
Sample Temperature	25 ± 2 °C			
Detector	Fixed Wavelength Absorbance Detector with 220 nm			
Injection Volume	5 µL			
Run time	20 Minutes			
Mobile phase	Mobile phase A (0.1 % v/v Trifluoroacetic acid in Milli Q water) and Mobile phase B (0.085% v/v Trifluoroacetic acid in Acetonitrile)			
Gradient System	Time 0 min 10 min 11 min 13 min 13.5 min 20 min	Flow 0.35 mL/min 0.35 mL/min 0.35 mL/min 0.35 mL/min 0.35 mL/min end run	% A 90 75 10 10 90	% B 10 25 90 90 10

6. Preparation of the HPLC Standard Calibration Curve: Standards were prepared in a solution of 20% Acetonitrile: phosphate buffer (Cysteine peptide) and/or ammonium acetate buffer (Lysine peptide).
   Standard Preparation
7. STD 1 preparation (0.534 mM) was prepared in acetonitrile. Further serial dilution prepared till standard 6. A blank of dilution buffer as STD 7 was included. Before starting the analysis HPLC system was equilibrated for around 2 h.
8. Cysteine and Lysine peptide solutions were incubated in glass auto sampler vial with the test item (100 mM and neat chemical) at 1:10 and 1:50 ratio, respectively. The reaction solution and standards prepared were left in the dark at 22.5-30 ºC for 24 ± 2 hours before the HPLC analysis. Test item was tested in triplicate for both peptides. Samples were visually inspected for precipitation prior to HPLC analysis. Depletion of the peptide in the reaction mixture was measured by HPLC using UV detection.
9. Reference Control is a peptide solution where the test item is replaced by the solvent used to dissolve test item. Reference control A (0.5 mM Cysteine/Lysine peptide).
   used to verify the accuracy of the calibration curve for peptide quantification.

   And Reference Control B (0.5 mM Cysteine/Lysine peptide): run before and after 24 ± 2 hours incubation at 22.5-30 °C in the dark (to verify the stability of the peptide over the analysis time).

10. Samples were prepared in triplicate in 1 mL auto sampler vials for both peptides on separate days. Vials were capped, vortexed and were placed in the HPLC auto sampler at 22.5-30 °C in dark for 24 ± 2 hours. HPLC analysis of the batch of samples was started 24 ± 2 h after the test item was added to the peptide solution.
11. Co-elution control was prepared without peptide, to verify whether the test item absorbs at 220 nm and has a similar retention time as a peptide and/or interfere with the data analysis.
12. Data from individual test item replicates and Reference Control replicates, e.g., mean peak area, mean percent peptide depletion, mean peptide concentration of Reference Controls, standard deviation, and relative coefficient of variability for test item were determined.

II. RESULTS AND DISCUSSION

A. **HPLC Standard Calibration Curve** – A standard calibration curve was generated on each day of the HPLC analysis, and the value of r^2 obtained was 0.99991 for Cysteine and 0.99999 for Lysine containing peptides. It showed that the system was suitable for the assay.

B. **Cysteine Peptide Stability in Acetonitrile**- The stability of Cysteine peptide in acetonitrile was checked at 0, 24, and 48 h. The relative coefficient of variability (RCV) for the stability of Cysteine peptide in acetonitrile was 2.97% against the set standard of <15%. This showed that Cysteine peptide was stable in acetonitrile

C. **Reference Controls-** The mean peptide concentration of the reference controls in acetonitrile was within the expected concentrations, fulfilling the criteria for a valid test.

Peptide	Mean Reference Control A (mM)	Mean Reference Control B (mM)	Expected Concentration (mM)
Cysteine	0.52	0.50	0.45-0.55
Lysine	0.50	0.50	0.45-0.55

D. **Positive Controls-** The mean percent peptide depletion value of the positive control- Cinnamaldehyde (at a concentration of 100 mM in acetonitrile) was 76% for Cysteine peptide and 62% for Lysine peptide and the Maximum Standard Deviation (SD) for the positive control replicate was 1.85% for percent Cysteine depletion and 2.83% for percent Lysine depletion. These values met the acceptance criteria for the positive control.

E. **Peptide Reactivity Assay-**

The % peptide depletion following incubation of the test item in cysteine and lysine peptide solutions (1:10 and 1:50 ratio, respectively) was determined by HPLC analysis. On each day, before the HPLC analysis, samples were inspected visually for precipitation or phase separation. Precipitation was not observed in any vial after 24 h of incubation, except for neat test item and neat co-elution control with lysine peptide. The same samples were centrifuged at 400 x g for 10 minutes at 25 ºC to remove the precipitation before analysis.
Following the incubation with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) (100 mM and neat chemical), the percent peptide depletion obtained for Cysteine and Lysine is as tabulated below. The maximum standard deviation of the test item (100 mM and neat chemical) replicates met the acceptability criteria of <14.9% for percent cysteine depletion and <11.6% for percent lysine depletion. Co-elution of test item did not occur with either Cysteine or Lysine peptides. So, the Cysteine 1:10/lysine 1:50 prediction model was used to assign a reactivity category.

TABLE 1: Percent Peptide Depletion:

Sample Name and Date	Percent Peptide Depletion (Cysteine)			Percent Peptide Depletion (Lysine)		
	%	SD	Expected	%	SD	Expected

	Depletion		Depletion	Depletion		Depletion
Cinnamaldehyde (Positive control)	**76**	1.85	60.8-100	**62**	2.83	40.2-69
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) **100 mM**	**0**	0.1	-	**0**	0.75	-
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) **Neat**	**29**	2.37	-	**73**	0.63	-

TABLE 2: % Mean Depletion of Peptides and DPRA prediction

Test Item Name	Actual % Depletion (Cysteine)	Actual % Depletion (Lysine)	% Mean Depletion (Cysteine and Lysine)	Maximum Standard Deviation (Cysteine)	Maximum Standard Deviation (Lysine)	DPRA Prediction
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) **100 mM**	0	0	0	0.1	0.75	No Reactivity (Negative)
ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) **Neat**	29	73	51	2.37	0.63	High Reactivity (Positive)

F. **Data Evaluation**- The reactivity category assigned to the test item (100 mM) was "No Reactivity" using the Cysteine 1:10/lysine 1:50 prediction model, based on the mean Cysteine and Lysine % depletion. This gives a prediction of Negative for skin sensitisation. In contrast, the reactivity category assigned to the neat test item was "High Reactivity" using the Cysteine 1:10/lysine 1:50 prediction model, based on the mean Cysteine and Lysine % depletion. The Non-concordant results between 100 mM and Neat Test item leads to a prediction that the test item is inconclusive for skin sensitisation.

G. **CONCLUSION-** From the results of this study, under the specified experimental conditions, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is predicted by DPRA to be inconclusive.

A 1.7.4 Study 4

Comments of zRMS:	~~From the scientific point of view study Skin Sensitisation Study of ADM.03503.F.1.A Ramesh Verma, 2021 is valid however due to the availability of toxicity assessment based on calculation method described in Regulation (EC) No 1272/2008, study has been considered as a supplementary.~~ *In Vivo* studies has been considered as primary source of information regarding hazard classification, thus classification based on the complete composition of the plant protection or *in vitro* studies has not been taken into account (please refer zRMS consideration in the preface to this dRR reflecting cMS comments). Study has been reviewed for compliance with the current guidelines, resulting from scientific progress. Results of the study and conclusions are adequate for risk assessment and classification purpose. Study accepted.

Reference **KCP 7.1.6/03**

Report author:	[redacted]
Report year ;	2021
Report title:	Skin Sensitisation Study of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) by Local Lymph Node Assay in Mice

Report no:	[redacted]
Reference number:	[redacted]
Guidelines followed in study:	OECD 429 (2010)
Deviations from current test guideline:	No
GLP/Officially recognised testing facilities:	Yes

EXECUTIVE SUMMARY: This study was conducted to evaluate the skin sensitisation potential of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) in CBA/J strain mice, using the local lymph node assay (LLNA). The test item was formulated in 1% Pluronic®L92 (1% L92) following a solubility assessment. A preliminary assay was conducted to assess toxicity and to determine suitable concentrations for the main study based on signs of irritation (ear thickness and erythema).

For the main LLNA, five groups of mice (each comprising 5 females) were treated with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) at concentrations of 50% and 75% (v/v) in 1% L92 and 100% (undiluted), alongside a vehicle control group treated with 1% L92 and a positive control group treated with HCA (α-hexylcinnamaldehyde) at a concentration of 25% (v/v) in 1% L92. The test or control item was applied to the dorsum of both ears (25 µL per ear) on day 0, 1, and 2. On day 5, a dose ^{3}H-methyl thymidine was injected intravenously into each mouse, and 5 hours later, the auricular (local) lymph nodes were removed. Pairs of nodes from each mouse were pooled, and suspensions of the cellular components were prepared in 5% w/v trichloroacetic acid and processed through a scintillation counter. The mean DPM value for each test group was divided by the mean DPM for the control group to provide the stimulation index (SI) value for each test group. Stimulation Index (SI) values of greater than 3 are considered as a positive indicator for skin sensitisation potential.

