Assessment : Safety assessment of the application for L-lysine sulfate produced by *Corynebacterium glutamicum* KCCM 80227 as a feed additive for all animal species, from Daesang Europe B. V.

Reference Number RP 1126

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Contents

1.Executive summary

The FSA/FSS have undertaken an assessment of application RP 1126 for the use of L-lysine sulfate (≥ 69%, of which ≥ 52% L-lysine and ≥ 17.3 sulfate [1]) produced by *Corynebacterium glutamicum* KCCM 80227 as a feed additive for all animal species, from Daesang Europe B. V. Van Heuven Goedhartlaan 935, 1181 LD Amstelveen, Netherlands (category: nutritional additives; functional group: amino acids, their salts and analogues, to satisfy the nutritional needs of the target species with this essential amino acid).

A number of feed additive applications have been received by Great Britain (GB) where EFSA, prior to the end of the transition period, evaluated an application for the product. FSA/FSS have reviewed the EFSA opinion (EFSA Journal 2021;19(7):6706) and confirm that it is adequate and relevant for GB risk analysis and used this to form the basis of the GB opinion. A full risk assessment of this application was not considered necessary by FSA and FSS.

The FSA/FSS risk assessors concluded that the EFSA opinion is adequate and relevant for GB risk analysis. Therefore, the use of the L-lysine sulfate additive, as described in this application, is safe and is not liable to have an adverse effect on the target species, worker safety, environmental safety and human health at the intended concentrations of use.

There are no specific conditions or restrictions in relation to handling, labelling, post-market monitoring requirements and use of this additive as described in this application. Maximum Residue Limits (MRLs) are not required for this additive.

2.Background and purpose of review

EFSA Journal 2021; 19(7): 6706,

Question number: EFSA-Q-2020-00708

In accordance with Retained EU Regulation 1831/2003 on feed additives, the application RP 1126 for the use of L-lysine sulfate produced by fermentation with *Corynebacterium glutamicum* KCCM 80227 as a feed additive for all animal species from Daesang Europe B. V. has been submitted for authorisation in each nation of Great Britain (GB).

Whilst it was a Member State of the EU, the UK accepted the assessments of EFSA in respect of authorisations for regulated food and feed products. Since the end of the transition period, FSA/FSS has adopted equivalent technical guidance and quality assurance processes to be able to undertake GB risk assessments for regulated product applications.

[1] The analytical method for L-lysine was method EN ISO 17180:2013 based on ion-exchange chromatography (IEC) coupled with optical (visible – VIS or fluorescence – FLD) detection. The analytical method for the identification of sulfate was the European Pharmacopoeia monograph (Ph. Eur. 01/2008:20301) based on colourimetry.

However, a number of applications have been received by GB where EFSA, prior to the end of the transition period, evaluated an application for the product for which an application is now made to GB. FSA/FSS has decided to make use of the EFSA risk assessment, where this is appropriate, in forming its opinion. Therefore, FSA/FSS risk assessors have reviewed the EFSA opinion[2] for the application below in the context of intended GB use and have concluded that the intended uses are safe.

In reviewing the EFSA risk assessment opinion the reviewers have verified that the standard approach as outlined in the relevant guidance[3] has been followed and the arguments made are consistent with the data summarised in the opinion. Consideration has been given to the processes undertaken to ensure the opinions are robust and whether there are any aspects that would require further review such as specific issues for the countries of the GB. The result of the assessment is that the EFSA scientific opinions are adequate also for GB risk analysis. Therefore, a full risk assessment has not been performed by FSA/ FSS.

3.Details of the EFSA assessment

Methodology applied in the EFSA opinion

EFSA FEEDAP guidance: Guidance on the assessment of the safety of feed additives for the environment (**2019**), Guidance on the assessment of the safety of feed additives for the target species (**2017a**), Guidance on the identity, characterisation and conditions of use of feed additives (**2017b**), Guidance on the assessment of the safety of feed additives for the consumer (**2017c**), Guidance on the characterisation of microorganisms used as feed additives or as production organisms (**2018a**), Guidance on studies concerning the safety of use of the additive for users/workers (**2012**) and principles in Regulation (EC) No 429/2008.

3.1 Source/organism

The additive contains L-lysine sulfate produced by fermentation with the non-genetically modified *Corynebacterium glutamicum* KCCM 80227. The application is for the additive as a solid as L-lysine sulfate with a water content of ≤ 4%.

