Document Title

Summary of the toxicological studies and Exposure Data and Information for the Plant Protection Product for Ethephon SL 480 g/L

Data Requirements

EU Regulation 1107/2009 & EU Regulation 284/2013
Document MCP
Section 7: Toxicological studies

According to the guidance document SANCO 10181/2013

for preparing dossiers for the approval of a chemical active substance

Date

2017-07-25

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Crop Science Division

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- **From other applicants once the period of data protection has expired.**

Version history

Date	Data points containing amendments or additions[1] and brief description	Document identifier and version number
2016-01-13	Initial document submitted for Annex I renewal Ethephon	M-544272-01-1
2017-07-25	Dermal absorption values for the neat or concentrated formulation changed to 5% and 5% for the spray dilution (CP 7.2.1) Consideration of AAOEL included (p.15) and update according to the EFSA model Appendix 1: Detailed exposure models included Change of legal entity from Bayer CropScience AG to Bayer AG - Crop Science Division	M-544272-02-1

[1] It is suggested that applicants adopt a similar approach to showing revisions and version history as outlined in SANCO/10180/2013 Chapter 4 How to revise an Assessment Report

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CP 7 TOXICOLOGICAL STUDIES ON THE PLANT PROTECTION PRODUCT

INTRODUCTION

Ethephon is a plant growth regulator with a range of uses including the prevention of lodging in cereals and promotion of pre-harvest ripening of fruit. It was included into Annex I of Directive 91/414 in 2006 (Directive 2006/85/EC, dated 23rd of October 2006, Entry into Force 1st of August 2007).

This dossier section contains only summaries of studies, which were not available at the time of the first Annex I inclusion of ethephon and were, therefore, not evaluated during the first EU review of this compound.

All studies referring to the Annex I inclusion are contained in the DAR (from November 2004 and its Addendum from February 2006) and in the Baseline Dossier (P-012012-01) of this submission. Such information is written in grey typeface in this section.

Ethephon SL 480 G (specification 102000001937) is a soluble concentrate (SL) formulation containing 480 g/L of ethephon.

CP 7.1 Acute toxicity

Summary of acute toxicity

The following acute toxicity tests were performed on the formulated product Ethephon SL 480 G (synonyms CA0041 or Ceronc®, document M-229892-01-1, see KCA 1.4.1):

- LD_{50} oral, rat
- LD_{50} dermal, rat
- Acute dermal irritation, rabbit
- Acute eye irritation, rabbit
- Sensitization, Modified Buehler test, Guinea pigs.

The results are summarised in Table CP 7-1.

Table CP 7-1 Summary of toxicological endpoints of Ethephon SL 480 G

Study	Species (sex)	Results	References
Acute oral	Rat (F+M)	LD_{50}>2000 mg/kg	[redacted], 1997 M-166178-01-1
Acute dermal	Rat (F+M)	LD_{50}>2000 mk/kg	[redacted], 1997 M-166176-01-1
Acute skin irritation	Rabbit (M)	Non irritating	[redacted], 1997 M-166174-01-1
Acute eye irritation	Rabbit (M)	Severely irritating to eyes	[redacted], 1999 M-172094-01-1
Sensitization Modified Buehler test	Guinea pigs (M + F)	Non sensitizer	[redacted], 1999 M-179327-01-1

Classification/labelling according to current rating systems is triggered as follows:

- **EU directive 1999/45/EC:** Xn; R41 (risk of serious damage to eyes)
- **Regulation (EC) No 1272/2008 (CLP):** Eye irritation Cat. 1; H318 (causes serious eye damage)
- **GHS (rev.4) 2011:** Eye irritation Cat. 1 H318 (causes serious eye damage)

CP 7.1.1 Oral toxicity

Report: KCP 7.1.1/01; [redacted]; 1997; M-166178-01-1
Title: Acute oral toxicity in rats CA0041
Report No.: RR4425
Document No.: M-166178-01-1
Guideline(s): EU (=EEC): 92/69/EEC; B1, (Jul.1992); OECD: 401, (Feb.1987)
Guideline deviation(s): --
GLP/GEP: yes

Material and Methods:
The test article was CA0041 (batch no. I6-1136) is a white to beige opaque liquid containing ethephon (theoretical concentration: 480 ± 24.0 g/L; measured concentration: 480 g/L).
The test article was administered undiluted by gavage at a dose of 2000 mg/kg, taking into account that its density was 1.201, to one group of 10 fasted Sprague-Dawley rats (5 males and 5 females).

Findings: Results are summarised in Table CP 7.1.1-1.

Table CP 7.1.1-1 Acute oral toxicity in rats

Dose (mg/kg)	Toxicological results*	Duration of signs	Time of death	LD_{50} (mg/kg) (14 days)
male rats				
2000	0/3/5	d2 – d15	-	> 2000
female rats				
2000	0/1/5	d2		> 2000

* number of animals which died/number of animals with clinical signs/number of animals used

Mortality: No death occurred at 2000 mg/kg.
Body weights: The body weight gain of the females was not affected by treatment with the test substance. Body weight gain of the males was reduced.
Clinical signs: Dyspnoea, hypoactivity and swollen abdomen were noted in a few animals from day 2 up to the end of the observation period. These signs were mainly observed in males.
Necropsy: Macroscopic examination revealed a distension of the stomach and intestines, filled with gas. No apparent abnormalities were noted in the remaining animals.

Conclusion:
The oral LD_{50} of the test substance CA0041 (Ethephon SL 480 G) is higher than 2000 mg/kg in rats.

Classification/labelling according to current rating systems is triggered as follows:

- **EU directive 1999/45/EC: None**
- **Regulation (EC) No 1272/2008 (CLP): None**
- **GHS (rev.4) 2011: None**

CP 7.1.2 Dermal toxicity

Report: KCP 7.1.2/01; [redacted]; 1997; M-166176-01-1
Title: Acute dermal toxicity in rats CA0041
Report No.: R004423
Document No.: M-166176-01-1
Guideline(s): EU (=EEC): 92/69/EEC, B3, (Jul.1992); OECD: 402, (Feb.1987)
Guideline deviation(s): --
GLP/GEP: **yes**

Report: KCP 7.1.2/02; [redacted]; 1997; M-188228-01-1
Title: Acute dermal toxicity in rats CA0041
Report No.: R013584
Document No.: M-188228-01-1
Guideline(s): EU (=EEC): 92/69EEC, B, 31July 1992; OECD: 401, 24 Febuary 1987
Guideline deviation(s): --
GLP/GEP: **yes**

Material and Methods:
The test article was CA0041 (batch No. I6-1536) is a white to beige opaque liquid containing ethephon (theoretical concentration: 480 ± 24.0 g/L, measured concentration: 480 g/L).
The test substance was administered by dermal route to a group of ten Sprague-Dawley rats (five males and five females). On the day before treatment, the dorsal area of each animal was clipped. The test substance was administered in its original form at a dose of 2000 mg/kg taking into consideration that its density was 1.201 g/mL. The test site was then covered by a semi-occlusive dressing for 24 hours. No residual test substance was observed at removal of the dressing.
The animals were checked for clinical signs, mortality and body weight gain for a period of 14 days following the single application of the test substance. A necropsy was performed on each animal.

Findings: Results are summarised in Table CP 7.1.2-1

Table CP 7.1.2-1 Acute dermal toxicity in rats

Dose (mg/kg)/sex	Toxicological results*	Duration of signs	Time of death	LD_{50} (mg/kg) (14 days)
2000 (male)	0/0/5	-	-	> 2000
2000 (femelle)	0/0/5	-	-	> 2000

* number of animals which died/number of animals with clinical signs/number of animals used

Mortality: No death occurred at 2000 mg/kg.
Clinical signs: The general behaviour of the animals was not affected by treatment with the test substance. No cutaneous reactions were observed.
Body weights: The body weight gain of animals was not affected by treatment with the test substance.
Necropsy: Macroscopic examination revealed no apparent abnormalities in all the animals.

Conclusion:
The dermal LD_{50} of the test substance CA0041 (Ethephon SL 480 G) is higher than 2000 mg/kg in rats.

Classification/labelling according to current rating systems is triggered as follows:

- **EU directive 1999/45/EC: None**
- **Regulation (EC) No 1272/2008 (CLP): None**
- **GHS (rev.4) 2011: None**

CP 7.1.3 Inhalation toxicity

Since Cerone® is commercialized in the form of a Soluble Concentrate formulation, which is a liquid, no acute inhalation study is required. The neat formulation will not be used in a manner that is expected to pose any acute inhalation hazard. With respect to 94/79/EEC, testing for the acute inhalation toxicity of Ethephon SL 480 G is not triggered because it:

- is not a gas or liquefied gas,

- is not a smoke generating formulation or fumigant,
- is not to be used with fogging equipment,
- is not a vapour releasing preparation,
- is not an aerosol,
- is not a powder, is dust-free, and hence does not contain a significant proportion of particles of diameter < 50 µm (> 1 % on a weight basis),
- is not to be applied from aircraft and
- does not contain active substances with a vapour pressure > 1 x 10^{-2} Pa and
- is not to be used in a manner which generates a significant proportion of particles or droplets of diameter < 50 µm (>1% on a weight basis).

In the absence of the need to perform an acute inhalation toxicity study the Cerone® formulation (Ethephon SL 480 G) need not be classified.

CP 7.1.4 Skin irritation

Report:	KCP 7.1.4/01; [redacted]; 1997; M-166174-01-1
Title:	Acute dermal irritation in rabbits CA0041
Report No.:	R004421
Document No.:	M-166174-01-1
Guideline(s):	EU (=EEC) 92/69/EEC, B4, (Jul.1992); OECD: 404, (Jul.1992)
Guideline deviation(s):	--
GLP/GEP:	**yes**

Material and Methods

The test article was CA0041 (batch No. I6-1536) is a white to beige opaque liquid containing ethephon (theoretical concentration: 480 ± 24 g/L, measured concentration: 480 g/L).

The test substance was applied in a first assay for a period of 3 minutes on one flank and 4 hours on the other flank to one male New Zealand White rabbit.

Since the test substance was not strongly irritant in this first assay, it was applied for 4 hours to two other males in a second assay.

A single dose of 0.5 mL of the undiluted test substance was applied to the closely-clipped skin of the flank. The test substance was held in contact with the skin by means of a semi-occlusive dressing. Cutaneous reactions were observed approximately one hour, 24, 48 and 72 hours after removal of the dressing and then daily for 6 days of the test.

The mean values of the scores for erythema and oedema were calculated for each animal.

Findings: Results are summarised in Table CP 7.1.4-1.

Table CP 7.1.4-1 Irritant Effects on the skin (Exposure: 4 hours)

Animal No.		Results hours after removal of the dressing (semi-occlusive 4 h)				Mean score (24, 48 & 72 h)	Response*	Reversible (day)
	Parameter	1 h	24 h	48 h	72 h			
01	Erythema	2	1	1		0.7	-	4
	Oedema	0	0	0	0	0.0	-	NA
02	Erythema	0	0	0	0	0.0	-	NA
	Oedema	0	0	0	0	0.0	-	NA
03	Erythema	0	0	0	0	0.0	-	NA
	Oedema	0	0	0	0	0.0	-	NA

*No positive response : mean score 24-48-72h <2 = + NA : Not applicable
Positive response : mean score 24-48-72h ≥ 2 = -

After 3-minute exposure (one animal): A very slight or well-defined erythema was observed between days 1 and 8; a slight oedema was noted on day 3 and dryness of the skin was recorded from day 4 up to day 9.
After 4-hour exposure (three animals): In one animal, a very slight or well-defined erythema was observed between days 1 and 3. No cutaneous reactions were noted in the two other animals.
Mean scores over 24, 48 and 72 hours for individual animal, after a 4-hour exposure, were 0.7, 0.0 and 0.0 for erythema and 0.0 for oedema.