Following the treatment with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC), there were no signs of systemic toxicity or local irritation in treated mice. At all tested concentrations of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated groups, the SI values obtained showed a lower than three-fold increase compared to the vehicle control value, as shown below:

Concentration of Test Item in Vehicle	**50% (v/v)**	**75% (v/v)**	**100% (undiluted)**	**25% (v/v) HCA Positive Control**
Stimulation Index	1.86	1.92	2.39	8.53

In all mice treated with 25% (v/v) HCA, a local reaction consisting of very slight erythema (score of 1) was observed from day 1 to 4. A positive response for HCA (SI = 8.53) confirmed the reliability of the test procedure.

Therefore, based on the results of this local lymph node assay, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) did not demonstrate skin sensitisation potential at the tested concentrations

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Unclassified**

Under the experimental conditions, ADM.03503.F.1.A is not a skin sensitiser. Thus, **no classification is required according to Regulation (EC) No. 1272/2008.**

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements are provided. There was no deviation from regulatory requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1.	**Test Item:**	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)
	Description:	Yellowish to brownish, transparent liquid

	Lot/Batch #:	1162-230719-011
	Analysed Concentration:	Fluxapyroxad: 7.17% w/w (SD ± 0.06) (77.4 g/L) (SD ± 0.6) Prothioconazole : 13.7% w/w (SD ± 0.1) (148 g/L) (SD ± 1)
	Date of Expiry	05 September 2021

2. <u>Vehicle and/or Positive Control</u>: Test item formed homogenous emulsion in 1% Pluronic®L92 (up to 75%). Based on the results of the solubility and in consultation with the Sponsor 1% Pluronic®L92 was selected as vehicle. α-hexylcinnamaldehyde was used as positive control at the concentration of 25% in 1% Pluronic®L92

3.	**<u>Test Animals</u>:**		
	Species:	Mice	
	Strain:	(CBA/J)	
	Age/Weight at Dosing:	8 to 11 weeks, Weight (g) Minimum: 18.0, Maximum: 24.1 g	
	Source:	Animal Breeding Facility, Jai Research Foundation	
	Housing:	One to two mice/cage	
	Feed:	Teklad certified Global High Fiber Rat and Mice Feed manufactured by Harlan, U.S.A. *ad libitum* with the exception of overnight fasting, prior to dosing, until three hours post-dosing.	
	Water:	UV sterilized water filtered through Kent Reverse Osmosis water filtration system was provided *ad libitum*	
	Environmental Conditions:	**Temperature:** **Humidity:** **Air Changes:** **Photoperiod:**	19 to 23 °C 56 to 65% Minimum 15 air changes/h 12 h dark/ 12 h light
	Acclimation Period:	5 days	

B. <u>STUDY DESIGN AND METHODS</u>

2. <u>In-life Dates</u> –

Start: 02 April 2021 End: 04 May 2021

1. Preliminary assay: A Preliminary assay was performed on CBA/J mice using four doses (2 females/dose), at test item concentrations of 25, 50 and 75 % (w/v) in 1% L92, and undiluted test item for three consecutive days. The 100% formulation was considered to be a suitable maximum dose level for the study. The application of the material and the local effects on the animals were considered acceptable for a valid LLNA.

2. <u>Animal Assignment and Treatment</u> - Based on the preliminary assay, the concentrations of ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) chosen for the main study were 50% and 75% (v/v) in 1% L92 and 100% (undiluted).

Groups of five female mice were treated topically for three consecutive days (days 0, 1 and 2) on the dorsal surface of both ears (25 µL/ear) using a calibrated micropipette, as follows:

Group N°	Dose Concentration (%)	Animal Numbers
G1	1% L92 (Vehicle Control)	1–5
G2	25% (v/v) HCA in 1% L92 (Positive Control)	6–10
G3	50% (v/v) test item in 1% L92	11–15
G4	75% (v/v) test item in 1% L92	16–20
G5	100% test item (undiluted)	21–25

Mice from the vehicle control group (G1) and positive control group (G2) were handled in the same manner but received 25 µL/ear of vehicle (1% L92) or 25% α-Hexylcinnamaldehyde (v/v) in vehicle (1% L92), respectively.

On day 5 all mice were injected, via the tail vein, with 250 µL of sterile phosphate buffered saline (PBS) containing approximately 20 µCi of ^{3}H-methyl thymidine.

3. Proliferation assay: On day 5, 5 hours post-administration of ^{3}H-methyl thymidine, all mice were euthanised. The draining auricular lymph nodes from each individual mouse were excised and collected in separate petri-dishes containing phosphate buffered saline (PBS).

Preparation of cell suspension: A single cell suspension of lymph node cells was prepared by gentle mechanical disaggregation. Lymph node cells were pelleted by centrifugation, the supernatants were discarded. The washing step was repeated two times. After the final wash, the supernatant was removed leaving a minimal volume (approximate 0.5 mL) of supernatant above each pellet.

Each pellet was agitated before re-suspending with 3 mL of 5% trichloroacetic acid (TCA) and kept in the refrigerator for approximately 18 hours for precipitation of macromolecules. After incubation with 5% TCA at 4 °C, each precipitate was recovered by centrifugation for 10 minutes and the supernatant was removed. The precipitate was re-suspended in 1 mL of 5% TCA. Each precipitate was transferred to a scintillation vial with 10 mL of scintillation fluid and thoroughly mixed. The vials were loaded into a β-scintillation counter to measure the 3H-methyl thymidine incorporation. Background 3H-methyl thymidine level was measured from 1 mL aliquots of 5% TCA.

4. Data evaluation: Disintegrations per minute (DPM) were measured for each mouse and expressed as DMP/mouse corrected for background DPM.

The DPM/mouse, along with an appropriate measure of inter-animal variability (i.e., mean ± standard deviation), were calculated for each test group and vehicle and positive control groups. Final results were expressed as the Stimulation Index (SI) which is calculated as a ratio of the mean DPM of test group divided by mean DPM of vehicle control group. The test item is regarded as a skin sensitiser if the SI for a dose group is 3 or above, together with consideration of a dose-response relationship, providing the application sites have not shown excessive irritation. The test article is regarded as a sensitiser when the maximum value of the SI is greater than 3.0.

II. RESULTS AND DISCUSSION

No mortality or signs of systemic toxicity was observed during the study.

No erythema was observed at the site of the application in vehicle control, and groups treated with ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) at concentrations of 50% and 75% (v/v) in 1% L92 and 100% (undiluted) from day 0 to day 5. A local reaction consisting of very slight erythema (score of 1) was observed from day 1 to 4 in all mice treated with 25% (v/v) HCA.

D. Group Mean DPM

Proliferative responses in the draining lymph nodes were monitored by measuring the incorporation of ^{3}H-methyl thymidine. Group mean DPM values for the treated and control groups are shown below:

Vehicle (1% L92)	**Positive control (25% HCA)**	**ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)**		
		50% (v/v) in 1% L92	**75% (v/v) in 1% L92**	**100% (undiluted)**
482.60	4118.00**	896.60	927.20	1151.60*

* Significantly higher than control (p≤0.05), ** Significantly higher than control (p≤0.01)

A statistically significant increase in DPM was observed in 25% (v/v) HCA and in the 100% ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) treated groups when compared to control group values.

E. Stimulation Index (SI Value) and EC_3 Value

Stimulation index (SI) values (i.e. the ratios of the mean scintillation counts obtained from the test groups relative to the corresponding mean scintillation count obtained from controls) for groups treated with

ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) and for the positive control group are shown below:

Positive control (25% HCA)	ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC)		
	50% (v/v) in 1% L92	75% (v/v) in 1% L92	100% (undiluted)
8.53	1.86	1.92	2.39

The SI obtained for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) at all tested concentrations showed a less than three-fold increase over the control value. Therefore, the EC_3 value was not determined.

E. Conclusion:

Based on the results of this local lymph node assay, ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) did not demonstrate skin sensitisation potential at the tested concentrations.

Based on the results of this study, an indication of the classification for ADM.03503.F.1.A (Fluxapyroxad 75 Prothioconazole 150 g/L EC) is as follows:

Globally Harmonized System of Classification and Labelling of Chemicals (GHS 2019): **Unclassified**

Under the experimental conditions, ADM.03503.F.1.A is not a skin sensitiser. Thus, **no classification is required according to Regulation (EC) No. 1272/2008.**

A 1.8 Supplementary studies for combinations of plant protection products (KCP 7.1.7)

Not required.

A 1.9 Data on co-formulants (KCP 7.4)

A copy of the material safety data sheet of the product ADM.03503.F.1.A is located under KCP 7.4.

A 1.9.1 Material safety data sheet for each co-formulant

Information regarding material safety data sheets of the co-formulants can be found in the confidential dossier of this submission (Registration Report - Part C).

A 1.9.2 Available toxicological data for each co-formulant

Available toxicological data for each co-formulant can be found in the confidential dossier of this submission (Registration Report - Part C).

