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**EURL Evaluation Report on the Analytical Methods
submitted in connection with the Application for the
Authorisation of Feed Additives according to
Regulation (EC) No 1831/2003**

Dossier related to:	FAD-2010-0358 - CRL/100221
Feed additive:	Copper complex of Chlorophyllins
Active Substance(s):	Copper complex of Chlorophyllins
Rapporteur Laboratory:	European Union Reference Laboratory for Feed Additives (EURL-FA) Geel, Belgium
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Date:	28/10/2011

EXECUTIVE SUMMARY

In the current application authorisation is sought under articles 4(1) and 10(2) for *Copper complexes of chlorophyllins - E141(ii)* - under the "sensory additives", functional group 2(a) "colourants", according to the classification system of Annex I of Regulation (EC) No 1831/2003. Authorisation is sought for the use of the *feed additive* for all species and categories.

Copper complexes of chlorophyllins - E141(ii) is readily water-soluble dark green/black free flowing powder, extracted from *Festuca arundinacea* plants. The active substance of the additive has a minimum purity of 95 %. The *additive* is intended to be incorporated directly in dry or moist *feedingstuffs*, with no recommended minimum or maximum levels.

For the determination of *Copper complexes of chlorophyllins* in the *feed additive*, the Applicant proposed the internationally recognised FAO JECFA monograph for food additives. Identification is based on a spectrophotometric analysis at 405 and 630 nm in aqueous phosphate buffer and a test for copper presence, while quantification of *Copper complexes of chlorophyllins* in the *feed additive* is based on spectrophotometry at 405 and 630 nm in aqueous phosphate buffer, as recommended by Commission Directive 2008/128/EC laying down specific purity criteria concerning colours for use in foodstuffs. Even though no performance characteristics are provided, the EURL recommends for official control the JECFA monograph based on spectrophotometry for the quantification of the *Copper complexes of chlorophyllins* in the *feed additive*.

The Applicant did not provide any experimental method or data for the determination of *Copper complexes of chlorophyllins* in *premixtures*, *feedingstuffs* and *water*. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Copper complexes of chlorophyllins* in *premixtures*, *feedingstuffs* and *water*.

Further testing or validation of the methods to be performed through the consortium of National Reference Laboratories as specified by Article 10 (Commission Regulation (EC) No 378/2005) is not considered necessary.

KEYWORDS

Copper complexes of chlorophyllins, sensory additive, colourants, all species and categories

1. BACKGROUND

In the current application authorisation is sought under articles 4(1) (new use in water) and 10(2) (re-evaluation of additives already authorised under the provisions of the Council Directive 70/524/EEC) for *Copper complexes of chlorophyllins* under the "sensory additives", functional group 2(a) "colourants" [1], according to the classification system of Annex I of Regulation (EC) No 1831/2003. Authorisation is sought for the use of the *feed additive* for all animal species and categories [2].

Copper complexes of chlorophyllins - E141(ii) is readily water-soluble dark green/black free flowing powder, extracted from *Festuca arundinacea* plants [3]. The active substance of the additive has a minimum purity of 95 % [3]. The Applicant states that the purity criteria set in the Commission Directive 2008/128/EC for the food additive apply to the requirement for the *feed additive* [3]. The *additive* is intended to be incorporated directly in dry or moist *feedingstuffs*, with no recommended minimum or maximum levels [2].

Note – An ambiguous nomenclature:

The chemical formulas provided by the Applicant to describe the active substance ($C_{34}H_{32}CuN_4O_5$ and $C_{34}H_{30}CuN_4O_6$) are authorised as: - E141 (ii) *Copper Complexes of Chlorophyllins* in the Commission Directive 2008/128/EC (laying down specific purity criteria concerning colours for use in **foodstuffs**), and – E 141 *Chlorophyll Copper Complex* in the Directive 70/525/EC and in Commission Regulation 358/2005 (additive... authorisation in feedingstuffs)

2. TERMS OF REFERENCE

In accordance with Article 5 of Regulation (EC) No 378/2005, as last amended by Regulation (EC) No 885/2009, on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the duties and the tasks of the European Union Reference Laboratory concerning applications for authorisations of feed additives, the EURL is requested to submit a full evaluation report to the European Food Safety Authority for each application or group of applications. For this particular dossier, the methods of analysis submitted in connection with *Copper complexes of chlorophyllins*, and their suitability to be used for official controls in the frame of the authorisation, were evaluated.

