ENTOMELA 50SL/ENT50

DOCUMENT M-CP, Section 7

TOXICOLOGICAL STUDIES ON THE PLANT PROTECTION PRODUCT

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number

[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

Table of Contents

CP 7 TOXICOLOGICAL STUDIES ON THE PLANT PROTECTION PRODUCTS

Introduction

PHYTOPHYL manufactures “Hydrolysed Protein” which is made of Beet molasses and Urea. Both of them are used very widely for many years and have not ever classified as dangerous substances.

Beet molasses are a natural by-product of the sugar industry, defined as the end product of sugar manufacture or refining from which no more sugar may be economically crystallized by conventional means.

Beet molasses mainly used for two purposes, Animal feed additive and Alcohol Production.

There is no evidence in bibliography that Beet molasses are for some reason toxic, irritant or ecologically unsafe.

Urea and beet molasses are substances widely used as feed additives for decades without problems and are permitted in EC, US and many other countries.

PHYTOPHYL & FORESTRY COMISSION notified urea according to 91/414 and the substance is now approved under Reg. (EC) No 1107/2009. No toxicity studies were submitted but literature data about the toxicity of urea indicated limited toxicological potential.

During this first notification and inclusion Urea was not registered to ECHA but now has a full registration, the dossier is evaluated and there are 163 active registrants as a high volume chemical (production of 10.000 000 – 100.000.000 TONNES per year).

PHYTOPHYL & FORESTRY COMISSION submitted also confirmatory data about the risk for operators, workers and bystanders concerning the application of urea to low volume bait sprays in olive trees in mixture with insecticides to control Olive Fruit Fly Bactrocera oleae.

In this report performed a preliminary and indicative exposure assessment for urea based on all available data for urea in literature and ECHA registration database. All application scenarios and application rates as detailed in GAP have concluded in acceptable exposure when appropriate Personal Protective Equipment is assigned as required.

An open literature review on MC-A Section 9 which will give more data about scientific knowledge during the last decade for hydrolysed proteins concerning, toxicity studies, relevant data and the potential risk for man and the environment.

More detailed literature information on urea had given in the first assessment report of the substance and will be found also in the Urea renewal dossier which will be submitted at the same time with this dossier by PHYTOPHYL and FORESTRY COMISSION.

In next pages of this section presenting data about urea and particularly all the end point summaries found on Urea registration dossier from ECHA site and Safety Assessment of Urea by CIR expert panel to support the limited toxicological potential of Beet molasses - Urea hydrolysates and particularly of ENTOMELA 50SL which has a 17% w / w urea content.

About the risk for operators, workers and bystanders concerning the application of urea to low volume bait sprays in olive trees in mixture with insecticides to control Olive Fruit Fly *Bactrocera oleae* we present data from the report of Barkwitth Associates presented on January 2016 as confirmatory data about urea.

CP 7.1 Acute Toxicity

Summary of acute toxicity

ECHA endpoint summary for urea on acute toxicity:

"**Acute toxicity: via oral route**

Endpoint conclusion

Dose descriptor: **LD50 14 300 mg/kg bw**

Acute oral toxicity

Urea is of very low acute oral toxicity in the rat and mouse. Sato et al (1977) report LD50 values of 14.3 (12.9 -15.9) and 15.0 (13.4 -16.8) g/kg bw in male and female rats; LD50 values of 11.5 (10.6 -12.5) and 13.0 (11.0 -15.4) g/kg bw in the mouse. Urea is of generally low acute oral toxicity in most species but higher toxicity is noted in ruminants due to the generation of ammonia by gastric flora. Stiles et al (1970) report an LDlo of approximately 600 mg/kg bw in cattle.

Acute dermal toxicity

No data are available: a waiver is proposed for this endpoint. Urea is demonstrated to be of very low acute toxicity by the oral, subcutaneous and intravenous routes in the rat and mouse. Testing for acute dermal toxicity is not justified on scientific grounds and for reasons of animal welfare. Specifically, the very low toxicity of urea by the subcutaneous and intravenous routes indicates that dermal toxicity would also be very low, even assuming rapid and total dermal penetration.