A 1.10 Studies on dermal absorption (KCP 7.3)

A 1.10.1 Study 1 – Fluxapyroxad in ADM.03503.F.1.A

Comments of zRMS:	Dermal absorption study on fluxapyroxad has been conducted according to the OECD TG 428 revision 2004. The dermal penetration of fluxapyroxad formulated as ADM.03503.F.1.A through human dermatomed skin was determined *in vitro*. ~~The amount of applied dose penetrating within 24 hours was determined to be 3.28 ± 1.74, 7.61 ± 1.69 and 9.06 ± 2.80 for the formulation concentrate, spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 5.0%, 9.0 and 12% for the formulation concentrate, spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).~~ zRMS: Residues found in unexposed skin are not normally included in the absorption esti-

	mate. Unexposed skin is not subject to skin washing during the study and tape stripping of that area is not likely to have been conducted effectively. Therefore, it is likely that any residue detected is as a result of leakage under the cell flange and not lateral diffusion. For the final absorption estimates [^{14}C]-Fluxapyroxad detected in unexposed skin has been included since the possibility of lateral diffusion cannot be excluded. However, it should be noted that this is considered highly conservative since the unexposed skin will not have been subject to skin washing during the study and tape stripping is unlikely to have been effective. Unexposed skin is normally only considered when recovery is low and leakage under the cell flange is suspected, as was the case here for Cells 22 and 23. The dermal penetration of fluxapyroxad formulated as ADM.03503.F.1.A through human dermatomed skin was determined *in vitro*. The amount of applied dose penetrating within 24 hours was determined to be 3.37 ± 1.79, 7.98 ± 1.85 and 9.81 ± 3.59 for the formulation concentrate, spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 5.2%, 9.5 and 13% for the formulation concentrate, spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).

Reference	**KCP 7.3/01**
Report	The *In Vitro* Percutaneous Absorption of Radiolabelled Fluxapyroxad in a Concentrate and Two In-Use Dilutions of the Fluxapyroxad 75 g/L + Prothioconazole 150 g/L EC Formulation (ADM.03503.F.1.A) Through Human Split-Thickness Skin, [redacted], [redacted]
Guidelines	OECD Guideline for Testing of Chemicals, Guideline 428: Skin Absorption: *In Vitro* Method (2004). OECD Environmental Health and Safety Publications Series on Testing and Assessment No. 28. Guidance Document for the Conduct of Skin Absorption Studies (2004). European Commission Guidance Document on Dermal Absorption – Sanco/222/2000/Rev.7 (19 March 2004).
Deviations	No
GLP	Yes
Acceptability	Yes
Duplication (if vertebrate study)	Not applicable

Summary

The test item, Fluxapyroxad, is an active ingredient used in an emulsifiable concentrate (EC) formulation (ADM.03503.F.1.A). The concentration of Fluxapyroxad in the concentrate formulation was ca 75 g/L. The highest concentration in-use spray dilution was ca 0.234375 g/L. The lowest concentration in-use spray dilution was ca 0.1875 g/L.

As part of the safety evaluation of Fluxapyroxad, this study was conducted to assess the rate and extent of absorption of Fluxapyroxad following topical application of the concentrate formulation and two in-use spray dilutions to human split-thickness skin.

Split-thickness human skin membranes were mounted into flow-through diffusion cells in vitro. Receptor fluid was pumped underneath the skin at a flow rate of 1.5 mL/h ± 0.15 mL/h. The skin surface temperature was maintained at 32 °C ± 1 °C throughout the experiment. An electrical resistance barrier integrity assessment was performed and any skin sample exhibiting resistance lower than 7.7 kΩ was excluded from subsequent absorption measurements.

The test preparations were applied (10 µL/cm^2) to human split-thickness skin membranes from four different donors and the cells were left open to the atmosphere. Test item stability during dosing was con-

firmed by high performance liquid chromatography (HPLC).

Absorption was assessed by collecting receptor fluid in hourly fractions from 0 to 8 h post dose and then 2 hourly fractions from 8 to 24 h post dose. At 8 h post dose, the exposure period was terminated by washing the skin surface with a concentrated commercial hand wash soap followed by rinsing with a dilute soap solution (2%, v/v) and drying the surface with tissue paper. At 24 h post dose, the skin was washed as previously described and the underside of the skin was rinsed with receptor fluid for all cells. The cells were dismantled and the donor chamber and receptor chamber were retained separately for analysis. The stratum corneum was tape stripped and the skin divided into exposed and unexposed skin. The skin samples were dissolved with Solvable™ tissue solubilizer. All samples were analysed by liquid scintillation counting.

For the concentrate, Cells 5 and 6 showed high donor wash, unexposed skin and receptor wash values compared to other cells which is indicative of leaking of the formulation from the exposed area therefore these cells were excluded from mean results.

For spray dilution 1, all cells are included in the mean calculation.

For spray dilution 2, Cell 22 exhibited high donor wash and receptor wash values compared to other cells which is indicative of leaking from the exposed area. Cell 23 also showed a high receptor wash value with an atypical absorption profile, suggesting damage during the 8 h wash. For these reasons Cells 22 and 23 were excluded from mean results.

In conclusion, following topical application of [^{14}C]-Fluxapyroxad in concentrate, spray dilution 1 and spray dilution 2 to human skin in vitro, the total absorbed dose was 0.43% (3.28 µg equiv./cm^2), 2.11% (49.5 ng equiv./cm^2) and 1.87% (35.2 ng equiv./cm^2) of the applied dose, respectively. The dermal delivery was 1.71% (13.1 µg equiv./cm^2), 5.55% (130 ng equiv./cm^2) and 5.96% (112 ng equiv./cm^2) of the applied dose, respectively. The potentially absorbable dose was 3.29% (25.2 µg equiv./cm^2), 7.61%(178 ng equiv./cm^2) and 9.07% (171 ng equiv./cm^2) of the applied dose, respectively. The mass balance was 100.46% (770 µg equiv./cm^2), 96.37% (2256 ng equiv./cm^2) and 97.63% (1841 ng equiv./cm^2) of the applied dose, respectively.

Materials and methods

Test material	Name (Lot/Batch No.)	[14C]-Fluxapyroxad (900-2301)
	Test preparation	Spiking
	Specific activity	5.69 MBq/mg (153.8 µCi/mg)
	Radiochemical purity	98.8%
Product	Name (Lot/Batch No.)	Fluxapyroxad 75 + Prothoconazole 150 g/L EC (1162-230719-011)
	Company code	ADM.03503.F.1.A
	Concentration a.s.	75 g/L
	Formulation type	Emulsifiable concentrate (EC)
Blank product	Name (Lot/Batch No.)	Blank Formulation of Fluxapyroxad 75 + Prothoconazole 150 EC (3438-190420-02)
	Concentration a.s.	0 g/L

Test system		
Diffusion cell	Cell type	dynamic
	(if dynamic) Flow rate	1.5 mL/h
	Exposed skin area	0.64 cm²
	Cover	occlusive
Membrane	Skin type	dermatomed
	Skin thickness range	200-400 µm
	Skin donors age	42-72
	Skin donors sex	Male and female
	Location	Abdomen and buttocks
	Source	Unknown

	Integrity test	Yes, electrical resistance
Receptor	Receptor medium	phosphate buffered saline containing polyoxyethylene 20 oleyl ether (PEG, ca 6%, w/v), sodium azide (ca 0.01%, w/v), streptomycin (0.1 mg/mL) and penicillin (ca 100 units/mL).
	Solubility in receptor medium	Yes – 0.744 g/L tested
Sample Time	Exposure time	8 hours
	Observation time	24 hours
Sampling	Sample intervals	hourly for first 8 hours then every two hours until 24 hours.
Washing		Post exposure - washed with soap (50 µL) and then rinsed with soap solution (10 × 0.5 mL)
Final Procedure	Tape stripping	y
	TS1-2 analysed separately	y
Remarks:		

Tested doses	Concentrate	Spray dilution 1	Spray dilution 2
Target concentration [mg/mL]	75	0.234375	0.1875
Area dose [µg/cm²]	750	2.34375	1.875
Total dose [µg/cell]	480	1.5	1.2
Specific activity	1.84 µCi/mg	153.8 µCi/mg	153.8 µCi/mg
No. of donors	4	4	4
No of cells used/valid cells*	8/6	8/8	8/6

* Concentrate: Cell 5 and Cell 6 excluded due to high values for donor wash, unexposed skin and receptor wash. Dilution 2: Cell 22 excluded due to high donor wash and receptor wash values. Cell 23 excluded due to high receptor wash and atypical absorption profile suggesting damage at 8h wash.

