Hydrolysed Proteins

DOCUMENT N1

OVERALL CONCLUSIONS

Version history[1]

Date	Data points containing amendments or additions and brief description	Document identifier and version number
28 February 2018	Initial DRAR, Document N1	
May 2018	Amended Document N1 in light of comments from the completeness check.	
February 2020	Amended Document N1 in light of RMS comments	

[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

1.1.1 Common Name Proposed or ISO-accepted and synonyms

ISO-common name:	Animal tissue Hydrolysate (under the general term Hydrolysed Protein)
Synonym:	Hydrolysed Protein

1.1.2 Chemical Name (IUPAC and CA nomenclature)

IUPAC:	Not applicable
CA:	Not applicable

1.1.3 Producer's Development Code Numbers

Proalan S.A. Code Number:	NORLAN AMPL

1.1.4 CAS, EC and CIPAC Numbers

CAS No:	Not applicable
CIPAC No:	901
EC No:	Not applicable

1.1.5 Molecular and Structural Formula, Molar Mass

Molecular formula:	Not applicable
Structural formula:	Not applicable
Molecular mass:	Not applicable

2 PHYSICAL AND CHEMICAL PROPERTIES

2.1 Summary of physical and chemical properties of the active substance

The active substance Hydrolysed Proteins is a homogeneous brown opaque liquid with a boiling point of 102.1°C and a mean surface tension of 39.9 mN/m at 19.9°C and is neither explosive, nor oxidising. The active substance is completely soluble in water and miscible in dichloromethane and methanol and immiscible in acetone, ethyl acetate, toluene and heptane.

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

The appearance of the product SVMA14-004 is that of a homogeneous brown opaque liquid. It is not explosive, has no oxidising properties. No flash point was observed up to 120°C and no self-ignition temperature was observed up 599 °C. The pH value of the neat aqueous formation is around 4.9 at 21°C. The effect of low temperature on the stability of the formulation, after 7 days at 0 °C was the presence of brown/beige deposit in suspension. For this reason, the phrase “Protection from frost” is proposed to be added on the label. Based on the available data there is no effect of high temperature on the stability of the formulation, since after 14 days at 54 °C, the technical properties were not changed in a HDPE packaging HDPE. The stability data proved a shelf life of at least 2 years at ambient temperature when stored in a PET packaging and HDPE packaging. Its technical characteristics are acceptable for a *soluble concentrate* formulation.
The intended concentration of use depends on the choice of the insecticides to be used in mixture. The maximum intended concentration of use is however estimated to be 1.5% v/v whereas the minimum intended concentration of use is estimated to be 0.15% v/v.
The product should be mixed in the tank together with insecticides authorized for the intended uses. Studies regarding the combination with authorized insecticides containing different active substances (Reldan-E - chlorpyrifos-methyl; Nuprid - Imidacloprid; Karate zeon+ - Lambda cyhalothrin; Decis 2.5 EC – Deltamethrin; Imidan 50 WP – Phosmet) were submitted and the application as tank mixture is acceptable.

3 DATA ON APPLICATION AND EFFICACY

3.1 Summary of effectiveness

Hydrolysed Proteins are intended to be used in agriculture in baits for trapping the insects and in mixture with different insecticides to increase their efficacy.

Hydrolysed Proteins have an attractant effect against:

- Mediterranean Fruit flies *(ceratitis capitata)*
- Flies of olive trees (*Bactrocera oleae*)
- Flies of the *Rhagoletis* type

The mode of action of the attractant effect is based on the decomposition and putrefaction of the organic matter process. In this process, a compound called as putrescine is released, among others, and has a characteristic odour able to attract the insects.
The control and elimination of insects is carried out by the insecticide or because the insects cannot escape from the traps.
The environmental temperature is the most important factor that affects the speed of the decomposition.

3.2 Summary of information on the development of resistance

The nature of the Hydrolysed Proteins rules out any possibility of the development of resistance. The proteins are necessary elements for the nourishment of living beings. The digestive system hydrolyses the protein for their assimilation. The process is continuous and does not generate resistance in the living beings.

3.3 Summary of adverse effects on treated crops

No adverse effects on treated crops reported or anticipated due to the nature of the active substance Hydrolysed Proteins. The active substance Hydrolysed Proteins is a natural substance biodegradable. The metabolites resulting from the decomposition process are volatiles substances (ammonia, carbon dioxide) or organic molecules highly biodegradables that do not have harmful effect.

