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Safety and efficacy of a feed additive consisting of manganese(II)-betaine complex for all animal species (Biochem Zusatzstoffe Handels- und Produktionsges. mbH)

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Abstract

Following a request from the European Commission, EFSA was asked to deliver a scientific opinion on the safety and efficacy of manganese(II)-betaine complex as a nutritional feed additive for all animal species and categories. Based on the tolerance study performed in chickens, the FEEDAP Panel concluded that the additive is safe for chickens for fattening when used up to the maximum proposed level of manganese in feed; this conclusion was extrapolated to all animal species and categories at the corresponding maximum manganese levels in complete feed (100 mg Mn/kg feed for fish and 150 mg Mn/kg feed for all other animal species). The FEEDAP Panel concluded that the use of additive in animal nutrition does not represent a concern for consumer safety or for the environment. The additive is irritant to the eyes, but not irritant to skin. Owing to the presence of nickel the additive is considered to be a dermal and respiratory sensitiser. The Panel concluded that the additive is efficacious as a nutritional additive for all animal species under the proposed conditions of use.

Keywords: manganese(II)-betaine, manganese, nutritional additive, feed additive, complex

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1. Introduction

1.1. Background and Terms of Reference

Regulation (EC) No 1831/2003[1] establishes the rules governing the Community authorisation of additives for use in animal nutrition. In particular, Article 4(1) of that Regulation lays down that any person seeking authorisation for a feed additive or for a new use of feed additive shall submit an application in accordance with Article 7.

The European Commission received a request from Biochem Zusatzstoffe Handels- und Produktionsges. mbH[2] for the authorisation of the additive consisting of manganese(II)-betaine complex, when used as a feed additive for all animal species (category: nutritional additives; functional group: compounds of trace elements).

According to Article 7(1) of Regulation (EC) No 1831/2003, the Commission forwarded the application to the European Food Safety Authority (EFSA) as an application under Article 4(1) (authorisation of a feed additive or new use of a feed additive). EFSA received directly from the applicant the technical dossier in support of this application. The particulars and documents in support of the application were considered valid by EFSA as of 18 January 2023.

According to Article 8 of Regulation (EC) No 1831/2003, EFSA, after verifying the particulars and documents submitted by the applicant, shall undertake an assessment in order to determine whether the feed additive complies with the conditions laid down in Article 5. EFSA shall deliver an opinion on the safety for the target animals, consumers, user and the environment and on the efficacy of the feed additive consisting of manganese(II)-betaine complex, when used under the proposed conditions of use (see **Section 3.1.5**).

1.2. Additional information

The additive, manganese(II)-betaine complex is intended to be used as a source of manganese in all animal species. It has not been previously authorised as a feed additive in the European Union.

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of a technical dossier[3] in support of the authorisation request for the use of manganese(II)-betaine complex as a feed additive. The dossier was received on 13 September 2022 and the general information and supporting documentation is available at https://open.efsa.europa.eu/questions/EFSA-Q-2022-00556

The confidential version of the technical dossier was subject to a target consultation of the interested Member States from 23 January 2023 to 23 April 2023 for which received comments were considered for the assessment.

In accordance with Article 38 of the Regulation (EC) No 178/2002[4] and taking into account the protection of confidential information and of personal data in accordance with Articles 39 to 39e of the same Regulation, and of the Decision of EFSA's Executive Director laying down practical arrangements concerning transparency and confidentiality,[5] a non-confidential version of the dossier has been published on Open.EFSA.[6]

The FEEDAP Panel used the data provided by the applicant together with data from other sources, such as previous risk assessments by EFSA or other expert bodies, peer-reviewed scientific papers, other scientific reports and experts' knowledge, to deliver the present output.