Results and discussions

Table A 1: *In-vitro* dermal penetration of fluxapyroxad formulated as ADM.03503.F.1.A through human skin - Recovery data

	Fluxapyroxad					
Dose group	Formulation concentrate		Spray dilution 1		Spray dilution 2	
Target concentration [mg/mL]	75		0.234375		0.1875	
Target dose [µg/cm²]	750		2.34375		1.875	
Mean actual applied dose [µg/cm²]	767		2.34		1.89	
	Recovery [%]		Recovery [%]		Recovery [%]	
	Mean	S.D.	Mean	S.D.	Mean	S.D.
Dislodgeable dose						
Skin washing after 8 h + 24 h	92.80	5.39	87.18	3.50	85.47	7.05
Donor chamber wash	2.53	2.59	0.51	0.32	1.10	0.63
Dose associated to skin						
Tape strips: 1st sample, strips 1 + 2	1.76	1.09	0.70	0.41	1.26	0.78
Tape strips: 2nd sample; strips 3 - n	1.58	0.84	2.06	0.89	3.11	1.44
Skin preparation	1.28	0.69	3.43	1.23	4.09	1.75
Unexposed skin	0.08	0.16	0.38	0.27	0.74	0.98
Absorbed dose						
Receptor fluid	0.03	0.02	1.61	0.68	1.37	0.60
Receptor chamber wash	0.39	0.70	0.50	0.12	0.49	0.19
Total recovery[1]	100.46	0.59	96.38	2.53	97.63	4.70
Absorption essentially complete at end of study (>75% absorption within half the study duration) [%Absorption at $t_{0.5}$]	No [23.37 ± 10.65]		No [50.70 ± 6.78]		No [46.69 ± 7.13]	
If no: Absorption estimates = absorbed dose + skin preparation + tape strips sample 2)[2]	3.37	1.79	7.98	1.85	9.81	3.56
If yes: Absorption estimates = absorbed dose + skin preparation)[2]	N/A	N/A	N/A	N/A	N/A	N/A

Absorption estimate normalised[3]	3.37	1.79	7.98	1.85	9.81	3.56
Relevant absorption estimate[4]	5.155		9.534		13.364	
Absorption estimates used for risk assessment[5]	5.2		9.5		13	

	Fluxapyroxad					
Dose group	Formulation concentrate		Spray dilution 1		Spray dilution 2	
Target concentration [mg/mL]	75		0.234375		0.1875	
Target dose [µg/cm²]	750		2.34375		1.875	
Mean actual applied dose [µg/cm²]	767		2.34		1.89	
	Recovery [%]		Recovery [%]		Recovery [%]	
	Mean	S.D.	Mean	S.D.	Mean	S.D.
Dislodgeable dose						
Skin washing after 8 h + 24 h	92.88	5.41	87.56	3.41	86.21	6.62
Donor chamber wash	2.53	2.59	0.51	0.32	1.10	0.63
Dose associated to skin						
Tape strips: 1st sample, strips 1 + 2	1.76	1.09	0.70	0.41	1.26	0.78
Tape strips: 2nd sample; strips 3 – n	1.58	0.84	2.06	0.89	3.11	1.44
Skin preparation	1.28	0.69	3.43	1.23	4.09	1.75
Absorbed dose						
Receptor fluid	0.03	0.02	1.61	0.68	1.37	0.60
Receptor chamber wash	0.39	0.70	0.50	0.12	0.49	0.19
Total recovery[1]	100.46	0.59	96.38	2.53	97.63	4.70
Absorption essentially complete at end of study (>75% absorption within half the study duration) [%Absorption at $t_{0.5}$]	No [23.37 ± 10.65]		No [50.70 ± 6.78]		No [46.69 ± 7.13]	
If no: Absorption estimates = absorbed dose + skin preparation + tape strips sample 2)[2]	3.28	1.74	7.61	1.69	9.06	2.80
If yes: Absorption estimates = absorbed dose + skin preparation)[2]	N/A	N/A	N/A	N/A	N/A	N/A
Absorption estimate normalised[3]	3.28	1.74	7.61	1.69	9.06	2.80
Relevant absorption estimate[4]	5.027		9.023		11.865	
Absorption estimates used for risk assessment[5]	**5.0**		**9.0**		**12**	

1 Values may not calculate exactly due to rounding of figures

2 In accordance with the EFSA Guidance on Dermal Absorption (EFSA Journal 2017;15(6):4873) the radioactivity in the second tape-strip pool (3rd to nth tape strip) is considered potentially absorbable if less than 75% of the absorption occurred in the first half of the study (see Table 7.6.2-1) Finally, the skin preparation is also considered potentially absorbable.

3 According to the EFSA Guidance on Dermal Absorption, cells with insufficient recovery (< 95%) can be corrected by normalisation of absorption estimate to 100% recovery; explanation should be included.

4. In accordance with the EFSA Guidance on Dermal Absorption, the standard deviation (modified by the appropriate multiplication factor) was added to the mean% dermal penetration.

5 Relevant absorption estimate was rounded to the required number of significant figures.

N/A: not applicable

Remarks

For the concentrate, Cells 5 and 6 showed high donor wash, unexposed skin and receptor wash values compared to other cells which is indicative of leaking of the formulation from the exposed area therefore these cells were excluded from mean results.

For spray dilution 2, Cell 22 exhibited high donor wash and receptor wash values compared to other cells which is indicative of leaking from the exposed area. Cell 23 also showed a high receptor wash value with an atypical absorption profile, suggesting damage during the 8 h wash. For these reasons Cells 22 and 23 were excluded from mean results.

For the final absorption estimates [^{14}C]-Fluxapyroxad detected in unexposed skin has been included since the possibility of lateral diffusion cannot be excluded. However, it should be noted that this is considered highly conservative since the unexposed skin will not have been subject to skin washing during the study

and tape stripping is unlikely to have been effective. Unexposed skin is normally only considered when recovery is low and leakage under the cell flange is suspected, as was the case here for Cells 22 and 23.

Conclusion/endpoint:

~~The dermal penetration of fluxapyroxad formulated as ADM.03503.F.1.A through human dermatomed skin was determined *in vitro*. The amount of applied dose penetrating within 24 hours was determined to be 3.28 ± 1.74, 7.61 ± 1.69 and 9.06 ± 2.80 for the formulation concentrate, spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 5.0%, 9.0 and 12% for the formulation concentrate, spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).~~

The dermal penetration of fluxapyroxad formulated as ADM.03503.F.1.A through human dermatomed skin was determined *in vitro*. The amount of applied dose penetrating within 24 hours was determined to be 3.37 ± 1.79, 7.98 ± 1.85 and 9.81 ± 3.59 for the formulation concentrate, spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 5.2%, 9.5 and 13% for the formulation concentrate, spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).

A 1.10.2 Study 2 – Prothioconazole-desthio in ADM.03503.F.1.A

Comments of zRMS:	Dermal absorption study on prothioconazole-desthio has been conducted according to the OECD TG 428 revision 2004. For testing human split thickness skin has been used. There were no deviations from the TG. ~~The amount of applied dose penetrating within 24 hours was determined to be 9.71 ± 2.58 and 11.08 ± 2.20 for spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 12% and 13% for spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).~~ Results of the DA study and conclusions are adequate for risk assessment (NDE) Study accepted. zRMS: Residues found in unexposed skin are not normally included in the absorption estimate. Unexposed skin is not subject to skin washing during the study and tape stripping of that area is not likely to have been conducted effectively. Therefore, it is likely that any residue detected is as a result of leakage under the cell flange and not lateral diffusion. For the final absorption estimates [^{14}C]-Prothioconazole-desthio detected in unexposed skin has been included since the possibility of lateral diffusion cannot be excluded. However, it should be noted that this is considered highly conservative since the unexposed skin will not have been subject to skin washing during the study and tape stripping is unlikely to have been effective. Unexposed skin is normally only considered when recovery is low and leakage under the cell flange is suspected. The dermal penetration of prothioconazole formulated as ADM.03503.F.1.A through human dermatomed skin was determined in vitro. The amount of applied dose penetrating within 24 hours was determined to be 9.82 ± 2.60 and 11.83 ± 1.65 for spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 12% and 13% for spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).

Reference	**KCP 7.3/02**
Report	The *In Vitro* Percutaneous Absorption of Radiolabelled Prothioconazole-desthio in Two In-Use Dilutions of the Fluxapyroxad 75 g/L + Prothioconazole 150 g/L EC Formulation (ADM.03503.F.1.A) Through Human Split-Thickness Skin, [redacted], [redacted]
Guidelines	OECD Guideline for Testing of Chemicals, Guideline 428: Skin Absorption: *In Vitro* Method (2004). OECD Environmental Health and Safety Publications Series on Testing and Assessment No. 28. Guidance Document for the Conduct of Skin Absorption Studies (2004).

	European Commission Guidance Document on Dermal Absorption – Sanco/222/2000/Rev.7 (19 March 2004).
Deviations	No
GLP	Yes
Acceptability	Yes
Duplication (if vertebrate study)	Not applicable

Summary

The test item, Prothioconazole-desthio, is the main degradation product of the active ingredient Prothioconazole used in an emulsifiable concentrate (EC) formulation (ADM.03503.F.1.A). The concentration of Prothioconazole-desthio in the highest concentration in-use spray dilution was ca 0.46875 g/L. The lowest concentration in-use spray dilution was ca 0.375 g/L.

As part of the safety evaluation of Prothioconazole-desthio, this study was conducted to assess the rate and extent of absorption of Prothioconazole-desthio following topical application of two in-use spray dilutions to human split-thickness skin.

Split-thickness human skin membranes were mounted into flow-through diffusion cells in vitro. Receptor fluid was pumped underneath the skin at a flow rate of 1.5 mL/h ± 0.15 mL/h. The skin surface temperature was maintained at 32 °C ± 1 °C throughout the experiment. An electrical resistance barrier integrity assessment was performed and any skin sample exhibiting resistance lower than 7.7 kΩ was excluded from subsequent absorption measurements.