Acute inhalation toxicity

No data are available: a waiver is proposed for this endpoint. The substance is a non-volatile solid and is produced as crystals with a particle size of >100 µm. There is therefore no potential for inhalation exposure. In addition, the substance has been demonstrated to be of very low toxicity by other routes of exposure. Testing for acute inhalation toxicity is therefore not justified on scientific grounds or based on exposure considerations.

Acute toxicity by other routes of exposure

Urea is also of very low acute toxicity by the subcutaneous route. Sato et al (1977) report LD50 values of 9.4 (8.2 -10.8) and 8.2 (7.1 -9.5) g/kg bw in male and female rats and LD50 values of 9.2 (8.6 -9.8) and 10.7 (9.5 -12.1) g/kg bw in male and female mice. It is notable that the

subcutaneous LD50 values are greatly in excess of the limit dose for acute dermal toxicity testing.

Urea is of very low toxicity following intravenous administration. Sato et al (1977) report LD50 values of 5.4 (4.9 -5.9) and 5.3 (4.8 -5.7) g/kg bw in male and female rats and LD50 values of 4.6 (4.3 -4.9) and 5.2 (4.8 -5.6) g/kg bw in male and female mice, respectively.

Justification for classification or non-classification

Urea is of inherently very low toxicity by all routes investigated. No data are available for the inhalation route, however low toxicity can also be predicted for this route. No classification is proposed for acute toxicity according to CLP."

CP 7.1.1 Oral toxicity

Refer to Summary of acute toxicity.

CP 7.1.2 Dermal toxicity

Refer to Summary of acute toxicity.

CP 7.1.3 Inhalation toxicity

Refer to Summary of acute toxicity.

CP 7.1.4 Skin irritation

ECHA endpoint summary for urea on irritation/corrosion for skin irritation:

"*Endpoint conclusion:*

no adverse effect observed (not irritating)

Skin irritation

Frosch & Kligman (1977) (cited in WHO/JECFA evaluation) exposed human volunteers to three daily applications of urea (dissolved in water) at concentrations of between 7.5 -30%; applications were made to intact and scarified skin. On abraded skin, slight irritation was seen with 7.5% urea; marked irritation was seen with 30% urea. No effects were seen on intact skin. In a study by Lashmar *et al* (1989) application of 10% urea for 24 hours induced no discernible change in the histological appearance of the skin. No evidence of skin irritation was seen in a modern guideline study (Hooiveld, 2003).

Justification for classification or non-classification

No classification is proposed. There is no evidence from animal studies or from human experience that urea is a skin irritant."

CP 7.1.5 Eye irritation

ECHA endpoint summary for urea on irritation/corrosion for eye irritation:

"*Key value for chemical safety assessment*

Endpoint conclusion: no adverse effect observed (not irritating)

Eye irritation

Urea was found to be a mild eye irritant in a guideline-compliant study (Kirsch & Kersebohm, 1988), which would not require a classification according to DSD, however require a classification according to CLP.

Medical surveillance data of 10 urea producing facilities were collected, which showed no cases of eye irritation or related adverse eye effects resulting from exposure to urea. (Borealis Agrolinz Melamine, 2013).

Justification for classification or non-classification

Eye irritation:

No classification is proposed. Although formally a classification as eye irritant would be required according to CLP, medical surveillance data show no adverse effects on eyes following direct contact."

CP 7.1.6 Skin sensitization

ECHA endpoint summary for urea on irritation/corrosion for skin sensitation:

Endpoint conclusion: no adverse effect observed (not sensitising)

No data are available: a waiver is proposed for this endpoint. Urea is naturally present at relatively high concentrations in human skin (up to 1% by weight) and is widely used in skin creams for the treatment of dry and irritant skin conditions without any reports of sensitisation reactions (Loden et al, 2002). A survey of 1905 patients does not reveal any reports of sensitisation (Stuttgen, 1992). A human volunteer study (Alchangian et al, 1986) does not report any sensitisation reactions. It is therefore considered to be very unlikely to be a skin sensitiser.

Justification for classification or non-classification

No classification is proposed. There is no indication that urea is a skin sensitiser or a respiratory sensitiser.

