Hydrolysed proteins

DOCUMENT N1

OVERALL CONCLUSIONS

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

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1 IDENTITY

1.1 Summary of identity

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Name: Hydrolysed proteins (Beet Mollasses – Urea Hdrolysate)

Chemical Name:
Not applicable

CAS, EC and CIPAC Numbers
Not applicable

Molecular and Structural Formula, Molar Mass
Not applicable

Specification of Purity of the Active Substance in g/kg
Minimum Crude Protein Equivalent: 360gr/kg

2 PHYSICAL AND CHEMICAL PROPERTIES

2.1 Summary of physical and chemical properties of the active substance

In case of Hydrolysed proteins the a.s. and the ppp are identical, the physical and chemical properties of a.s. are similar to the properties of formulated product. Hydrolysed protein is a dark brown liquid with characteristic odour soluble in water not flammable and cannot be explosive or an oxidising agent. It has a surface tension of 70.6 (mN/m), at 20^{o} C. For more detailed properties of the ppp see also next paragraph.

2.2 Summary of physical and chemical properties of the plant protection product

ENTOMELA 50SL is a dark brown liquid with characteristic odour, is not anticipated to have explosive or oxidizing properties and cannot be flammable. The pH of the formulation in its initial, unopened packaging is increased over time with no other effect on specifications and no significant effect on application, since when diluted in application rates gives lower pH. The maximum pH value for the "fresh" formulation (within one month) is 7,10. The range in this case is 6.20-7.10. The pH is max 7.3 after 1 year from its production date, and pH is max 8.0 after 2 years from its production date.

ENTOMELA 50SL has a viscosity of 1148 cSt (=mm2/sec), surface tension of 70.6 (mN/m) at 20^o C and density of 1.35 gr/ml at 20^o C. The product is not heat sensitive, only pH may rise if we use temperatures over 50^o C. Stability tests performed in original HDPE and in glass bottle with a metallic specimen inside. In both cases all product values where inside limits and no deterioration to packing material observed. The stability data indicate a shelf life of at least 2 years when stored in its original unopened container at normal ambient temperature of 1- 40^o C. No persistence of foaming observed and no material separated after diluted in water after 24 hours standing. In physical and chemical compatibility tests the product found compatible with four different insecticides used in bait sprays.

3 DATA ON APPLICATION AND EFFICACY

3.1 Summary of effectiveness

Beet molasses –Urea Hydrolysates are in agricultural use for more than 40 year and are used as attractant in suppression control operations by spot bait sprays. Beet mollases urea hydrolysate affects the instinctive behavioural activity of olive-fly (Bactrocera (dacus) oleae) in seeking food. This is achieved by the volatile degradation products of the diluted preparation, which attract the insect to a toxicant.

Hydrolysed proteins attractants in Greece have a definitive place in annual collective programs of bait sprays organized by the Greek Ministry to protect the ripening olive fruit against gravid fertilised females of Bactrocera oleae and prevent extensive damage to crops all over the country.

In bait sprays by terrestrial spray the recommended quantity of bait is 300ml per tree foliage and the recommended attractant dosage for the preparation of bait is 2% w/w in Low Volume (LV) applications which are the mainly used.

Maximum number of applications depends on the insecticide used and in practice it is 6 applications per crop season. Usually according to insect population they are necessary one or two applications during summer and two or three applications during autumn. The timing of the first application aim at destroying the adults of previous year and as preventing the oviposition on the earliest fruits, and it is of particular importance. The principal criterion for the timing of applications is the capture of adult-flies and specially of the females. Capture of five and more adults per trap and per week indicates the need for bait spraying. The last application before harvest depends on the insecticide used.

3.2 Summary of information on the development of resistance

Not applicable

3.3 Summary of adverse effects on treated crops

Not applicable

3.4 Summary of observations on other undesirable or unintended side-effects

Not applicable

4 FURTHER INFORMATION

4.1 Summary of methods and precautions concerning handling, storage, transport or fire

Handling: Good industrial practice in housekeeping and personal hygiene should be followed. When using do not eat, drink or smoke. Wash hands thoroughly after handling or contact. Thoroughly clean equipment after use. The product should not be dumped, spilled, rinsed or washed into sewers or public waterways.