The production strain of the L-lysine sulfate is from the species *C.glutamicum* which is designated as eligible for the Qualified Presumption of Safety (QPS) assessment approach.

For this application, the production strain was sequenced using a whole genome approach to confirm identity and evaluated for susceptibility to antibiotics. From this it was concluded that the strain is not phenotypically resistant to any of the antibiotics tested.

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[2] EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2021. Safety and efficacy of a feed additive consisting of L-lysine sulfate produced by *Corynebacterium glutamicum* KCCM 80227 for all animal species (Daesang Europe BV).EFSA Journal 2021; 19(7):6706. https://doi.org/10.2903/j.efsa.2021.6706

[3] See reference list for the full set of guidance applied.

3.2 Genetic modification step
Not applicable as the production strain is not genetically modified.

3.3 Specification
Information provided on the identity, composition and specifications of the production species does not raise safety concerns (the detail is redacted in the EFSA opinion whilst confidentiality claims are determined.)

Physiochemical properties, homogeneity data and stability data (as shelf life of the products) were presented for the additive. The solid additive L-lysine sulfate is homogenous (for total lysine) within a pelleted final feed ration. Stability data for the additive within six batches of exemplar feed rations (three batches for mash and three for pelleted forms) and three batches of feed pre-mix were provided. Maximum losses in feed mash after 3 months were 4%, in pre-mix after 6 months 20%, with no losses in pellet feed.

Three batches of L-lysine sulfate were analysed for heavy metals (mercury, lead, cadmium and arsenic), dioxins, dioxin-like polychlorinated biphenyls (PCBs), mycotoxins (aflatoxins, citrinin, ochratoxin A, zearalenone, fumonisins and deoxynivalenol) and microbiological contamination (*E.coli*, total *Enterobacteriaceae*, *Salmonella* spp, yeasts and filamentous fungi). The concentration of deoxynivalenol of 876 – 2,750 µg/kg was considered high. However, the results of these analyses were considered to raise no safety concerns.

This application included the request for use within all species. No recommended inclusion rates were discussed in the application due to the varying concentrations that may be required due to the animal species, nutritional specification of the feed ration, environmental conditions and the current health state of the animal.

3.4 Exposure assessment
Not relevant as L-lysine and sulfur are natural components of any consumed animal products. The applicant is seeking to use L-lysine sulfate as a nutritional feed additive and the product has been assessed on this basis.

3.5 Toxicological data
L-lysine and its salts are currently authorised as a feed additive for all species as a nutritional additive, using different production species than the one used in this application. This application is for the use of L-lysine sulfate produced by fermentation with *Corynebacterium glutamicum* KCCM 80227.

Regarding the high intrinsic nature of sulfate within the additive, a tolerance study in chickens for fattening administered magnesium sulfate was provided. These results along with results obtained from the applicant's literature review led to EFSA concluding that no negative effects are expected from the sulfate concentrations administered with the intended use of this additive. However, this is only if the total sulfur intake complies with the recommendations established by scientific bodies, which includes the background sulfur/sulfate content in the compound feed.

The additive has low dusting potential, therefore the exposure of the user/worker to the additive is expected to be limited. An acute inhalation toxicity study was provided which showed a lack of respiratory toxicity. This study was using L-lysine sulfate produced from a different species strain, *C. glutamicum* ATCC 13032 evaluated in a

previous opinion (EFSA FEEDAP Panel 2016b). EFSA concluded that due to the similarity of this production strain with *C. glutamicum* KCCM 80227 and the manufacturing process of the additive in this application, that the conclusions on inhalation toxicity are relevant for this application.

The skin irritancy potential of the additive was tested in a GLP study performed according to OECD TG 404. The additive was concluded to be non-irritant to skin.

A skin sensitisation test was undertaken which found the additive is not a skin sensitiser. Again, this was from a study using a different production strain of *C. glutamicum*. Due to the similarity of the strain and the manufacturing processes EFSA concluded that the results were still applicable for this application.

The potential eye irritancy / corrosion of the additive was tested in a GLP study performed according to OECD TG 405. The results showed the additive was not an irritant or corrosive to eyes.