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

No undesirable or unintended side effects observed.

4 FURTHER INFORMATION

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

Precautions for safe handling: The usual precautions for the handling of a non-dangerous product must be observed. Avoid contact with skin and eyes. Do not eat or drink during the handling of the product.

Conditions for safe storage: The packaging should remain closed, in a cool, aired and dry place and should be protected from frost. Avoid contact with sun light.

Fire-fighting measures:

- Extinguishing media: All conventional means of extinction are appropriate (water, carbon dioxide, etc.)
- Special hazards arising from the substance or mixture: None.
- Advice for fire-fighters: No specific fire-fighting protection is required.

4.2 Summary of procedures for destruction or decontamination

Hydrolysed proteins are of low toxicity and are biodegradable. Thus, no particular procedure for destruction or decontamination is needed. Contaminated material may be rinsed with clear water.

4.3 Summary of emergency measures in case of an accident

The usual precautions for a non-dangerous product must be observed. Use sand or soil to avoid leakage expansion. Collect as much released product as possible. Absorb spill with inert material (soil, sand).

First aid measures:

- Eye contact: Rinse thoroughly with water.
- Inhalation: Go to fresh air. If symptoms appear, seek medical attention.
- Ingestion: Rinse the mouth with a lot of water. Consult a doctor if you feel unwell.
- Skin contact: Wash the affected area with water and soap, rinse thoroughly.
- Most important symptoms and effects, both acute and delayed: There is no knowledge of important symptoms and effects
- Indication of any immediate medical attention and special treatment needed: Not known.

5 METHODS OF ANALYSIS

5.1 Methods used for the generation of pre-authorisation data

5.1.1 Analysis of the active substance as manufactured

The total nitrogen and ammoniacal nitrogen have been determined by the method of Kjeldahl. Then, the content of organic nitrogen is calculated by the difference between the content of total nitrogen and the content of ammonium nitrogen. From that, the content of pure hydrolysed proteins is expressed by multiplying the organic nitrogen content by the conventional factor of 6.25

In response to the RMS request, further information regarding the methods used for the determination of amino acids have been provided under the form of a summary of the method PNT-M-109 used for the determination of amino acids.
The determination of primary and secondary amino acids was performed using an automated precolumn derivatization with OPA for primary amino acids and Fmoc for secondary amino acids, followed by a High-Performance Liquid Chromatography.
A schedule of accreditation for the laboratory involved in this study is also provided to argue that this laboratory is accredited by ENAC and that the used methods are based on the Official Methods established by the Spanish Real Decreto 1110/1991 Anexo num. 18.

5.1.2 Formulation analysis

The total nitrogen and ammoniacal nitrogen have been determined by the method of Kjeldahl. Then, the content of organic nitrogen is calculated by the difference between the content of total nitrogen and the content of ammonium nitrogen. From that, the content of pure hydrolysed proteins is expressed by multiplying the organic nitrogen content by the conventional factor of 6.25. Full details are provided in Section 5 of the MCP summaries.

5.1.3 Methods for Risk Assessment

According to the EFSA Journal 2012;10(2):2545, the requirement for methods of analysis for residues was waived as no residue definitions were proposed due to the nature of the Hydrolysed proteins.

Indeed, as demonstrated in Documents M-CA 5, M-CA 6, M-CA 7 and M-CA 8, Hydrolysed proteins are *per se* of low toxicity and not classified, and are therefore considered to pose a low risk to human health, to the environment and to non-target organisms. Furthermore, no maximum residue levels (MRLs) is required for the Hydrolysed proteins.

In this context, method for the risk assessment is not required, nor relevant.

5.2 Methods for post-authorisation control and monitoring purposes

According to the EFSA Journal 2012;10(2):2545, the requirement for methods of analysis for residues was waived as no residue definitions were proposed due to the nature of the Hydrolysed proteins.

Indeed, as demonstrated in Documents M-CA 5, M-CA 6, M-CA 7 and M-CA 8, Hydrolysed proteins are *per se* of low toxicity and not classified and are considered to pose a low risk to human health, to the

environment and to non-target organisms. Furthermore, no maximum residue levels (MRLs) is required for the Hydrolysed proteins.

In this context, method for the risk assessment is not required, nor relevant.

