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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-2010-0217** **CRL/100087**
Product Name:	**Disodium guanosine 5'-monophosphate**
Active Substance(s):	**Disodium guanosine 5'-monophosphate**
Rapporteur Laboratory:	**European Union Reference Laboratory for Feed Additives (EURL-FA)** **Geel, Belgium**
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EXECUTIVE SUMMARY

In the current application authorisation is sought under articles 4(1) and 10(2) for *Disodium guanosine 5'-monophosphate* under the category "sensory additives", functional group 2(b) "flavouring compounds", 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.

According to the Applicant the *feed additive* consists of *disodium guanosine 5'-monophosphate* (GMP) **and** *disodium inosine 5'-monophosphate* (IMP) with a minimum content of the sum of constituents of 97%, with the content of each constituent ranging from 47 to 53% relative to dry mass.

The *feed additive* is intended to be incorporated only into *feedingstuffs* or drinking *water*, in combination with other flavouring substances as constituents of *flavouring mixtures*. The Applicant suggested no minimum or maximum levels for the *feed additive*, but normal contents of the flavouring compound in *feedingstuffs* range up to from 0.1 to 100 mg/kg.

For the identification of *GMP and IMP* in the *feed additive*, the Applicant proposed the internationally recognised FAO JECFA monograph for disodium 5'-ribonucleotides. For the determination of ING and GMP in raw materials, *mixtures of flavourings* and *water*, the Applicant submitted a High Performance Liquid Chromatography method coupled to UV detection (HPLC-UV) based on the above mention JECFA protocol.

Even though no performance characteristics are provided, the EURL recommends for official control the HPLC-UV method submitted by the Applicant, for the quantification of the *IMP and GMP* in the *feed additive* and *water*.

As no experimental data were provided by the Applicant for the determination of the product in *feedingstuffs*, the EURL could not evaluate nor recommend the method for official control to determine *GMP* and *IMP* in *feedingstuffs*.

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

Disodium guanosine 5'-monophosphate, Chemically Defined Flavourings, flavouring mixtures, sensory additives, 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 Directive 70/524/EC) for *Disodium guanosine 5'-monophosphate* under the category "sensory additives", functional group 2(b) "flavouring compounds" [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 species and categories [2].

In general the additive is produced either by decomposing microbial RNA with 5'-phosphodiesterase to form 5'-nucleotides, or by the combination of fermentation employing sugar as a starting material and resulting in the production of nucleosides and chemical synthesis by phosphorylation into 5'-nucleotides [3].

According to the Applicant the *feed additive* consists of *disodium guanosine 5'-monophosphate* (GMP) **and** *disodium inosine 5'-monophosphate* (IMP) with a minimum content of the sum of constituents of 97%, with the content of each constituent ranging from 47 to 53% relative to dry mass [2].

The *feed additive* is intended to be incorporated only into *feedingstuffs* or drinking *water*, in combination with other flavouring substances as constituents of *flavouring mixtures* [3]. The Applicant suggested no minimum or maximum levels for the *feed additive* [2], but normal contents of the flavouring compound in *feedingstuffs* range up to from 0.1 to 100 mg/kg [3].

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 *Disodium guanosine 5'-monophosphate*, and their suitability to be used for official controls in the frame of the authorisation, were evaluated.

3. EVALUATION

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, 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 identification of *GMP and IMP* in the *feed additive,* the Applicant proposed the internationally recognised FAO JECFA monograph for disodium 5'-ribonucleotides [5], based on the following tests for: solubility, ribose, sodium organic phosphate, inosine and guanosine, as described in the JECFA monographs [6].

For the determination of *GMP* and *IMP* in raw materials, *mixtures of flavourings* and *water*, the Applicant submitted a High Performance Liquid Chromatography method coupled to UV detection (HPLC-UV) [7] based on the above mention JECFA protocol, having a limit of detection of 5 mg/kg.

Even though no performance characteristics are provided, the EURL recommends for official control the HPLC-UV method submitted by the Applicant, for the quantification of the *GMP* and *IMP* in the *feed additive* and *water*, for concentrations above 5 mg/kg .

As no experimental data were provided by the Applicant for the determination of the product in *feedingstuffs*, the EURL could not evaluate nor recommend the method for official control to determine *GMP* and *IMP* in *feedingstuffs*.

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 the HPLC-UV method submitted by the Applicant for the identification and determination of *GMP* and *IMP* in the *feed additive and water*, for concentration levels above 5 mg/kg.

The Applicant provided no experimental data for *feedingstuffs*, therefore the EURL is unable to recommend a method for the determination of *GMP* and *IMP* in *feedingstuffs*.

Recommended text for the register entry (analytical method)

For the identification of the *disodium guanosine 5'-monophosphate* **and** *disodium inosine 5'-monophosphate* in the *feed additive* and *water*:

- JECFA monograph, Specifications for food additives: Disodium 5'-Ribonucleotides

For the determination of the *disodium guanosine 5'-monophosphate* **and** *disodium inosine 5'-monophosphate* in the *feed additive* and *water*:

- High Performance Liquid Chromatography coupled to UV detection (HPLC-UV)

5. DOCUMENTATION AND SAMPLES PROVIDED TO EURL

In accordance with the requirements of Regulation (EC) No 1831/2003, reference samples of *Disodium guanosine 5'-monophosphate* 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. 1831/0130-2010

[2] *Application, Proposal for Register Entry – Annex A

[3] *Technical dossier, Section II – Sect_II_Identity.pdf: 2.1. Identity of the additives - 2.5. Conditions of use of the additive – 2.6. Method of analysis and reference samples

[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] *Technical dossier, Section II – Annex_II_8_FAO 2006c

[6] *Technical dossier, Section II – Annex_II_9_FAO 2006d

[7] *Technical dossier, Section II – Annex_II_14_HPLC method analysis premixtures

* Refers to Dossier No. FAD-2010-0217

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:

- Centro di referenza nazionale per la sorveglianza ed il controllo degli alimenti per gli animali (CReAA), Torino (IT)