4. EFSA assessment and conclusions

L-lysine sulfate produced by fermentation with *Corynebacterium glutamicum* KCCM 80227 raises no concerns for the consumer of the products obtained from animals fed the additive. *Corynebacterium glutamicum* is designated as eligible for the QPS assessment approach.

The additive is not a skin or eye irritant or deemed a skin sensitiser. It has a low dusting potential and therefore the exposure to workers handling the product is expected to be low and represent no safety concerns.

No risks to the environment are expected and no further environmental risk assessment is required. The additive is present as a natural component of both plants and animals. If given to animals, L-lysine is excreted as urea/uric acid, indole-related compounds and carbon dioxide. The use of this additive in animal nutrition would not lead to any increases in the concentration of L-lysine or sulfate in the environment. The application does not raise safety concerns for the environment regarding the production strain *Corynebacterium glutamicum* KCCM 80227.

Regarding safety for the target species, there are no safety concerns noted relating to the amino acid L-lysine. In relation to the high sulfate concentration in the additive it is concluded that adverse effects are not expected at the concentrations of typical use, provided the total sulfur intake complies with recommendations. This includes that any other background sulfur in the final feed is also taken into account.

The production strain and its DNA were not detected in the final additive product.

5. Caveats and uncertainties

There are no further caveats or uncertainties to highlight.

6. FSA Conclusion on reliability and applicability

The application has been assessed in line with the applicable guidance and is partially based on considerations of detailed proprietary information available to the EFSA Panel, whilst this is only briefly summarised this description is consistent with the conclusions.

6.1 Analytical Method Review

FSA/FSS accepts the EURL analytical method evaluation report[4]. FSA/FSS determined the analytical method as appropriate for official controls for this feed additive.

7. Outcome of assessment

FSA/FSS has reviewed the EFSA opinions and consider them adequate and relevant for GB risk analysis. Therefore, the opinions were used to form the basis of the GB opinion. A full risk assessment of this application was not considered necessary by FSA and FSS.

FSA/FSS had access to all supporting documentation that was provided to the EFSA Panel by the applicant, and subsequently used to form the EFSA opinion. FSA/FSS agree with the safety conclusions outlined in the EFSA opinions.

Following the principles outlined in the background for making use of the EFSA opinions, the FSA/FSS opinion is that the L-lysine sulfate additive, as described in this application, is safe and is not liable to have an adverse effect on the target species, worker safety, environmental safety and human health at the intended concentrations of use.

8. References

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2012. Guidance on studies concerning the safety of use of the additive for users/workers. EFSA Journal 2012;10(1):2539, 5 pp. https://doi.org/10.2903/j.efsa.2012.2539

[4] https://joint-research-centre.ec.europa.eu/system/files/2021-07/finrep-fad-2020-00820085-lysine-sulfate.pdf

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2016b. Scientific opinion on the safety and efficacy of concentrated liquid L-lysine (base), L-lysine monohydrochloride and L-lysine sulphate produced using different strains of *Corynebacterium glutamicum* for all animal species based on a dossier submitted by AMAC/EEIG. EFSA Journal 2016;14(3):4346, 3 pp. https://doi.org/10.2903/j.efsa.2016.4346

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2017a. Guidance on the assessment of the safety of feed additives for the target species. EFSA Journal 2017;15(10):5021, 19 pp. https://doi.org/10.2903/j.efsa.2017.5021

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2017b. Guidance on the identity, characterisation and conditions of use of feed additives. EFSA Journal 2017;15(10):5023, 12 pp. https://doi.org/10.2903/j.efsa.2017.5023

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2017c. Guidance on the assessment of the safety of feed additives for the consumer. EFSA Journal 2017;15(10):5022, 17 pp. https://doi.org/10.2903/j.efsa.2017.5022

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2018a. Guidance on the characterisation of microorganisms used as feed additives or as production organisms. EFSA Journal 2018;16(3):5206, 24 pp. https://doi.org/10.2903/j.efsa.2018.5206

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2019. Guidance on the assessment of the safety of feed additives for the environment. EFSA Journal 2019;17(4):5648, 78 pp. https://doi.org/10.2903/j.efsa.2019.5648

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2021. Safety and efficacy of a feed additive consisting of L-lysine sulfate produced by *Corynebacterium glutamicum* KCCM 80227 for all animal species (Daesang Europe BV).EFSA Journal 2021; 19(7):6706. https://doi.org/10.2903/j.efsa.2021.6706