6 IMPACT ON HUMAN AND ANIMAL HEALTH

6.1 Effects Having Relevance to Human and Animal Health

According to EFSA Journal 2012; 10(2):2545, it was concluded that "*hydrolysed proteins per se are likely to be of low toxicological concern provided hydrolysed proteins of animal origin are pathogen-free. On this basis no risks to human health could be expected from its use as a plant protection product and data waivers for specific toxicological studies were initially supported. However, due to the fact that a specification to include the main components in the active substances is still outstanding, a final conclusion cannot be drawn whether the technical specification is of toxicological concern and whether data waivers can be accepted.*" A data gap was identified.

Considering the nature, origin, manufacturing process, composition and technical specifications of the Hydrolysed proteins as provided in Document J, there is no component of toxicological concern, thus confirming that hydrolysed proteins are of low toxicity.

Furthermore, Hydrolysed proteins are naturally occurring compounds of degradation from the hydrolysis of living organisms' tissues that can have vegetable or animal origin. The degradation of the hydrolysed proteins results in more simple metabolites called amino acids. Proteins and amino acids are abundant organic molecules in living cells. They can be found in every single cell, since they are fundamental in all aspects of the cell structure and function, and intervene in the most essential biochemical processes.
The proteins are one of the three basic principal nourishment of living beings. The proteins that are found in food and eaten by human beings and mammals are normally degraded metabolically by means of enzymatic processes and results in amino acids, that are then used by the living cells for the biosynthesis of new specific proteins.
Therefore, the hydrolysed proteins and resulting metabolites are not expected to cause any danger to human beings and mammals in general because as explained, these compounds take place in every living cells and are therefore essential for life.

Moreover, it should be noted that the review of the scientific literature within the last 10 years did not give any results indicating a hazardous effect or a potential risk for humans and mammals in general. Please refer to Document M-CA 9.

Furthermore, it should be noted that according to the notifications provided to ECHA in REACH registrations and CLP notifications, no hazards have been classified for the active substance Hydrolysed proteins, referred as "*Protein hydrolyzates, animal*" by ECHA. Please refer to the summary from ECHA provided in Document M-CA 10.

For all these reasons, the use of Hydrolysed proteins is considered to pose a low risk to operators, workers, bystanders and residents and no testing toxicity data are required.

6.1.1 Summary of adsorption, distribution, metabolism and excretion

No data submitted, not required. Please refer to point 6.1.

6.1.2 Summary of acute toxicity

No data submitted, not required. Please refer to point 6.1.

6.1.3 Summary of short-term toxicity

No data submitted, not required. Please refer to point 6.1.

6.1.4 Summary of genotoxicity

No data submitted, not required. Please refer to point 6.1.
Due to its nature, origin and composition, Hydrolysed proteins are *per se* of low toxicological concern and since their components have an essential role in living cells, it is deemed acceptable to consider that the active substance Hydrolysed proteins have no genotoxic potential.

6.1.5 Summary of long-term toxicity and carcinogenicity

No data submitted, not required. Please refer to point 6.1.
Due to its nature, origin and composition, Hydrolysed proteins are *per se* of low toxicological concern and since their components have an essential role in living cells, it is deemed acceptable to consider that the active substance Hydrolysed proteins have no carcinogenic potential.

6.1.6 Summary of reproductive toxicity

No data submitted, not required. Please refer to point 6.1.
Due to its nature, origin and composition, Hydrolysed proteins are *per se* of low toxicological concern and since their components have an essential role in living cells, it is deemed acceptable to consider that the active substance Hydrolysed proteins have no reproductive toxicity.

6.1.7 Summary of neurotoxicity

No data submitted, not required. Please refer to point 6.1.
Due to its nature, origin and composition, Hydrolysed proteins are *per se* of low toxicological concern and since their components have an essential role in living cells, it is deemed acceptable to consider that the active substance Hydrolysed proteins are not neurotoxic.

6.1.8 Summary of further toxicological studies on the active substance

No data submitted, not required. Please refer to point 6.1.

6.1.9 Summary of toxicological data on impurities and metabolites

No data submitted, not required. Please refer to point 6.1.

6.1.10 Summary of medical data and information

No data submitted, not required. Please refer to point 6.1.

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

No ADI available, not required. Please refer to point 6.1.

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

No ARfD available, not required. Please refer to point 6.1.

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

No AOEL available, not required. Please refer to point 6.1.

6.5 Summary of product exposure and risk assessment

No EU endpoints available for the active substance. No AOEL was defined due to the nature of the Hydrolysed proteins considered *per se* of low toxicological concern
Exposure assessments and risk evaluations for operators, workers, bystanders and residents are therefore not required.
The use of Hydrolysed proteins as plant protection product is considered to pose a low risk to operators, workers, bystanders and residents.