CP 7.1.7 Supplementary studies on the plant protection product

Not available

CP 7.1.8 Supplementary studies for combinations of plant protection product

Not available

CP 7.2 Data on Exposure

From BARKWITH ASSOCIATES report:

"Summary

Using available toxicity literature sourced from the Urea active substance peer review and the REACH registration database toxicity values were identified, the most appropriate exposure guidance available from the European Food Safety Authority and Biocide Technical guidance notes were selected and the following acceptable predicted exposure values for urea concluded;

Scenario	Model	PPE required	Predicted % of AOEL
Application to Olive Trees			
Operator Exposure- Tractor mounted – upwards spray	AOEM	No	2.35
Operator Exposure– Manual handheld – upwards spray	AOEM	No	67.62
Operator Exposure– Manual Knapsack – Upwards spray	AOEM	No	67.22
Bystander Exposure – Spray Drift	Europoem II	n/a	0
Bystander Exposure – Spray Drift	UK – (Lloyd et al)	n/a	4.4
Resident Exposure (Toddler) - Fallout	USA EPA	n/a	0.2
Worker Exposure – Re-entry	Europoem II	No	5.0
Application to Cut Tree Stump			
Operator Exposure– Manual Knapsack – Downwards spray	AOEM	Yes*	49.44
Operator Exposure – Brush – Stump Drench	Biocide TNsG	No	94.67
Operator exposure - Harvesting Machine	Exposure risk considered negligible, mixing loading element considered covered by other application scenarios.		
Bystander Exposure – Spray Drift	Europoem II	n/a	8
Bystander Exposure – Spray Drift	UK – (Lloyd et al)	n/a	4.6
Resident Exposure (Toddler) - Fallout	USA EPA	n/a	1.8
Worker Exposure – Re-entry	Europoem II	Yes~	36

* Protective gloves are required during mixing/loading and application
~ Gloves are required for re-entry.

As noted above all application scenarios and application rates as detailed in Appendix 1 have concluded in acceptable exposure when appropriate Personal Protective Equipment is assigned as required. " (page 10).

CP 7.2.1 Operator exposure

Risk assessment for operator

CP 7.2.1.1 Estimation of operator exposure

From BARKWITH ASSOCIATES report below is the table 2 (page 11) with the estimated operator exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to GAP and AOEM model:

Estimated operator exposure to Urea applied as a low volume bait spray (10-30L water volume)

Model data	Level of PPE	Total absorbed dose (mg/kg/day) (RVNAS)	% of AOEL (RVNAS)	Calculation Sheets
FIELD APPLICATIONS - Tractor Mounted Upward Spraying *Application rate: 0.159 kg Urea/ha*				
AOEM • 10 ha/day • 60 kg operator	no PPE	0.53	**2.35**	Appendix 2, Figure 3.
	mixing/loading and application: working clothing (long sleeved shirt and trousers) + Gloves	0.04	**0.17**	
FIELD APPLICATIONS – Manual Handheld Upward Spraying *Application rate: 0.159 kg Urea/ha*				
AOEM • 50 ha/day • 60 kg operator	no PPE	15.21	**67.62**	Appendix 2, Figure 4.
	mixing/loading and application: working clothing (long sleeved shirt and trousers) + Gloves	0.77	**3.41**	
FIELD APPLICATIONS – Manual Knapsack Upward Spraying *Application rate: 0.159 kg Urea/ha*				
AOEM • 50 ha/day • 60 kg operator	no PPE	15.35	**67.22**	Appendix 2, Figure 5.
	mixing/loading and application: working clothing (long sleeved shirt and trousers) + Gloves	0.77	**3.41**	

In conclusion (page 13) refer that the estimated exposures calculated by the AOEM are acceptable when the operator wears standard PPE (gloves during mixing loading and application).