[1] Regulation (EC) No 1831/2003 of the European Parliament and of the council of 22 September 2003 on the additives for use in animal nutrition. OJ L 268, 18.10.2003, p. 29.
[2] Biochem Zusatzstoffe Handels- und Produktionsges. mbH, Küstermeyerstr. 16, 49393 Lohne, Germany.
[3] Dossier reference: FEED-2022-4790.
[4] Regulation (EC) No 178/2002 of the European Parliament and of the Council of 28 January 2002 laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety. OJ L 31, 1.2.2002, p. 1–48.
[5] Decision available online: https://www.efsa.europa.eu/en/corporate-pubs/transparency-regulation-practical-arrangements
[6] Available online: https://open.efsa.europa.eu/questions/EFSA-Q-2022-00556

EFSA has verified the European Union Reference Laboratory (EURL) report as it relates to the methods used for the control of the manganese(II)-betaine complex in animal feed.[7]

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of manganese (II)-betaine complex is in line with the principles laid down in Regulation (EC) No 429/2008[8] and the relevant guidance documents: Guidance on studies concerning the safety of use of the additive for users/workers (EFSA FEEDAP Panel, 2012), Guidance on the assessment of the safety of feed additives for the consumer (EFSA FEEDAP Panel, 2017a), Guidance on the identity, characterisation and conditions of use of feed additives (EFSA FEEDAP Panel, 2017b), Guidance on the assessment of the safety of feed additives for the target species (EFSA FEEDAP Panel, 2017c), Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018), Guidance on the assessment of the safety of feed additives for the environment (EFSA FEEDAP Panel, 2019).

3. Assessment

The additive under assessment is manganese(II)-betaine complex. The additive corresponds to the active compound. The additive manganese(II)-betaine complex is intended to be used as a nutritional additive (functional group: compounds of trace elements) in feed for all animal species.

3.1. Characterisation

3.1.1. Characterisation of the additive

The additive is a complex defined by the applicant with a chemical formula $[Mn(H_2O)_2((CH_3)_3NCH_2COO)(SO_4)]_n$, corresponding to a molecular weight of 304.15 g/mol for the repeat unit. International Union of Pure and Applied Chemistry (IUPAC) chemical name is catena-[μ3-sulfato-(trimethylammonio)acetato-manganese(II)]; Chemical Abstracts Service (CAS) number or EC number are unavailable.

Proof of complex formation between manganese, betaine and sulfate was provided by using X-ray powder diffraction.[9] X-ray patterns of the starting materials [redacted] and of one batch of manganese(II)-betaine complex were measured. Results showed that the starting materials were no longer present, and that the conversion into the new product appeared to be complete.

The Panel notes that the data (single crystal X-ray analyses) indicate that the additive is a coordination polymer, [redacted]

By specification, manganese(II)-betaine complex contains minimum 17% of manganese, minimum 42% of betaine, sulfur content is 9–12% (equivalent to 27–36% of sulfate[11]) and a maximum 5% of moisture.[12]

Five independent batches[13] of the additive from the small-scale production were analysed for manganese, sulfur and betaine content. Content of manganese was on average 19% (range from 17.6% to 19.6%), betaine 42.7% (range from 42.5% to 43.1%), sulfur 11.4%% (range from 10.9% to 11.8%; expressed as sulfate 32.7% to 35.3%) and moisture from 0.7% to 1.9% which confirms the specification. Crystallised water[14] ranged from 1.43% to 3.19%. Content of manganese, sulfate,

[7] Evaluation report received on 27/03/2023 and available on the EU Science Hub https://joint-research-centre.ec.europa.eu/eurl-fa-eurl-feed-additives/eurl-fa-authorisation/eurl-fa-evaluation-reports_en

[8] Commission Regulation (EC) No 429/2008 of 25 April 2008 on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the preparation and the presentation of applications and the assessment and the authorisation of feed additives. OJ L 133, 22.5.2008, p. 1.

[9] Annex_II_12.

[10] Annex II_11.

[11] Sulfate content (%) is recalculated from analytically determined sulfur percentage by using a multiplication factor 2.996.

[12] Moisture content determined after 4 h at 103°C.