6.5.1 Operators

No exposure assessment performed, not required.

6.5.2 Bystander and resident exposure

No exposure assessment performed, not required.

6.5.3 Workers

No exposure assessment performed, not required.

7 RESIDUES

According to EFSA Journal 2012; 10(2):2545, "*hydrolysed proteins as a plant protection product is likely to be of low toxicity and a quantitative consumer risk assessment is not needed unless the required technical specification raises a toxicological concern.*"

Considering the nature, origin, manufacturing process, composition and technical specifications of the Hydrolysed proteins as provided in Document J, there is no component of toxicological concern, thus confirming that hydrolysed proteins are of low toxicity.

Furthermore, it should be noted that Hydrolysed proteins are natural compounds derived by the hydrolysis of the animal tissue proteins. The proteins are macromolecules constituted by the union of amino acids. In the nature, there is a restricted number of amino acids, the proportion of these amino acids and their order, among other characteristics, are the origin of variability of the different proteins.
The hydrolysis consists on breaking the peptide links which join the amino acids of a protein. Depending on the intensity of the process, a variable composition of free amino acids, peptides and polypeptides will be obtained. The hydrolysis does not mean the appearance of new molecules but its defragmentation of treated proteins, these fragments do not lose their biological capacity, so they can be re-used by the cells to construct new protein tissues. This characteristic is the base of the complex metabolic process of all the animals feeding, through a part of the consumed protein feed, is transformed in new proteins.
According to their nature, it cannot be said that hydrolysed proteins generate residues but materials that can be re-used because they structurally have the same composition as the cells. Hydrolysed protein components, free amino acids and peptides, are natural substances and consequently completely biodegradable.

Besides, since proteins, free amino acids and peptides occur naturally in plants and animals, it would not possible to distinguish the naturally components from those resulting from the use of plant protection products.

Therefore, hydrolysed proteins can be exempted from the requirements of residues data.

7.1 Summary of storage stability of residues

No data submitted, not required. Please refer to point 7.

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

7.3 Definition of the residue

No residue definition is recommended. Please refer to point 7.

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

No data submitted, not required. Please refer to point 7.

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

No data submitted, not required. Please refer to point 7.

7.6 Summary of effects of processing

No data submitted, not required. Please refer to point 7.

7.7 Summary of residues in rotational crops

No data submitted, not required. Please refer to point 7.

7.8 Summary of other studies

No data submitted, not required. Please refer to point 7.

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

Acceptable Daily Intake (ADI) and Dietary Exposure Calculation

No ADI available, dietary exposure calculation not required. Please refer to point 7.

Acute Reference Dose (ARfD) and Dietary Exposure Calculation

No ARfD available, dietary exposure calculation not required. Please refer to point 7.

7.10 Proposed MRLs and compliance with existing MRLs

According to the argumentation presented under Point 7, Hydrolysed proteins should be exempted from the requirements of residues data and thus included in Annex IV of the Regulation (EC) No 396/2005. No maximum residue levels (MRLs) should be required for Hydrolysed proteins.

7.11 Proposed import tolerances and compliance with existing import tolerances

Not required.

8 FATE AND BEHAVIOUR IN THE ENVIRONMENT

Hydrolysed proteins are naturally occurring compounds of degradation from the hydrolysis of living organisms' tissues that can have vegetable or animal origin. The degradation of the hydrolysed proteins results in more simple metabolites called amino acids. Proteins and amino acids are abundant organic molecules in living cells. They can be found in every single cell, since they are fundamental in all aspects of the cell structure and function, and intervene in the most essential biochemical processes.
The proteins are one of the three basic principal nourishment of living beings. The proteins that are found in food and eaten by human beings and mammals are normally degraded metabolically by means of enzymatic processes and results in amino acids, that are then used by the living cells for the biosynthesis of new specific proteins.
Thus, the hydrolysed proteins are biodegradable, so their persistence in the environment is very short without any tendency for bioaccumulation.
Due to the nature of the hydrolysed proteins and their characteristics regarding the fate and behaviour in the environment, it is deemed very unlikely the existence of relevant residues resulting from applications as plant protection product in the soil, surface water or sediment and even more unlikely the existence of relevant residues reaching the groundwater.