The test preparations were applied (ca 10 µL/cm^2) to human split-thickness skin membranes from four different donors and the cells were left open to the atmosphere. Test item stability during dosing was confirmed by high performance liquid chromatography (HPLC).

Absorption was assessed by collecting receptor fluid in hourly fractions from 0 to 8 h post dose and then 2 hourly fractions from 8 to 24 h post dose. At 8 h post dose, the exposure period was terminated by washing the skin surface with a concentrated commercial hand wash soap followed by rinsing with a dilute soap solution (2%, v/v) and drying the surface with tissue paper. At 24 h post dose, the skin was washed as previously described and the underside of the skin was rinsed with receptor fluid for all cells. The cells were dismantled and the donor chamber and receptor chamber were retained separately for analysis. The stratum corneum was tape stripped and the skin divided into exposed and unexposed skin. The skin samples were dissolved with Solvable™ tissue solubilizer. All samples were analysed by liquid scintillation counting.

For the spray dilution 1, Cell 7 and Cell 8 were excluded from the mean and standard deviation (SD) results, due to atypical absorption profiles and consequently high values for receptor fluid. The justifications for exclusion that follow are derived from the examination of the results within the Spray dilution 1 and between Spray dilution 1 and Spray dilution 2 as the concentration of the test item in both preparations are very similar (0.46875 g/L versus 0.375 g/L). Both cells 7 and 8 are from the same donor (1090). Cells 15 and 16, dosed with Spray dilution 2, are also from donor 1090. Receptor fluid values for Cell 7 and Cell 8 are 58.02% and 40.22%, respectively, which are around 10 × higher than seen for all other cells in the group as well as Cell 15 and Cell 16, i.e. 5.99% and 4.23%, respectively. Across the 16 cells dosed with [^{14}C]-Prothioconazole-desthio, cells 7 and 8 clearly show a different absorption profile. Cells 7 and 8 reach 50% absorption in the first 2 h following dosing. For all other cells, 50% absorption is reached between 6 and 12 h following dosing. The observed difference in absorption profile starting at hour 0 could have been caused by a damage that occurred during dosing. The arguments mentioned above highlight that Cell 7 and Cell 8 are clear outliers, with absorption profiles very different to all other cells across both groups, which follow the definition of an outlier “an observation which deviates so much

from other observations as to arouse suspicions that it was generated by a different mechanism" (Hawkins 1980). For this reason, it was deemed appropriate to exclude Cell 7 and Cell 8 from the mean and SD results.

In conclusion, following topical application of [^{14}C]-Prothioconazole-desthio in Spray dilution 1 and Spray dilution 2 to human skin *in vitro*, the total absorbed dose was 5.53% (0.28 µg equiv./cm^2) and 6.58% (0.26 µg equiv./cm^2) of the applied dose, respectively. The dermal delivery was 8.79% (0.44 µg equiv./cm^2) and 10.29% (0.40 µg equiv./cm^2) of the applied dose, respectively. The potentially absorbable dose was 9.70% (0.49 µg equiv./cm^2) and 11.09% (0.44 µg equiv./cm^2) of the applied dose, respectively. The mass balance was 98.55% (4.94 µg equiv./cm^2) and 99.15% (3.89 µg equiv./cm^2) of the applied dose, respectively.

Materials and methods

Test material	Name (Lot/Batch No.)	[^{14}C]-Prothioconazole-desthio (XXVI/5/C/1)
	Test preparation	Spiking
	Specific activity	2.425 MBq/mg (65.5 µCi/mg)
	Radiochemical purity	99.45%
Product	Name (Lot/Batch No.)	Fluxapyroxad 75 + Prothoconazole 150 g/L EC (1162-230719-011)
	Company code	ADM.03503.F.1.A
	Concentration a.s.	150 g/L
	Formulation type	Emulsifiable concentrate (EC)
Blank product	Name (Lot/Batch No.)	Blank Formulation of Fluxapyroxad 75 + Prothoconazole 150 EC (1162-230719-011)
	Concentration a.s.	0 g/L

Test system		
Diffusion cell	Cell type	dynamic
	(if dynamic) Flow rate	1.5 mL/h
	Exposed skin area	0.64 cm²
	Cover	occlusive
Membrane	Skin type	dermatomed
	Skin thickness range	200-400 µm
	Skin donors age	35-56
	Skin donors sex	Female
	Location	Abdomen
	Source	Unknown
	Integrity test	Yes, electrical resistance
Receptor	Receptor medium	phosphate buffered saline containing polyoxyethylene 20 oleyl ether (PEG, ca 6%, w/v), sodium azide (ca 0.01%, w/v), streptomycin (0.1 mg/mL) and penicillin (ca 100 units/mL).
	Solubility in receptor medium	Yes – 0.0137 g/L tested
Sample Time	Exposure time	8 hours
	Observation time	24 hours
Sampling	Sample intervals	hourly for first 8 hours then every two hours until 24 hours.
Washing		Post exposure - washed with soap (50 µL) and then rinsed with soap solution (10 × 0.5 mL)
Final Procedure	Tape stripping	y
	TS1-2 analysed separately	y
Remarks:		

Tested doses	Spray dilution 1	Spray dilution 2
Target concentration [mg/mL]	0.46875	0.375
Area dose [µg/cm²]	4.6875	3.75
Total dose [µg/cell]	3.0	2.4
Specific activity	65.5 µCi/mg	65.5 µCi/mg
No. of donors	4	4

No of cells used/valid cells*	8/6	8/8

* Spray dilution 1: Cells 7 and 8 were excluded due to high values for receptor fluid and absorption profiles not consistent with the other cells.

Results and discussions

Table A 2: *In-vitro* dermal penetration of prothioconazole-desthio formulated as ADM.03503.F.1.A through human skin - Recovery data

		Prothioconazole-desthio			
Dose group		Spray dilution 1		Spray dilution 2	
Target concentration	[mg/mL]	0.46875		0.375	
Target dose	[µg/cm²]	4.6875		3.75	
Mean actual applied dose	[µg/cm²]	4.99		3.88	
		Recovery [%]		Recovery [%]	
		Mean	S.D.	Mean	S.D.
Dislodgeable dose					
Skin washing after 8 h + 24 h		87.70	2.32	86.40	3.80
Donor chamber wash		0.70	0.22	0.64	0.60
Dose associated to skin					
Tape strips: 1st sample, strips 1 + 2		0.34	0.21	0.29	0.26
Tape strips: 2nd sample; strips 3 - n		0.91	0.66	0.80	0.71
Skin preparation		3.27	1.14	3.70	1.68
Unexposed skin		0.11	0.08	0.74	1.22
Absorbed dose					
Receptor fluid		4.77	1.89	5.51	2.51
Receptor chamber wash		0.77	0.31	1.07	0.63
Total recovery[1]		98.55	1.79	99.15	2.61
Absorption essentially complete at end of study (>75% absorption within half the study duration) [%Absorption at $t_{0.5}$]		No [65.36 ± 3.78]		No [58.95 ± 6.23]	
If no: Absorption estimates = absorbed dose + skin preparation + tape strips sample 2)[2]		9.82	2.60	11.83	1.65
If yes: Absorption estimates = absorbed dose + skin preparation)[2]		N/A	N/A	N/A	N/A
Absorption estimate normalised[3]		9.82	2.60	11.83	1.65
Relevant absorption estimate[4]		12.411		13.212	
Absorption estimates used for risk assessment[5]		**12**		**13**	

		Prothioconazole-desthio			
Dose group		Spray dilution 1		Spray dilution 2	
Target concentration	[mg/mL]	0.46875		0.375	
Target dose	[µg/cm²]	4.6875		3.75	
Mean actual applied dose	[µg/cm²]	4.99		3.88	
		Recovery [%]		Recovery [%]	
		Mean	S.D.	Mean	S.D.
Dislodgeable dose					
Skin washing after 8 h + 24 h		87.81	2.33	87.14	4.23
Donor chamber wash		0.70	0.22	0.64	0.60
Dose associated to skin					
Tape strips: 1st sample, strips 1 + 2		0.34	0.21	0.29	0.26
Tape strips: 2nd sample; strips 3 - n		0.91	0.66	0.80	0.71
Skin preparation		3.27	1.14	3.70	1.68
Absorbed dose					
Receptor fluid		4.77	1.89	5.51	2.51
Receptor chamber wash		0.77	0.31	1.07	0.63
Total recovery[1]		98.55	1.79	99.15	2.61
Absorption essentially complete at end of study (>75% absorption within half the study duration) [%Absorption at $t_{0.5}$]		No [65.36 ± 3.78]		No [58.95 ± 6.23]	
If no:		9.71	2.58	11.08	2.20

Absorption estimates = absorbed dose + skin preparation + tape strips sample 2)[2]				
If yes: Absorption estimates = absorbed dose + skin preparation)[2]	N/A	N/A	N/A	N/A
Absorption estimate normalised[3]	9.71	2.58	11.08	2.20
Relevant absorption estimate[4]	12.287		12.933	
Absorption estimates used for risk assessment[5]	**12**		**13**	