[13] Annex II_1 and Annex II_3. Same batches were tested for manganese and sulfur in two different laboratories.

[14] Annex II_2: crystalized water determined at 190°C.

betaine, crystalised water, zinc[15] and ashes insoluble in hydrochloric acid, all summed up to 99.52% on average.

Three batches of the additive were analysed for impurities.[16] Content of lead, cadmium, mercury, arsenic and fluorine (detected as fluoride) were all below the corresponding limits of quantification (LOQ).[17] Nickel[18] was on average 18.4 mg/kg (range 18.0–18.7 mg/kg), which was in line with the manufacturer's specification for Ni < 100 mg/kg.

The levels of polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/F) and the sum of PCDD/F and dioxin-like polychlorinated biphenyls (PCBs) were 0.16 ng WHO-PCDD/F-TEQ/kg and 0.30 ng WHO-PCDD/F-PCB-TEQ/kg, respectively, in all three batches tested. Non-dioxin-like PCBs (upper-bound) were 1.7 μg/kg in all three batches.[19]

Levels of the mycotoxins aflatoxin B1 and ochratoxin A analysed in three batches were below the LOQ.[20]

Three batches of the additive were analysed for microbiological contamination.[21] *Salmonella* spp. was not detected in 25 g, and Enterobacteriaceae and *Escherichia coli* were < 100 colony forming units (CFU)/g and < 10 CFU/g, respectively.

The FEEDAP Panel considers that the microbial contamination and the amounts of the detected impurities do not raise safety concerns, except for nickel which is further addressed in Section 3.2.3.

3.1.2. Physical properties of the additive

The additive appears as a white to slightly pink granular product with a bulk density[22] of 822 kg/m^3 (average from three batches).

Solubility[23] in water (20°C) was reported to be in a range 100–105 g/L. Considering the solubility is above 33.3 g/L, there is no need to further characterise the fraction of small particles/nanoparticles potentially present in the additive (EFSA Scientific Committee, 2021). The additive is insoluble in organic solvents.

Dusting potential[22] of the additive was determined by the Stauber-Heubach method in three batches and was in a range of 320–600 mg/m^3 (mg airborne dust per m^3 of air). The average content of manganese[24] in dust samples separated during testing of dusting potential was from 14.7% to 16.9%.

Particle size distribution[25] of the additive was tested in three batches by using the laser-diffraction method. The results showed that, in the batches tested, 10% of the particles had an average diameter smaller than 380 μm (range: 365–393 μm), 50% a diameter < 630 μm (range: 616–643 μm) and 90% a diameter < 912 μm (range: 905–922 μm), and that no particles below 100 μm were present.

3.1.3. Manufacturing process

Full details of the manufacturing process for manganese(II)-betaine complex are described in the dossier.[26] [redacted]

[15] Annex II_1 Residues of zinc may be present due to the cross-contamination from previous small-scale production containing zinc salts. Zinc ranged from 0.23% to 0.74%.
[16] Annex_II_1.
[17] LOQ(Pb) = 0.5 mg/lg; LOQ(Cd) = 0.2 mg/kg; LOQ(Hg) = 0.02 mg/kg and LOQ(As) = 0.5 mg/kg; LOD(F) = 40 mg/kg.
[18] Annex II_6.
[19] Annex II_5.
[20] LOQ (Aflatoxin B1) = 0.3 μg/kg; LOQ (Ochratoxin A) =0.5 μg/kg.
[21] Annex II_4.
[22] Annex II_7.
[23] 2023-02-24_ADR(1)/Annex II.
[24] Annex II_9.
[25] Annex II_8.
[26] 2.3 Manufacturing process.

3.1.4. Stability and homogeneity

Stability studies are normally not required for mineral-based additives.