Warehouse storage: Store in a dry place.

User level storage: Keep out of the reach of children. Keep away from food, drink and animal feeding stuffs. Keep only in original container. Do not store above 40ºC for prolonged time. It is stable for more than 2 years.

Transport: Not classified as hazardous under transport regulations.

Fire: Is not flammable. Urea decomposition at high temperatures (>133oC) emits toxic fumes of nitrogen oxide, ammonia and isocyanic acid.

4.2 Summary of procedures for destruction or decontamination

In case of controlled incineration for the package product wastes. Close and label waste receptacles. Dispose of them at a suitable waste incineration plant in accordance with the official regulations. Where large quantities are concerned, consult the supplier.In case of no controlled incineration wash or dilute with large quantities of water, while avoiding streaming into water-courses.

4.3 Summary of emergency measures in case of an accident

In case of a spillage first we have to stop it and after wash with water. In case of decontamination wash with water Damaged packaging emptying into suitable containers and then rinsed thoroughly with water and deposited at a collection point for recycling or energy recovery. Avoid contact with eyes. Wash with water for cleaning up. It is incombustible but Fire/explosion fumes should not be inhaled and in case of fire wear a self-contained breathing apparatus. If swallowed, seek medical advice immediately and show this container or label.

5 METHODS OF ANALYSIS

5.1 Methods used for the generation of pre-authorisation data

Analytical methods for determination of hydrolysed proteins content as crude protein equivalent, for urea content and for the determination of the Product Specifications (physicochemical properties) were not evaluated as part of the first EU review of urea and hydrolysed proteins, but evaluated as part of the registration report of ENT50 . Therefore all relevant data are provided now and are considered adequate.
Below is the table with the methods used for a.s. content and all parameters of the specification and their limits.

Test parameter	**Method of analysis**	**Minimum**	**Maximum**
Total nitrogen (x 6.25 = Crude protein equivalent)	AOAC 2001.11	80 gr/kg (500gr/kg)	92.4gr/kg (577.5 gr/kg)
Ureic nitrogen (x 60/28=Urea)	Modified AOAC 959.03	74.6gr/kg (159.8gr/kg)	84.1gr/kg (180.2gr/kg)
Ammoniacal Nitrogen as NH_4Cl	Modified EN 15475:2009 - Similar method to 2.6.2 section 7.5 EC Reg. 2003/2003		5.30 % w/w
Chlorine salts expressed as NaCl	In house ISO 457/1983		2.00 % w/w
Amino-acids index	Modified AOAC 965.31		2.00 meq/10gr
Dry matter	In house ISO 2920 at 105^{o}C	74%w/w	82%w/w
Insoluble in water	Modified CIPAC MT.10.2		0.7 % w/w
PH	CIPAC 75.3	6.20	7.30 after 1 year 8.00 after 2 years
Density	CIPAC 3.3.2	1.31 g/ml	1.39 g/ml
Appearance	Macroscopic examination	Surupy liquid	
Color	Macroscopic examination	Deep reddish-brown	
Odor	Sensory evaluation	Characteristic	

5.1.1 Analysis of the active substance as manufactured

In case of Hydrolysed proteins the a.s. and the ppp are identical, the physical and chemical properties of a.s. are similar to the properties of formulated product. For more detailed information refer on MC-A Section 4 and document J.

5.1.2 Formulation analysis

In case of Hydrolysed proteins the a.s. and the ppp are identical, the physical and chemical properties of a.s. are similar to the properties of formulated product. For more detailed

information refer on formulation process on Document J - PHY and for detailed physical and chemical properties refer to MC-P Section 2 – ENT50.

5.1.3 Methods for Risk Assessment

Not applicable

Plants and plant products

Food of animal origin

Soil

Water

Air

5.2 Methods for post-authorisation control and monitoring purposes

Not applicable

Plants and plant products

Food of animal origin

Soil

Water

Air

6 IMPACT ON HUMAN AND ANIMAL HEALTH

6.1 Effects Having Relevance to Human and Animal Health

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 is 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. There are not any new studies submitted, only 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 MC-A section 6 also presented the end point summaries found on Urea registration dossier from ECHA site to support the limited toxicological potential of Beet molasses - Urea hydrolysates.

