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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-2009-0056 - CRL/090028
FAD-2010-0160 - CRL/100239
FAD-2010-0186 - CRL/100237
FAD-2010-0310 - CRL/100340

Feed Additive Name: Cassia gum

Active Substance(s): --

Rapporteur Laboratory: European Union Reference Laboratory
for Feed Additives (EURL-FA)
Geel, Belgium

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Date: 23/05/2012

EXECUTIVE SUMMARY

In the current grouped application authorisation is sought under article 10(2) for *Cassia gum* under the category/functional groups 1(c), 1(d), 1(e) and 1(f) 'technological additives' / 'emulsifiers'⁽¹⁾, 'stabilisers'^(1,2), 'thickeners'^(1,3), 'gelling agents'^(1,2,3,4), according to the classification system of Annex I of Regulation (EC) No 1831/2003. Specifically, authorisation is sought for the use of the *feed additive* for cats and dogs. *Cassia gum* is already approved as *feed additive* under Directive 93/55/EEC, amending Council Directive 70/524/EEC concerning additives in *feedingstuffs*.

Cassia gum is a yellowish powder produced from purified endosperm of the seeds of *Cassia tora* and *Cassia obtusifolia* belonging to the family *Leguminosae*. The *feed additive* consists of a minimum of 75 % of galactomannan with a galactose/mannose ratio close to 1:5. The Applicants^(3,4) proposed maximum inclusion level of 1.76 % (17600 mg/kg) of *Cassia gum* in the complete *feedingstuffs*.

For the determination of *Cassia gum* in the *feed additive* Applicants^(3,4) proposed the internationally recognised FAO JECFA monographs for food additives, recommended by Commission Directive 2010/67/EU, amending the Commission Directive 2008/84/EC. Identification is based on several tests including - solubility; - gel formation with borate and xanthan gum; - viscosity; - pH; while the quantification of galactomannan is based on the "gum by difference" method. Furthermore, Applicants^(1,4) submitted additional methods based on high performance anion exchange chromatography with pulsed amperometric detection (HPAEC-PAD) and high performance liquid chromatography with refractive index detection (HPLC-RI) and reported galactose/mannose ratios ranging from 1:8.5 to 1:6.6, determined with a precision of *ca.* 6 %.

Based on the experimental evidence and the performance characteristics presented the EURL recommends for official control the methods recommended by Commission Directive 2010/67/EU and described in the FAO JECFA monographs for the identification of *Cassia gum*. Furthermore, the EURL recommends the additional methods submitted by the Applicants^(1,4), based on HPAEC-PAD or HPLC-RI - to determine the galactose/mannose ratio in the *feed additive*.

⁽¹⁾ FAD-2010-0310; ⁽²⁾ FAD-2010-0186; ⁽³⁾ FAD-2010-0160; ⁽⁴⁾ FAD-2009-0056

The Applicants provided no experimental data or any analytical methods for the determination of *Cassia gum* in *premixtures* and *feedingstuffs*, as the accurate determination of *Cassia gum* in these matrices is not achievable experimentally. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Cassia gum* in *premixtures* and *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

Cassia gum, *galactomannan*, galactose, mannose, technological additives, emulsifiers, stabilisers, thickeners, gelling agents, dogs and cats

1. BACKGROUND

In the current grouped application authorisation is sought under article 10(2) for *Cassia gum* under the category/functional groups 1(c), 1(d), 1(e) and 1(f) 'technological additives' / 'emulsifiers'⁽¹⁾, 'stabilisers'^(1,2), 'thickeners'^(1,3), 'gelling agents'^(1,2,3,4), according to the classification system of Annex I of Regulation (EC) No 1831/2003 [1-4]. Specifically, authorisation is sought for the use of the *feed additive* for cats and dogs [4-7]. *Cassia gum* is already approved as *feed additive* under Directive 93/55/EEC, amending Council Directive 70/524/EEC concerning additives in *feedingstuffs* [8].

Cassia gum is a yellowish powder produced from purified endosperm of the seeds of *Cassia tora* and *Cassia obtusifolia* belonging to the family *Leguminosae*. The *feed additive* consists of a minimum of 75 % of galactomannan with a galactose/mannose ratio close to 1:5 [9-12]. The Applicants^(3,4) proposed maximum inclusion level of 1.76 % (17600 mg/kg) of *Cassia gum* in the complete *feedingstuffs* [5,6].

Note:

This report is a joint evaluation of the information provided in a group of dossiers linked to the above mentioned four applications⁽¹⁻⁴⁾. Therefore it replaces a previously published EURL report, which covered exclusively the information from the dossier FAD 2009-0056.

⁽¹⁾ FAD-2010-0310; ⁽²⁾ FAD-2010-0186; ⁽³⁾ FAD-2010-0160; ⁽⁴⁾ FAD-2009-0056

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 *Cassia gum* 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, aflatoxin B1 and dioxins) are available from the respective European Union Reference Laboratories [13].

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

For the determination of *Cassia gum* in the *feed additive* the Applicants (FAD-2010-0160 & FAD-2009-0056) proposed the internationally recognised FAO JECFA monographs for food additives [14,15], recommended by Commission Directive 2010/67/EU, amending the Commission Directive 2008/84/EC, where identification is based on several tests including - solubility; - gel formation with borate and xanthan gum; - viscosity; - pH; and the determination of galactomannan by the "gum by difference" method [9]. The galactomannan content is calculated by subtracting the values of moisture, crude protein, crude fat, crude fiber and crude ash, from the initial weight of the *feed additive*.

The average composition of *Cassia gum* is presented in Table 1.