The capacity for homogeneous distribution of the additive in mash and pelleted feed for chickens for fattening was studied in 10 subsamples.[27] Average content of manganese in the mash feed was 93.3 mg/kg and in pelleted feed was 96.14 mg/kg (intended 100 mg Mn/kg); corresponding coefficients of variations (CV) were 9.12% and 5.24%.

3.1.5. Conditions of use

The additive is intended to be used in feed – directly or via a premixture or complementary feed – for all animal species up to a maximum authorised total manganese content in complete feed[28]: 100 mg Mn/kg complete feed for fish and 150 mg Mn/kg complete feed for all other animal species.

3.2. Safety

The Panel notes that the additive is a coordination polymer formed by manganese, sulfate and betaine. Based on the data available uncertainty remains on the size of the coordination polymer. Although there is uncertainty on how the size of the molecule would affect the dissociation rate, considering the nature of the coordination bonds between manganese(II), betaine and sulfate ions, the FEEDAP Panel considers it likely that, under physiological conditions in the animal gastrointestinal tract, the additive dissociates into its components.

The safety of betaine and manganese sulfate when used as feed additives has been previously assessed and established by the FEEDAP Panel (EFSA FEEDAP, 2013, 2016).

3.2.1. Safety for the target species

The applicant provided a combined tolerance and efficacy study in chickens for fattening to support the safety for the target animals.

3.2.1.1. Safety for the chickens for fattening

A total of 840 one-day-old male chickens for fattening (Ross 308) were distributed in 42 pens in groups of 20 chickens and allocated to seven dietary treatments (six replicates per treatment).[29] Three basal diets (starter from d1 to d10, grower from d11 to d21 and finisher from d22 to d35), based on maize and soyabean meal, were either not supplemented (control) or supplemented with manganese sulfate or with manganese-betaine complex, to provide 100, 150 (1× maximum recommended level) or 900 (6×) mg Mn/kg feed (confirmed by analysis).[30] Manganese content (intended and analysed) in experimental diets for each treatment is provided in Table 1.

Table 1: Manganese content in the experimental diets offered to the different experimental groups

Treatment	Source of manganese	Total Mn (mg/kg)			
		Intended	Analysed		
			Starter	Grower	Finisher
T1	None	–	47	44	42
T2	$MnSO4 \times H_2O$	100	106	100	106
T3		150	148	144	150
T4		900	1,052	926	972

[27] Annex II_15 and 16.

[28] Commission Implementing Regulation (EU) 2017/1490 of 21 August 2017 concerning the authorisation of manganous chloride tetrahydrate, manganese (II) oxide, manganous sulfate monohydrate, manganese chelate of amino acids hydrate, manganese chelate of protein hydrolysates, manganese chelate of glycine hydrate and dimanganese chloride trihydroxide as feed additives for all animal species.

[29] Annex III_4a and Annex III_4b.

[30] Annex III_4e Appendix 6.

Treatment	Source of manganese	Total Mn (mg/kg)			
		Intended	Analysed		
			Starter	Grower	Finisher
T5	Mn-betaine complex	100	98	105	77
T6		150	144	166	128
T7		900	1155	890	905

Mn: manganese.

Diets, in mash form and drinking water were offered on *ad libitum* basis for a total of 35 days. Mortality and health status were checked daily. Chickens and feed were weighed at start and on days 21 and 35 on a pen basis, and average daily weight gain, average daily feed intake and feed to gain ratio were calculated. All the performance parameters were corrected for mortality. At the end of the study (day 35), two birds per pen were randomly selected (12 per group), blood sampled, necropsied and organs/tissues[31] collected. The blood was subjected for haematology[32] and biochemistry analysis.[33] The organs were weighed and evaluated for gross pathology. The breast, liver, kidney, skin and abdominal fat of one bird per pen (six birds per group) were analysed for the content of manganese (see section 3.2.2.1).[34]

Data[35] were statistically analysed via the generalised linear model (GLM), with the pen as the statistical unit and the diet as fixed effect. Group means were compared with Tukey's test.

