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Institute for Reference Materials and  
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European Union Reference Laboratory  
for Feed Additives



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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-2010-0244  
CRL/100363

Feed additive: *Bentonite-Montmorillonite*

Active Substance(s): *Bentonite-Montmorillonite*

Rapporteur  
Laboratory: European Union Reference  
Laboratory  
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Date: 16/05/2012

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## EXECUTIVE SUMMARY

In the current application authorisation is sought under articles 4(1) and 10(2) for *Bentonite-Montmorillonite* as *feed additive* under the category/functional groups 1(g) and 1(m) "technological feed additives"/"binders" and "substances for the reduction of the contamination of feed by mycotoxins" according to the classification system of Annex I of Regulation (EC) No 1831/2003. The authorisation is sought for the use for all animal species and categories.

The active substance is *Bentonite-Montmorillonite*, a fine light grey powder, originated from volcanic ash in the Scandinavian region. The main clay constituents are the mineral *Montmorillonite* (25 - 34 mass %) and *Illite* (26 - 29 mass %).

The *feed additive* is intended to be included in *premixtures* or added into *feedingstuffs*. The Applicant suggested an inclusion level of *Bentonite-Montmorillonite* in complete *feedingstuffs* ranging from 4 to 20 g/kg.

For the determination of the mineralogical and geological parameters of *Bentonite-Montmorillonite* as *feed additive* the Applicant listed several crystallographic methods, including X-Ray Diffraction (XRD), Fourier transformed infrared spectroscopy (FTIR), and the several chemical analysis methods (i.e. Inductively coupled plasma atomic emission spectroscopy ICP-AES; Inductively coupled plasma mass spectrometry (ICP-MS) ICP-MS and Atomic absorption spectroscopy (AAS). The EURL recommends for official control the crystallographic characterisation by X-ray diffraction (XRD) together with the elemental analysis by ICP-AES for the determination of *Bentonite-Montmorillonite* in the *feed additive*.

The Applicant is aware that the direct determination of *Bentonite-Montmorillonite* added to *premixtures* or *feedingstuffs* is not achievable by analysis and provided no experimental data. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Bentonite-Montmorillonite* 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

*Bentonite-Montmorillonite*, technological additives, binders, substances for the reduction of the contamination of feed by mycotoxins, all animal species and categories.

## 1. BACKGROUND

In the current application authorisation is sought under articles 4(1) and 10(2) for *Bentonite-Montmorillonite* as *feed additive* under the category/functional groups 1(g) and 1(m) "technological feed additives"/"binders" and "substances for the reduction of the contamination of feed by mycotoxins" according to the classification system of Annex I of Regulation (EC) No 1831/2003 [1,2]. The authorisation is sought for the use for all animal species and categories [2].

The active substance is *Bentonite-Montmorillonite*, a fine light grey powder, originated from volcanic ash in the Scandinavian region. The main clay constituents are the mineral *Montmorillonite* (25 - 34 mass %) and *Illite* (26 - 29 mass %) [3].

The *feed additive* is intended to be included in *premixtures* or added into *feedingstuffs*. The Applicant suggested an inclusion level of *Bentonite-Montmorillonite* in complete *feedingstuffs* ranging from 4 to 20 g/kg [2].

## 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 *Bentonite-Montmorillonite* 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*

#### *Quantitative and quantitative composition of impurities in the additive*

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

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

For the determination of the mineralogical and geological parameters of *Bentonite-Montmorillonite* as *feed additive* the Applicant listed some crystallographic methods, including X-Ray Diffraction (XRD) [5], Fourier transformed infrared spectroscopy (FTIR), and the several chemical analysis methods (i.e. Inductively coupled plasma atomic emission spectroscopy ICP-AES; Inductively coupled plasma mass spectrometry (ICP-MS) ICP-MS and Atomic absorption spectroscopy (AAS).