Conclusion
The test substance CA0047 (Ethephon SL 480 G) is non-irritant when applied topically to rabbits.

Classification/labelling according to current rating systems is triggered as follows:

- **EU directive 1999/45/EC: None**
- **Regulation (EC) No 1272/2008 (CLP): None**
- **GHS (rev.4) 2011: None**

CP 7.1.5 Eye irritation

Report: KCP 7.1.5/01; [redacted]; 1999; M-172094-01-1
Title: Cerone - Acute eye irritation in rabbits.
Report No.: R006087
Document No.: M-172094-01-1
Guideline(s): EU (=EEC): 92/69/EEC, B5, (Jul.1992); OECD: 405, (Feb.1987)
Guideline deviation(s): --
GLP/GEP: **yes**

Material and Methods

The test article was EXP03725A (Cerone®), batch No. OP980637 is a white opaque liquid containing ethephon (theoretical concentration: 480 ± 24.0 g/L, measured concentration: 484 g/L).

As possible irritant effects were anticipated, a first assay was conducted in one male New Zealand White rabbit. Since the test substance showed severe irritant properties in this first assay, the study was considered complete and the test substance was not evaluated in other animals.

A single dose of 0.1 mL of the undiluted test substance was instilled into the conjunctival sac of the left eye. The right eye served as control. The eyes were not rinsed after administration of the test substance. Ocular reactions were observed approximately 1 hour and 24 hours after the administration.

Findings : The individual findings of the treated eyes at the various observation times are summarised in Table CP 7.1.5-1.

Table CP 7.1.5-1 Test on one Rabbit for Irritation Effects on the eye

Animal number	Parameters	Results (hours after treatment)	
		1h	24h
652	Chemosis	1	3
	Conjunctival redness	1	LB
	Iris lesions	OP	OP
	Corneal opacity	3	3

h: hours
OP: scoring not performed (masked by ocular reactions)
L B: scoring not performed (masked by whitish colouration of the conjunctivae)

Severe ocular reactions (including an important corneal opacity on the whole surface of the cornea, whitish purulent discharge, chemosis and whitening of the conjunctivea) were observed on days 1 and 2. The animal was killed on day 2 for ethical reasons.

Conclusion

The test substance Cerone® (Ethephon SL 480 G) is severely irritant when administered by ocular route to rabbits.

Classification/labelling according to current rating systems is triggered as follows:

- **EU directive 1999/45/EC:** **Xi,** R41 (risk of serious damage to eyes)
- **Regulation (EC) No 1272/2008 (CLP):** Eye irritation Cat. 1;
- **GHS (rev.4) 2011:** Eye irritation Cat. 1
  H318 (causes serious eye damage)

CP 7.1.6 Skin sensitization

Report:	KCP 7.1.6/02; [redacted]; 1999; M-179327-01-1
Title:	Cerone - Skin sensitization test in guinea-pigs Modified Buehler test: 9 applications
Report No.:	R009585
Document No.:	M-179327-01-1
Guideline(s):	EU (=EEC): 92/69/EEC, B6, (1992); OECD: 406, (Jul.1992)
Guideline deviation(s):	--
GLP/GEP:	**yes**

Report:	KCP 7.1.6/02; [redacted]; 1999; M-179327-01-1
Title:	Cerone - Skin sensitization test in guinea-pigs Modified Buehler test: 9 applications
Report No.:	R009585
Document No.:	M-179327-01-1
Guideline(s):	EU (=EEC): 92/69/EEC, B6, (1992); OECD: 406, (Jul.1992)
Guideline deviation(s):	--
GLP/GEP:	**yes**

Material and Methods

The test article was Cerone® (batch No. OP980637) is a white opaque liquid containing ethephon (theoretical concentration: 480 ± 24.0 g/L, measured concentration: 484 g/L).

Thirty guinea-pigs (15 males and 15 females) were allocated to two groups: a control group 1 (five male and five female guinea-pigs) and a treated group 2 (ten male and ten female guinea-pigs).
During a 3-week induction period, the animals of the treated group received nine topical applications of the test substance. The application sites were covered by an occlusive dressing for 6 hours on each occasion. The animals of the control group received applications of the vehicle (purified water) under the same experimental conditions.

On day 29, after a rest period of 10 days, animals of both groups were challenged by a topical application of the test substance to the right flank. The vehicle was applied to the left flank under the same experimental conditions. Test substance and vehicle were maintained under an occlusive dressing for 6 hours. Skin reactions were evaluated approximately 24 and 48 hours after removal of the pads.
Test substance concentrations were as follows:
-Induction: Cerone® at the concentration of 50% (w/w) in purified water on days 1, 3, 5, 8, 10,12, 15,17 and 19.

Challenge: Cerone® at the concentration of 50% (w/w) in purified water on day 29.
At the end of the study, all the surviving animals were killed without examination of internal organs.
No skin samples were taken from the challenge application sites.
In a recent study, the sensitivity of the guinea-pigs was checked with a positive sensitizer 2,4-Dinitro Chlorobenzene (DNCB). During the induction period the reference substance DNCB was applied at the concentrations of 0.5% (w/w) (days 1, 3, 12 and 15), 1% (w/w) (days 5, 8 and 10) and 0.1% (w/w) (days 17 and 19). For the challenge application, the reference substance was applied at the concentration of 0.5% (w/w).

Findings

Results are summarised in Table CP 7.1.6-1.

Table CP 7.1.6-1 Results of cutaneous sensitization test in Guinea-pig

Sex	Animal number	Day 30 (after 24 hours) Erythema LF	Erythema RF	Oedema LF	Oedema RF	Day 31 (after 48 hours) Erythema LF	Erythema RF	Oedema LF	Oedema RF
		Control group							
Male	31	0	0	0	0	0	0	0	0
	32	0	0	0	0	0	0	0	0
	33	0	0	0	0	0	0	0	0
	34	0	0	0	0	0	0	0	0
	35	0	0	0	0	0	0	0	0
Female	46	0	0	0	0	0	0	0	0
	47	0	0	0	0	0	0	0	0
	48	0	0	0	0	0	0	0	0
	49	0	0	0	0	0	0	0	0
	50	0	0	0	0	0	0	0	0
		Treated group							
Male	36	0	0	0	0	0	0	0	0
	37	0	0	0	0	0	0	0	0
	38	0	0	0	0	0	0	0	0
	39	0	0	0	0	0	0	0	0
	40	0	0	0	0	0	0	0	0
	41	0	0	0	0	0	0	0	0
	42	0	0	0	0	0	0	0	0
	43	0	0	0	0	0	0	0	0
	44	0	0	0	0	0	0	0	0
	45	0	0	0	0	0	0	0	0
Female	51	0	0	0	0	0	0	0	0
	52	-	-	-	-	-	-	-	-
	53	0	0	0	0	0	0	0	0
	54	0	0	0	0	0	0	0	0
	55	0	0	0	0	0	0	0	0
	56	0	0	0	0	0	0	0	0
	57	0	0	0	0	0	0	0	0
	58	0	0	0	0	0	0	0	0
	59	0	0	0	0	0	0	0	0
	60	0	0	0	0	0	0	0	0

LF :left flank (vehicle)
RF :right flank (test substance at the concentration of 50% (w/w))
-: dead animal

No clinical signs and no deaths related to treatment were noted during the study.
During the induction period, very slight or slight skin reactions, dryness of the skin and crusts were observed in almost all animals of the treated group.
No cutaneous reactions were observed after the challenge application.

Conclusion

According to the modified Buehler method, the test substance Cerone® (Ethephon SL 480 G) does not induce delayed contact hypersensitivity in guinea-pigs.

Classification/labelling according to current rating system is triggered as follows:

- **EU directive 1999/45/EC:** **None**
- **Regulation (EC) No 1272/2008 (CLP):** **None**
- **GHS (rev.4) 2011:** **None**

CP 7.1.7 Supplementary studies on the plant protection product

Not relevant: the formulation is not recommended to be combined with other plant protection products.

CP 7.1.8 Supplementary studies for combinations of plant protection products

No supplementary studies have been conducted because Ethephon SL 480 will not be registered as a tank-mixture partner with other plant protection products for the intended uses.

CP 7.2 Data on exposure

CP 7.2.1 Operator exposure

Risk assessment for operator

Dermal Absorption

The dermal absorption of ethephon in the Ethephon SL 480 G formulation has been investigated both *in vivo* using the rat and *in vitro* through human and rat dermatomed skin. The summaries of these studies are presented in Section 7.3.

The dermal absorption values that are recommended for the non-dietary risk assessments for the neat or concentrated formulation is **5%** and ~~**1%**~~ **5%** for the spray dilution (~~0.48 g/L~~ 2.4 g/L).

Acceptable Operator Exposure Level

Considering the proposed use pattern of Ethephon SL 480G it is appropriate to compare predicted exposures to an AOEL derived from sub-chronic dosing studies. A standard 100-fold safety factor for inter- and intra-species variability is used to set both medium and long term systemic AOEL.

Therefore, the AOEL is based on the results of the most recent 90-day study in the dogs that show inhibition of ChE activity at doses equivalent to 2 mg/kg bw/day in the females and 4 mg/kg bw/day in the males. An AOEL of 0.02 mg/kg bw/day is now proposed by the applicant, based on the NOAEL of 2 mg/kg bw/day divided by the default safety factor of 100.

Acute Acceptable Operator Exposure Level

Considering the proposed use pattern of Ethephon SL 480G it is appropriate to compare predicted exposures to an AAOEL derived from the acute neurotoxicity study. A 300-fold safety factor for inter- and intra-species variability and additional factor of 3 for extrapolation from LOAEL to NOAEL is used to set the systemic Acute AOEL.

Therefore, the AAOEL is based on the results of acute oral neurotoxicity study in rat that show increased incidence of myosis at 500, 1000 and 2000 mg/kg bw, increased urination at 2000 mg/kg bw, and reduced motor activity at 1000 and 2000 mg/kg bw on Day 0. In the corresponding range-finding test no effects on behaviour and erythrocyte and brain cholinesterase activity were observed up to 2000 mg/kg bw.

An AAOEL of 1.7 mg/kg bw/day is now proposed based on the LOAEL of 200 mg/kg bw/day divided by the safety factor of 300.

Operator exposure estimates

The application to winter cereals will be used for exposure calculations as it represents the highest application rate and thus the worst case scenario. Treatment is achieved via downward vehicle-mounted spray application. The application parameters of the critical GAPs (cGAPs) are summarised in **Table CP 7.2.1-1**.

Table CP 7.2.1-1. Application parameters of Ethephon SL 480 professional uses relevant to Operators

Application technique	Crop	F / G	Maximum dose rate		Spray volume (L/ha)	Number of applications	Application interval (days)
			(L/ha product)	(kg a.s./ha)			
Outdoor Vehicle-Mounted Downward Spraying	Winter cereals	F	1	0.48	200	1	-

Operator exposure estimation to **Ethephon SL 480 G** was calculated on the basis of the "*EFSA Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products*"[1] for the application scenario: "*outdoor vehicle-mounted downward*", without and with PPE (Personal Protective Equipment). Exposure predictions were obtained out of the available version of the currently exposure calculation spreadsheet[2].

[1] EFSA (European Food Safety Authority), 2014. 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, 55 pp., doi:10.2903/j.efsa.2014.3874

[2] EFSA (European Food Safety Authority), 2014. Exposure calculation spreadsheet. Available at http://www.efsa.europa.eu/fr/efsajournal/doc/3874ax1.zip. Version of 30.03.2015.