6.1.1 Summary of adsorption, distribution, metabolism and excretion

Only data for urea from ECHA presented here.

Urea is widely distributed within the natural world, as a by-product in protein synthesis in ureotelic animals, including mammals whence it is excreted in urine (humans 25g - 30g/day).

All humans are exposed to this material from birth, and its toxicity has been studied over many years. A lot of literature studies exist about the metabolism of urea in mammals.

Urea is produced in large quantities by the human body as a product of normal metabolism and is excreted unchanged in the urine. Further studies characterising the toxicokinetics of urea are not required.

6.1.2 Summary of acute toxicity

No extra data for the acute toxicity of hydrolysed protein presented. Because of very low toxicity of beet molasses only data for a.s. urea from ECHA presented.

Urea is of generally low acute oral toxicity in most species only in ruminants is noted higher toxicity due to the generation of ammonia by gastric flora. Urea is demonstrated to be of very low acute toxicity by the oral, subcutaneous and intravenous routes in the rat and mouse. 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. Urea found to be slightly irritant to skin and mild irritant to eye. 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. It is therefore considered to be very unlikely to be a skin sensitiser.

6.1.3 Summary of short-term toxicity

No extra data for the short term toxicity of hydrolysed protein presenting. Because of very low toxicity of beet molasses only data for a.s. urea from ECHA presented.

Dose descriptor: NOAEL 2 250 mg/kg bw/day

6.1.4 Summary of genotoxicity

No extra data for the genotoxicity of hydrolysed protein presented. Because of very low toxicity of beet molasses only data for a.s. urea from ECHA presented.

Positive results obtained in vitro are associated with concentrations well in excess of the recommended limit concentrations are not considered to be of biological relevance. A positive result in vivo is also associated with an excessive dose level. Considering the physiological role and presence of substantial quantities of urea in the human body, it is not considered likely that this substance is genotoxic. Further testing for genotoxicity is not proposed.

6.1.5 Summar of long-term toxicity and carcinogenicity

No extra data for the long term toxicity and carcinogenicity of hydrolysed protein presented. Because of very low toxicity of beet molasses only data for a.s. urea from ECHA presented.

No classification is proposed for carcinogenicity. There is no evidence from animal studies that urea is carcinogenic. The physiological role of urea and level of production by the human body indicates that the substance is not carcinogenic.

6.1.6 Summary of reproductive toxicity

No extra data for the reproductive toxicity of hydrolysed protein presented. Because of very low toxicity of beet molasses only data for a.s. urea from ECHA presented.

Developmental toxicity testing in rats dosed orally up to 1000 mg/kg bw did not result in adverse effects. There are no studies in animals showing clear evidence of reproductive effects. The results of the available studies do not trigger classification according to Directive 67/548/EEC.

6.1.7 Summary of neurotoxicity

6.1.8 Summary of further toxicological studies on the active substance

6.1.9 Summary of toxicological data on impurities and metabolites

6.1.10 Summary of medical data and information

6.2 Toxicological end point for assessment of risk following long-term dietary exposure - ADI

6.3 Toxicological end point for assessment of risk following acute dietary exposure - ARfD (acute reference dose)

6.4 Toxicological end point for assessment of occupational, bystander and residents risks – AOEL

6.5 Summary of product exposure and risk assessment

6.5.1 Operators

In report of Barkwith Associates estimated operator exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to GAP and AOEM model. It is referred that the estimated exposures calculated by the AOEM are acceptable when the operator wears standard PPE (gloves during mixing loading and application).

6.5.2 Bystander and resident exposure

In report of Barkwith Associates estimated bystander exposure to urea when used ENTOMELA 75SL insect attractant which has a 25% urea content according to GAP and Europoem II Model, or according to GAP and UK Model. It is referred it is referred that 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 referred 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.

6.5.3 Workers

In report of Barkwith Associates 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. It is referred that the predicted worker exposures are below the AOEL value without PPE when considering application to olive trees.

7 RESIDUES

A quantitative consumer dietary risk assessment for Beet molasses Urea hydrolysate is not necessary due to the specific methods of application, the low toxicity of beet molasses and urea and particularly for urea as the natural exposure to that substance is far higher than the one linked to the use of urea as a plant protection product.