Overall mortality was low (1.67%), and not treatment related. The results on the zootechnical performance are presented in Table 2. No significant differences among treatments were observed for organ weights. There were no differences in blood haematology and biochemistry parameters among groups. As the gross pathology evaluation did not show any relevant findings, no microscopic examination of organs was carried out.

Table 2: Effect of manganese-betaine complex on performance parameters and mortality in chickens for fattening after 35 days

Treatment	Source of added manganese	Total manganese intended (mg/kg diet)	Average daily feed intake (g/bird)	Final body weight (g)	Feed to gain ratio	Mortality and culling (%)
T1	None	–	94.0^{ab}	$2{,}328^{a}$	1.45^{b}	1.7
T2		100	93.2^{ab}	$2{,}260^{ab}$	1.49^{ab}	2.5
T3	$MnSO_4 \times H_2O$	150	94.9^{ab}	$2{,}302^{ab}$	1.48^{ab}	1.7
T4		900	96.0^{a}	$2{,}235^{ab}$	1.54^{a}	0.8
T5		100	93.9^{ab}	$2{,}273^{ab}$	1.48^{ab}	0.8
T6	Mn-betaine	150	91.2^{b}	$2{,}286^{ab}$	1.44^{b}	1.7
T7		900	90.3^{b}	$2{,}214^{b}$	1.48^{ab}	2.5

Mn: manganese.
[a,b]: Means in a column not sharing a common letter are statistically different ($p < 0.05$).

At the maximum authorised level of manganese in feed (150 mg Mn/kg feed) from both manganese sources, no significant differences were observed for feed to gain ratio in comparison to the control group. At the overdose of $MnSO_4$, there was a significant increase in feed to gain ratio compared to the control group. There was a significant decrease of final body weight at the 6×

[31] Liver, kidneys, spleen, adrenal gland, lung, stomach, pancreas, small intestine, colon, caecum, thymus, thyroid gland, heart, intestinal lymph nodes, testes.
[32] Total red blood cell count, packed cell volume, haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, total & differential counts for leukocytes, platelet counts, prothrombin time & fibrinogen, sodium, potassium, chloride, calcium, phosphate, magnesium, total protein, albumin, globulin, glucose, urea/uric acid, cholesterol, creatinine, bilirubin, acute phase proteins (ovotransferrin & C-reactive protein), amylase, alanine aminotransferase, aspartate aminotransferase, lactate dehydrogenase, gamma-glutamyltransferase, alkaline phosphatase and creatine kinase).
[33] Annex_III_4.d_Appendix 6.
[34] Annex III_4.f_Appendix 6.
[35] Annex III_4g_Appendix 7.

overdose group (900 mg Mn/kg feed) for manganese-betaine compared to the control group. This decrease of the final body weight at the overdose level with manganese-betaine is accompanied by a numerical (although not statistically different) decrease in average daily intake; the feed to gain ratio was not influenced by these reductions. Therefore, the FEEDAP Panel does not consider it as an adverse effect.

3.2.1.2. Conclusions on safety for the target species

Based on the results of the tolerance study, the FEEDAP Panel concludes that the additive is safe for chickens for fattening when used up to the maximum authorised levels of manganese in feed. This conclusion can be extrapolated to all animal species and categories provided that the maximum authorised levels in the EU for total manganese in feed are not exceeded: 100 mg Mn/kg complete feed for fish and 150 mg Mn/kg complete feed for all other animal species.

3.2.2. Safety for the consumer

The sources used for the manufacturing of the manganese(II)-betaine complex, are authorised in the EU as feed additives and their safety has been established. The complex is expected to dissociate under physiological conditions into betaine, manganese(II) and sulfate ions. Therefore, the FEEDAP Panel retains that only exposure to manganese is of interest concerning the consumer safety.

