Safety and efficacy of a feed additive consisting of ferric citrate chelate (CI-FER™) for poultry species for fattening or reared up to the point of lay (Akeso Biomedical, Inc.)

EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP), Vasileios Bampidis, Giovanna Azimonti, Maria de Lourdes Bastos, Henrik Christensen, Birgit Dusemund, Mojca Fasmon Durjava, Maryline Koub, Marta López-Alonso, Secundino López Puente, Francesca Marcon, Baltasar Mayo, Alena Pechova, Mariana Petkova, Fernando Ramos, Yolanda Sanz, Roberto Edoardo Villa, Ruud Woutersen, Jaume Galobart, Gloria López Gálvez and Montserrat Anguita

Abstract

Following a request from the European Commission, the Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) was asked to deliver a scientific opinion on the safety and efficacy of ferric citrate chelate (CI-FER™) as a zootechnical additive for poultry species for fattening or reared up to the point of lay. The product had been already assessed by the FEEDAP Panel for use in suckling and weaned piglets and minor porcine species. The application was for an extension of use to poultry species for fattening or reared up to the point of lay. The FEEDAP Panel considers that the new use would not raise safety concerns for the consumers and the environment and retained the previous conclusions as regards to the user: CI-FER™ does not pose a risk by inhalation, it is non-irritant to the skin but should be considered as an eye irritant and as a skin sensitisier. Owing to the limitations identified in the tolerance trial submitted, the FEEDAP Panel could not conclude on the safety of the additive for the target species. Regarding the efficacy, three studies were submitted but two of them were not considered further in the assessment due to the husbandry conditions to which the animals were subject to. Therefore, the FEEDAP Panel could not conclude on the efficacy of the additive.

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Keywords: zootechnical additives, gut flora stabiliser, ferric citrate chelate, CI-FER™, safety, efficacy, poultry

Requestor: European Commission

Question number: EFSA-Q-2020-00843

Correspondence: feedap@efsacomm.eu
Panel members: Giovanna Azimonti, Vasileios Bampidis Maria de Lourdes Bastos, Henrik Christensen, Birgit Dusemund, Mojca Fasmon Durjava, Maryline Kouba, Marta López-Alonso, Secundino López Puente, Francesca Marcon, Baltasar Mayo, Alena Pechová, Mariana Petkova, Fernando Ramos, Yolanda Sanz, Roberto Edoardo Villa and Ruud Woutersen.

Declarations of interest: The declarations of interest of all scientific experts active in EFSA’s work are available at https://ess.efsa.europa.eu/doi/doiweb/doisearch.

Acknowledgements: The Panel wishes to acknowledge the contribution of the Working Group on Animal Nutrition to this opinion.

Suggested citation: EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Bampidis V, Azimonti G, Bastos ML, Christensen H, Dusemund B, Fasmon Durjava M, Kouba M, López-Alonso M, López Puente S, Marcon F, Mayo B, Pechová A, Petkova M, Ramos F, Sanz Y, Villa RE, Woutersen R, Galobart J, López Gálvez G and Anguita M, 2022. Scientific Opinion on the safety and efficacy of a feed additive consisting of ferric citrate chelate (CI-FER™) for poultry species for fattening or reared up to the point of lay (Akeso Biomedical, Inc.). EFSA Journal 2022;20(2):7155, 8 pp. https://doi.org/10.2903/j.efsa.2022.7155

ISSN: 1831-4732

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The EFSA Journal is a publication of the European Food Safety Authority, a European agency funded by the European Union.
1. Introduction

1.1. Background and Terms of Reference as provided by the requestor

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 a feed additive shall submit an application in accordance with Article 7.

The European Commission received a request from Akeso Biomedical Inc. represented in the EU by Pen & Tec Consulting SLU\(^2\) for the authorisation of the product ferric citrate chelate (CI-FERTM) when used as a feed additive for chickens for fattening, chickens reared for laying, turkeys for fattening, turkeys reared for breeding, and minor poultry species reared up to slaughter or up to the point of lay (category: zootechnical additives; functional groups: gut flora stabilisers, other zootechnical additives).

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). The particulars and documents in support of the application were considered valid by EFSA as of 17 February 2021.

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, consumer, user and the environment and on the efficacy of the product ferric citrate chelate (CI-FERTM), when used under the proposed conditions of use.