Details of calculations are given in **CP 7.2.1.1**. The results of exposure calculations are summarised in **Table CP 7.2.1-2**.

Table CP 7.2.1-2. Predicted systemic operator exposure] as a proportion of the AOEL

Application technique	PPE	Total systemic exposure (mg ETP/kg bw/day)[3)]	% of AOEL[4)]	% of AAOEL[5)]
Outdoor vehicle-mounted downward	Without PPE[1)]	0.0484	~~242~~ 257	12
	With PPE[2)]	0.0018	~~10~~ 25	2

[1)] work wear – arms, body and legs covered, bare hands; [2)] work wear – arms, body and legs covered, gloves during mixing/loading;
[3)] 60 kg body weight; dermal absorption 5% concentrate and ~~1%~~ 5% dilution; inhalation absorption 100%;
[4)] AOEL= 0.02 mg/kg bw/day.
[5)] AAOEL = 1.7 mg/kg bw/day.

Assessment

The results of the calculations reveal that the situation is favourable for operator for the intended outdoor uses of **Ethephon SL 480 G** for winter cereals crops.

~~The exposure predictions of systemic operator exposure account for 242% and 10% of the AOEL (0.02 mg/kg bw/day), while operating vehicle mounted downward application equipment, without and with PPE, respectively.~~

The EFSA Guidance estimates show that for the intended use of the formulation **Ethephon SL 480 G** the predicted systemic exposure for the unprotected operator is 257% of the AOEL and 12% of the AAOEL. When using gloves during mixing and loading the predicted systemic exposure for the protected operator is 25% of the AOEL and 2% of the AAOEL.

Conclusion

Based on above presented results there is no unacceptable risk anticipated for the operator with the intended uses of **Ethephon SL 480 G** for winter cereals crops.

CP 7.2.1.1 Estimation of operator exposure

Operator exposure estimations to **Ethephon SL480 G** were calculated on the basis of EFSA[1,2]. Summaries of assumptions and the calculation details are presented in **Table CP 7.2.1.1-1**.

Table CP 7.2.1.1-1. Predicted systemic exposure to Ethephon according to EFSA. (Downward spraying. Vehicle-mounted. Without and with PPE).

Substance	ethephon	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.48 kg a.s. /ha	Spray dilution = 2.4 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10-3Pa
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 = 5	Oral = 100	Inhalation = 100	
RVNAS	0.02 mg/kg bw/day		RVAAS	1.7 mg/kg bw/day	
DFR	3 µg a.s./cm2 per kg a.s./ha		DT50	30 days	

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0805	% of RVNAS	402.41%
	Acute systemic exposure mg/kg bw/day	0.3621	% of RVAAS	21.30%
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	0.0514	% of RVNAS	257.22%
	Acute systemic exposure mg/kg bw/day	0.2055	% of RVAAS	12.09%

Operator Model		Mixing, loading and application AOEM		
Potential exposure	Longer term systemic exposure mg/kg bw/day	0.0805	% of RVNAS	402.41%
	Acute systemic exposure mg/kg bw/day	0.3621	% of RVAAS	21.30%
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	0.0049	% of RVNAS	24.65%
	Acute systemic exposure mg/kg bw/day	0.0333	% of RVAAS	1.96%

PPE = Personal Protective Equipment
RVNAS = Reference value non acutely toxic active substance
RVAAS = Reference value acutely toxic active substance

CP 7.2.1.2 Measurement of operator exposure

Since the risk assessments carried out indicated that the AOEL for **ethephon** was not be exceeded under practical conditions of use, a study to provide a measure of operator exposure under field conditions was not necessary and was therefore not carried out.

CP 7.2.2 Bystander and resident exposure

The EFSA guidance has proposed a number of changes to current practice in assessing exposure to plant protection products. These changes include the introduction of acute risk assessments and the application of an AAOEL value (Acute Acceptable Operator Exposure Level) - a term used to describe a reference value against which acute non-dietary exposures (i.e. those that might be incurred in a single day) could be assessed. Currently, however, no methodology is available for setting an AAOEL. Non-dietary risk exposure is primarily via dermal and inhalation routes. Thus, the derivation of an AAOEL will differ from the procedure of setting an ARfD, which is used in dietary risk assessments where oral exposure is relevant. It will require careful evaluation, expert judgment or even additional data to determine which toxicological information should be used for AAOEL setting. ~~It is therefore proposed that an acute risk assessment is made when an agreed guideline is available for establishing an AAOEL. The following risk assessment therefore considers the longer term exposure which will be compared with the AOEL. In this context only resident exposure is calculated using the EFSA model3 and is considered as covering the bystander exposure~~.

Even though, no agreed guideline is available for establishing an AAOEL, an AAOEL of 1.7 mg/kg bw/day has been derived and an acute risk assessment is performed.

The following risk assessment considers short- and long-term exposures which will be compared with the AAOEL and AOEL, respectively. In this context both bystander and resident exposure is calculated using the EFSA model.

The intended outdoor uses comprise cereals crops. Treatment is achieved via downward vehicle-mounted spray application. The application parameters of the critical GAPs (cGAPs), following a risk envelope approach, are summarised in **Table 7.2.2-1**

Table 7.2.2-1 Critical GAP for bystander and resident exposure assessment

Application technique	Crop	F / G	Maximum dose rate		Spray volume (L/ha)	Number of applications	Application interval (days)
			(L/ha product)	(kg a.s./ha)			
Outdoor Vehicle-Mounted Downward Spraying	Winter cereals	F	1	0.48	200	1	-

Consideration on estimation of bystander/resident exposure

Bystander/resident exposure estimations to Ethephon SL 480 G are estimated using the EFSA model[3] with the relevant scenario "Tractor-mounted/trailed boom sprayer: hydraulic nozzles". Details of calculations are given in **CP 7.2.2.1**. The results of exposure calculations are summarised in **Table 7.2.2-2**.

Table 7.2.2-2. Predicted systemic bystander and resident exposure [mg Ethephon/kg bw/day] as a proportion of the AOEL

Application Scenario	Exposure Scenario		Total systemic exposure (mg ETP[#]/kg bw/day)[*]	% of AOEL[**]	% of AAOEL[***]
Field Crop	Resident Child	All pathways (mean)	0.0067	33	-
	Resident Adult	All pathways (mean)	0.0025	13	-
	Bystander Child	Spray drift	0.0076	-	0.44
		Vapour	0.0011	-	0.06
		Surface deposits	0.0021	-	0.12
		Entry into treated crops	0.0041	-	0.24
	Bystander Adult	Spray drift	0.0020	-	0.12
		Vapour	0.0002	-	0.01
		Surface deposits	0.0005	-	0.03
		Entry into treated crops	0.0023	-	0.13

[*] 60 and 10 kg body weight for adult and children respectively; dermal absorption 1% 5% diluted spray; inhalation absorption 100%; [**] 0.02 mg/kg bw/day; [***] 1.7 mg/kg bw/day
[#] ETP = Ethephon

Assessment

The EFSA Guidance estimates show that for the intended use of the formulation **Ethephon SL 480 G** the predicted systemic exposure is maximally 33% and 13% for the child and adult resident, respectively, and <1% for the child and adult bystander.

The results of the calculations reveal that the situation is favourable for bystanders and residents for the intended outdoor uses of **Ethephon SL 480 G** for winter cereals.

Conclusion

Based on above presented results there is no unacceptable risk anticipated for bystanders and residents, both adults and children, exposed to ethephon with the intended uses of **Ethephon SL 480 G**.

CP 7.2.2.1 Estimation of bystander and resident exposure

The following definitions and assumptions for bystanders/residents may be applied.

[3] EFSA Panel on Plant Protection Products and their Residues (PPR); Guidance on Pesticides Exposure Assessment of Operators, Worker, Residents and Bystanders, EFSA Journal 2014;12(10):3874.

Bystanders and residents are not involved in application or handling plant protection products or the professional handling of treated crops.
Bystander/resident exposure may occur following foliar spray application outdoors. Bystander/resident exposure is calculated regarding the application scenario leading to the highest drift value. Application scenarios causing lower spray drift will be covered by this calculation and separate evaluations are not made. Exposure is calculated for adult and child residents.

Data used for the calculation

The following assumptions have been made in calculating resident exposure:

- The application rate is 1.0 L/ha of Ethephon SL 480 G resulting in 0.48 kg of ethephon.

Table 7.2.2.1-1: Detailed calculations of resident exposure to ethephon, absorbed dose and % of AOEL

Substance	ethephon	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.48 kg a.s. /ha	Spray dilution = 2.4 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10-3Pa	
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 = 5	Oral = 100	Inhalation = 100		
RVNAS	0.02 mg/kg bw/day		RVAAS	1.7 mg/kg bw/day		
DFR	3 µg a.s./cm2 per kg a.s./ha		DT50	30 days		

Resident - child	Spray drift (75th percentile) mg/kg bw/day	0.0033	% of RVNAS	16.35%
	Vapour (75th percentile) mg/kg bw/day	0.0011	% of RVNAS	5.35%
	Surface deposits (75th percentile) mg/kg bw/day	0.0007	% of RVNAS	3.70%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0041	% of RVNAS	20.25%
	All pathways (mean) mg/kg bw/day	0.0067	% of RVNAS	33.26%
Resident - adult	Spray drift (75th percentile) mg/kg bw/day	0.0008	% of RVNAS	3.87%
	Vapour (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	1.15%
	Surface deposits (75th percentile) mg/kg bw/day	0.0002	% of RVNAS	0.82%
	Entry into treated crops (75th percentile) mg/kg bw/day	0.0023	% of RVNAS	11.25%
	All pathways (mean) mg/kg bw/day	0.0025	% of RVNAS	12.57%

Bystander - child	Spray drift (95th percentile) mg/kg bw/day	0.0076	% of RVAAS	0.44%
	Vapour (95th percentile) mg/kg bw/day	0.0011	% of RVAAS	0.06%
	Surface deposits (95th percentile) mg/kg bw/day	0.0021	% of RVAAS	0.12%
	Entry into treated crops (95th percentile) mg/kg bw/day	0.0041	% of RVAAS	0.24%
Bystander - adult	Spray drift (95th percentile) mg/kg bw/day	0.0020	% of RVAAS	0.12%
	Vapour (95th percentile) mg/kg bw/day	0.0002	% of RVAAS	0.01%
	Surface deposits (95th percentile) mg/kg bw/day	0.0005	% of RVAAS	0.03%
	Entry into treated crops (95th percentile) mg/kg bw/day	0.0023	% of RVAAS	0.13%

RVNAS = Reference value non acutely toxic active substance (equivalent to AOEL)
RVAAS = Reference value acutely toxic active substance

CP 7.2.2.2 Measurement of bystander and resident exposure

Since the exposure estimate carried out indicated that the health-based limit values (AOEL) will not be exceeded under practical conditions of use, a study to provide a measure of bystander and resident exposure was not necessary and was therefore not carried out.

CP 7.2.3 Worker exposure

Risk assessment for worker

The greatest potential for worker exposure following re-entry will be contamination *via* the skin. Risk of inhalation exposure during re-entry is generally confined to a brief period after application, while the product is drying, which will be rapid under outdoor conditions and would generally be avoided according to good agricultural practices.

Consideration on dermal exposure of workers:

Worker exposure estimation to **Ethephon SL 480 G** was calculated on the basis of the "*EFSA Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products*"[2]. Exposure predictions were obtained out of the available version of the exposure calculation spreadsheet[3].

Dermal exposure from contact with residues on foliage should be estimated as the product of the dislodgeable foliar residue (DFR), the transfer coefficient (TC) and the task duration (T):

Potential dermal exposure (PDE) in mg a.s./day = (DFR [µg/cm²] × TC [cm²/h] × T [h/day])/1 000

The default value for time of exposure should be taken as two hours for crop inspection and irrigation-type activities.

