Safety of zinc chelate of methionine sulfate for the target species

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Abstract

Zinc chelate of methionine sulfate is intended to be used as a nutritional additive (functional group: compounds of trace elements). The additive is zinc chelated with methionine in a molar ratio 1:1. It is intended to supply zinc as a nutritional additive to all animal species/categories. In 2017, the EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) adopted an opinion on the safety and efficacy of zinc chelate of methionine sulfate for all animal species. In that opinion, the Panel could not conclude on the safety of the additive for the target species. The Commission gave the applicant the possibility of submitting additional information to allow the FEEDAP Panel to complete its assessment. For this reason, additional data have been provided related to the safety of the additive for the target species and this new information is the subject of this opinion. The new tolerance study on chickens for fattening has been assessed; the results of this study indicate that the additive does not promote adverse effects in the birds as compared to an inorganic source of zinc (zinc oxide). The FEEDAP Panel concluded that the additive under assessment ‘zinc chelate of methionine sulfate’ is safe for chickens for fattening when used up to the maximum content of zinc in complete feed authorised in the EU; this conclusion is extended to all animal species.

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1. Introduction

1.1. Background and Terms of Reference as provided by the European Commission

Regulation (EC) No 1831/2003 establishes rules governing the Community authorisation of additives for animal nutrition and, in particular, Article 9 defines the terms of the authorisation by the Commission.

The applicant, Norel S.A., is seeking a Community authorisation of Zinc chelate of methionine sulfate as a feed additive to be used as a compound of trace elements for all animal species (Table 1).

| Table 1: Description of the substances |
|---------------------------------------|
| **Category of additive**              | Nutritional additive |
| **Functional group of additive**      | Compounds of trace elements |
| **Description**                       | Zinc chelate of methionine sulfate |
| **Target animal category**            | All animal species |
| **Applicant**                         | Norel, S.A. |
| **Type of request**                   | New opinion |

On 18 May 2017, the Panel on Additives and Products or Substances used in Animal Feed of the European Food Safety Authority (“Authority”), in its opinion on the safety and efficacy of the product, could not conclude on the safety of Zinc chelate of methionine sulfate as a feed additive for all animal species.

The Commission gave the possibility to the applicant to submit complementary information in order to complete the assessment and to allow a revision of Authority’s opinion. The new data have been received on 26 March 2018.

In view of the above, the Commission asks the Authority to deliver a new opinion on Zinc chelate of methionine sulfate as a feed additive for all animal species based on the additional data submitted by the applicant.

1.2. Additional information

The FEEDAP Panel, in 2017, delivered an opinion on the safety and efficacy of the zinc chelate of methionine sulfate as a nutritional additive for all animal species (EFSA FEEDAP Panel, 2017). In that opinion, the Panel concluded that the additive was safe for the consumer and the environment while safety issues for the user were identified; the additive was also considered efficacious as an available source of zinc for all animal species. However, regarding the safety for the target animals, due to the limitations of the tolerance study provided, the FEEDAP Panel was unable to conclude.

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of additional information1 to a previous application of the same product.2

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety of zinc chelate of methionine sulfate is in line with the principles laid down in Regulation (EC) No 429/2008 and the relevant guidance documents: Guidance on nutritional additives (EFSA FEEDAP Panel, 2012) and Technical guidance: Tolerance and efficacy studies in target animals (EFSA FEEDAP Panel, 2011).

3. Assessment

The additive under application, ‘zinc chelate of methionine sulfate’, is zinc chelated with methionine in a molar ratio 1:1. It is intended to supply zinc as a nutritional additive for all animal species/categories up

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1 FEED dossier reference: FAD-2018-0014.
2 FEED dossier reference: FAD-2015-0025.
to a maximum total content in complete feedingstuffs of: 200 mg Zn/kg for dogs and cats, 180 mg Zn/kg for salmonids and milk replacers for calves, 150 mg Zn/kg for piglets, sows, rabbits and all fish species other than salmonids, and 120 mg Zn/kg for other species and categories. These values are in agreement with the maximum total content of zinc in feed set in Commission implementing Regulation (EU) 2016/1095.

A formulated additive is intended to be manufactured and marketed as BIOMET® Zn 15%, 10% and 2%, with a zinc concentration of 15%, 10% and 2%, respectively; calcium carbonate is used as an excipient in the formulation.

The FEEDAP Panel has delivered an opinion on the additive under assessment (EFSA FEEDAP Panel et al., 2017). In the current application opinion, the applicant has submitted a new tolerance study to support the safety of the additive for the target species and this information is the subject of this opinion.