Table 1. Average composition of *Cassia gum*.

Analyte	Average composition [9]
galactomannan	$\geq 75\%$
moisture	$12\% \leq$
crude protein	$7\% \leq$
crude fats	$2\% \leq$
crude fiber	$4\% \leq$
crude ash	$2\% \leq$

Furthermore, Applicants (FAD-2010-0310 & FAD-2009-0056) submitted additional methods based on high performance anion exchange chromatography with pulsed amperometric detection (HPAEC-PAD) [16] and high performance liquid chromatography with refractive index detection (HPLC-RI) [17]. Both methods are based on acidic hydrolysis of the galactomannan and further determination of galactose and mannose contents using the chromatographic techniques mentioned above. The reported galactose/mannose ratios were ranging from 1:8.5 to 1:6.6 [16,17], determined with a precision of *ca.* 6 % [16].

Based on the experimental evidence and the performance characteristics presented the EURL recommends for official control the methods recommended by Commission Directive 2010/67/EU and described in the FAO JECFA monographs for the identification of *Cassia gum*. Furthermore, the EURL recommends the additional methods submitted by the Applicants (FAD-2010-0310 & FAD-2009-0056), based on HPAEC-PAD or HPLC-UV/RI - to determine the galactose/mannose ratio in the *feed additive*.

The Applicants provided no experimental data or any analytical methods for the determination of *Cassia gum* in *premixtures* and *feedingstuffs*, as the accurate determination of *Cassia gum* in these matrices is not achievable experimentally. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Cassia gum* in *premixtures* and *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 of *Cassia gum* in the *feed additive*, the analytical methods recommended by Commission Directive 2010/67/EU (amending Commission Directive 2008/84/EC) and described in FAO JECFA monographs (No. 1, Vol. 4) and '*Cassia gum*' monograph No. 10 (2010).

The Applicants provided no experimental data or any analytical methods for the determination of *Cassia gum* in *premixtures* and *feedingstuffs*, as the accurate determination of *Cassia gum* in these matrices is not achievable experimentally. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Cassia gum* in *premixtures* and *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.

Recommended text for the register entry (analytical method)

For the determination of *Cassia gum* in the *feed additive*:

- FAO JECFA Monograph No. 10 (2010) '*Cassia gum*' as referred in Commission Directive 2010/67/EU

5. DOCUMENTATION AND SAMPLES PROVIDED TO EURL

In accordance with the requirements of Regulation (EC) No 1831/2003, reference samples of *Cassia gum* 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/G/1 Forw. Appl. 1831/0056-2009
(Previous Reference - SANCO/D/2 Forw. Appl. 1831/035-2009)
- [2] +Application, Reference SANCO/G/1 Forw. Appl. 1831/0114-2011
- [3] –Application, Reference SANCO/G/1 Forw. Appl. 1831/0113-2011
- [4] ±Application, Reference SANCO/G/1 Forw. Appl. 1831/0112-2011
- [5] *Application, Proposal for Register Entry – Annex A
- [6] +Application, Proposal for Register Entry – Annex A
- [7] –Application, Proposal for Register Entry – Annex A
- [8] European Union Register of Feed Additives (pursuant to Regulation (EC) No 1831/2003)
http://ec.europa.eu/food/food/animalnutrition/feedadditives/comm_register_feed_additives_1831-03.pdf (last visited on 23/05/2012)
- [9] *Technical dossier, Section II – Identity, characterisation and conditions of use of the additive; Methods of analysis
- [10] +Technical dossier, Section II – Identity, characterisation and conditions of use of the additive; Methods of analysis
- [11] –Technical dossier, Section II – Identity, characterisation and conditions of use of the additive; Methods of analysis
- [12] ±Technical dossier, Section II – Identity, characterisation and conditions of use of the additive; Methods of analysis
- [13] 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
- [14] FAO JECFA Combined Compendium for Food Additive Specifications - *Analytical methods, test procedures and laboratory solutions used by and referenced in the food additive specifications*, Monographs No. 1, Vol. 4
<http://www.fao.org/docrep/009/a0691e/a0691e00.htm>
(last visited on 23/05/2012)
- [15] FAO JECFA Combined Compendium of Food Additive Specifications, *Cassia gum*, Monograph No. 10 (2010)
<http://www.fao.org/ag/agn/jecfa-additives/specs/monograph10/additive-513-m10.pdf> (last visited on 23/05/2012)
- [16] ±Technical dossier, Section II – Appendix 8
- [17] *Technical dossier, Section II – Annex II_10
- *Refers to Dossier No. FAD-2009-0056
+ Refers to Dossier No. FAD-2009-0160
– Refers to Dossier No. FAD-2009-0186
± Refers to Dossier No. FAD-2009-0310

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:

- Schwerpunktlabor Futtermittel des Bayerischen Landesamtes für Gesundheit und Lebensmittelsicherheit (LGL), Oberschleißheim (DE)
- Skúšobné laboratórium – Oddelenie analýzy krmív, Ústredný kontrolný a skúšobný ústav poľnohospodársky, Bratislava (SK)
- Ústřední kontrolní a zkušební ústav zemědělský (ÚKZÚZ), Praha (CZ)
- Plantedirektoratet, Laboratorium for Foder og Gødning, Lyngby (DK)
- Thüringer Landesanstalt für Landwirtschaft (TLL), Abteilung Untersuchungswesen, Jena (DE)
- Univerza v Ljubljani, Veterinarska fakulteta. Nacionalni veterinarski inštitut, Enota za patologijo prehrane in higieno okolja, Ljubljana (SI)