The Applicant provided only one set of analytical results related to chemical composition of the product: SiO<sub>2</sub> (59.5 %); Al<sub>2</sub>O<sub>3</sub> (14.9 %) and Fe<sub>2</sub>O<sub>3</sub> (6.8 %) [6]. These results are in agreement with the data obtained in the frame of characterisation study of mineralogical properties of several bentonites by Karland *et al.* [7] where the following mineralogical compositions and the elemental concentrations were reported:

- Montmorillonite (32 %); Illite (26 %) and Kaolin (11.7%) – determined using XRD; and
- SiO<sub>2</sub> (61 %); Al<sub>2</sub>O<sub>3</sub> (17.3 %) and Fe<sub>2</sub>O<sub>3</sub> (6.4 %) – determined by ICP-AES.

The Applicant provided in Section IV *in vitro* experimental data demonstrating the efficacy of the product related to the reduction of mycotoxin contamination by *Bentonite-Montmorillonite*. However, he did not suggest using this *in vitro* method for the characterisation of the *feed additive* in the frame of official control, and did not provide any validation and verification data. Therefore, the EURL could not evaluate nor recommend this method for official control.

Based on the experimental evidence provided, the EURL recommends for official control the crystallographic characterisation by X-ray diffraction (XRD) together with the elemental analysis by ICP-AES for the determination of *Bentonite-Montmorillonite* in the *feed additive*.

The Applicant is aware that the direct determination of *Bentonite-Montmorillonite* added to *premixtures* or *feedingstuffs* is not achievable by analysis and provided no experimental data. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Bentonite-Montmorillonite* 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.

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#### 4. CONCLUSIONS AND RECOMMENDATIONS

Based on the experimental evidence provided, the EURL recommends for official control the crystallographic characterisation by X-ray diffraction (XRD) together with the elemental analysis by ICP-AES for the determination of *Bentonite-Montmorillonite* in the *feed additive*.

The Applicant is aware that the direct determination of *Bentonite-Montmorillonite* added to *premixtures* or *feedingstuffs* is not achievable by analysis and provided no experimental data. Therefore the EURL cannot evaluate nor recommend any method for official control to determine *Bentonite-Montmorillonite* in *premixtures* and *feedingstuffs*.

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#### ***Recommended text for the register entry (analytical method)***

For the characterisation of the preparation *Bentonite-Montmorillonite* in the *feed additive*:

- X-Ray Diffraction (XRD) and
- Inductively coupled plasma atomic emission spectroscopy (ICP-AES)

#### 5. DOCUMENTATION AND SAMPLES PROVIDED TO EURL

In accordance with the requirements of Regulation (EC) No 1831/2003, reference samples of *Bentonite-Montmorillonite* 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. 1831/00145 (10541)-2010
  - [2] \*Application, Proposal for Register Entry – Annex A
  - [3] \*Technical dossier, Section II: 2.2 Characterisation Of The Active Substances
  - [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\_Schulze\_2001.pdf
  - [6] \* Technical dossier, Section II: Annex\_II\_Schomburg\_2005\_DURTEC.pdf
  - [7] \*Technical dossier, Section II : Annex\_II\_Karland\_2006\_SKB\_TR-06-30.pdf
- \*Refers to Dossier No. FAD-2010-0244

## 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

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- Państwowy Instytut Weterynaryjny, Puławy, PL
- Skúšobné laboratórium – Oddelenie analýzy krmív, Ústredný kontrolný a skúšobný ústav poľnohospodársky, Bratislava, SK
- Fødevarestyrelsen, Ringsted, DK
- Ústřední kontrolní a zkušební ústav zemědělský (ÚKZÚZ), Praha, CZ
- RIKILT-Instituut voor Voedselveiligheid, Wageningen, NL
- Centro di referenza nazionale per la sorveglianza ed il controllo degli alimenti per gli animali (CReAA), Torino, IT
- Schwerpunktlabor Futtermittel des Bayerischen Landesamtes für Gesundheit und Lebensmittelsicherheit (LGL), Oberschleißheim, GE
- Univerza v Ljubljani, Veterinarska fakulteta. Nacionalni veterinarski inštitut, Enota za patologijo prehrane in higieno okolja, Ljubljana, SI
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