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Safety and efficacy of a feed additive consisting of L-valine produced by *Corynebacterium glutamicum* CGMCC 7.366 for all animal species (Ningxia Eppen Biotech Co., Ltd.)

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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 the feed additive consisting of L-valine produced by fermentation using a non-genetically modified strain of *Corynebacterium glutamicum* (CGMCC 7.366). The additive is intended to be used in feed and water for drinking for all animal species and categories. The production strain is considered suitable for the qualified presumption of safety (QPS) approach to safety assessment. The FEEDAP Panel concludes that L-valine produced using *C. glutamicum* CGMCC 7.366 is safe for the target species when supplemented in appropriate amounts to the diet according to the nutritional needs of the target species. The FEEDAP Panel has concerns on the use of amino acids in water for drinking for hygienic reasons, and due to the risk of imbalances when administered simultaneously via feed. The use of L-valine produced using *C. glutamicum* CGMCC 7.366 in animal nutrition is considered safe for the consumer and for the environment. No conclusion could be drawn on the potential of L-valine produced using *C. glutamicum* CGMCC 7.366 to be toxic by inhalation, irritant to the skin or eyes, or a dermal sensitiser due to the lack of data. The additive L-valine produced by fermentation using *C. glutamicum* CGMCC 7.366 is regarded as an efficacious source of the essential amino acid L-valine for non-ruminant nutrition. For the supplemental L-valine to be as efficacious in ruminants as in non-ruminant species, it requires protection against degradation in the rumen.

Keywords: nutritional additive, amino acids, L-valine, *Corynebacterium glutamicum* CGMCC 7.366, safety, efficacy

Requestor: European Commission

Question number: EFSA-Q-2020-00375

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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 Ningxia Eppen Biotech Co., Ltd., represented in the EU by Welding GmbH & Co. KG[2] for authorisation of the feed additive consisting of L-valine produced by *Corynebacterium glutamicum* CGMCC 7.366, when used as a feed additive for all animal species (category: nutritional additives; functional group: amino acids, their salts and analogues).

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 22 June 2020.

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 feed additive consisting of L-valine (≥ 98.0%), produced by fermentation using *Corynebacterium glutamicum* CGMCC 7.366 for all animal species, when used under the proposed conditions of use (see Section 3.1.4).

1.2. Additional information

The subject of the present assessment is the product consisting of L-valine (minimum 98.0%) produced by fermentation using *C. glutamicum* CGMCC 7.366. This product has not been previously assessed as a feed additive in the European Union. L-Valine produced by different microbial strains is authorised as feed additive for all animal species.[3]

The FEEDAP Panel has issued several scientific opinions on the safety and efficacy of L-valine produced by fermentation using different strains of *C. glutamicum* or *Escherichia coli* (EFSA 2008a,b; EFSA FEEDAP Panel, 2013, 2014a, 2015a,b, 2019a,b, 2020a,b) or when used as a feed flavouring compound (EFSA FEEDAP Panel, 2014b).

L-Valine is authorised for use in food.[4,5] L-Valine and DL-valine are also authorised as sensory additives, belonging to the functional group flavouring compounds (FLAVIS No 17.028 and 17.023, respectively).[6]

The Cosmetic Ingredient Review (CIR) Expert Panel (2012) issued a safety assessment of alpha amino acids as used in cosmetics.

The European Pharmacopoeia (2020) has a monograph dedicated to L-valine (01/2017:0796).

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[7] in support of the authorisation request for the use of the product consisting of L-valine

[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] Ningxia Eppen Biotech Co., Ltd., represented by Welding GmbH & Co. Kg, Esplanade 39, 20354, Hamburg, Germany.
[3] Regulation (EC) No 403/2009 of 14 May 2009/Amended by Commission Implementing Regulation (EU) No 848/2014 of 4 August 2014/Amended by Commission Implementing Regulation (EU) No 2015/1114 of 9 July 2015; Commission Implementing Regulation (EU) No 848/2014 of 4 August 20144/Amended by Commission Implementing Regulation (EU) No 2015/1114 of 9 July 2015; Commission Implementing Regulation (EU) No 1236/2014 of 18 November 2014/Amended by Commission Implementing Regulation (EU) No 2015/1114 of 9 July 2015; Commission Implementing Regulation (EU) No 2015/1114 of 9 July 2015; and Commission Implementing Regulation (EU) 2019/1289 of 31 July 2019.
[4] Commission Directive 2006/141/EC on infant formulae and follow-on formulae, OJ L 401, 30.12.2006, pp. 1–33.
[5] Commission Directive 2001/15/EC of 15 February 2001 on substances that may be added for specific nutritional purposes in foods for particular nutritional uses. OJ L 52, 22.2.2001, pp. 1–7.
[6] Commission list of the authorised additives in feedingstuffs published in application of Article 9t (b) of Council Directive 70/524/EEC concerning additives in feedingstuffs (2004/C 50/01) OJ C/50, 25.2.2004, pp. 1–144.
[7] FEED dossier reference: FAD-2020-0033.