1.2. Additional information

The safety and efficacy of ferric citrate chelate (CI-FERTM) for use in suckling and weaned piglets and minor porcine species has been evaluated by the FEEDAP Panel (EFSA FEEDAP Panel, 2019a, 2021). The additive is currently authorised for use in suckling and weaned piglets and minor porcine species.\(^3\)

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\(^4\) in support of the authorisation request for the use of ferric citrate chelate (CI-FERTM) as a feed additive for chickens for fattening, chickens reared for laying, turkeys for fattening, turkeys reared for breeding, and minor poultry species. The FEEDAP Panel used the data provided by the applicant together with data from other sources, such as previous risk assessments by EFSA.

The European Union Reference Laboratory (EURL) considered that the conclusions and recommendations reached in the previous assessment regarding the methods used for the control of ferric citrate chelate (CI-FERTM) in animal feed are valid and applicable for the current application.\(^5\)

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of ferric citrate chelate (CI-FERTM) is in line with the principles laid down in Regulation (EC) No 429/2008\(^6\) and the relevant guidance documents: Guidance on the identity, characterisation and conditions of use of feed

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\(^1\) REGULATION (EC) No 1831/2003 of the European Parliament and of the Council of 22 September 2003 on additives for use in animal nutrition. OJ L 268, 18.10.2003, p. 29.

\(^2\) Akeso Biomedical Inc., USA, represented in the EU by Pen & Tec Consulting S.L.U. Pl. Ausias March 1, 4th Floor D01. ES-08195, Sant Cugat del Vallès, Spain.

\(^3\) COMMISSION IMPLEMENTING REGULATION (EU) 2021/1412 of 27 August 2021 concerning the authorisation of Iron(III) citrate chelate as a feed additive for piglets and minor porcine species (holder of the authorisation: Akeso Biomedical, Inc. USA, represented in the Union by Pen & Tec Consulting SLU). OJ L 304, 30.8.2021, p. 14.

\(^4\) FEED dossier reference: FAD-2020-0092.

\(^5\) The full report is available on the EURL website: https://ec.europa.eu/jrc/sites/jrcsh/files/finrep-fad-2018-0065-ferric-citrate.pdf

\(^6\) 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.
additives (EFSA FEEDAP Panel, 2017a), Guidance on the assessment of the safety of feed additives for the target species (EFSA FEEDAP Panel, 2017b), Guidance on the assessment of the safety of feed additives for the consumer (EFSA FEEDAP Panel, 2017c), 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 environment (EFSA FEEDAP Panel, 2019b) and Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018).

3. Assessment

The present application concerns ferric citrate chelate, hereinafter referred to as CI-FERTM, to be used as a feed additive (category: zootechnical additives; functional groups: gut flora stabilisers and other zootechnical additives (performance enhancer)) for chickens for fattening, chickens reared for laying, turkeys for fattening, turkeys reared for breeding, and minor poultry species reared up to slaughter or up to the point of lay.

3.1. Characterisation

The additive, including its manufacturing process, was fully characterised in previous opinions of the FEEDAP Panel (EFSA FEEDAP Panel, 2019a, 2021). CI-FERTM is specified to contain total iron ≤ 23%, iron (III) ≥ 16.5% and moisture ≤ 10.0%. The FEEDAP Panel is not aware of any new information that would modify the previous characterisation of the additive. However, the Panel notes that the authorisation for the use in piglets specifies that the total iron content is 20%, the minimum iron III content is 15% and moisture ≤ 10.0%.3

The product is intended to be used in feed for chickens for fattening and reared for laying, turkeys for fattening and reared for breeding and all minor poultry species up to slaughter or point of lay, at a minimum level of 200 mg additive/kg complete feed. The applicant states that CI-FERTM can be used with other iron sources up to the maximum iron content permitted in poultry feeds by EU legislation (750 mg iron/kg complete feed).

3.2. Safety

The safety of the product for the consumer, user and environment was evaluated in the context of previous opinions that regarded the use in piglets (EFSA FEEDAP Panel, 2019a, 2021). The Panel concluded that the use of the additive in feed for piglets under the conditions of use proposed would not raise concerns for consumers and environment. Concerning the safety for the users, the FEEDAP Panel considered in 2021 that CI-FERTM does not pose a risk by inhalation and it is classified as non-irritant to the skin but should be considered as an eye irritant and as a skin sensitiser.