Consideration on Dislodgeable Foliar Residues (DFR):

As experimentally determined DFR data are not available, the initial DFR (DFR0 is the DFR just after application, it assumes that no dissipation will take place and that everything is dislodgeable) in a first tier assessment should assume 3 µg active substance/cm² of foliage/kg a.s. applied/ha; the value provided was regarded as highly conservative (EUROPOEM II, 2002)[4].

Transfer Coefficients:

The indicative TC values are based and modified from EUROPOEM II (2002)[5] and in consideration of US EPA values. US Re-entry Agricultural Transfert Factor (TF) data were used, recalculated by Health and Safety Executive to account for 75th percentile instead of arithmetic mean.

For crop inspection, the TC values of 12 500 cm²/h (total potential exposure) and 1 400 cm²/h (assuming arms, body and legs covered (bare hands)) were considered.

[4] [redacted] et al (2002) Post-application exposure of workers to pesticides in agriculture. Report of the re-entry working group. EUROPOEM II project. FAIR3 CT96-1406.

Predicted exposures are compared with the AOEL of ethephon. Systemic exposure values assume the highest dermal absorption values. A body weight of 60 kg is assumed for the re-entry worker. Exposure estimates based proportions of the systemic AOEL accounted for by the estimates are summarised in the following table. Detailed calculations are presented below.

Table CP 7.2.3-1: Summary of predicted worker exposures arising from the use of ethephon in the Ethephon SL 480 G formulation and comparison with the AOEL

Active substance	Exposure scenario	Systemic exposure[3)] (mg/kg bw/day)	AOEL (mg/kg bw/day)	% of AOEL**
Ethephon	Without PPE[1)]	0.0300	0.02	**150**
	With PPE[2)]	0.0034		**17**

[1)] without working clothes- bare hands; [2)] with working clothes-bare hands;
[3)] 60 kg body weight; dermal absorption 5% concentrate and 5% dilution; inhalation absorption 100%
** 0.02 mg/kg bw/day;

Assessment

The EFSA OPEX estimates show that for the intended use of the formulation **Ethephon SL 480 G** the predicted systemic exposure during crop inspection for the unprotected worker wearing working clothing is 17%.

The exposure of workers entering treated areas is well within acceptable limits **Ethephon SL 480 G**.

Conclusion

Based on above presented results there is no unacceptable risk anticipated for workers with the intended uses of **Ethephon SL 480 G** entering in cereals fields for inspection. Working clothes must be worn during re-entry activities for safety reasons, according to good agricultural practices.

CP 7.2.3.1 Estimation of worker exposure

Details of calculations are presented in table CP 7.2.3.1-1.

Table CP 7.2.3.1-1 Predicted systemic worker exposure to ethephon according to EFSA without and with PPE (cereals inspection)

Substance	ethephon	Formulation = Soluble concentrates, emulsifiable concentrate, etc.	Application rate- 0.48 kg a.s. /ha	Spray dilution = 2.4 g a.s./l	Vapour pressure = low volatile substances having a vapour pressure of <5*10-3Pa
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 = 5	Oral = 100	Inhalation = 100	
RVNAS	0.02 mg/kg bw/day		RVAAS	1.7 mg/kg bw/day	
DFR	3 µg a.s./cm2 per kg a.s./ha		DT50	30 days	

Worker - Inspection, irrigation	Potential exposure mg/kg bw/day	0.0300	% of RVNAS	150.00%
	Working clothing mg/kg bw/day	0.0034	% of RVNAS	16.80%

RVNAS = Reference value non acutely toxic active substance (equivalent to AOEL)
RVAAS = Reference value acutely toxic active substance

CP 7.2.3.2 Measurement of worker exposure

Since the exposure estimate carried out indicated that the acceptable operator exposure level (AOEL) will not be exceeded under practical conditions of use, a study to provide a measure of worker exposure was not necessary and was therefore not carried out.

CP 7.3 Dermal absorption

As the technical material is known to cause severe skin irritation and is classified as being corrosive and the formulation was essentially a water dilution of the active ingredient with a pH value that is less than 2 there were concerns that there would be skin damage leading to increased dermal absorption when testing the neat formulation. Therefore a skin tolerance study was performed before choosing the dose levels for the *in vivo* study leading to a 1:10 dilution of 48 g/L being used as the high dose and 0.48 g/L being the representative spray dilution.

Table CP 7.3-1 presents the worst case data from the rat *in vivo* study along with those from the comparative *in vitro* human and rat skin study and the estimated *in vivo* human dermal absorption results based on the application of the "triple pack" approach. The values are obtained from the sum of the directly absorbed fraction and the skin in both cases. The stratum corneum was not included as the absorption profiles met the EFSA guidance criteria for exclusion.

Table CP 7.3-1: Percentage dermal absorption values for ethephon in the SL formulation from the *in vivo* and *in vitro* studies and the estimated human *in vivo* values

Concentration	*In vivo* rat results* (%)	*In vitro* rat skin results* (%)	*In vitro* Human skin results* (%)	*In vitro* human/rat ratio	Estimated human *in vivo* dermal absorption (%)
480 g/L	n.a.	77.3	93.3	1.21	n.a.
48 g/L	4.27	53.3	56.9	1.07	4.6
4.8 g/L	n.a.	16.0	10.7	0.67	n.a.
0.48 g/L	4.04	19.0	3.4	0.18	0.72

*EFSA guidance rules applied apart from the "rounding" of the values.

The dermal absorption values that are recommended for the non-dietary risk assessments for the neat or concentrated formulation is **5%**, **5%** for the in-use dilution of 2.4 g/L and **1%** for the spray dilution (0.48 g/L).

Dermal absorption, in vivo in the rat

Report: KCP 7.3/01; [redacted]; 2004; M-228640-01-1
Title: [14C]-Ethephon: In vivo dermal absorption in the male rat
Report No.: C040292
Document No.: M-228640-01-1
Guideline(s): OECD Guideline for Testing of Chemicals, Toxicokinetics, 417, Adopted April 1984 using the latest draft of the OECD Test Guideline 427 and the respective OECD Guidance Documents for the conduct of in vivo skin adsorption studies (April 2002)
Guideline deviation(s): not specified
GLP/GEP: yes

Material and methods

Rat:
Species, strain: Sprague Dawley CRL:CD (SD) BR.
Source: [redacted].
Sex: Male.
Body weights: 164 - 243 g.
Age: Approx. 6 to 8 weeks.
Acclimatisation & Housing: The animals were housed in individual metabolism cages following treatment.
Animal identification: Tail marking.
Environmental conditions: Temperature: 21 ± 2°C.
Humidity: 40-70%.
Photoperiod: 12 hour light/dark cycles.
Air changes Ca. 15/hour.

Food: Free access to pelleted laboratory rodent diet.
Water: Free access to mains water.

Test Material:
Non-radiolabelled: Batch: 1263X.

Purity = 98%.

Radiolabelled: [^{14}C]-Ethephon.
Batch: CFQ13111 and CFQ13687.

Specific activity: 13.5 & 13.3 MBq/mg.

Radiopurity of the formulation: 98.26% & 99.2%.

Formulation: The formulation used in this experiment was the Cerone SL 480 formulation EXP03725B formulation (Spec [redacted] 102000001957) containing ethephon. It was used at 2 nominal concentrations; 48 g/L and 0.48 g/L. Since the commercial formulation is irritant to skin the dose levels were based on results of a skin tolerance study in male rats (Esdaile D., Cerone: Skin tolerance in male rats, [redacted]), January 10th, 2003).

Treatment & Sampling: Prior to the two main experiments a preliminary experiment was performed to obtain an indication of the proportion of the test substance absorbed through the skin and excreted, expired, retained in the skin, or remaining on the skin surface following topical application of both the low and high dose formulations. The results from the preliminary study were used to determine the sacrifice times used in the main study, the need to trap expired air and material evaporating from the dose site, the need to investigate the remaining material in the skin and its localisation, and any requirement to examine the tissue distribution of radioactivity.

An area of dorsal skin (at least 3x4 cm) was clipped approximately 16-24 hours prior to dosing. At dosing, a silicone rubber saddle was secured in place on the clipped area of skin using cyanoacrylate adhesive. The dose formulation was applied to the clipped area using a calibrated pipette. Each rat received 120 µL of the dose solution (equivalent to 10 µL/cm^2) which was spread over the application site (3 x 4 cm) using the pipette tip as a spreader. The pipette tip was retained for analysis to determine the residual dose.

In the main study, a second detachable saddle was placed over the first and covered with a stainless steel gauze and taped into place with surgical tape and Vetrap™ bandage, to prevent loss of test substance while permitting air circulation over the application site. Following dose application, the animals were placed in metabolism cages that allowed the separate collection of urine, faeces and expired air. No filters were used.

At 8 hours the gauze and tape were removed and retained for analysis. The treated area was washed with cotton wool swabs soaked in 1% v/v Tween 80 in aqueous sodium chloride (0.9 g/L) in order to remove and retain non-absorbed dose. The swabs were taken for analysis.

In the main study, animals that were required to provide samples beyond 8 hours were then fitted with a clean gauze cover and tape/bandage and replaced in the metabolism cage. Urine and faeces were collected separately from each animal into containers cooled in solid carbon dioxide at 0 to 8, 8 to

24, and at 24-hour intervals up to sacrifice. The expired air was collected from 0 to 8 hours into four traps in series, the first containing methanol, the second and third containing 0.2 M mercury (II) acetate in methanol and the fourth containing 2M aqueous potassium hydroxide. Similar collection of expired air from 8 to 24 hours was carried out for rat 38 only. However, this was not continued in the remainder of the main study because of the risk of precipitation of the mercury acetate in the air pipes overnight which could block the air supply to the rats. The interior of each metabolism cage was washed with water at each sample point and the washings retained for analysis. A terminal blood sample was taken, from each animal, approximately 2 mL, via the orbital sinus into vials containing lithium heparin.

The treated area of skin was tape stripped following dissection. The area of skin, approximately 1 cm in width surrounding the site of dose application was removed to investigate leaching of the dose through the skin. The untreated skin and the residual carcass were also taken for analysis. Dressings and gauze covers removed from the animals were retained and taken for analysis.

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Radioassy: Radioactivity was measured by liquid scintillation counting (LSC), using either a LKB Backbeta 1219 (Perkin Elmer Life Sciences, Milton Keynes, Bucks, UK) or a Tricarb 2300 TR (Packard, Pangbourne, Berks, UK) liquid scintillation counter. Quench correction was achieved via an automatic external standard ratio method. Samples were generally counted for a total of 4 minutes or 900 000 counts, whichever occurred first. The limit of detection was derived statistically from the background counts so that there was a 99% certainly that samples with a mean value greater than the limit of detection will contain radioactivity from the [^{14}C]-Ethephon.

The limit of detection throughout the study was approximately 6 dpm, which is equivalent to approximately 0.939 ng Ethephon in the high dose formulation and 0.011 ng in the low dose formulation. This is equivalent to 0.00002% and 0.00002% at high and low dose levels respectively.

Findings:

The radioactivity in aliquots taken from the top, middle and bottom of the dose formulations were within 3% of the mean activity for the three regions. No appreciable concentration gradient was observed. Therefore, the dose formulations were considered to be homogeneous.

48 g/L dose group

The mean total recoveries of radioactivity were in the range 96.29% to 98.55%.