1 Values may not calculate exactly due to rounding of figures
2 In accordance with the EFSA Guidance on Dermal Absorption (EFSA Journal 2017;15(6):4873) the radioactivity in the second tape-strip pool (3rd to nth tape strip) is considered potentially absorbable if less than 75% of the absorption occurred in the first half of the study (see Table 7.6.2-1) Finally, the skin preparation is also considered potentially absorbable.
3 According to the EFSA Guidance on Dermal Absorption, cells with insufficient recovery (< 95%) can be corrected by normalisation of absorption estimate to 100% recovery; explanation should be included.
4. In accordance with the EFSA Guidance on Dermal Absorption, the standard deviation (modified by the appropriate multiplication factor) was added to the mean% dermal penetration.
5 Relevant absorption estimate was rounded to the required number of significant figures.
N/A: not applicable

Remarks

For the spray dilution 1, Cell 7 and Cell 8 were excluded from the mean and standard deviation (SD) results, due to atypical absorption profiles and consequently high values for receptor fluid. The justifications for exclusion that follow are derived from the examination of the results within the Spray dilution 1 and between Spray dilution 1 and Spray dilution 2 as the concentration of the test item in both preparations are very similar (0.46875 g/L versus 0.375 g/L). Both cells 7 and 8 are from the same donor (1090). Cells 15 and 16, dosed with Spray dilution 2, are also from donor 1090. Receptor fluid values for Cell 7 and Cell 8 are 58.02% and 40.22%, respectively, which are around 10 × higher than seen for all other cells in the group as well as Cell 15 and Cell 16, i.e. 5.99% and 4.23%, respectively. Across the 16 cells dosed with [^{14}C]-Prothioconazole-desthio, cells 7 and 8 clearly show a different absorption profile. Cells 7 and 8 reach 50% absorption in the first 2 h following dosing. For all other cells, 50% absorption is reached between 6 and 12 h following dosing. The observed difference in absorption profile starting at hour 0 could have been caused by a damage that occurred during dosing. The arguments mentioned above highlight that Cell 7 and Cell 8 are clear outliers, with absorption profiles very different to all other cells across both groups, which follow the definition of an outlier “an observation which deviates so much from other observations as to arouse suspicions that it was generated by a different mechanism” (Hawkins 1980). For this reason, it was deemed appropriate to exclude Cell 7 and Cell 8 from the mean and SD results.

For the final absorption estimates [^{14}C]-Prothioconazole-desthio detected in unexposed skin has been included since the possibility of lateral diffusion cannot be excluded. However, it should be noted that this is considered highly conservative since the unexposed skin will not have been subject to skin washing during the study and tape stripping is unlikely to have been effective. Unexposed skin is normally only considered when recovery is low and leakage under the cell flange is suspected.

Conclusion/endpoint:

~~The dermal penetration of prothioconazole formulated as ADM.03503.F.1.A through human dermatomed skin was determined in vitro. The amount of applied dose penetrating within 24 hours was determined to be 9.71 ± 2.58 and 11.08 ± 2.20 for spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 12% and 13% for spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).~~

The dermal penetration of prothioconazole formulated as ADM.03503.F.1.A through human dermatomed skin was determined in vitro. The amount of applied dose penetrating within 24 hours was determined to be 9.82 ± 2.60 and 11.83 ± 1.65 for spray dilution 1 and spray dilution 2, respectively. The dermal penetration estimates to be used for risk assessment were set at 12% and 13% for spray dilution 1 and spray dilution 2 based on the EFSA guidance criteria (EFSA Journal 2017; 15(6):4873).

A 1.11 Other/Special Studies

None.

Appendix 2 Exposure calculations

A 2.1 Operator exposure calculations (KCP 7.2.1.1)

A 2.1.1 Calculations for Fluxapyroxad

Table A 3: Input parameters considered for the estimation of operator exposure- cereals

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5	Dermal for in use dilution = 9.5	Oral = 68	Inhalation = 100	
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 4: Estimation of longer term operator exposure towards fluxapyroxad according to EFSA guidance – cereals

No PPE

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0251	% of RVNAS	62.69%
	Acute systemic exposure mg/kg bw/day	0.1654	% of RVAAS	
Mixing and Loading	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0154	% of RVNAS	38.50%
	Acute systemic exposure mg/kg bw/day	0.0651	% of RVAAS	

Gloves worn for mixing and loading

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0251	% of RVNAS	62.69%
	Acute systemic exposure mg/kg bw/day	0.1654	% of RVAAS	
Mixing and Loading	Gloves = Yes	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No

Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0017	% of RVNAS	4.14%
	Acute systemic exposure mg/kg bw/day	0.0145	% of RVAAS	

A 2.1.2 Calculations for Prothioconazole

Table A 5: Input parameters considered for the estimation of operator exposure – cereals

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 6: Estimation of longer term operator exposure towards prothioconazole according to EFSA guidance – cereals (100% prothioconazole)

No PPE (100% prothioconazole)

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.2129	% of RVNAS	106.47%
	Acute systemic exposure mg/kg bw/day	1.1961	% of RVAAS	Not required.
Mixing and Loading	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.1332	% of RVNAS	66.58%
	Acute systemic exposure mg/kg bw/day	0.5807	% of RVAAS	Not required.

Gloves worn for mixing and loading (100% prothioconazole)

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.2129	% of RVNAS	106.47%
	Acute systemic exposure mg/kg bw/day	1.1961	% of RVAAS	Not required.
Mixing and Loading	Gloves = Yes	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0204	% of RVNAS	10.22%
	Acute systemic exposure mg/kg bw/day	0.1649	% of RVAAS	Not required.

Table A 7: Input parameters considered for the estimation of operator exposure – cereals (50% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.094 kg a.s. /ha	Spray dilution = 0.752 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

No PPE (50% prothioconazole)

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.1248	% of RVNAS	62.39%
	Acute systemic exposure mg/kg bw/day	0.8312	% of RVAAS	
Mixing and Loading	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0766	% of RVNAS	38.31%
	Acute systemic exposure mg/kg bw/day	0.3405	% of RVAAS	

Gloves worn for mixing and loading (50% prothioconazole)

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.1248	% of RVNAS	62.39%
	Acute systemic exposure mg/kg bw/day	0.8312	% of RVAAS	
Mixing and Loading	Gloves = Yes	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0104	% of RVNAS	5.20%
	Acute systemic exposure mg/kg bw/day	0.0969	% of RVAAS	

A 2.1.3 Calculations for Prothioconazole-desthio

Table A 8: Input parameters considered for the estimation of operator exposure – cereals

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

No PPE (100% prothioconazole-desthio)

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0879	% of RVNAS	879.46%
	Acute systemic exposure mg/kg bw/day	0.4940	% of RVAAS	
Mixing and Loading	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0547	% of RVNAS	546.64%
	Acute systemic exposure mg/kg bw/day	0.2205	% of RVAAS	

First tier estimates of exposure assume as a theoretical worst case that there is 100% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. For this conversion 1 kg prothioconazole yields 0.907 kg prothioconazole-desthio. This conversion can occur during the drying process on clothing and skin so for spray operators the exposure assessment only considers exposure from application of the spray solution.

Gloves for mixing and loading (100% prothioconazole-desthio)

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0879	% of RVNAS	879.46%
	Acute systemic exposure mg/kg bw/day	0.4940	% of RVAAS	
Mixing and Loading	Gloves = Yes	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0045	% of RVNAS	44.89%
	Acute systemic exposure mg/kg bw/day	0.0355	% of RVAAS	

Table A 9: Input parameters considered for the estimation of operator exposure – cereals (50% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.085 kg a.s. /ha	Spray dilution = 0.2125 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 23	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

No PPE (50% prothioconazole-desthio)

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0537	% of RVNAS	537.20%
	Acute systemic exposure mg/kg bw/day	0.3647	% of RVAAS	
Mixing and Loading	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0329	% of RVNAS	328.50%
	Acute systemic exposure mg/kg bw/day	0.1397	% of RVAAS	

Gloves for mixing and loading (50% prothioconazole-desthio)

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0537	% of RVNAS	537.20%

	Acute systemic exposure mg/kg bw/day	0.3647	% of RVAAS	
Mixing and Loading	Gloves = Yes	Clothing = Work wear - arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear - arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0034	% of RVNAS	34.37%
	Acute systemic exposure mg/kg bw/day	0.0316	% of RVAAS	

A 2.2 Worker exposure calculations (KCP 7.2.3.1)

A 2.2.1 Calculations for Fluxapyroxad

Input parameters considered for the estimation of worker exposure – cereals

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5.2	Dermal for in use dilution = 9.5	Oral = 68	Inhalation = 100	
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 10: Estimation of longer term worker exposure towards fluxapyroxad according to EFSA guidance - cereals

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.0111	% of RVNAS	27.83%
	Working clothing mg/kg bw/day	0.0012	% of RVNAS	3.12%

A 2.2.2 Calculations for Prothioconazole

Table A 11: Input parameters considered for the estimation of worker exposure – cereals (100% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 12: Estimation of longer term worker exposure towards prothioconazole according to EFSA guidance – cereals (100% prothioconazole)

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.1641	% of RVNAS	82.03%
	Working clothing mg/kg bw/day	0.0184	% of RVNAS	9.19%

Table A 13: Input parameters considered for the estimation of worker exposure – cereals (50% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.094 kg a.s. /ha	Spray dilution = 0.752 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 14: Estimation of longer term worker exposure towards prothioconazole according to EFSA guidance – cereals (50% prothioconazole)