Metabolism, deposition and toxicology of manganese have been reviewed in previous FEEDAP opinions (EFSA FEEDAP Panel, 2016, 2020). In those assessments, the Panel concluded that the use of the manganese compounds in animal nutrition is of no concern for the safety of consumers, provided that the current authorised maximum total contents of manganese in feed are respected (100 mg total Mn/kg for fish and 150 mg total Mn/kg for other species).

3.2.2.1. Deposition study

The tolerance study (see Section 3.2.1.1) provided data on manganese deposition in tissues and organs (kidney, liver, breast muscle, skin and fat) of chickens for fattening supplemented with either manganese sulfate or manganese-betaine complex (Table 3). Tissue samples were taken on day 35 of age (one bird per replicate).

Table 3: Total manganese content in edible tissue (reported as fresh matter,[36] one bird per replicate)

Treatment	Manganese source	Mn in feed intended (mg/kg)	Kidney (mg/kg)	Liver (mg/kg)	Breast muscle (mg/kg)	Skin (mg/kg)	Abdominal fat (mg/kg)
T1	None	–	1.8^{c}	2.2^{c}	< LOD	1.2^{b}	< LOD
T2	$MnSO_4 \times H_2O$	100	2.2^{c}	2.5^{bc}	< LOD	1.9^{b}	< LOD
T3		150	2.3^{c}	2.6^{bc}	< LOD	4.0^{b}	< LOD
T4		900	3.9^{ab}	3.4^{ab}	< LOD	13.3^{ab}	< LOD
T5	Mn-betaine complex	100	2.6^{c}	2.8^{bc}	< LOD	6.6^{b}	< LOD
T6		150	2.8^{bc}	2.8^{bc}	< LOD	4.1^{b}	< LOD
T7		900	4.2^{a}	4.5^{a}	< LOD	24.7^{a}	< LOD

Mn: manganese.
^{a,b,c}: Means in a column not sharing the same superscript are statistically different ($p < 0.05$); LOD = 1 mg Mn/kg.

Manganese deposition in breast muscle and abdominal fat was below the LOD in all treatments. No differences were observed in the deposition of manganese in kidney, liver and skin among the treatments supplemented with manganese at 100 or 150 mg/kg (regardless of the source) and the unsupplemented control. The results obtained are in line with the FEEDAP Panel's observation from the review of the residue and metabolic studies of different manganese compounds (EFSA FEEDAP Panel, 2016), that the muscle tissue is not an important deposition pathway for manganese, irrespective of the supplementation source (inorganic or organic).

Based on the submitted residue data the Panel concludes that the use of the additive in animal nutrition at proposed use level will not increase the background manganese levels in animal edible

[36] 2023-07-20_ADR(3).

tissue, and therefore the new manganese source manganese(II)-betaine complex will not significantly increase the exposure of consumers to manganese via animal tissues.

3.2.2.2. Conclusions on safety for the consumer

The FEEDAP Panel considers that the use of additive in feed for animal species at the proposed use levels is safe for the consumer provided that the maximum authorised levels in the EU for total manganese in feed are not exceeded.

3.2.3. Safety for the user

3.2.3.1. Effect on respiratory system

No specific inhalation toxicity studies for the additive under assessment were provided by the applicant. The highest measured dusting potential[37] of manganese(II)-betaine complex was 600 mg/m^3, therefore the FEEDAP Panel considers that the exposure through inhalation is likely. With a maximum concentration of 16.92% manganese in the dust, 101.5 mg Mn/m^3 may be released by the dust during handling of the additive. No data on the particle size of the dust was available, therefore the Panel cannot estimate exposure to respirable particles and compare it to the threshold limit value (TLV) for manganese of 0.02 mg/m^3 (ACGIH, 2012).

The nickel content in the additive per specification must be below 100 mg/kg. Considering a dusting potential of 600 mg/m^3, the highest level of nickel in the additive (100 mg Ni/kg, as per specifications) and assuming a similar proportion of nickel in the dust as in the additive, the nickel content in the dust would be up to 0.06 mg Ni/m^3. This value would not exceed the transitional limit value of 0.1 mg Ni/m^3 for the inhalable fraction and 8 h time-weighed average (8 h TWA) exposure established in Directive (EU) 2022/431[38]. However, owing to the presence of nickel, the additive is considered to be a respiratory sensitiser.