For all these reasons, it is deemed not necessary to conduct any studies with Hydrolysed proteins about the fate and behaviour in the environment.

In this context, Hydrolysed Proteins meet the criteria for the approval of low-risk active substance because not considered to be persistent nor to have potential for bioaccumulation.

8.1 Summary of fate and behaviour in soil

No data submitted, not required. Please refer to point 8.

8.2 Summary of fate and behaviour in water and sediment

In response to the RMS request regarding "Ready biodegradability" of Hydrolysed proteins, the applicant PROALAN S.A. has performed a study to assess the aerobic biodegradability of the product SVMA14-004. Furthermore, it is important to note that due to the composition of the product SVMA14-004, the results of this study can be extrapolated to the active substance Hydrolysed proteins.
On the basis of results obtained, interpreted in accordance to OECD 310:2014, the product is considered readily biodegradable in aerobic conditions.

8.3 Summary of fate and behaviour in air

The active substance Hydrolysed Proteins is a natural substance biodegradable. Some of the metabolites resulting from the decomposition process are volatiles substances, such as ammonia, carbon dioxide or putrescine.
The volatile compounds from Hydrolysed proteins are naturally occurring compounds of no concern.

The applicant wants to highlight that hydrolysed proteins are naturally occurring compounds whose degradation leads to simple metabolites called amino acids that are abundant
organic molecules in living cells.

Furthermore, amino acids from hydrolysis of animal proteins are used as fertilisers in Spain according to the Real Decreto 506/2013 (please refer to p 35, group 4.1 called "Aminoácidos"). In addition, hydrolysed proteins of category 3 materials is part of the proposal of the future new EU regulation for fertilisers products (please refer to the proposal of December, the 4th, page 83)
Therefore, these compounds are considered to be of low risk for soil and water compartments and considering that they are widely used as fertilisers in Europe, the amount of hydrolysed proteins added to the environmental compartments linked to the application as plant protection product is considered to be negligible in comparison to the amounts derived from the use as fertilisers.
The applicant is of the opinion that this information provides an argumentation strong enough to justify the exemption of further studies, considered as unnecessary from a scientific and rational point of view.
However, for completeness purposes, the applicant provided new data for vapour pressure and half-life in air for amino acids.

However, according to its composition, the active substance Hydrolyse Proteins contains a significant content of nitrogen, and therefore the eutrophication potential must be assessed. Please refer to the calculations provided below under point 8.6.

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

No data submitted, not required.
It should be noted that since proteins are naturally occurring in the environment, it would not possible to distinguish the naturally compounds from those resulting from the use of plant protection products. Thus, the concept of environmental monitoring is not applicable for Hydrolysed proteins.

8.5 Definition of the residues in the environment requiring further assessment

No residue definition for monitoring, not required. Please refer to point 8.
Furthermore, it should be noted that since proteins are naturally occurring in the environment, it would not possible to distinguish the naturally compounds from those resulting from the use of plant protection products.

8.6 Summary of exposure calculations and product assessment

Soil
Due to the nature of the active substance Hydrolysed Proteins, no DT_{50} is available and it can be considered that Hydrolysed Proteins are rapidly broken down into its constituent parts on contact with soil and/or crop material. Therefore, it is appropriate to calculate the PEC_S following a single application only, using the following equation:

$$PEC_S\,(mg/kg) = \frac{\text{Application rate}\,(g/ha) \times (1 - F)}{100 \times \text{Soil depth}\,(cm) \times \text{Soil dry bulk density}\,(g/cm^3)}$$

PEC_{soil} calculations

Active substance	Application rate (g/ha)	Crop interception (%)	PECs (mg/kg)
Hydrolysed Proteins	450	65	0.210

Groundwater

Due to the nature of the hydrolysed proteins and their characteristics regarding the fate and behaviour in the environment, it is deemed very unlikely the existence of relevant residues resulting from applications as plant protection product in the soil and even more unlikely the existence of relevant residues reaching the groundwater.
Therefore, no PECgw are required.

The applicant wants to highlight that hydrolysed proteins are naturally occurring compounds whose degradation leads to simple metabolites called amino acids that are abundant organic molecules in living cells.
Furthermore, amino acids from hydrolysis of animal proteins are used as fertilisers in Spain according to the Real Decreto 506/2013 (please refer to p 35, group 4.1 called "Aminoácidos"). In addition, hydrolysed proteins of category 3 materials is part of the proposal of the future new EU regulation for fertilisers products (please refer to the proposal of December, the 4th, page 83)
Therefore, these compounds are considered to be of low risk for soil and water compartments and considering that they are widely used as fertilisers in Europe, the amount of hydrolysed proteins added to the environmental compartments linked to the application as plant protection product is considered to be negligible in comparison to the amounts derived from the use as fertilisers.
The applicant is of the opinion that this information provides an argumentation strong enough to justify the exemption of further studies, considered as unnecessary from a scientific and rational point of view.