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.0823	% of RVNAS	41.13%
	Working clothing mg/kg bw/day	0.0092	% of RVNAS	4.61%

A 2.2.3 Calculations for Prothioconazole-desthio

Table A 15: Input parameters considered for the estimation of worker exposure – cereals (100% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 16: Estimation of longer term worker exposure towards prothioconazole-desthio according to EFSA guidance – cereals (100% prothioconazole-desthio)

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.0276	% of RVNAS	276.25%
	Working clothing mg/kg bw/day	0.0031	% of RVNAS	30.94%

Table A 17: Input parameters considered for the estimation of worker exposure – cereals (50% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.085 kg a.s. /ha	Spray dilution = 0.2125 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 23	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 18: Estimation of longer term worker exposure towards prothioconazole-desthio according to EFSA guidance – cereals (50% prothioconazole-desthio)

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.0244	% of RVNAS	244.38%
	Working clothing mg/kg bw/day	0.0027	% of RVNAS	27.37%

A 2.3 Resident and bystander exposure calculations (KCP 7.2.2.1)

A 2.3.1 Calculations for Fluxapyroxad

Table A 19: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5.2	Dermal for in use dilution = 9.5	Oral = 68	Inhalation = 100	
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 20: Estimation of longer term resident exposure towards fluxapyroxad according to EFSA guidance - Maximum spray volume

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0006	% of RVNAS	1.51%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	2.68%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0015	% of RVNAS	3.76%
	All pathways (mean) mg/kg bw/day	0.0027	% of RVNAS	6.83%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.36%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.58%

	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.15%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0008	% of RVNAS	2.09%
	All pathways (mean) mg/kg bw/day	0.0010	% of RVNAS	2.52%

Table A 21: Input parameters considered for the estimation of longer term resident exposure- Minimum spray volume

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.09375 kg a.s. /ha	Spray dilution = 0.75 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5.2	Dermal for in use dilution = 9.5	Oral = 68	Inhalation = 100	
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 22: Estimation of longer term resident exposure towards fluxapyroxad according to EFSA guidance - Minimum spay volume

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0019	% of RVNAS	4.82%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	2.68%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0015	% of RVNAS	3.76%
	All pathways (mean) mg/kg bw/day	0.0035	% of RVNAS	8.66%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0005	% of RVNAS	1.15%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.58%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.15%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0008	% of RVNAS	2.09%
	All pathways (mean) mg/kg bw/day	0.0012	% of RVNAS	2.90%

A 2.3.2 Calculations for Prothioconazole

Table A 23: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume (100% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.1875 kg a.s. /ha	Spray dilution = 0.46875 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 24: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance - Maximum spray volume (100% prothioconazole)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0088	% of RVNAS	4.40%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.03%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0221	% of RVNAS	11.07%
	All pathways (mean) mg/kg bw/day	0.0251	% of RVNAS	12.55%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.05%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0009	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0123	% of RVNAS	6.15%
	All pathways (mean) mg/kg bw/day	0.0117	% of RVNAS	5.85%

Table A 25: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume (50% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.094 kg a.s. /ha	Spray dilution = 0.235 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 26: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance - Maximum spray volume (50% prothioconazole)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0044	% of RVNAS	2.21%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0010	% of RVNAS	0.52%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0111	% of RVNAS	5.55%
	All pathways (mean) mg/kg bw/day	0.0131	% of RVNAS	6.56%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.53%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	0.22%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0062	% of RVNAS	3.08%
	All pathways (mean) mg/kg bw/day	0.0060	% of RVNAS	2.99%

Table A 27: Input parameters considered for the estimation of longer term resident exposure - Minimum spray volume (100% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 28: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance - Minimum spray volume (100% prothioconazole)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0282	% of RVNAS	14.09%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.03%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0221	% of RVNAS	11.07%
	All pathways (mean) mg/kg bw/day	0.0358	% of RVNAS	17.88%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0067	% of RVNAS	3.37%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0009	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0123	% of RVNAS	6.15%
	All pathways (mean) mg/kg bw/day	0.0139	% of RVNAS	6.95%

Table A 29: Input parameters considered for the estimation of longer term resident exposure - Minimum spray volume (50% prothioconazole)

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.094 kg a.s. /ha	Spray dilution = 0.752 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 30: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance - Minimum spray volume (50% prothioconazole)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0141	% of RVNAS	7.07%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0010	% of RVNAS	0.52%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0111	% of RVNAS	5.55%
	All pathways (mean) mg/kg bw/day	0.0185	% of RVNAS	9.23%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0034	% of RVNAS	1.69%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	0.22%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0062	% of RVNAS	3.08%
	All pathways (mean) mg/kg bw/day	0.0071	% of RVNAS	3.54%

A 2.3.3 Calculations for Prothioconazole-desthio

Table A 31: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume (100% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 32: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance - Maximum spray volume (100% prothioconazole-desthio)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0015	% of RVNAS	14.91%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0005	% of RVNAS	4.60%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0037	% of RVNAS	37.29%
	All pathways (mean) mg/kg bw/day	0.0052	% of RVNAS	52.03%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	3.56%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	1.51%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	20.72%
	All pathways (mean) mg/kg bw/day	0.0022	% of RVNAS	21.61%

Table A 33: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume (50% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product =12	Dermal for in use dilution = 23	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 34: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance - Maximum spray volume (50% prothioconazole-desthio)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0013	% of RVNAS	13.15%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	3.54%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0033	% of RVNAS	32.99%
	All pathways (mean) mg/kg bw/day	0.0047	% of RVNAS	46.84%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0003	% of RVNAS	3.14%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	1.33%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0018	% of RVNAS	18.33%
	All pathways (mean) mg/kg bw/day	0.0019	% of RVNAS	19.38%

Table A 35: Input parameters considered for the estimation of longer term resident exposure - Minimum spray volume (100% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.17 kg a.s. /ha	Spray dilution = 1.36 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 12	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 36: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance - Minimum spray volume (100% prothioconazole-desthio)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0044	% of RVNAS	44.06%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	4.35%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0034	% of RVNAS	34.43%
	All pathways (mean) mg/kg bw/day	0.0066	% of RVNAS	65.65%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.51%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	1.39%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0019	% of RVNAS	19.13%
	All pathways (mean) mg/kg bw/day	0.0024	% of RVNAS	23.56%

Table A 37: Input parameters considered for the estimation of longer term resident exposure - Minimum spray volume (50% prothioconazole-desthio)

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate-0.085 kg a.s. /ha	Spray dilution = 0.68 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 12	Dermal for in use dilution = 12	Oral = 100	Inhalation = 100	
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha		DT50	30 days	

Table A 38: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance - Minimum spray volume (50% prothioconazole-desthio)

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0022	% of RVNAS	22.03%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.18%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0017	% of RVNAS	17.21%
	All pathways (mean) mg/kg bw/day	0.0038	% of RVNAS	38.18%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0005	% of RVNAS	5.25%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.69%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0010	% of RVNAS	9.56%
	All pathways (mean) mg/kg bw/day	0.0013	% of RVNAS	12.93%

A 2.4 Combined exposure calculations for fluxapyroxad, prothioconazole and prothioconazole-desthio

See Section 6.6.5.1

A 2.5 Operator exposure calculations (KCP 7.2.1.1)

A 2.5.1 Calculations for Fluxapyroxad

Table A 39: Input parameters considered for the estimation of operator exposure – cereals

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5	Dermal for in use dilution = 9	Oral = 68	Inhalation = 100	-
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 40: Estimation of longer term operator exposure towards fluxapyroxad according to EFSA guidance – cereals

No PPE

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0241	% of RVNAS	60.23%
	Acute systemic exposure mg/kg bw/day	0.1589	% of RVAAS	Not required.
Mixing and Loading	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Soluble bags = No

Application	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0148	% of RVNAS	36.99%
	Acute systemic exposure mg/kg bw/day	0.0625	% of RVAAS	Not required.

Gloves worn for mixing and loading

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0241	% of RVNAS	60.23%
	Acute systemic exposure mg/kg bw/day	0.1589	% of RVAAS	Not required.
Mixing and Loading	Gloves = Yes	Clothing = Work wear – arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0016	% of RVNAS	3.95%
	Acute systemic exposure mg/kg bw/day	0.0139	% of RVAAS	Not required.

A 2.5.2 Calculations for Prothioconazole

Table A 41: Input parameters considered for the estimation of operator exposure – cereals

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in-use dilution = 70	Oral = 100	Inhalation = 100	-
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 42: Estimation of longer term operator exposure towards prothioconazole according to EFSA guidance – cereals

No PPE

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.2129	% of RVNAS	106.47%
	Acute systemic exposure mg/kg bw/day	1.1961	% of RVAAS	Not required.
Mixing and Loading	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Soluble bags = No

Application	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.1332	% of RVNAS	66.58%
	Acute systemic exposure mg/kg bw/day	0.5807	% of RVAAS	Not required.