The FEEDAP Panel is not aware of new information that would lead it to reconsider the previous conclusions on the safety for the consumers and users. It is noted that ferric citrate chelate contains a theoretical maximum content of iron of 23%, hence at the minimum recommended use level of 200 mg/kg feed, CI-FERTM would supply a maximum of 46 mg Fe/kg feed for poultry species. Considering that CI-FERTM is proposed to be used with other iron sources up to the max iron content allowed by EU legislation (750 mg Fe/kg feed), it is not expected that the use of ferric citrate in poultry feed would have any significant influence on the iron content of edible tissues/products from poultry receiving the additive. Therefore, the FEEDAP Panel considers that the proposed extension of use to poultry species would not introduce risks to consumers. Similarly, the Panel considers that the proposed extension to poultry species would not introduce additional risks to users. Consequently, the FEEDAP Panel retains the conclusions drawn in previous assessments for the consumer and user.

However, the extension of use to the new target species requires an assessment of the safety for the target species and for the environment.

3.2.1. Safety for the target species

The applicant submitted a tolerance/efficacy trial, done with a total of 480 one-day-old male chickens for fattening (Cobb 500) which were distributed in 24 pens and 3 dietary treatments were allocated to the pens (representing 8 replicates per treatment). Two basal diets with no supplemental iron (starter, 1–14 days; grower, 15–35 days) based on maize, soybean meal, and wheat, were either
not supplemented (control) or supplemented with CI-FERTM to provide 200 mg/kg feed (1× minimum recommended level) or 2,000 mg/kg feed (10×), confirmed by analysis of the microtracer and total iron in the diet. The birds were under study for 35 days and the feeds were offered in mash on ad libitum basis. General health status, behaviour, culls and mortality were monitored daily. During the study, the feed intake and the body weight were measured on weekly basis and by pen. The feed to gain ratio was calculated considering the dead/culled animals. Blood samples were collected on day 35 from 2 animals per pen and analysed for biochemical and haematological parameters. A one-way analysis of variance was done with the treatment as the factor considered and the group means were compared with Tukey's adjustment, significance level applied was of 0.05.

This study showed limitations that prevent the Panel to perform a complete assessment of the safety for the target species. The additive was included at an overdose level of 10× the minimum recommended level, but the battery of observations was not complete. For instance, some relevant blood parameters were not measured (gamma-glutamyl transferase, platelet counts and prothrombin time) and necropsy was not performed at the end of the study. Therefore, in the absence of an adequate tolerance study, the FEEDAP Panel cannot conclude on the safety of the additive for chickens for fattening, chickens reared for laying, turkeys for fattening, turkeys reared for breeding, and minor poultry species reared up to slaughter or up to the point of lay.

3.2.2. Safety for the environment

The components of the additive, iron and citric acid, are ubiquitous in the environment. Iron is a major element in soil with a median value of 2.1% (Rose et al., 1979; representing 21,000 mg/kg), while citric acid is a physiological and natural component of animals and plants.

Based on the calculation method provided in the technical guidance for assessing the safety of feed additives for the environment (EFSA FEEDAP Panel, 2019b), the highest increase of iron concentration in soil after a 1-year application of manure from chicken for fattening or for turkey for fattening would be around 12 mg/kg, assuming that 100% of the highest level (750 mg Fe/kg feed) will be excreted. Therefore, any additional load from the use of the product in feed for poultry for fattening or reared for laying/breeding is not expected to pose an environmental risk.

3.2.3. Conclusions on the safety of the additive

The FEEDAP Panel cannot conclude on the safety of the additive for the target species. The additive when used in feed for poultry at the minimum recommended level does not pose a risk for the consumers or for the environment.

The additive CI-FERTM does not pose a risk by inhalation and is non-irritant to the skin but should be considered as an eye irritant and a skin sensitiser.

3.3. Efficacy

The applicant submitted an in vitro study and three in vivo studies to support the efficacy of the additive as a zootechnical additive. The applicant states that the effects of the additive are as a gut flora stabiliser, evidenced by reduction in carriage of enteropathogens, and as a performance enhancer, evidenced by and improved zootechnical performance of the birds.

The in vitro study was already evaluated in a previous opinion of the FEEDAP Panel (EFSA FEEDAP Panel, 2019a) in which it was not considered relevant for the assessment, owing to the lack of mimicking to the in vivo conditions.