### 3.1. Safety for the target species

A total of 288 one-day-old male chickens for fattening (Ross 308) were housed in cages of six birds per pen and randomly distributed to the experimental treatments. The experimental design was completely randomised with two zinc sources (zinc chelate of methionine sulphate 'BIOMET® Zn 15%' and zinc oxide 'ZnO') and three zinc levels in feed (40 (lowest requirement level), 120 (highest recommended dose) and 500 mg/kg feed (overdose, ×4.2)), organised in six dietary treatments: T1, BIOMET® Zn (40 mg/kg feed); T2, BIOMET® Zn (120 mg/kg feed); T3, BIOMET® Zn (500 mg/kg); T4, ZnO (40 mg/kg feed); T5, ZnO (120 mg/kg feed) and T6, ZnO (500 mg/kg feed). Treatments were replicated eight times. The study lasted 35 days.

Mash feeds were fed ad libitum, and were based on wheat and soybean meal. Background zinc concentrations were analysed in the basal diet (30 mg/kg); the different treatment diets were prepared by adding the zinc sources (BIOMET® Zn or ZnO) up to the intended experimental zinc concentration. Treatments T1, T2 and T3 were adjusted in formulation by the total methionine content of the BIOMET® Zn added to the diets. The intended zinc concentrations in the diets were confirmed by analysis (see Table 2). Starter feeds were fed from 0 to 21 days and grower feeds from 22 to 35 days of age. General health and mortality were recorded. Feed consumption and body weight were measured on days 21 and 35. At the end of the study, blood was obtained from two birds per pen (n = 96, i.e. 16/treatment) and analysed for blood chemistry and haematology parameters. At the same time, one bird per pen (n = 48, i.e. 8/treatment) was killed and the following organs and tissues were examined grossly (including weight of the organs) and preserved for microscopic evaluation (in case of findings in the gross pathology were found): liver, kidneys, spleen, bursa of Fabricius, lungs, proventriculus, gizzard, pancreas, small intestine, caecum, thymus, heart, ovaries/testes. Samples of pancreas and tibia were also collected for zinc analysis.

The statistical unit was the pen for animal performance and the individual bird for blood and tissue deposition. An analysis of variance (ANOVA) was applied to the data, considering as the main effects treatment alone (one-way ANOVA) or zinc source and zinc level (two-way ANOVA). Group means were compared with Tukey test. Statistical significance was considered at p ≤ 0.05.

Overall, the health of the animals was considered normal throughout the study, and no adverse events were noted. Mortality was low and not related to treatment (Table 2). No significant differences between treatments were observed in the body weight of birds and in any of the zootechnical performance parameters (Table 2); neither zinc source and zinc dose, nor their interaction were significant in the analysis.

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3 Commission Implementing Regulation (EU) 2016/1095 of 6 July 2016 concerning the authorisation of Zinc acetate dihydrate, Zinc chloride anhydrous, Zinc oxide, Zinc sulphate heptahydrate, Zinc sulphate monohydrate, Zinc chelate of amino acids hydrate, Zinc chelate of protein hydrolysates, Zinc chelate of glycine hydrate (solid) and Zinc chelate of glycine hydrate (liquid) as feed additives for all animal species and amending Regulations (EC) No 1334/2003, (EC) No 479/2006, (EU) No 335/2010 and Implementing Regulations (EU) No 991/2012 and (EU) No 636/2013. OJ L 182, 7.7.2016, p. 7.

4 Technical Dossier/Annex 05_NEW_Annex_III_01_Annex_IV_02

5 Sodium, potassium, chloride, calcium, phosphate, magnesium, total protein, albumin, globulin, glucose, urea/uric acid, cholesterol, creatinine, bilirubin, acute phase proteins, amylase, alanine aminotransferase, aspartate aminotransferase, lactate dehydrogenase, gamma-glutamyl transferase, alkaline phosphatase and creatine kinase.

6 Total count for red blood cells, haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, total and differential counts for leucocytes, prothrombin time and fibrinogen.
Similar to the performance parameters, no significant differences between treatments were observed in any of the haematology parameters, metabolites, blood enzymes or electrolytes analysed. Furthermore, no significant differences between treatments were observed for the weight of any of the organs analysed.