After a single application of the high level dose formulation of [^{14}C]-Ethephon to four groups of male rats, the mean total amount of non-absorbed radiolabelled material (skin swabs; surface dose located in shavings, razor head wash and the first two tape strips; and gauze wash) was 73.76% to 92.64%. The majority of this material was removed from the treated site, by swabbing, after 8 hours exposure to the formulation (mean values were in the range 53.07% to 61.59% applied dose). The radiolabelled material lost by desquamation was recovered in the terminal gauze wash plus surface dose (shavings, razor head wash and the first two tape strips). The mean amount of radiolabelled material lost by desquamation increased from 11.73% at 24 hours to approximately 35% at 72 and 120 hours after dosing.

Following tape stripping of the treated skin, mean amounts of radioactivity recovered in the stratum corneum decreased from 22.54% at 8 hours to 4.56% by 120 hours.

From measurements of total radioactivity in excreta, carcasses, untreated skin (including skin surrounding the dose site) and treated skin (after tape stripping), it was shown that the amount of material absorbed varied between 0.51% and 2.31% of applied dose at the four timepoints. Excreted radioactivity was in the range 0.45 % to 2.04%. The amount of radioactivity remaining in the tissues (untreated skin, skin surrounding the dose site and carcass) was in the range 0.07% to 0.27%. The material remaining in the treated skin, following removal of both residual dose and stratum corneum, decreased from 0.69% at 8 hours to 0.32% at 120 hours.

Table CP 7.3.1-1.: The mean distribution of radioactivity 8, 24, 72 and 120 hours after a single topical application of [^{14}C]-ethephon in the 48 g/L dilution of the 480 SL formulation

Results expressed as % of applied dose:

Dose Group 48 g/L	Hours post application							
	8		24		72		120	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Skin Swabs 8 hour	59.17	11.33	61.59	5.44	53.02	3.29	56.96	5.56
Total Gauze Washes	2.40	2.05	1.21	1.08	2.28	2.20	1.05	0.97
Stratum Corneum Strips 1& 2	11.62	6.17	11.52	6.25	3.57	1.34	4.33	1.67
Fur: razor wash	n.s.	n.a.	n.s.	n.a.	0.60	0.09	0.84	0.37
Fur: shavings	n.s.	n.a.	n.s.	n.a.	30.11	5.76	28.52	2.93
Non Absorbed	**73.19**	**10.23**	**75.03**	**2.61**	**90.85**	**2.58**	**97.63**	**12.60**
Stratum Corneum	22.54	6.66	21.38	3.46	2.68	1.00	4.50	3.57
Skin	**0.69**	**0.29**	**0.69**	**0.24**	**0.46**	**0.42**	**0.32**	**0.13**
Dose site	23.29	6.51	22.13	3.36	3.26	0.99	4.90	3.73
Carcass	0.15	0.10	0.07	0.03	0.27	0.17	0.10	0.08
Faeces	0.02	0.02	0.01	0.00	0.01	0.01	0.02	0.01
Urine	0.89	0.84	0.29	0.25	0.98	0.69	0.33	0.11
Cage wash	0.35	0.36	0.09	0.05	0.66	0.47	0.14	0.14
Expired air	0.15	0.12	0.06	0.02	0.39	0.19	0.08	0.03
Directly Absorbed	**1.49**	**1.39**	**0.81**	**0.33**	**2.19**	**1.24**	**0.59**	**0.28**
Directly Absorbed + Dose Site	24.78	7.66	22.57	3.49	5.44	1.33	5.49	3.83
Directly Absorbed + Skin	***2.24***	***1.41***	***1.19***	***0.43***	***2.77***	***1.50***	***1.00***	***0.40***
Total Recovered	98.54	4.86	97.60	2.75	96.29	3.04	98.20	7.69
Evaluation according to EFSA Guidance								
% of total absorption occurring within half of the duration of the total sampling period	The mean % total absorption occurring by the mid-point of the sampling period (72h) 79% and therefore the stratum corneum can be excluded from the absorbed fraction							
standard deviation >±5%	Yes		Yes		Yes		Yes	
recovery <95%	No		No		No		No	
adjusted: Total % Potentially Absorbable	3.65		1.62		4.27		1.4	

SD: standard deviation, n.s. = no sample n.d. = not detected n.a. = not applicable.

0.48 g/L dose group

The mean total recoveries of radioactivity were in the range 92.07% to 94.58%.

After a single application of the low level dose formulation of [^{14}C]-Ethephon to four groups of male rats, the mean total amount of non-absorbed radiolabelled material (skin swabs; surface dose located in shavings, razor head wash and the first two tape strips; and gauze wash) was 69.97% to 91.05%. The majority of this material was removed from the treated site, by swabbing, after 8 hours exposure to the formulation (mean values were in the range 57.41% to 66.77% applied dose). The radiolabelled material lost by desquamation was recovered in the terminal gauze wash plus surface dose (shavings,

razor head wash and the first two tape strips). The mean amount of radiolabelled material lost by desquamation increased from 5.90% at 24 hours to 18.62% at 120 hours after dosing.
Following tape stripping of the treated skin, mean amounts of radioactivity in the stratum corneum increased from 17.06% at 8 hours to 22.28% at 24 hours after dosing, then decreased to 2.06% at 120 hours after dosing.
From measurements of total radioactivity in excreta, carcasses, untreated skin (including skin surrounding the dose site) and treated skin (after tape stripping), it was shown that the amount of material absorbed varied between 1.16% and 2.59% of applied dose at the four timepoints. Excreted radioactivity was in the range 0.90 % to 2.11%. The amount of radioactivity remaining in the tissues (untreated skin, skin surrounding the dose site and carcass) was in the range 0.19% to 0.75%. The material remaining in the treated skin, following removal of both residual dose and stratum corneum, decreased from 0.59% at 8 hours to 0.31% at 120 hours.

Table CP 7.3.1-2.: The mean distribution of radioactivity 8, 24, 72 and 120 hours after a single topical application of [^{14}C]-ethephon in the 0.48 g/L dilution of the 480 SL formulation

Results expressed as % of applied dose:

Dose Group 0.48 g/L	Hours post application							
	8		24		72		120	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Skin Swabs 8 hour	57.41	4.32	61.96	3.69	72.06	62.37	66.77	4.67
Total Gauze Wipes	8.13	6.05	2.51	2.17	0.62	2.45	7.87	4.72
Stratum Corneum Strips 1& 2	7.08	5.35	5.71	3.41	3.17	5.96	2.12	0.90
Fur: razor wash	n.s.	n.a.	n.s.	n.a.	0.14	0.15	0.27	0.17
Fur: shavings	n.s.	n.a.	n.s.	n.a.	8.93	11.46	14.03	2.90
Non Absorbed	73.22	4.66	69.97	3.57	84.92	82.39	91.05	4.91
Stratum Corneum	9.86	5.17	22.28	2.25	4.46	6.81	2.06	2.17
Skin	1.15	0.95	0.30	0.06	0.56	0.74	0.48	0.39
Dose site	18.21	5.26	22.58	2.28	5.02	7.54	2.54	2.07
Carcass	0.13	0.1	0.00	0.00	0.00	0.01	0.02	0.03
Faeces	0.00	0.01	0.01	0.01	0.00	0.14	0.00	0.00
Urine	0.59	0.64	0.19	0.05	0.06	0.17	0.04	0.03
Cage wash	0.35	0.36	0.06	0.02	0.03	0.27	0.05	0.03
Expired air	0.15	0.13	1.40	0.11	3.24	1.63	0.81	0.49
Directly Absorbed	1.41	0.28	1.66	0.11	3.33	2.22	0.99	0.47
Directly Absorbed + Dose Site	19.62	5.20	24.24	2.21	8.35	9.76	3.53	2.46
Directly Absorbed + Skin	2.60	1.13	1.96	0.09	3.89	2.95	1.47	0.52
Total Recovered	92.88	1.51	94.20	1.44	93.29	92.07	94.58	3.22
Evaluation according to EFSA Guidance								
% of total absorption occuring within half of the duration of the total sampling period	The mean %total absorption occurring by the mid-point of the sampling period (72h) 88% and therefore the stratum corneum can be excluded from the absorbed fraction							
standard deviation >25%	Yes		No		Yes		Yes	
Adjusted to SD Total % potentially absorbable	3.73		2.03		3.72		1.98	
recovery <95%	Yes		Yes		Yes		Yes	
adjusted: Total % Potentially Absorbable	4.02		2.15		4.04		2.09	

SD: standard deviation, n.s. = no sample n.d. = not detected n.a. = not applicable.

Conclusion:
The distribution patterns of radioactivity following a single topical application of high and low dose level formulations of [^{14}C]-Ethephon (nominally 48.0 g/L and 0.48 g/L respectively) to male rats were investigated during this study.

In both dose groups a large proportion of the radioactivity was removed from the skin by swabbing with a mild detergent solution at 8 hours to remove the residual surface dose. This accounted for 53.07% to 61.59% and 57.41% to 66.77% of the applied dose at the high and low dose levels, respectively.

In both dose groups a significant amount of radioactivity was recovered in the stratum corneum, this accounted for 2.68% to 22.54% and 2.06% to 22.28% of the applied dose at the high and low dose levels, respectively. At both high and low dose levels the activity remaining in the stratum corneum decreased with time. The distribution profiles of radioactivity observed following the tape stripping procedure showed that the majority of the [^{14}C]-Ethephon remained in the upper layers of the stratum corneum at all timepoints in both dose groups. This suggests that there is little migration of ethephon through the stratum corneum with time. After swabbing, the amount of radioactive material lost by desquamation (terminal gauze wash plus surface dose) increased with time from 11.72% to 34.62% and from 5.90% to 18.62% at high and low dose levels, respectively. As the activity is lost from the stratum corneum with time so the activity lost by desquamation increases correspondingly. The amount of activity remaining in the tissues and lost by excretion are comparatively low. This indicates that any activity found in the stratum corneum will be lost mainly by desquamation with time and so should be considered as non-absorbed material.

According to the new EFSA guidance[5] there is the provision that when the sampling period is 24 hours or longer (which is the case for this study) and over 75% of the total absorption (material in excreta, exhaled gasses and in the carcass at the end of the study) occurred within half of the duration of the total sampling period then the absorption will be taken as the sum of the excreta, exhaled air, carcass and skin (excluding the stratum corneum). These criteria were met for both dose groups in this study. Additionally a standard deviation equal to or larger than 25% of the mean of the absorption requires the use of an alternative value on rejection of the study. The guidance prefers the approach of adding the standard deviation to the mean to cover the upper 80th percentile value of the results. Additionally where an overall recovery of less than 95% occurs, a normalisation procedure is to be used by preference. Albeit that the notifier considers that both the value of 25% for the standard deviation limit and the 95% recovery limit to be too conservative, the application of the guidance results in the following values for ethephon in the ETP SL 480 formulation taking the worst case 72 hour values:

- 4.27% for the 1/10 dilution of the neat formulation (48 g/L)
- 4.04% for the 1/1000 dilution (0.48 g/L).

The dermal absorption of ethephon in **Ethephon SL 480 G** in rats was 4.27% for the 1:10 dilution of the concentrate 48 g/L and 4.04% for the 1/1000 dilution (0.48 g/L).

[5] EFSA Panel on Plant Protection Products and their Residues (PPR); Guidance on Dermal Absorption. EFSA Journal 2012;10(4):2665. [30 pp.] doi:10.2903/j.efsa.2012.2665.