7.1 Summary of storage stability of residues

7.2 Summary of metabolism, distribution and expression of residues in plants, poultry, lactating ruminants, pigs and fish

7.3 Definition of the residue

7.4 Summary of residue trials in plants and identification of critical GAP

7.5 Summary of feeding studies in poultry, ruminants, pigs and fish

7.6 Summary of effects of processing

7.7 Summary of residues in rotational crops

7.8 Summary of other studies

7.9 Estimation of the potential and actual exposure through diet and other sources

7.10 Proposed MRLs and compliance with existing MRLs

7.11 Proposed import tolerances and compliance with existing import tolerances

8 FATE AND BEHAVIOUR IN THE ENVIRONMENT

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

Urea is stable in aqueous solution. Hydrolysis is not seen and is not predicted based on a theoretical assessment of the structure of the molecule. Urea is considered to be readily biodegradable: the substance will be rapidly degraded by microorganisms present in the environment (as a nutrient and N-source) and subsequently incorporated into the nitrate cycle. Urea is additionally utilised as a N-source by terrestrial and aquatic plants.

There is no evidence of any adverse effects of Hydrolysed protein in the environment.

8.1 Summary of fate and behaviour in soil

8.2 Summary of fate and behaviour in water and sediment

8.3 Summary of fate and behaviour in air

8.4 Summary of monitoring data concerning fate and behaviour of the active substance, metabolites, degradation and reaction products

8.5 Definition of the residues in the environment requiring further assessment

8.6 Summary of exposure calculations and product assessment

9 EFFECTS ON NON-TARGET SPECIES

9.1 Summary of effects on birds and other terrestrial vertebrates

There is no evidence of any adverse effects of hydrolysed proteins on birds or other terrestrial vertebrates.

9.2 Summary of effects on aquatic organisms

There is no evidence of any adverse effects of hydrolysed proteins on aquatic organisms.

9.3 Summary of effects on arthropods

9.4 Summary of effects on non-target soil meso- and macrofauna

9.5 Summary of effects on soil nitrogen transformation

9.6 Summary of effects on terrestrial non-target higher plants

9.7 Summary of effects on other terrestrial organisms (flora and fauna)

9.8 Summary of effects on biological methods for sewage treatment

9.9 Summary of product exposure and risk assessment

10 CLASSIFICATION AND LABELLING

Proposed classification according to Regulation (EC) No 1272/2008 on the classification, labelling and packaging of substances and mixtures:

GHS Pictogram:

GHS07

Signal word:

WARNING

According to Regulation (EC) 1272/2008 as last amended by Regulation (EC) 286/2011:

Hazard statements (because of the use with insecticides on bait application**):**

H317	May cause an allergic skin reaction (Cat. 1)
EUH401	To avoid risks to human health and the environment, comply with the instructions for use.

Precautionary statements:

P102+405	Keep out of the reach of Children. Store locked up
P270	Do not eat, drink or smoke when using this product
P280	Wear protective gloves/protective clothing/eye protection/face protection
P301+312	IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell
P302 + 352	IF ON SKIN: Wash with plenty of soap and water
P333 + P313	If skin irritation or rash occurs: Get medical advice/attention
P305+351+ 338	IF IN EYES: Rinse cautiously with water for several minutes Remove contact lenses, if present and easy to do. Continue rinsing
P363	Wash contaminated clothing before reuse
P370+260	In case of fire: Do not breathe dust/fume/gas/mist/vapours/spray

Other phrases

Other precautionary phrases because of the use with insecticide:

- Take all the necessary measures for the insecticide.
- Do not spray against the wind.
- After use wash with water and soap.
- Do not spill in watercourse or water irrigating systems.

Labelling: Signal word:
Hazard statements:
Precautionary statements:

11 RELEVANCE OF METABOLITES IN GROUNDWATER

11.1 Summary

Not applicable.

11.2 Conclusion

12 CONSIDERATION OF ISOMERIC COMPOSITION IN THE RISK ASSESSMENT

12.1 Summary

Not applicable.

12.2 Conclusion

FURTHER INFORMATION TO BE SUBMITTED

Not applicable.

Appendix 1: Metabolites formed from Active Substance and their occurrence

Code Number	Description	• Compound found in	Structure
		•	
		•	

Appendix 2: Proposed Metabolic Pathway