The three in vivo studies were conducted in chickens for fattening in two different locations. In two of the studies, the birds were subject to a ‘mild stress’ situation and challenge. In the studies the pens were bedded with fresh and used bedding (50:50) since the start and the animals received 24 h of continuous light throughout the study period. On day 7, feed was withdrawn for 3 h, and 2 kg of litter spiked with different microbial strains was added to the pens. The husbandry conditions in which

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7 Technical dossier/Section III/Annex III.1.1.1 and supplementary information August 2021.
8 Total cholesterol, triglycerides, bilirubin, creatine, urea, glucose, albumins, globulins, total protein, enzymes (α-amylase, aspartate-aminotransferase, alanine aminotransferase, glutamate-dehydrogenase, alkaline phosphatase, L-lactate dehydrogenase), and serum amyloid A, sodium, potassium, chloride, calcium, magnesium, phosphate.
9 Erythrocytes, leukocytes, lymphocytes, monocytes, eosinophils, basophils, neutrophils, haemoglobin, haematocrit, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration.
10 Technical dossier/Section IV/Annex IV.3.1 and IV.3.2 and supplementary information August 2021 and October 2021.
the birds were kept do not reflect the conditions in which the animals would be raised in a farm in the EU and are not in line with the Council Directive 2007/43/EC\cite{11} as regards to the lightning conditions and bedding. Therefore, these two trials were not considered further.

The third trial is the tolerance/efficacy trial that has been described above in Section 3.2.1. The results for the zootechnical performance for the control group and the 200 mg/kg group are provided in Table 1. Mortality including culling was 2.5% and no differences were found between treatments. Feeding the birds with the additive resulted in a significantly higher final body weight and in a significant improved feed to gain ratio.

### Table 1: Effect of CI-FER\textsuperscript{TM} on the performance of chickens for fattening from day 1 to 35 of life

| mg CI-FER\textsuperscript{TM} per kg feed | mg total iron/kg feed | Daily feed intake (g/bird)\textsuperscript{(1)} | Final weight (g) | Feed to gain ratio | Mortality (%) |
|----------------------------------------|-----------------------|-----------------------------------------------|-----------------|-------------------|---------------|
| Intended                               | Analysed\textsuperscript{(1)} |                               |                |                   |               |
| 0                                      | –/–                   | 78/80                                        | 86              | 2,236\textsuperscript{a} | 1.38\textsuperscript{b} | 3.1          |
| 200                                    | 184/168               | 116/125                                      | 87              | 2,332\textsuperscript{b} | 1.33\textsuperscript{a} | 2.5          |

\textsuperscript{a,b}: Values within one trial and within one column with different superscripts as significantly different (p < 0.05).

\textsuperscript{(1)}: Calculated values from the analysis of the microtracer added to the additive.

#### 3.3.1. Conclusions on efficacy

The results from the study considered in the assessment showed improvements on the zootechnical performance of the birds fed with the additive at the minimum recommended level. However, in the absence of enough evidence, the FEEDAP Panel cannot conclude on the efficacy of the additive as a zootechnical additive.

#### 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\cite{12} and Good Manufacturing Practice.

#### 4. Conclusions

The FEEDAP Panel cannot conclude on the safety and the efficacy of the additive CI-FER\textsuperscript{TM} for the target species.

The additive when used in feed for poultry at the proposed conditions of use does not pose a risk for the consumers or for the environment.

The additive CI-FER\textsuperscript{TM} does not pose a risk by inhalation and it is classified as non-irritant to the skin but should be considered as an eye irritant and as a skin sensitiser.

#### 5. Documentation as provided to EFSA/Chronology

| Date       | Event                                                                                     |
|------------|--------------------------------------------------------------------------------------------|
| 12/11/2020 | Dossier received by EFSA. Ferric citrate chelate as a zootechnical feed additive for suckling and weaned piglets. Submitted by AKESO BIOMEDICAL, INC. |
| 30/11/2020 | Reception mandate from the European Commission                                              |
| 17/02/2021 | Application validated by EFSA – Start of the scientific assessment                         |
| 17/05/2021 | Comments received from Member States                                                       |
| 22/06/2021 | Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. Issues: Characterisation, safety for the target species and efficacy |
| 05/08/2021 | Reception of supplementary information from the applicant - Scientific assessment re-started |

\textsuperscript{11} DIRECTIVE 2007/43/EC of 28 June 2007 laying down minimum rules for the protection of chickens kept for meat production. OJ L 182, 12.7.2007, p. 19.

\textsuperscript{12} REGULATION (EC) No 183/2005 of the European Parliament and of the Council of 12 January 2005 laying down requirements for feed hygiene. OJ L 35, 8.2.2005, p. 1.
CI-FERTM for poultry species

Date | Event
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29/09/2021 | Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. Issues: efficacy
29/10/2021 | Reception of supplementary information from the applicant - Scientific assessment re-started
26/01/2022 | Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

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Abbreviation

FEEDAP EFSA Panel on Additives and Products or Substances used in Animal Feed