Significant differences between treatments were observed in zinc deposition in pancreas and tibia (Table 3). In both tissues zinc deposition showed a linear dose-related increase; however, statistical significant differences were only observed for zinc concentrations in the pancreas between broilers fed at 500 mg Zn from BIOMET®Zn/kg and those fed at 40 mg Zn from ZnO/kg, and for zinc tibia concentrations between broilers fed at 500 mg/kg Zn with ZnO and those fed at 40 mg/kg Zn with ZnO. When applying a two-way ANOVA, no significant differences in zinc deposition were observed between sources (BIOMET®Zn and ZnO), or for the interaction zinc source-zinc level.7

The tolerance study indicates that zinc chelate of methionine sulfate is safe for chickens for fattening. The study has been performed according to the established requirements to allow the extension of the conclusion from chickens for fattening to all animal species.

### 4. Conclusions

Based on a tolerance study, the FEEDAP Panel concludes that the additive under assessment ‘Zinc chelate of methionine sulfate’ is safe for chickens for fattening when used up to the maximum content of zinc in complete feed authorised in the EU (i.e. 120 mg Zn/kg complete poultry feed). This conclusion is extended to all animal species when the additive is used in feed at the respective authorised zinc maximum levels: 200 mg Zn/kg for dogs and cats, 180 mg Zn/kg for salmonids and milk replacers for calves, 150 mg Zn/kg for piglets, sows, rabbits and all fish species other than salmonids, and 120 mg Zn/kg for other species and categories.

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**Table 2:** Effects of various dosages of zinc chelate of methionine sulfate and zinc oxide on performance of chickens (35 days)

| Zinc source | Zinc chelate of methionine sulfate | Zinc oxide |
|-------------|----------------------------------|------------|
| Zinc content, intended (1) | T1  | T2  | T3  | T4  | T5  | T6  |
| 40          | 120 | 500 | 51  | 114 | 433 |
| 51          | 137 | 518 | 43  | 120 | 470 |
| Mortality (%) | 2.1 | 2.1 | 2.1 | 0.0 | 0.0 | 2.1 |
| Body weight (g) | 1901 | 1952 | 1926 | 1962 | 1950 | 2000 |
| Feed intake (g/day) | 76.4 | 75.9 | 75.6 | 75.7 | 76.7 | 77.8 |
| Daily weight gain (g) | 54.5 | 56.0 | 55.2 | 56.3 | 55.9 | 57.4 |
| Feed to gain ratio | 1.40 | 1.36 | 1.37 | 1.35 | 1.37 | 1.36 |

(1), (2): (mg/kg). Background zinc concentrations were analysed in the basal diet: 30 mg/kg.

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**Table 3:** Effects of various dosages of zinc chelate of methionine sulfate and zinc oxide on zinc content in pancreas and tibia (mg/kg fresh matter) in chicken for fattening (35 days; eight birds per treatment)

| Zinc source | Zinc chelate of methionine sulfate | Zinc oxide |
|-------------|----------------------------------|------------|
| Zinc content, intended (1) | T1  | T2  | T3  | T4  | T5  | T6  |
| 40          | 120 | 500 | 51  | 114 | 433 |
| 51          | 137 | 518 | 43  | 120 | 470 |
| Mortality (%) | 2.1 | 2.1 | 2.1 | 0.0 | 0.0 | 2.1 |
| Body weight (g) | 1901 | 1952 | 1926 | 1962 | 1950 | 2000 |
| Feed intake (g/day) | 76.4 | 75.9 | 75.6 | 75.7 | 76.7 | 77.8 |
| Daily weight gain (g) | 54.5 | 56.0 | 55.2 | 56.3 | 55.9 | 57.4 |
| Feed to gain ratio | 1.40 | 1.36 | 1.37 | 1.35 | 1.37 | 1.36 |

(1): (mg/kg). Background zinc concentrations were analysed in the basal diet: 30 mg/kg.

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a,b,c: Different superscripts within a given row indicate statistical differences (\(P<0.05\)).

The tolerance study indicates that zinc chelate of methionine sulfate is safe for chickens for fattening. The study has been performed according to the established requirements to allow the extension of the conclusion from chickens for fattening to all animal species.

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7 Technical dossier/05_NEW_Annex_III_01_Annex_IV_02.
**Documentation provided to EFSA**

Zinc chelate of methionine sulfate. March 2018. Submitted by Norel S.A.

**Chronology**

| Date       | Event                                                                                       |
|------------|---------------------------------------------------------------------------------------------|
| 03/04/2018 | Dossier received by EFSA                                                                    |
| 16/04/2018 | Reception mandate from the European Commission                                             |
| 19/04/2018 | Application validated by EFSA                                                               |
| 04/10/2018 | Opinion adopted by the FEEDAP Panel. End of the Scientific assessment                      |

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**Abbreviations**

ANOVA    analysis of variance  
FEEDAP   EFSA Panel on Additives and Products or Substances used in Animal Feed