Surface water and sediment

Due to the differing and unknown dissipation times of the constituents of Hydrolysed proteins in aquatic systems, it was only possible to calculate the maximum instantaneous PEC_{SW} value from entry through spray-drift that occurred immediately after a single application. The PEC_{SW} was calculated using the following equation:

$$\mathrm{PEC_{SW}}\,(\mu\mathrm{g/L}) = \frac{\%\,\mathrm{Drift}_{90\text{th }\%\mathrm{ile}} \times \mathrm{Application\ rate}\,(g/ha)}{\mathrm{Water\ depth\ (cm)} \times 10}$$

Potential of eutrophication in surface water

According to its composition, the active substance Hydrolyse Proteins contains a significant content of nitrogen, and therefore the eutrophication potential must be assessed. As there is no current guideline to assess the eutrophication potential, the following approach have been considered here:
According to composition of the active substance Hydrolysed Proteins and the product SVMA14-004 (please refer to documents J), a maximum nitrogen content of 6% w/w in the product SVMA14-004 is considered as an appropriate worst case for the calculations. Therefore, the corresponding application rate would be 103.95 g of nitrogen/ha, considering a density of 1.155.
By using the formula above, the resulting maximum instantaneous PEC_{SW} value for nitrogen is equal to 5.440 µg/L.
This PECsw is far below the environmental water quality standard for NO3 of 50 mg/l (equivalent to 11.3 mg N/l), indicating that Hydrolysed Proteins do not have any eutrophication potential.

9 EFFECTS ON NON-TARGET SPECIES

Hydrolysed proteins are naturally occurring compounds of degradation from the hydrolysis of living organisms' tissues that can have vegetable or animal origin. The degradation of the hydrolysed proteins results in more simple metabolites called amino acids. Proteins and amino acids are abundant organic molecules in living cells. They can be found in every single cell, since they are fundamental in all aspects of the cell structure and function, and intervene in the most essential biochemical processes.
Thus, the hydrolysed proteins are biodegradable, so their persistence in the environment is very short without any tendency for bioaccumulation.
Due to the nature of the hydrolysed proteins and their characteristics regarding the fate and behaviour in the environment, it is deemed very unlikely the existence of relevant residues resulting from applications as plant protection product in the soil, surface water or sediment.
Furthermore, the proteins are one of the three basic principal nourishment of living beings. The proteins that are found in food and eaten by human beings and mammals are normally degraded metabolically by means of enzymatic processes and results in amino acids, that are then used by the living cells for the biosynthesis of new specific proteins.
Therefore, the hydrolysed proteins and resulting metabolites are not expected to cause any danger to human beings and mammals in general because as explained, these compounds take place in every living cells and are therefore essential for life.

Moreover, it should be noted that the review of the scientific literature within the last 10 years did not give any results indicating a hazardous effect or a potential risk for the terrestrial and aquatic wildlife and ecosystems in general. Please refer to Document M-CA 9.

The applicant would like to highlight that Hydrolysed proteins are naturally occurring compounds whose degradation leads to simple metabolites called amino acids that are abundant organic molecules in living cells and used for the biosynthesis of new specific proteins and therefore essential for life.
Furthermore, in addition to the use as fertilisers as explained for environmental fate and behaviour section, hydrolysed proteins are also used as raw material for feed in aquaculture or animal feeding market.
On request, the applicant can provide several articles or examples of final products containing hydrolysed proteins in the market linked to the aquaculture and animal feeding uses, to reinforce this argumentation.
Moreover, the active substance hydrolysed proteins is not classified according to the ECHA.
The applicant is of the opinion that this information provide an argumentation strong enough to justify the exemption of further studies or risk assessments, considered as unnecessary from a scientific and rational point of view.
Moreover, for completeness purposes, the applicant provides a study performed with Hydrolysed proteins to prove that the administration of such compounds to non-target organisms has no harmful effects.
The conclusion of this study is that the use of Hydrolysed proteins resulted in significant increases in the concentration of monocytes and platelets and in mean platelet volume, but without having any significant effect on the growth and feed conversion index. Moreover, it was clearly highlighted that the increased concentration of monocytes observed after administration reveals an immune efficacy, inducing increased defences against various pathogenic microorganisms and therefore having non harmful effects to non-target organisms.