(minimum 98.0%) produced by fermentation using *C. glutamicum* CGMCC 7.366, as a feed additive intended to be used in feed and water for drinking for all animal species.

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, to deliver the present output.

EFSA has verified the European Union Reference Laboratory (EURL) report as it relates to the methods used for the control of the product consisting of L-valine produced by fermentation using *C. glutamicum* CGMCC 7.366 in animal feed. The Executive Summary of the EURL report can be found in Annex A.[8]

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of the additive under assessment is in line with the principles laid down in Regulation (EC) No 429/2008[9] and the relevant guidance documents: Guidance on the identity, characterisation and conditions of use of feed additives (EFSA FEEDAP Panel, 2017a), Guidance on the characterisation of microorganisms used as feed additives or as production organisms (EFSA FEEDAP Panel, 2018a), 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 feed additives for user/workers (EFSA FEEDAP Panel, 2012), Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018b) and Guidance on the assessment of the safety of feed additives for the environment (EFSA FEEDAP Panel, 2019c).

3. Assessment

The subject of the assessment is a product consisting of L-valine (minimum 98.0%) produced by *C. glutamicum* CGMCC 7.366, intended to be used as a nutritional additive (functional group amino acids, their salts and analogues) in feed and water for drinking for all animal species.

3.1. Characterisation

3.1.1. Characterisation of the production organism

L-Valine is produced by a non-genetically modified strain of *C. glutamicum* which is deposited in the China General Microbiological Culture Collection Center with accession number CGMCC 7.366.[10]

The taxonomic identification of the production strain CGMCC 7.366 as *C. glutamicum* was confirmed by analysis of the whole genome sequence (WGS). [redacted][11]

The production strain is derived [redacted] the mutant strain producing the highest level of L-valine was selected and named/deposited as CGMCC 7.366.[12]

The susceptibility of the production strain to the battery of antibiotics recommended by the FEEDAP Panel (EFSA FEEDAP Panel, 2018a,b) was tested by broth microdilution following the method of the Clinical and Laboratory Standards Institute (CLSI). All the minimum inhibitory concentration (MIC) values were equal to or fell below the corresponding cut-off values for *Corynebacterium* and other Gram-positives (EFSA FEEDAP Panel, 2018a).[13] Therefore, the production strain is considered susceptible to all relevant antibiotics.

[8] The full report is available on the EURL website: https://ec.europa.eu/jrc/sites/jrcsh/files/finrep_fad-2020-0033_l-valine.pdf
[9] 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.
[10] Technical Dossier/Section II/Annex 2.2.1.2.a.
[11] Technical Dossier/Section II/Annex 2.2.1.2.c and Annex 2.2.1.2.d.
[12] Technical Dossier/Section II/Annex 2.2.1.2.b
[13] Technical Dossier/Section II/Annex 2.2.2.2.

The WGS of the production strain was interrogated for the presence of antimicrobial resistance (AMR) genes [redacted] No genes of concern were identified.[14]

3.1.2. Manufacturing process

L-Valine is produced by fermentation using *C. glutamicum* CGMCC 7.366. [redacted][15]

The applicant stated that no antimicrobials are used during the manufacturing process of the additive.[16]

3.1.3. Characterisation of the active substance/additive

L-Valine (International Union of Pure and Applied Chemistry (IUPAC)) name: (2*S*)-2-amino-3-methylbutanoic acid; synonyms: α-amino isovaleric acid, 2-amino-3-methylbutyric acid), a compound identified by Chemical Abstracts Service (CAS) No 72-18-4 and European Inventory of Existing Commercial Chemical Substances (EINECS) No 200-773-6, has a molecular weight of 117.15 g/mol; the molecular formula is $C_5H_{11}NO_2$ and its structural formula is given in Figure 1.