3. EVALUATION

Identification /Characterisation of the feed additive

Qualitative and quantitative composition of impurities in the additive

When required by EU legislation, analytical methods for official control of undesirable substances in the additive (e.g. arsenic, cadmium, lead, mercury, mycotoxins, and dioxins) are available from the respective European Union Reference Laboratories [4].

Description of the analytical methods for the determination of the active substance in feed additive, premixtures and feedingstuffs

For the determination of *Copper complexes of chlorophyllins* in the *feed additive*, the Applicant proposed the internationally recognised FAO JECFA monograph for food additives [5,6]:

- Identification of *Copper complexes of chlorophyllins* in the *feed additive* is based on – a solubility test in water, a spectrophotometric measurement at 405 and 630 nm in aqueous phosphate buffer, and qualitative tests to identify the presence of copper.
- Quantification of *Copper complexes of chlorophyllins* in the *feed additive* is based on spectrophotometry at 405 and 630 nm in aqueous phosphate buffer, as recommended by Commission Directive 2008/128/EC.

Even though no performance characteristics are provided, the EURL recommends for official control the JECFA monograph based on spectrophotometry for the quantification of the *Copper complexes of chlorophyllins* in the *feed additive*.

The Applicant did not provide any experimental method or data for the determination of *Copper complexes of chlorophyllins* in *premixtures, feedingstuffs* and *water*. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Copper complexes of chlorophyllins* in *premixtures, feedingstuffs* and *water*.

Further testing or validation of the methods to be performed through the consortium of National Reference Laboratories as specified by Article 10 (Commission Regulation (EC) No 378/2005) is not considered necessary.

4. CONCLUSIONS AND RECOMMENDATIONS

In the frame of this authorisation the EURL recommends for official control of the *feed additive* the identification tests and the quantification assay recommended by Commission Directive 2008/128/EC and described in the JECFA monographs n. 1 (Vol. 4), Combined Compendium for Food Additive Specifications.

The Applicant did not provide any experimental method or data for the determination of *Copper complexes of chlorophyllins* in *premixtures, feedingstuffs* and *water*. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Copper complexes of chlorophyllins* in *premixtures, feedingstuffs* and *water*.

Recommended text for the register entry (analytical method)

For the quantification of *Copper complexes of chlorophyllins* in the *feed additive*:

- spectrophotometry at 405 or 630 nm (Commission Directive 2008/128/EC referring to FAO JECFA monographs n. 1 (Vol. 4))

5. DOCUMENTATION AND SAMPLES PROVIDED TO EURL

In accordance with the requirements of Regulation (EC) No 1831/2003, reference samples of *Copper complexes of chlorophyllins* have been sent to the European Union Reference Laboratory for Feed Additives. The dossier has been made available to the EURL by EFSA.

6. REFERENCES

- [1] *Application, Reference SANCO/D/2 Forw. Appl. 1831/(00163)/(10283)-2010
 - [2] *Application, Proposal for Register Entry – Annex A
 - [3] *Technical dossier, Section II: Identity, characterisation and conditions of use of the additive; Methods of analysis
 - [4] Commission Regulation (EC) No 776/2006 amending Annex VII to Regulation (EC) No 882/2004 of the European Parliament and of the Council as regards to Community Reference Laboratories
 - [5] FAO JECFA monographs n. 1 (Vol. 4), Combined Compendium for Food Additive Specifications
 - [6] FAO JECFA monograph 1 (2005) – corrected in 2008: Chlorophyllins
- * Refers to Dossier No. FAD-2010-0358

7. RAPPORTEUR LABORATORY & NATIONAL REFERENCE LABORATORIES

The Rapporteur Laboratory for this evaluation was European Union Reference Laboratory for Feed Additives, IRMM, Geel, Belgium. This report is in accordance with the opinion of the consortium of National Reference Laboratories as referred to in Article 6(2) of Commission Regulation (EC) No 378/2005, as last amended by Regulation (EC) No 885/2009.

8. ACKNOWLEDGEMENTS

The following National Reference Laboratories contributed to this report:

- Österreichische Agentur für Gesundheit und Ernährungssicherheit (AGES), Wien (AT)
- Thüringer Landesanstalt für Landwirtschaft (TLL), Abteilung Untersuchungswesen. Jena (DE)
- Plantedirektoratet, Laboratorium for Foder og Gødning, Lyngby (DK)
- Państwowy Instytut Weterynaryjny, Puławy (POL)
- Centro di referenza nazionale per la sorveglianza ed il controllo degli alimenti per gli animali (CReAA), Torino (IT)
- Ústřední kontrolní a zkušební ústav zemědělský (ÚKZÚZ), Praha (CZ)
- Kmetijski inštitut Slovenije, Ljubljana (SLO)