For all these reasons, the use of Hydrolysed proteins is considered to pose a low risk to non-target organisms (birds, aquatic organisms, wild mammals, bees, non-target arthropods, earthworms and other soil-macroorganisms, soil microorganisms and non-target plants) and no testing toxicity data are required.

9.1 Summary of effects on birds and other terrestrial vertebrates

No data submitted, not required. Please refer to point 9.

9.2 Summary of effects on aquatic organisms

No EU data/endpoints available. According to the EFSA Journal 2012;10(2):2545, studies on aquatic organisms were considered necessary to fulfil the Annex II requirements directly related to classification and labelling. A data gap was identified.

Therefore, confirmatory data were presented and the designated RMS (Greece) in the Addendum IV to the Draft Assessment Report (Volume 3, Annex B-9: Ecotoxicology, September 2014), has concluded the following:

"Although no specific testing toxicity data on either of the hydrolysed proteins notified have been submitted, by taking into account:

(i) the lack of any information or evidence in the scientific literature related to the aquatic toxicity potential of hydrolysed proteins,

(ii) the indication of low hazard and risk entailed by the use of hydrolysed proteins (e.g. beet molasses-urea hydrolysate) in insect attractants for bait spray applications compared to other nitrogen compounds and

(iii) the nature of the active substance and its characteristics regarding the fate and behavior in the environment (biodegradable, non-persistent, non bioaccumulative),

it can be concluded that the use of hydrolysed proteins is of low danger for the aquatic ecosystems in general and for the aquatic organisms in particular. In consequence, from the RMS's point of view, hydrolysed proteins should not be assigned any classification for aquatic hazards and should be deemed as non-dangerous for the environment substances."

Furthermore, it should be noted that according to the notifications provided to ECHA in REACH registrations and CLP notifications, no hazards have been classified for the active substance Hydrolysed proteins, referred as "*Protein hydrolyzates, animal*" by ECHA. Please refer to the summary from ECHA provided in Document M-CA 10.

According to the EFSA Conclusion (2012) a data gap was identified concerning toxicity studies for aquatic organisms: "Studies on aquatic organisms that are necessary to fulfil the Annex II requirements directly related to classification and labelling".
Therefore, and in agreement with the current data requirements related to Regulation (EU) 283/2013 the following toxicity studies have been submitted for aquatic organisms:

- Acute toxicity to fish (*Danio rerio*).
- Acute toxicity for aquatic invertebrates (*Daphnia magna*).
- Growth inhibition test for freshwater alga (*Pseudokirchneriella subcapitata*).

Risk assessment for aquatic organisms

No risk assessment performed, not required. No adverse effect of Hydrolysed proteins to aquatic organisms is expected following the use of SVMA14-004.

Nevertheless, the following assumption is presented instead. An unacceptable risk is identified when PEC/RAC > 1 and by taking into account the maximum PEC_{sx} of 23.550 µg/L calculated in Document M-CP 9, this would result in a RAC < 23.55 µg/L. This would happen if the corresponding acute LC_{50}/EC_{50} would be lower than 2.355 mg/L (with an AF of 100).
Such a low LC_{50}/EC_{50} is highly unexpected considering the nature of the active substance. In addition, it should be noted that according to the notifications provided to ECHA in REACH registrations and CLP notifications, no hazards have been classified for the active substance Hydrolysed proteins, referred as "*Protein hydrolyzates, animal*" by ECHA. Please refer to the summary from ECHA provided in Document M-CA 10.
Moreover, it should be noted that the review of the scientific literature within the last 10 years did not give any results indicating a hazardous effect or a potential risk for aquatic wildlife. Please refer to Document M-CA 9.