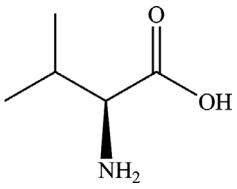

Figure 1: Molecular structure of L-valine

The additive is specified to contain ≥ 98% L-valine on a dry matter (DM) basis and ≤ 2.0% moisture.

The analysis of five batches showed an average of 99.4% valine (range 99.0–99.8%) on DM.[17] Moisture was on average 0.3% (range 0.1–0.6%).[18]

The specific optical rotation measured in three batches ranged from +27.5 to +28.2° which fall within the reference range (+26.5 to +29.0°) set in the European Pharmacopoeia (10th edition) (2020) and confirms the L-enantiomer of valine.[19]

3.1.3.1. Undesirable substances

Three batches of the additive were analysed for heavy metals (cadmium, lead, mercury) and arsenic. Lead and arsenic were < 10 μg/kg, and cadmium and mercury < 2 μg/kg in all batches. In relation to mycotoxins, aflatoxins B1, ochratoxin A, zearalenone, fumonisins (B1, B2 and B3), deoxynivalenol (DON) and citrinin were analysed in three batches of the additive and found to be below < 0.1 μg/kg, < 5.0 μg/kg, < 17.0 μg/kg, < 25.0 μg/kg, < 134.0 μg/kg, and < 15.0 μg/kg, respectively.[20] No clear information on the limit of detection/quantification (LOD/LOQ) of the methods have been provided.[21]

Three batches were analysed for polychlorinated dibenzodioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), dioxin-like polychlorinated biphenyls (DL-PCBs) and non DL-PCBs and the results were found below the LOD. The sum of dioxins in all the three batches calculated considering

[14] Technical Dossier/Section II/Annex 2.2.1.2.c.
[15] Technica Dossier/Section II/Annex 2.3.1.a and Supplementary Information November 2020/Annex 1.
[16] Technical Dossier/Section II/Annex 2.3.1.b.
[17] Technical Dossier/Section II/Annex 2.1.3. L-Valine analysed using the VDLUFA method.
[18] Technical dossier/Supplementary information November 2020/Annex 3 and FAD-2020-0033 Sin 110920 Answers.
[19] Technical Dossier/Section II/Annex 2.2.2.1.
[20] Technical Dossier/Section II/Annex 2.1.3.
[21] Technical Dossier/Supplementary Information August and November 2020.

the upper bond was 137 ng/kg for dioxins, the sum of DL-PCBs was 132 ng/kg, and the sum of dioxin and DL-PCBs was 269 ng/kg for wet weight.[22] The detected amounts of these undesirable substances do not raise safety concerns.

The microbial quality of the additive was analysed in three batches. *Salmonella* spp., Enterobacteriaceae, *Escherichia coli*, yeasts and filamentous fungi were not detected in 25-g samples.[23]

The presence of viable cells of the production strain was investigated in three batches of the additive analysed in triplicate. For each replicate a 10-g sample was diluted into 290 mL of 0.9% NaCl and 30 ml of this solution (corresponding to 1 g of the original sample) was passed through a 0.45 μm mixed cellulose esters (MCE) filter. After filtration, the membrane was transferred to a selective medium (128 mg/L fosfomycin) and incubated at 30°C for 6 days to recover possible stressed cells. Positive and negative controls were included. A total of forty colonies were recovered which were tested by polymerase chain reaction (PCR) and did not show the DNA fragment expected from the production strain. Therefore, the final product does not contain cells of the production strain.[24]

3.1.3.2. Physico-chemical properties

The additive is a white solid crystalline powder,[25] soluble in water (stated to be 54.3 g/L at 25°C, but not analytical data provided),[26] with a typical density of 500–600 kg/m^3.

The dusting potential was analysed by the Stauber–Heubach method in three batches of the final product obtaining values of 0.2 g/m^3 for two batches and 0.3 g/m^3 in the third one.[27]

The particle size distribution was measured in three batches of the additive by laser diffraction. The fractions of particles having diameters < 100 μm, < 50 μm and < 10 μm were 64%, 40% and 3% (w/v), respectively.[28]

3.1.3.3. Stability and homogeneity

The shelf life of the additive (three batches) was tested when stored at 25°C and 40°C in tightly closed bags protected from light for six months. On average, the losses were ≤ 1% after storage in both conditions.[20]

The stability of the additive (three batches) in a vitamin–mineral premixture containing choline chloride (92,000 mg/kg) was studied when supplemented with 4% valine. The samples were stored in sealed plastic bags protected from light at 25°C for 6 months.[26] Losses ranged from 0% to 8% depending on the batch considered.[29]