Gloves worn for mixing and loading

Operator Model	Mixing, loading and application AOEM			
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.2129	% of RVNAS	106.47%
	Acute systemic exposure mg/kg bw/day	1.1961	% of RVAAS	Not required.
Mixing and Loading	Gloves = Yes	Clothing = Work wear – arms, body and legs covered	RPE = None	Soluble bags = No
Application	Gloves = No	Clothing = Work wear – arms, body and legs covered	RPE = None	Closed cabin = No
Exposure (including PPE options above)	Longer term systemic exposure mg/kg bw/day	0.0204	% of RVNAS	10.22%
	Acute systemic exposure mg/kg bw/day	0.1649	% of RVAAS	Not required.

Table A 43: Estimation of longer term operator exposure towards prothioconazole from mixing and loading according to EFSA guidance – Gloves worn

Spray Application	**Systemic exposure [µg a.s./kg bw/day]**
Clothing = Work wear – arms, body and legs covered + gloves	
Hands	0.6156947
Body	0.7199543
Head	2.0267026
Inhalation	0.1200902
Sum	3.4824418

A 2.5.3 Calculations for Prothioconazole-desthio

Table A 44: Input parameters considered for the estimation of operator exposure – cereals

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 0	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	-
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 45: Estimation of longer term operator exposure towards prothioconazole-desthio according to EFSA guidance – cereals

Spray Application	Systemic exposure [µg a.s./kg bw/day]
Clothing = Work wear – arms, body and legs covered	
Hands	2.7316222
Body	0.0418976
Head	0.0721875
Inhalation	0.0504288
Sum	2.8961362

First tier estimates of exposure assume as a theoretical worst case that there is 100% conversion from the parent prothioconazole to the metabolite prothioconazole-desthio. For this conversion 1 kg prothioconazole yields 0.907 kg prothioconazole-desthio. This conversion can occur during the drying process on clothing and skin so for spray operators the exposure assessment only considers exposure from application of the spray solution.

A 2.6 Worker exposure calculations (KCP 7.2.3.1)

A 2.6.1 Calculations for Fluxapyroxad

Input parameters considered for the estimation of worker exposure – cereals

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5	Dermal for in use dilution = 9	Oral = 68	Inhalation = 100	-
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 46: Estimation of longer term worker exposure towards fluxapyroxad according to EFSA guidance – cereals

Worker –	Potential exposure mg/kg bw/day	0.0105	% of RVNAS	26.37%

Inspection, irrigation	Working clothing mg/kg bw/day	0.0012	% of RVNAS	2.95%

A 2.6.2 Calculations for Prothioconazole

Table A 47: Input parameters considered for the estimation of worker exposure – cereals

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	-
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 48: Estimation of longer term worker exposure towards prothioconazole according to EFSA guidance – cereals

Worker – Inspection, irrigation	Potential exposure mg/kg bw/day	0.1641	% of RVNAS	82.03%
	Working clothing mg/kg bw/day	0.0184	% of RVNAS	9.19%

A 2.6.3 Calculations for Prothioconazole-desthio

Table A 49: Input parameters considered for the estimation of worker exposure – cereals

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 0	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	-
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 50: Estimation of longer term worker exposure towards prothioconazole-desthio according to EFSA guidance – cereals

Worker – Inspection, irrigation	Potential exposure mg/kg bw/day	0.0276	% of RVNAS	276.25%
	Working clothing mg/kg bw/day	0.0031	% of RVNAS	30.94%

A 2.7 Resident and bystander exposure calculations (KCP 7.2.2.1)

A 2.7.1 Calculations for Fluxapyroxad

Table A 51: Input parameters considered for the estimation of longer term resident exposure – Maximum spray volume

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.09375 kg a.s. /ha	Spray dilution = 0.234375 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5	Dermal for in use dilution = 9	Oral = 68	Inhalation = 100	-
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 52: Estimation of longer term resident exposure towards fluxapyroxad according to EFSA guidance – Maximum spray volume

Resident – child	Spray drift (75th percentile) mg/kg bw/day	0.0006	% of RVNAS	1.43%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	2.68%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.44%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0014	% of RVNAS	3.56%
	All pathways (mean) mg/kg bw/day	0.0026	% of RVNAS	6.62%
Resident – adult	Spray drift (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.34%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.58%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.14%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0008	% of RVNAS	1.98%
	All pathways (mean) mg/kg bw/day	0.0010	% of RVNAS	2.42%

Table A 53: Input parameters considered for the estimation of longer term resident exposure – Minimum spray volume

Substance	Fluxapyroxad	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.09375 kg a.s. /ha	Spray dilution = 0.75 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 5	Dermal for in use dilution = 9	Oral = 68	Inhalation = 100	-
RVNAS	0.04 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm² per kg a.s./ha	-	DT50	30 days	-

Table A 54: Estimation of longer term resident exposure towards fluxapyroxad according to EFSA guidance – Minimum spay volume

Resident –	Spray drift (75th percentile) mg/kg bw/day	0.0018	% of RVNAS	4.57%

child	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	2.68%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.44%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0014	% of RVNAS	3.56%
	All pathways (mean) mg/kg bw/day	0.0033	% of RVNAS	8.36%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	1.09%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.58%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	0.14%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0008	% of RVNAS	1.98%
	All pathways (mean) mg/kg bw/day	0.0011	% of RVNAS	2.77%

A 2.7.2 Calculations for Prothioconazole

Table A 55: Input parameters considered for the estimation of longer term resident exposure - Maximum spray volume

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.1875 kg a.s. /ha	Spray dilution = 0.46875 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$ Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in-use dilution = 70	Oral = 100	Inhalation = 100	-
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 56: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance – Maximum spray volume

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0088	% of RVNAS	4.40%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.03%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0221	% of RVNAS	11.07%
	All pathways (mean) mg/kg bw/day	0.0251	% of RVNAS	12.55%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.05%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0009	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0123	% of RVNAS	6.15%
	All pathways (mean) mg/kg bw/day	0.0117	% of RVNAS	5.85%

Table A 57: Input parameters considered for the estimation of longer term resident exposure – Minimum spray volume

Substance	Prothioconazole	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.1875 kg a.s. /ha	Spray dilution = 1.5 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle mounted			Buffer = 2 3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 25	Dermal for in use dilution = 70	Oral = 100	Inhalation = 100	-
RVNAS	0.2 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required.
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 58: Estimation of longer term resident exposure towards prothioconazole according to EFSA guidance – Minimum spray volume

Resident – child	Spray drift (75th percentile) mg/kg bw/day	0.0282	% of RVNAS	14.09%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	0.54%
	Surface deposits (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	1.03%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0221	% of RVNAS	11.07%
	All pathways (mean) mg/kg bw/day	0.0358	% of RVNAS	17.88%
Resident – adult	Spray drift (75th percentile) mg/kg bw/day	0.0067	% of RVNAS	3.37%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.12%
	Surface deposits (75th percentile) mg/kg bw/day	0.0009	% of RVNAS	0.45%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0123	% of RVNAS	6.15%
	All pathways (mean) mg/kg bw/day	0.0139	% of RVNAS	6.95%

A 2.7.3 Calculations for Prothioconazole-desthio

Table A 59: Input parameters considered for the estimation of longer term resident exposure – Maximum spray volume

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.17 kg a.s. /ha	Spray dilution = 0.425 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10^{-3}Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle mounted			Buffer = 2 3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 0	Dermal for in use dilution = 13	Oral = 100	Inhalation = 100	-
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 60: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance – Maximum spray volume

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0015	% of RVNAS	14.91%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0005	% of RVNAS	4.60%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0037	% of RVNAS	37.29%
	All pathways (mean) mg/kg bw/day	0.0052	% of RVNAS	52.03%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	3.56%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	1.51%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0021	% of RVNAS	20.72%
	All pathways (mean) mg/kg bw/day	0.0022	% of RVNAS	21.61%

Table A 61: Input parameters considered for the estimation of longer term resident exposure – Minimum spray volume

Substance	Prothioconazole-Desthio	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate 0.17 kg a.s. /ha	Spray dilution = 1.36 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of $<5*10^{-3}$Pa
Scenario	Cereals / Outdoor / Downward spraying / Vehicle-mounted			Buffer = 2-3	Number applications = 1, Application interval = 365 days
Percentage Absorption	Dermal for product = 0	Dermal for in use dilution = 12	Oral = 100	Inhalation = 100	-
RVNAS	0.01 mg/kg bw/day		RVAAS	mg/kg bw/day	Not required
DFR	3 µg a.s./cm^2 per kg a.s./ha	-	DT50	30 days	-

Table A 62: Estimation of longer term resident exposure towards prothioconazole-desthio according to EFSA guidance – Minimum spray volume

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0044	% of RVNAS	44.06%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.70%
	Surface deposits (75th percentile) mg/kg bw/day	0.0004	% of RVNAS	4.35%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0034	% of RVNAS	34.43%
	All pathways (mean) mg/kg bw/day	0.0066	% of RVNAS	65.65%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	10.51%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	2.30%
	Surface deposits (75th percentile) mg/kg bw/day	0.0001	% of RVNAS	1.39%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0019	% of RVNAS	19.13%
	All pathways (mean) mg/kg bw/day	0.0024	% of RVNAS	23.56%

A 2.8 Combined exposure calculations for for fluxapyroxad, prothioconazole and prothioconazole-desthio

See Section 6.6.5.1