Comparative dermal absorption, in vitro using rat and human skin

Report:	KCP 7.3/02; [redacted]; 2004; M-228412-01-1
Title:	[14C]-Ethephon - Comparative in vitro dermal penetration study using human and rat skin
Report No.:	C040169
Document No.:	M-228412-01-1
Guideline(s):	EU 91/414/EEC, Annex III, Section 7.3 (OECD: 417 + draft 428)
Guideline deviation(s):	not specified
GLP/GEP:	**yes**

Material and methods

Rat skin:

Species, strain:	Rat, Sprague-Dawley strain.
Source:	[redacted].
Sex:	Male.
Anatomical site:	Dorsal.
Rat Skin Preparation:	Each rat was sacrificed by cervical dislocation. After sacrifice, the body was shaved with electric clippers and the skin removed. Connective tissue, blood and any residual fat were removed from the dermis using absorbent tissue. The resulting full thickness skin membrane was then wiped briefly with 70% ethanol/water (v/v) and dried with absorbent tissue. The skin was then re-hydrated with distilled water ready for dermatoming. The full thickness skin sample was pinned out on a dermatome board (cork board with raised rubber cutting surface) and a mini-dermatome used to cut slices of skin which contained epidermis and some dermis (thickness measured using a digital calliper to be approximately between 300-400 um thick).

Human skin: Source: International Institute for the Advancement of Medicine, USA.

Number and sex: 4 donors, 1 male & 3 female.

Anatomical region: Back.

Preparation: Prior to use, human skin samples were thawed to room temperature. Each full thickness skin membrane was then swabbed with 70% v/v ethanol/water to remove residual fat and blood, wiped dry and re-hydrated with distilled water prior to dermatoming as per the rat skin.

Non-radiolabelled:	Batch: 1263X. Purity = 98%.
Radiolabelled:	[^{14}C]-Ethephon Batch: CFQ13111 and CFQ13687. Specific activity: 13.5 & 13.3 MBq/mg. Radiopurity of the formulation: 98.26% & 99.2%.

Formulation: The formulation used in this experiment was the ethephon (Cerone) SL 480 EXP03725B formulation (Spec. N° 102000001937). It was used at 4 nominal concentrations: 480 g/L, 48 g/L, 4.8 g/L and 0.483 g /L.

Test system: The Scott-Dick flow-through diffusion cell was constructed from PTFE and permitted the contents of the receptor chamber to be continuously stirred. The skin membranes were maintained at approximately 32°C using a water-heated manifold. The flow-rate of 1.5 mL/hr allowed approximately 6 receptor chamber content changes per hour. The receptor fluid used was phosphate buffered saline, (pH7.4).

Skin samples were cut from the dermatomed slice and placed onto the receptor chamber of the flow-through diffusion cell. The donor chamber was then fixed in place providing an exposure area of 0.64 cm^2 of skin and the assembled diffusion cell inserted in-line in the flow-through set-up.

Skin integrity: The integrity of the selected skin samples was checked by measuring the penetration of tritiated water (^3H_2O) through each membrane prior to application of [^{14}C]-ethephon.

Treatment: Each dose formulation was applied to the skin membrane with a calibrated positive displacement pipette at the rate of approximately 10 µL/cm^2 exposed skin area (6.4 µL dose, unoccluded). The actual amount of [^{14}C]-ethephon applied to the skin was determined from aliquots (6.4 µL) of each dose formulation (QC checks) taken before, during and after dosing each group of cells.

Immediately after dose administration, the donor chambers were covered with carbon filters (Groups 1 and 2) or mercuric acetate filters (Groups 3, 4, 5, 6, 7 and 8), in order to trap any evolved ethylene.

Sampling: The receptor fluid passing through the receptor chamber was collected into plastic scintillation vials held in a fraction collector. The fraction collector was moved on after dose application until each group was complete. Samples were then collected hourly for the duration of the experiment (24 hours).

After 8 hours, the skin was swabbed with 1% v/v Tween 80 in aqueous saline until no further radioactivity was removed (confirmed by monitoring the swabs with a Geiger-Müller mini-monitor). The carbon filters (Groups 1 and 2) or mercuric acetate filters (Groups 3, 4, *5,* 6, 7 and 8) were removed and replaced with new filters. These were subsequently collected at the end of the study.

After 24 hours exposure, the skin membranes were tape-stripped using 3 M Scotch 'Magic' tape. The tape strips were collected into glass vials. The remaining skin was retained and analysed separately.

The receptor fluid remaining in the cell and outlet tubing at the end of the experiment was retained and analysed for mass balance purposes. The diffusion cell components were also retained washed and the washings analysed for mass balance purposes.

All samples that were not analysed immediately after collection were stored at approximately -20°C as soon as possible after collection.

Radioassay: Radioactivity was measured by liquid scintillation counting (LSC). Generally radioactivity in gross amounts of less than twice background (4-minute counts) was considered to be below the limit of detection.

Findings:

The radioactivity in aliquots taken from the top, middle and bottom of the dose formulations was within 4% of the mean activity for the three regions, with no appreciable concentration gradient observed. Therefore, the dose formulations were considered to be homogeneous.

The target application rates of Ethephon were approximately; low dose equivalent to 4.8 µg/cm^2 (approximately 3.072 µg per skin sample); intermediate doses equivalent to 48 µg/cm^2 (approximately 30.72 µg per skin sample) and 480 µg/cm^2 (approximately 307.2 µg per skin sample), and high dose equivalent to 4800 µg/cm^2 (approximately 3072 µg per skin sample). These four dose levels are possible exposure levels in the field. The application rate was 6.4 µL of the dose per 0.64 cm^2 skin (semi-occluded) i.e. 10 µL/cm^2. Measurements of the achieved doses of the two concentrations of formulation applied indicated that it was acceptable.

<u>High dose (480 g/L)</u>

For human skin, 72.51% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained 3.497% of the applied dose, with 0.297% of the dose removed in the surface tape strips taken at 24 hours. The carbon filter extracts contained 0.116% of the applied dose. Tape strips of the stratum corneum accounted for 1.074% of the dose. The remaining skin contained 7.769% of the dose. The overall mean recovery of the dose was 83.51%.

For rat skin, 66.17% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained 18.00% of the applied dose, with 0.376% of the dose removed in the surface tape strips taken at 24 hours. The carbon filter extracts contained 0.106% of the applied dose. Tape strips of the stratum corneum accounted for 0.889% of the dose. The remaining skin accounted for 1.474% of the dose. The overall mean recovery of the dose was 87.59%.

<u>Intermediate Dose 1 (48 g/L)</u>

For human skin, 33.26% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained a large proportion of the applied dose (20.98%), with 4.400% of the dose removed in the surface tape strips taken at 24 hours. No radioactivity was detected in the mercuric acetate filter extracts. Tape strips of the stratum corneum accounted for 25.85% of the dose. The remaining skin contained *6.235%* of the dose. The overall mean recovery of the dose was 93.31%.

For rat skin, 40.81% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained a large proportion (35.51%) of the applied dose with 1.922% of the dose removed in the surface tape strips taken at 24 hours. No radioactivity was detected in the mercuric acetate filter extracts. Tape strips of the stratum corneum accounted for 16.75% of the dose. The remaining skin accounted for 0.629% of the dose. The overall mean recovery of the dose was 95.63%.

Intermediate Dose 2 (4.8 g/L)

For human skin, 4.700% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained a large proportion of the applied dose (54.56%), with 6.44% of the dose removed in the surface tape strips taken at 24 hours. The mercuric acetate filter extracts contained 1.931% of the applied dose. Tape strips of the stratum corneum accounted for 31.00% of the dose. The remaining skin contained 1.217% of the dose. The overall mean recovery of the dose was 102.3%.

For rat skin, 9.648% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained a large proportion (44.30%) of the applied dose with 16.39% of the dose removed in the surface tape strips taken at 24 hours. The mercuric acetate filter extracts contained 1.777% of the applied dose. Tape strips of the stratum corneum accounted for 28.71% of the dose. The remaining skin accounted for 0.881% of the dose. The overall mean recovery of the dose was 102.4%.

Low dose level (0.48 g/L)

For human skin, 1.145% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained the majority of the applied dose (71.17%), with 3.287% of the dose removed in the surface tape strips taken at 24 hours. The mercuric acetate filter extracts contained 0.006% of the applied dose. Tape strips of the stratum corneum accounted for 16.07% of the dose. The remaining skin contained 7.631% of the dose. The overall mean recovery of the dose was 92.90%.

For rat skin, 8.894% of the applied dose was recovered in the receptor fluid over the 24 hour period. The skin swabs taken at 8 hours contained a large proportion (55.56%) of the applied dose with 2.419% of the dose removed in the surface tape strips taken at 24 hours. No radioactivity was detected in the mercuric acetate filter extracts. Tape strips of the stratum corneum accounted for 22.71% of the dose. The remaining skin accounted for 2.632% of the dose. The overall mean recovery of the dose was 93.40%.

Table CP 7.3.2-1: Mean distribution of radioactivity at 24 hours after dose application of [^{14}C]-ethephon in an SL formulation at the rates of 480 g/L and 48 g/L to human and rat skin samples

Results expressed in terms of percentage of applied radioactivity:

Dose Levels	Neat formulation: High dose (480 g/L)				Dilution: Low dose (48 g/L)			
Species	Human (n=7)		Rat (n=5)		Human (n=7)		Rat (n=7)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Skin swabs (8h)[a]	3.50	0.60	18.00	2.05	20.69	6.[illegible]	35.51	5.36
Surface dose (tape strips 1 & 2)	0.30	0.22	0.38	0.38	4.[illegible]0	5.[illegible]0	1.[illegible]3	1.[illegible]0
Donor chamber	0.59	0.61	0.38	0.22	2.31	2.03	1.28	1.50
Filter	0.12	0.02	0.11	0.03	n.d.	n.a.	n.d.	n.a.
Total % non-absorbed	**4.50**	**1.22**	***18.86***	***2.41***	***27.[illegible]***	***9.62***	***37.[illegible]***	***6.07***
Skin [b]	***5.07***	***3.55***	***1.[illegible]***	***1.50***	***6.[illegible]***	***7.50***	***0.[illegible]***	***0.53***
Stratum corneum [c]	1.07	0.39	4.89	1.43	25.86	6.30	***16.75***	***3.36***
Total % at dose site	**6.14**	**3.84**	**2.30**	**1.91**	**32.09**	**8.79**	**17.38**	**8.71**
Receptor fluid (0-24h)	72.51	0.10	66.17	8.89	33.26	13.[illegible]	40.[illegible]	12.48
Receptor Chamber	0.25	0.51	0.14	0.10	0.[illegible]	0.[illegible]	0.[illegible]	0.36
Receptor fluid terminal	0.10	0.[illegible]	0.[illegible]	0.10	0.[illegible]	0.[illegible]	0.07	0.08
Total % Directly Absorbable [d]	***72.87***	***3.16***	***60.43***	***9.86***	***33.53***	***13.57***	***40.57***	***12.48***
Total % Potentially Absorbable [e]	**79.01**	**0.45**	**68.73**	**8.09**	**65.62**	**9.40**	**57.95**	**5.98**
Total % Direct + skin only	**77.94**	**13.16**	**67.84**	**8.33**	**39.[illegible]**	**13.[illegible]**	**41.20**	**12.16**
TOTAL % RECOVERY	**83.51**	**8.42**	**87.[illegible]**	**6.[illegible]**	**93.[illegible]**	**1.[illegible]**	**95.62**	**0.88**
Evaluation according to EFSA Guidance								
absorption >75% within half of study duration	**Yes**		**Yes**		**Yes**		**Yes**	
standard deviation >25%	**No**		**No**		**Yes**		**Yes**	
Adjusted to SD Total % potentially absorbable	77.94		67.73		53.1		53.34	
recovery <95%	**Yes**		**Yes**		**Yes**		**No**	
adjusted: Total % Potentially Absorbable [f]	93.33		77.33		56.91		53.34	

[a]: sum of radioactivity found in swabs at termination and in surrounding swabs.
[b]: sum of radioactivity found in skin after tape-stripping procedure and in surrounding skin.
[c]: tape-strips excluding numbers 1 & 2 which are considered to be non-absorbed dose.
[d]: sum of radioactivity found in receptor fluid (0-24h), receptor fluid terminal and receptor chamber.
[e]: total % directly absorbed + total % at dose site
[f]: values considered for the adjusted Total % Potentially Absorbable according to EFSA are in ***bold Italics***
SD: standard deviation
n.d.: not detected (below the limit of detection)
n.a. : not applicable
n: number of skin cells used for calculation
In the above table, the presented means do not always calculate exactly from the presented individual data. This is due to rounding-up differences resulting from the use of the spreadsheet program.