3.2.3.2. Effect on skin and eyes

The skin irritation/corrosion potential of the additive was investigated in a skin irritation study performed according to the OECD Testing Guideline (TG) 404.[39] The results of this study demonstrated that the additive is not a skin irritant.

The eye irritation/corrosion potential of the additive was investigated in an eye irritation study performed according to OECD TG 405.[40] Reversible signs of eye irritation were observed within 21 days, but the effects were not completely reversible after 7 days. The results of this study demonstrated that the additive is classified as an eye irritant (GHS category 2A)'.

No skin sensitisation studies were submitted. However, due to the presence of nickel in the additive (18.7 mg/kg), and given its well-known sensitisation potential, the additive should be considered a dermal sensitiser.

3.2.3.3. Conclusions on safety for the user

The FEEDAP Panel considers that exposure by inhalation is likely. Due to the presence of nickel, the additive is considered a dermal and respiratory sensitiser. The additive is irritant to the eyes, but not to skin.

3.2.4. Safety for the environment

The additive under assessment is a manganese(II)-betaine complex and contains manganese, betaine and sulfate. The environmental risk assessment is conducted with the assumption that the whole amount of the additive ingested by the animals is excreted.

Betaine is a physiological and natural component in animals and plants. For the betaine moiety, the FEEDAP Panel concluded that it is not of concern, since it is naturally present in the environment and its safety has already been assessed (EFSA FEEDAP Panel, 2013).

[37] Annex III_5.
[38] DIRECTIVE (EU) 2022/431 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 9 March 2022 amending Directive2004/37/EC on the protection of workers from the risks related to exposure to carcinogens or mutagens at work, 16.3.2022,OJ L88 pp.1–14. The limit value of 0.05 mg/m^3 for the inhalable fraction, measured as nickel, shall apply from 18 January 2025. Until then a limit value of 0,1 mg/m^3 shall apply.
[39] Annex III_9.
[40] Annex III_10.

Sulfates are naturally abundant in the environment and the use of the additive will not result in a substantial change in the natural background concentration under the current conditions of use.[41]

To assess the environmental risk of manganese from the additive, the Panel compared the worst-case predicted environmental concentrations (PECs),[42] calculated at the maximum authorised level of manganese, with the natural background concentration considered as the 90th percentile value from the FOREGS database.[43] If the PECs fall below 10% of this value, no further risk assessment is needed. The determination of natural background concentration for metals in water is described in the Guidance for implementing environmental quality standards (EQSs) for metals, as outlined in the Water Framework Directive implementation strategy.[44]

The 90th percentile concentration of manganese in topsoil obtained from 845 soil samples collected throughout Europe for the FOREGS survey was 1,200 mg Mn/kg dry weight (dw).

The 90th percentile concentration of manganese in freshwater obtained from 804 water samples collected throughout Europe for the FOREGS survey was 132 μg Mn/L.

3.2.4.1. Environmental safety from use in feeds for terrestrial farm animals

The worst-case PEC_{soil}[45] calculated using the additive up to the maximum authorised level of manganese in feed (150 mg Mn/kg dw) is 3.02 mg Mn/kg dw soil for pigs for fattening, while for the other species it ranged from 1.48 to 2.55 mg Mn/kg dw soil. This is below the 10% of the natural background concentration of manganese in topsoil (120 mg Mn/kg dw). No concern is expected for terrestrial compartment for all animal species at proposed conditions of use.

3.2.4.2. Environmental safety from use in aquaculture feeds

Calculating the predicted environmental concentration for surface water (PECswaq) for use of the additive manganese(II)-betaine complex at the maximum permitted level of 100 mg Mn/kg feed (for fish) results in a PECswaq of 0.12 μg/L to 0.25 μg/L (depending on species). PECswaq is below 10% of the 90th percentile of manganese concentration in European water (13.2 μg/L).