CP 7.2.1.2 Measurement of operator exposure

CP 7.2.2 Bystander and resident exposure

Risk assessment for bystander and resident

CP 7.2.2.1 Estimation of bystander and resident exposure

From BARKWITH ASSOCIATES report below is the table 8 (page 14) with the estimated bystander exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to GAP and Euorpoem II Model:

Table 8 - Estimated bystander exposure to Urea and % of the AOEL (Europoem II Model)

Crop	Dermal absorption	Dermal exposure (mg a.s day)	Inhalation exposure (mg a.s day)	Total systemic exposure (mg a.s day)	% of AOEL (1575 mg a.s day)
Olive Trees	100%	4.77	1.1925	5.963	0

Below is the table 9 (page 15) with the estimated bystander exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to GAP and UK Model:

Table 9 - Estimated bystander exposure to Urea and % of the AOEL (UK Model)

Crop	Dermal absorption	Dermal exposure (mg/kg day)	Inhalation exposure (mg/kg day)	Total systemic exposure (mg/kg bw/day)	% of AOEL (22.5 mg/kg bw/day)
Olive Trees	100%	58.83	0.0318	0.98	4.4

In conclusion (page 15) refer “All bystander exposures calculated using EUROPOEM II and UK derived information with No PPE result in acceptable values and require no further evaluation.”

For the estimation of potential resident exposure below is a part of the table 12 (page 18) and we can see that the systemic bystander exposure through spray drift is equivalent to 0.2% of the AOEL when used ENTOMELA 75SL insect attractant which has a 25% urea content.

Orchard application to Olive trees

Application	High level tree fruit
Parameter/Active	Urea
AR (kg a.s./ha)	0.954
Dabs (%)	100%
AOEL (mg/kg bw/d)	22.5
AR µg/cm²	9.54
DF %	11.01%
TTR % (d, HtM)	5%
TTR % (OtM)	20%
TC cm²/hour	5200
SE%	50%
SA cm²/event	20
Frequency events/hour	20
Hours	2
BW (kg)	15
IgR cm²/day	25
SE(d) ug/kg bw/d	36.41
SE(h) ug/kg bw/d	1.40
SE(o) ug/kg bw/d	0.35
Tot mg/kg bw/day	0.04
% AOEL	0.2

CP 7.2.2.2 Measurement of bystander and resident exposure

CP 7.2.3 Worker exposure

Risk assessment for worker

CP 7.2.3.1 Estimation of worker exposure

From BARKWITH ASSOCIATES report below is the tables 13 & 14 (page 19) with the estimated worker exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to Euorpoem II Model with and without PPE :

Table 13 - Europoem II worker exposure estimate without PPE

Application type	Dermal exposure (mg a.s /day)	Systemic Exposure (mg/kg bw/day)*	AOEL	% of AOEL
Low level/tree stump	2797.200	39.96	22.5	**178**
High level/olive tree	77.274	1.10	22.5	**5.0**

*bodyweight taken as 70kg (EUROPOEM default)

Table 14 - Europoem II worker exposure estimate with PPE

Application type	Dermal exposure with PPE (gloves) (mg a.s /day)	Systemic Exposure (mg/kg bw/day)*	AOEL	% of AOEL
Low level/tree stump	559.440	7.992	22.5	**36**
High level/olive tree	15.455	0.22	22.5	**1**

*bodyweight taken as 70kg (EUROPOEM default)

In conclusion refer that the predicted worker exposures are below the AOEL value without PPE when considering application to olive trees.

CP 7.2.3.2 Measurement of worker exposure

CP 7.3 Dermal Absorption

From ECHA site for urea:

Dermal absorption values of 7.2-9.5% is reported for urea.

Urea is present at appreciable levels in the human epidermis, where it may play a role as a humectant, maintaining hydration of the stratum corneum. At very high levels of exposure, urea may act as a denaturant and may enhance the dermal absorption of other compounds. Bronaugh et al (1982), report a dermal absorption value of 7.2%, based on the results of a study in the rat in vivo and comparable results in vitro.

The Cosmetic Ingredient Review (CIR) Expert Panel (2005) reviewed the available data on the dermal absorption of urea in vitro and vivo in various test systems and conclude that a value of 9.5% derived from a study in intact human skin in vitro is appropriate. The review also notes that the absorption of urea through abraded skin is considerably more extensive and that urea is known to enhance the dermal penetration of other substances.

CP 7.4 Available Toxicological Data Relating to Co-Formulants

CONFIDENTIAL information - data provided separately (Document J)