Table CP 7.3.2-2: Mean distribution of radioactivity at 24 hours after dose application of [^{14}C]-ethephon in an SL formulation at the rates of 4.8 g/L and 0.48 g/L to human and rat skin samples.

Results expressed in terms of percentage of applied radioactivity.

Dose Levels	Neat formulation: High dose (4.8 g/L)				Dilution: Low dose (0.48 g/L)			
Species	Human (n=7)		Rat (n=6)		Human (n=7)		Rat (n=4)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Skin swabs (8h)[a]	54.56	12.42	44.30	4.51	71.17	4.77	55.57	5.06
Surface dose (tape strips 1 & 2)	6.44	3.95	16.39	16.27	3.28	0.84	2.42	0.69
Donor chamber	1.94	0.47	0.69	0.47	0.28	0.09	1.35	0.19
Filter	1.93	0.29	1.78	0.34	0.01	0.01	n.d.	n.a.
Total % non-absorbed	**64.88**	**12.63**	***63.16***	***16.83***	***75.04***	***5.12***	***59.37***	***6.70***
Skin [b]	***1.22***	***1.63***	***0.88***	***0.33***	***0.61***	***0.68***	***2.23***	***2.76***
Stratum corneum [c]	31.00	6.94	28.73	16.34	16.07	8.20	22.14	13.76
Total % at dose site	**32.21**	**7.56**	**29.59**	**16.58**	**16.70**	**7.67**	**24.37**	**11.08**
Receptor fluid (0-24h)	4.70	3.14	0.65	0.01	1.15	0.01	8.90	4.31
Receptor Chamber	0.44	0.21	0.01	0.02	n.d.	n.a.	0.12	0.23
Receptor fluid terminal	0.03	0.02	0.01	0.00	0.01	0.00	0.08	0.11
Total % Directly Absorbable [d]	***5.17***	***3.74***	***0.67***	***0.36***	***1.15***	***0.69***	***9.07***	***4.49***
Total % Potentially Absorbable [e]	**37.38**	**10.46**	**30.26**	**17.69**	**17.85**	**7.28**	**32.04**	**8.95**
Total % Direct + skin only	**6.38**	**4.30**	**10.55**	**5.43**	**1.78**	**1.35**	**11.33**	**6.44**
TOTAL % RECOVERY	**102.3**	**2.60**	**102.42**	**3.23**	**92.90**	**2.56**	**93.40**	**2.93**
Evaluation according to EFSA Guidance								
absorption >75% within half of study duration	Yes		Yes		Yes		Yes	
standard deviation >25%	Yes		Yes		Yes		Yes	
Adjusted to SD Total % potentially absorbable	10.68		15.98		3.13		17.77	
recovery <95%	No		No		Yes		Yes	
adjusted: **Total % Potentially Absorbable [f]**	10.68		15.98		3.37		19.03	

[a]: sum of radioactivity found in swabs at termination and in surrounding swabs.
[b]: sum of radioactivity found in skin after tape-stripping procedure and in surrounding skin.
[c]: tape-strips excluding numbers 1 & 2 which are considered to be non-absorbed dose.
[d]: sum of radioactivity found in receptor fluid (0-24h), receptor fluid terminal and receptor chamber.
[e]: total % directly absorbed + total % at dose site
[f]: values considered for the adjusted Total % Potentially Absorbable according to EFSA are in ***bold Italics.***

SD: standard deviation
n.d.: not detected (below the limit of detection)
n.a. : not applicable
n: number of skin cells used for calculation
In the above table, the presented means may not always calculate exactly from the presented individual data.
This is due to rounding-up differences resulting from the use of the spreadsheet program.

Conclusion:

The dermal penetration of [^{14}C]-ethephon through human and rat dermatomed skin from the SL 480 formulation was investigated at four concentrations corresponding to the neat product (480 g /L) and 3 dilutions of 48 g/L, 4.8 g/L and 0.48 g/L respectively.

According to the new EFSA guidance[6] there is the provision that when the sampling period is 24 hours (which is the case for this study) and over 75% of the total absorption (material in the receptor fluid at the end of the study) occurred within half of the duration (12 hours) of the total sampling period that the absorption will be taken as the sum of receptor fluid, receptor chamber washes and the skin sample excluding all tape strips. These criteria were met for all the dose groups in this study. There is also the

[6] EFSA Panel on Plant Protection Products and their Residues (PPR); Guidance on Dermal Absorption. EFSA Journal 2012;10(4):2665. [30 pp.] doi:10.2903/j.efsa.2012.2665.

provision that a standard deviation equal to or larger than 25% of the mean of the absorption requires the use of an alternative value or rejection of the study. The guidance prefers the approach of adding the standard deviation to the mean to cover the upper 84th percentile value of the results. Additionally where an overall recovery of less than 95% occurs, a normalisation procedure is to be used by preference. Albeit that the notifier considers that both the value of 25% for the standard deviation limit and the 95% recovery limit to be too conservative, the application of the guidance results in the following values for [^{14}C]-ethephon in the SL 480 formulation:

The mean percentage of ethephon in the SL 480 formulation that was considered to be potentially absorbable *(directly absorbed plus total remaining at dose site)* over a period of 24 hours for the neat formulation was 79% and 69% for the human and rat skin, respectively. Applying the new EFSA guidance these values adjust to 93.33% and 77.33% for the human and rat skin respectively.
The mean percentage of ethephon in the SL 480 formulation that was considered to be potentially absorbable *(directly absorbed plus total remaining at dose site)* over a period of 24 hours for the 48 g/L dilution was 66% and 58% for the human and rat skin respectively. Applying the new EFSA guidance these values adjust to 56.91% and 58.34% for the human and rat skin respectively.
The skin irritating properties of the active substance might have damaged the barrier properties of the skin at these higher dose levels resulting in increased absorption. At these concentrations there was no relevant difference in absorption between human or rat skin.

The mean percentage of ethephon in the SL 480 formulation that was considered to be potentially absorbable *(directly absorbed plus total remaining at dose site)* over a period of 24 hours for the 4.8 g/L dilution was 37% and 39% for the human and rat skin, respectively. Applying the new EFSA guidance these values adjust to 12.68% and 15.98% for the human and rat skin respectively.
The mean percentage of ethephon in the SL 480 formulation that was considered to be potentially absorbable *(directly absorbed plus total remaining at dose site)* over a period of 24 hours for the 0.48 g/L dilution was 18% and 34% for the human and rat skin, respectively. Applying the new EFSA guidance these values adjust to 3.37% and 19.03% for the human and rat skin respectively.
Differences in skin absorption between rat and human skin were observed at a 1:100 dilution (ratio rat:human is 1.6:1) and at a 1:1000 dilution (ratio rat:human is 19:3).

CP 7.4 Available toxicological data relating to co-formulants

CONFIDENTIAL information - data provided separately (Document JCP)

Appendix 1-1: Detailed exposure models

Operator exposure for Ethephon SL 480 outdoor spray applications

Application rate of active substance	0.48	kg a.s./ha	*i_AppRate*
Assumed area treated	50	ha/day	*d_AreaTreated*
Amount of active substance applied	24	kg a.s./day	*i_AmoutAS*
Dermal absorption of the product	5.00%		*i_AbsorpProduct*
Dermal absorption of in-use dilution	5.00%		*i_AbsorInuse*
Formulation type	Soluble concentrates, emulsifiable concentrate, etc.		
Indoor or Outdoor application	Outdoor		
Application method	Downward spraying		
Application equipment	Vehicle-mounted		
Season	not relevant		

	Exposure values	µg exposure/day mixed and loaded 75th centile	95th centile	Reference	Comment
Mixing and loading	Hands	56090	211369	AOEM	
	Body	33306	281327	AOEM	
	Head	1245	6829	AOEM	
	Protected hands (gloves)	272	4763	AOEM	
	Protected body (workwear or protective garment and sturdy footwear)	398	3510	AOEM	
	Protected head (hood and face shield)	28	387	AOEM	
	Inhalation	10	31	AOEM	
	Protective Equipment		Select for inclusion	Penetration factor	Inhalation Protection factor
	Gloves		No		
	Clothing		Work wear - arms, body and legs covered	Incl. in AOEM model	
	Head and respiratory PPE		None	1	1
	Water soluble bag		No	1	

	Exposure values	µg exposure/day applied 75th centile	95th centile	Reference	Comment
Application	Hands	3560	25429	AOEM	
	Body	1990	16260	AOEM	
	Head	96	284	AOEM	
	Protected hands (gloves)	438	4828	AOEM	
	Protected body (workwear or protective garment and sturdy footwear)	55	134	AOEM	
	Inhalation	4	18	AOEM	
	Protective Equipment		Select for inclusion	Penetration factor	Inhalation Protection factor
	Gloves		No		
	Clothing		Workwear - arms, body and legs covered	Incl. in AOEM model	
	Head and respiratory PPE		None	1	1
	Closed cab		No	vehicle mounted upward spraying only	

1. Total

	Without RPE/PPE	With RPE/PPE	
Longer term			
Total systemic exposure from mixing, loading and application (mg a.s./day)	4.8289064	3.0866958	
Total systemic exposure from mixing, loading and application per kg body weight (mg/kg bw/day)	0.0804818	0.0514449	
% of RVNAS	402.41%	257.22%	
Acute			
Total systemic exposure from mixing, loading and application (mg a.s./day)	21.7274935	12.3303040	
Total systemic exposure from mixing, loading and application per kg body weight (mg/kg bw/day)	0.3621249	0.2055051	
% of RVAAS	21.30%	12.09%	

Operator exposure for Ethephon SL 480 outdoor spray applications

Application rate of active substance	0.48 kg a.s./ha	*i_AppRate*
Assumed area treated	50 ha/day	*i_AreaTreated*
Amount of active substance applied	24 kg a.s./day	*i_AmoutAS*
Dermal absorption of the product	5.00%	*i_AbsorpProduct*
Dermal absorption of in-use dilution	5.00%	*i_AbsorpIuse*
Formulation type	Soluble concentrates, emulsifiable concentrate, etc.	
Indoor or Outdoor application	Outdoor	
Application method	Downward spraying	
Application equipment	Vehicle-mounted	
Season	not relevant	

Outdoor|Soluble concentrates, emulsifiable concentrate, etc.|Downward sprayingVehicle-mounted

	Exposure values	µg exposure/day mixed and loaded 75th centile	95th centile	Reference	Comment
Mixing and loading	Hands	56090	211369	AOEM	
	Body	33905	181327	AOEM	
	Head	1245	6800	AOEM	
	Protected hands (gloves)	272	1054	AOEM	
	Protected body (workwear or protective garment and sturdy footwear)	398	3510	AOEM	
	Protected head (hood and face shield)	20	327	AOEM	
	Inhalation	10	31	AOEM	
	Protective Equipment	Select for inclusion		Penetration factor	Inhalation Protection factor
	Gloves	Yes		Incl. in AOEM model	
	Clothing	Work wear - arms, body and legs covered		Incl. in AOEM model	
	Head and respiratory PPE	None		1	1
	Water soluble bag	No		1	