Calculated PEC_{sed} in marine sediment is 220.5 mg Mn/kg dw for the use of the additive in marine aquaculture (at the use level 100 mg Mn/kg feed). No data are available for the background concentration of manganese in the marine sediment in FOREGS database. Considering that manganese, like iron, is widespread in relatively high amounts in the environment and taking into account its low toxicity, even without monitoring data on background concentration of manganese for the marine sediment, it can be concluded that the use of the additive in sea cages would not cause concern for the marine environment.

3.2.4.3. Conclusions on safety for the environment

The FEEDAP Panel concludes that the use of the additive in animal nutrition for all animal species is safe for the environment, provided that the current total contents of manganese authorised in feed are respected.

3.3. Efficacy

To demonstrate the efficacy of nutritional additives, one study in a single animal species or category, including laboratory animals, is generally considered sufficient for new forms of compounds of trace elements (EFSA FEEDAP Panel, 2018).

The applicant provided a combined tolerance/efficacy study in chickens for fattening (see Section 3.2.1.1). The experimental groups in the study are shown in Table 1. In this trial, the manganese concentration in edible tissues/organs was measured (Table 3).

[41] Based on the data available in FORGES database http://weppi.gtk.fi/publ/foregsatlas/ForegsData.php

[42] Predicted environmental concentration (PEC) is calculated by using FERA tool. Available online: https://www.efsa.europa.eu/en/applications/feedadditives/tools

[43] FOREGS database (2005). Available online: http://weppi.gtk.fi/publ/foregsatlas/

[44] Guidance document N38. Technical Guidance for implementing Environmental Quality Standards (EQS) for metals. Consideration of metal bioavailability and natural background concentrations in assessing compliance (2019). Common Implementation Strategy for the Water Framework Directive (2000/60/EC). Available online: https://metals-toolbox.com/uploads/pdf/environment/Guidance%2520No%252038%2520-%2520Technical%2520guidance%2520for%2520EQS%2520for%2520metals%2520(2).pdf

[45] ADR(2)-2023-06-14.

The deposition data for manganese from the treatment group with the additive under assessment at highest level tested (900 mg Mn/kg feed) showed a higher manganese content in kidney, liver and skin compared to the control.

Therefore, the FEEDAP Panel concludes that the additive is an efficient source of manganese for chickens. This conclusion can be extrapolated to all animal species and categories.

3.4. Post-market monitoring

The FEEDAP Panel considers that there is no need for specific requirements for a post-market monitoring plan other than those established in the Feed Hygiene Regulation[46] and Good Manufacturing Practice.

4. Conclusions

The FEEDAP Panel concludes that the use of manganese(II)-betaine complex in animal nutrition is safe for all animal species, the consumer and the environment provided that the maximum authorised levels in the EU for total manganese in feed are not exceeded.

Owing to the presence of nickel, the additive is considered a skin and respiratory sensitiser. It is an eye irritant, but it is not irritant to the skin.

The FEEDAP Panel concludes that the additive is efficacious as a source of manganese in all animal species and categories.

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Abbreviations

ADFI	average daily feed intake
ADG	average daily gain
BW	body weight
CAS	Chemical Abstracts Service
CD	Commission Decision
CV	coefficient of variation
DM	dry matter
DW	dry weight
EURL	European Union Reference Laboratory
FCR	feed conversion ratio
FEEDAP	EFSA Scientific Panel on Additives and Products or Substances used in Animal Feed
IUPAC	International Union of Pure and Applied Chemistry
LOD	limit of detection
LOQ	limit of quantification
MW	molecular weight
OECD	Organisation for Economic Co-operation and Development
PCBs	polychlorinated biphenyls
PCDD/F	polychlorinated dibenzo-p-dioxins and dibenzofurans
RH	relative humidity