According to the new aquatic toxicity studies performed, a new risk assessment was performed.
For the intended uses on citrus (both early and late), calculated PEC/RAC ratios did indicate an acceptable risk for the most sensitive groups of aquatic organisms (risk for fish as characterised by an EC50 for *Danio rerio* of 100000.00 µg/L in connection with an assessment factor of 100 and for invertebrates as characterised by an EC50 for *Daphnia magna* of 100000.00 µg/L in connection with an assessment factor of 100) for all the instantaneous PECsw calculations. Therefore, no further assessment is necessary.
For the intended uses on persimmon (both early and late), calculated PEC/RAC ratios did indicate an acceptable risk for the most sensitive groups of aquatic organisms (risk for fish as characterised by an EC50 for *Danio rerio* of 100000.00 µg/L in connection with an assessment factor of 100 and for invertebrates as characterised by an EC50 for *Daphnia magna* of 100000.00 µg/L in connection with an assessment factor of 100) for all the instantaneous PECsw calculations. Therefore, no further assessment is necessary.

Therefore, this confirms that the use of Hydrolysed proteins as plant protection product is considered to pose a low risk to aquatic organisms.

9.3 Summary of effects on arthropods

No data submitted, not required. Please refer to point 9.

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

No data submitted, not required. Please refer to point 9.

9.5 Summary of effects on soil nitrogen transformation

No data submitted, not required. Please refer to point 9.

9.6 Summary of effects on terrestrial non-target higher plants

No data submitted, not required. Please refer to point 9.

Furthermore, Hydrolysed proteins are not intended to be used as an herbicide or a plant growth regulator, and are not known to have any herbicidal activities.

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

No data submitted, not required. Please refer to point 9.

9.8 Summary of effects on biological methods for sewage treatment

No data submitted, not required. Please refer to point 9.

9.9 Summary of product exposure and risk assessment

Following application of SVMA14-004, no risk or unacceptable effects are expected for birds, mammals, fish, aquatic invertebrates, algae, higher aquatic plants, honeybees, non-target arthropods others than bees, non-target soil meso- and macrofauna, soil nitrogen transformation processes and non-target higher plants. Particular precautions to reduce the environmental concentrations resulting from SVMA14-004 applications are not required for the protection of terrestrial non-target organisms. Regarding application of SVMA14-004 close to surface waters, low risk to the aquatic environment are expected with no need for additional mitigation measures.

10 CLASSIFICATION AND LABELLING

Hydrolysed Proteins are *per se* of low toxicity and are therefore not classified according to CLP Regulation (EC) nº 1272/2008.

- Pictograms: none

- Signal words: none

- Hazard statements: none

- Precautionary statements: none

It should be noted that this is in compliance with the classification provided by ECHA, for which according to the notifications provided to ECHA in REACH registrations and CLP notifications, no hazards have been classified for the active substance Hydrolysed proteins, referred as "*Protein hydrolyzates, animal*" by ECHA. Please refer to the summary from ECHA provided in document M-CA 10.

11 ASSESSMENT AGAINST CRITERIA FOR APPROVAL OF LOW-RISK ACTIVE SUBSTANCE UNDER ANNEX II TO REGULATION 1107/2009/EC

In the assessment presented below, Hydrolysed proteins is compared against the criteria for approval of low-risk active substance under Annex II to Regulation 1107/2009/EC. Hydrolysed proteins is a naturally occurring substance which is not classified against any of the human health criteria in Annex II, paragraph 5 of regulation 1107/2009/EC and does not demonstrate endocrine disrupting properties that may cause adverse effects on non-target organisms.

Regulation 1107/2009/EC Annex II	**Criteria**	**Assessment**
Paragraph 5.1.1	Substance classified as a carcinogen category 1A, 1B or 2 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as a mutagen category 1A, 1B or 2 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as toxic for reproduction category 1A, 1B or 2 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as a skin sensitiser category 1 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as eye irritant category 1 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as a respiratory sensitiser category 1 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as acute toxic category 1,2 or 3 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as a specific target organ toxicant, category 1 or 2 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as toxic to aquatic life of acute and chronic category 1 according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance classified as explosive according to Regulation (EC) No 1272/2008	No
Paragraph 5.1.1	Substance considered to have endocrine disrupting properties	No
Paragraph 5.1.1	Substance considered to have neurotoxic or immunotoxic effets	No
Paragraph 5.1.2	Persistence: Soil DT_{50} > 60 days Or Bio-concentration factor > 100	Not relevant as Hydrolysed proteins is a naturally occurring active substance which does not correspond to any of points presented above.

12 RELEVANCE OF METABOLITES IN GROUNDWATER

Not required due to the nature of the active substance.

13 CONSIDERATION OF ISOMERIC COMPOSITION IN THE RISK ASSESSMENT

Not relevant due to the nature of the active substance.

FURTHER INFORMATION TO BE SUBMITTED

None.