	Exposure values	µg exposure/day applied 75th centile	95th centile	Reference	Comment
Application	Hands	3560	23499	AOEM	
	Body	1990	10260	AOEM	
	Head	94	284	AOEM	
	Protected hands (gloves)	238	4828	AOEM	
	Protected body (workwear or protective garment and sturdy footwear)	55	134	AOEM	
	Inhalation	5	18	AOEM	
	Protective Equipment	Select for inclusion		Penetration factor	Inhalation Protection factor
	Gloves	No			
	Clothing	Work wear - arms, body and legs covered		Incl. in AOEM model	
	Head and respiratory PPE	None		1	1
	Closed cab	No		vehicle mounted upward spraying only	

Document MCP: Section 7 Toxicological studies
Ethephon SL 480 g/L

1. Total

	Without RPE/PPE	With RPE/PPE	
Longer term			
Total systemic exposure from mixing, loading and application (mg a.s./day)	4.8289064	0.2958018	
Total systemic exposure from mixing, loading and application per kg body weight (mg/kg bw/day)	0.0804818	0.0049300	
% of RVNAS	402.41%	24.65%	
Acute			
Total systemic exposure from mixing, loading and application (mg a.s./day)	21.7274935	1.9995354	
Total systemic exposure from mixing, loading and application per kg body weight (mg/kg bw/day)	0.3621249	0.0333256	
% of RVAAS	21.30%	1.96%	

Worker exposure from residues on foliage for Ethephon SL 480

Crop type	Cereals		
Indoor or outdoor	Outdoor		
Application method	Downward spraying		
Application equipment	Vehicle-mounted		
Worker's task	Inspection, irrigation		
Main body parts in contact with foliage	Hand and body		
Application rate of active substance	0.48	kg a.s./ha	*i_AppRate*
Number of applications	1		*i_AppNo*
Interval between multiple applications	365	days	*i_AppInt*
Half-life of active substance	30	days	*d_HalflifeAS*
Multiple application factor	1.0		*d_MAF*
Dermal absorption of the product	5.00%		*i_AbsorpProduct*
Dermal absorption of the in-use dilution	5.00%		*i_AbsorpInuse*
Dislodgeable foliar residue (i_AppRate*i_DFR)	1.44	µg a.s./cm²	*d_DFR*
Working hours	2	hr	*d_WorkHr*
Dermal transfer coefficient - Total potential exposure	2500	cm²/hr	*d_DermTcUCV*
Dermal transfer coefficient - arms, body and legs covered	1400	cm²/hr	*d_DermTcCV1*
Dermal transfer coefficient - hands, arms, body and legs covered	no TC available for this assessment	cm²/hr	*d_DermTcCV2*
Inhalation transfer coefficient for automated applications	NA	ha/hr*10^(-3)	*d_InhalTcAut*
Inhalation transfer coefficient for cutting ornamentals	NA	ha/hr*10^(-3)	*d_InhalTcCut*
Inhalation transfer coefficient for sorting /bundling ornamentals	NA	ha/hr*10^(-3)	*d_InhalTcSort*

1. Total

	Potential exposure	Work wear - arms, body and legs covered	Working wear and gloves	Comments
Total systemic exposure (mg a.s./day)	1.8000000	0.2016000	no TC available for this assessment	
Total systemic exposure per kg body weight (mg/kg bw/day)	0.0300000	0.0033600		
% of RVNAS	150.00%	16.80%		

2. Details

	Systemic exposure [mg a.s./day]	Systemic exposure [mg a.s./kg bw/day]	Formula	Comments
Dermal - Potential	1.8000000	0.0300000	*d_DermTcUCV*d_WorkHr*i_DFR*i_MAF/1000*i_AbsorpInuse*	
Dermal - Work wear - arms, body and legs covered	0.2016000	0.0033600	*d_DermTcCV1*d_WorkHr*d_DFR*d_MAF/1000*i_AbsorpInuse*	
Dermal - Working wear and gloves	no TC available for this assessment		*d_DermTcCV2*d_WorkHr*d_DFR*d_MAF/1000*i_AbsorpInuse*	
Inhalation				Na for outdoor activities

Resident exposure for Ethephon SL 480

Croptype	Cereals		
Application method	Downward spraying		
Application equipment	Vehicle-mounted		i_AppEquip
Formulation type	Soluble concentrates, emulsifiable concentrate, etc.		i_FormVal
Buffer strip	2-3	m	i_Buffer
Application rate of the product	0.48	kg a.s./ha	i_AppRate
Concentration of active substance (in-use dilution for liquid applications)	2.4	g a.s./l	d_ConcAS
Dermal absorption of product	5.00%		i_AbsorpProduct
Dermal absorption of in-use dilution	5.00%		i_AbsorpInuse
Oral absorption	100.00%		i_AbsorpOralInuse
Dislodgeable foliar residue (i_AppRate*i_DFR)	1.44	µg a.s./cm^2	d_DFR
Vapour pressure of in-use dilution	low volatile substances having a vapour pressure of <5*10-3Pa	Pa	i_VapPr
Concentration in air	0.001	mg/m^3	d_AirCon
Resident dermal spray drift exposure 75th percentile - adult	0.47	ml spray dilution/person	
Resident dermal spray drift exposure 75th percentile - child	0.327	ml spray dilution/person	
Resident inhal. spray drift exposure 75th percentile - adult	0.00010	ml spray dilution/person	
Resident inhal. spray drift exposure 75th percentile - child	0.00022	ml spray dilution/person	
Resident dermal spray drift exposure mean - adult	0.22318	ml spray dilution/person	
Resident dermal spray drift exposure mean - child	0.16	ml spray dilution/person	
Resident inhal. spray drift exposure mean - adult	0.00005	ml spray dilution/person	
Resident inhal. spray drift exposure mean - child	0.00017	ml spray dilution/person	
Exposure duration dermal	2	hours	d_ReExpDur
Exposure duration inhalation	24	hours	d_ReExpDurInhal
Exposure duration entry into treated crops	0.25	hours	d_ExpDurEntrCrop
Light clothing adjustment factor	18.0%		d_ClothAF
Breathing rate adult	0.23	m^3/day/kg	d_BreathRAd
Breathing rate child (1-3 year old)	1.02	m^3/day/kg	d_BreathRCh
Drift percentage on surface (75th percentile)	5.60%		
Drift percentage on surface (mean)	4.10%		
Turf transferable residues percentage	5.00%		d_Turf
Transfer coeff. of surface deposits-adult	7300	cm^2/hour	d_ReTCAd
Transfer coeff. of surface deposits-child (1-3 year old)	2600	cm^2/hour	d_ReTCCh
Saliva extraction percentage	50.0%		d_SalExt
Surface area of hands mouthed	20	cm^2	d_AreaHM
Frequency of hand to mouth activity	9.5	events/hour	d_ReFreqHM
Ingestion rate for mouthing of grass per day	25	cm^2	d_MouthGrass
Dislodgeable residues percentage transferability for object to mouth	20.00%		d_DRP
Transfer coefficient for entry into treated crops (75th percentile) - ad	7500	cm^2/h	d_TcEntryAd
Transfer coefficient for entry into treated crops (75th percentile) - ch	2250	cm^2/h	d_TcEntryCh
Transfer coefficient for entry into treated crops (mean) - adult	5980	cm^2/h	d_TcEntryAd
Transfer coefficient for entry into treated crops (mean) - child	1794	cm^2/h	d_TcEntryCh

1. Total

1.1 1-3 year old child

	Spray drift (75th percentile)	Vapour (75th percentile)	Surface deposits (75th percentile)	Entry into treated crops (75th percentile)	All pathways (mean)
Total systemic exposure (mg a.s./day)	0.0327708	0.0107000	0.0073920	0.0405000	0.0665240
Total systemic exposure per kg body weight	0.0032705	0.0010708	0.0007392	0.0040500	0.0066524
% of RVNAS	16.35%	5.35%	3.70%	20.25%	33.26%

1.2 Adult

	Spray drift	Vapour	Surface deposits	Entry into treated crops	All pathways (mean)
Total systemic exposure (mg a.s./day)	0.0464880	0.0138000	0.0098112	0.1350000	0.1508001
Total systemic exposure per kg body weight	0.0007748	0.0002300	0.0001635	0.0022500	0.0025133
% of RVNAS	3.87%	1.15%	0.82%	11.25%	12.57%

Bystander exposure for Ethephon SL 480				
Croptype	Cereals			
Application method	Downward spraying			
Application equipment	Vehicle-mounted			i_AppEquip
Formulation type	luble concentrates, emulsifiable concentrate, etc.			
Application rate of the product	0.48	kg a.s./ha		i_AppRate
Buffer strip	2-3	m		i_Buffer
Concentration of active substance (in-use dilution for liquid applications)	2.4	g a.s./l		d_ConcAS
Dermal absorption of product	5.00%			d_AbsorpProduct
Dermal absorption of in-use dilution	5.00%			i_AbsorpInuse
Oral absorption	100.00%			i_AbsorpOralInuse
Dislodgeable foliar residue (i_AppRate*i_DFR)	1.44	µg a.s./cm^2		d_DFR
Vapour pressure of in-use dilution	low volatile substances having a vapour pressure of <5*10-3Pa	Pa		i_Volat
Concentration in air	0.001	mg/m^3		d_AirCon
Bystander dermal spray drift exposure - adult	1.21	ml spray dilution/person		
Bystander dermal spray drift exposure - child	0.74	ml spray dilution/person		
Bystander inhal. spray drift exposure - adult	0.00050	ml spray dilution/person		
Bystander inhal. spray drift exposure - child	0.00112	ml spray dilution/person		
Exposure duration	2	hours		d_ByExpDur
Exposure duration entry into treated crops	0.25	hours		d_ExpDurTreatCrop
Light clothing adjustment factor	18.0%			d_ClothAF
Breathing rate adult	0.23	m^3/kg bw/day		d_BreathRAd
Breathing rate child (1-3 year old)	1.07	m^3/kg bw/day		d_BreathRCh
Drift percentage on surface (90th percentile)	3.5%			
Turf transferable residues percentage	7.00%			d_Turf
Transfer coeff. of surface deposits-adult	14500	cm^2/hour		d_ByTCAd
Transfer coeff. of surface deposits-child (1-3 year old)	5200	cm^2/hour		d_ByTCCh
Saliva extraction percentage	50.00%			d_SalExt
Surface area of hands mouthed	20	cm^2		d_AreaHM
Frequency of hand to mouth activity	20	events/hour		d_ByFreqHM
Ingestion rate for mouthing of grass per day	25	cm^2		d_MouthGrass
Dislodgeable residues percentage transferability for object to mouth	20.00%			d_DRP
Transfer coefficient for entry into treated crops - a	7500	cm^2/h		d_TcEntryAd
Transfer coefficient for entry into treated crops	2250	cm^2/h		d_TcEntryCh

1. Total

1.1 1-3 year old child

	Spray drift	Vapour	Surface deposits	Entry into treated crops
Total systemic exposure (mg a.s./day)	0.0752040	0.0107000	0.0208080	0.0405000
Total systemic exposure per kg body weight (mg/kg bw/day)	0.0075504	0.0010700	0.0020808	0.0040500
% of RVAAS	0.44%	0.06%	0.12%	0.24%

1.2 Adult

	Spray drift	Vapour	Surface deposits	Entry into treated crops
Total systemic exposure (mg a.s./day)	0.1202640	0.0138000	0.0295800	0.1350000
Total systemic exposure per kg body weight (mg/kg bw/day)	0.0020044	0.0002300	0.0004930	0.0022500
% of RVAAS	0.12%	0.01%	0.03%	0.13%