The stability of the additive (three batches) was studied in a complete feed (mash and pelleted, containing 0.94% background valine) for chicken for fattening when supplemented at 0.2%. The complete feed consisted of maize, wheat, soybean meal and rapeseed meal, and contained 460 mg/kg choline chloride. The pelleting process at 73°C originated a loss that ranged from 0% to 4% depending on the batch considered. Mash and pelleted feed were tested after storage for 3 months as described above. Losses observed in mash feed ranged from 0% to 10% while in pelleted feed ranged from 4% to 6%.[30]

The stability of three batches of the additive at a concentration of 0.2% (w/w) in water was tested at room temperature for 48 h. No losses were observed in two batches analysed, while the third one showed a loss of 1.5%.[20]

The capacity of the additive to distribute homogeneously in feed was studied in one of the batches of pelleted feed for chickens for fattening described above by analysing 10 subsamples. Total valine was analysed and the background valine of the diet subtracted from each subsample, resulting in a coefficient of variation of 12%.[31]

[22] Technical dossier/Section II/Annex 2.1.4.a1; Annex 2.1.4.a2 and Annex 2.1.4.a3.
[23] Technical Dossier/Section II/Annex 2.1.3. and FAD-2020-0033 Sin 110920 Answers.
[24] Technical dossier/Supplementary information November 2020/Annex 4a; Annex 4b; Annex 4c and Annex 4d.
[25] Technical dossier/Section II/Annex 2.5.2.
[26] Technical dossier/Supplementary information August 2020.
[27] Technical dossier/Section II/Annex 2.1.5.d.
[28] Technical dossier/Section II/Annex 2.1.5.a1; Annex 2.1.5.a2; Annex 2.1.5.b1; Annex 2.1.5.b2; Annex 2.1.5.c1 and Annex 2.1.5.c2.
[29] Technical dossier/Section II/Annex 2.4.1.a and Annex 2.4.1.c.
[30] Technical dossier/Section II/Annex 2.4.1.b and Annex 2.4.1.c.
[31] Technical dossier/Section II/Annex 2.4.1.b for diet composition, and Annex 2.4.1.c for analytical results.

3.1.4. Conditions of use

L-Valine is intended to be used in feeds to achieve an adequate amino acid profile and to meet the L-valine requirements for all animal species. It can be added directly to complete feed, water, complementary feed or it can be supplemented via a premixture. No inclusion levels have been proposed, as the requirements, in quantitative terms, depend on the species, the physiological state of the animal, the performance level, the environmental conditions and the amino acid composition of the un-supplemented diet.

3.2. Safety

3.2.1. Safety for the target species, consumers and environment

The additive is highly purified (contains > 98% L-valine on DM basis). The use of the amino acid 'per se' will not raise safety concerns for the target animals provided it is supplemented in appropriate amounts to the diets. Safety concerns from the additive could derive from the residues of the fermentation process/production strain remaining in the final product. The production strain belongs to a species, *C. glutamicum*, that qualifies for the qualified presumption of safety (QPS) approach to safety assessment (EFSA, 2007) when used for production purposes (EFSA BIOHAZ Panel, 2020). The strain was unambiguously identified as *C. glutamicum* and was shown not to harbour acquired antimicrobial resistance determinants for antibiotics of human and veterinary importance, thus meeting the QPS requirements. The final product does not contain viable cells of the production strain. Consequently, no safety concerns for target animal, consumers and the environment are expected from the additive concerning the production strain and the fermentation residues that may be present in the final additive.

L-Valine requirements of different species (non-ruminant and ruminant) and animal categories, absorption and metabolic fate of L-valine, and tolerance to L-valine excess in the diet were described in previous opinions (EFSA FEEDAP Panel, 2013, 2014a). The FEEDAP Panel has concerns on the use of amino acids in water for drinking for hygienic reasons, and due to the risk of imbalances when administered simultaneously via feed (EFSA FEEDAP Panel, 2010).

The amino acid L-valine, supplemented to feed, will be incorporated into proteins of tissues and/or products of animal origin and any of their potential excess will be metabolised and excreted as urea/ uric acid and carbon dioxide. Therefore, the composition of tissues and products of animal origin will not be affected by the use of L-valine in animal nutrition.

The amino acid L-valine is a physiological and natural component of the proteins of living organisms. When consumed, it will be absorbed, and the non-absorbed fraction will be incorporated into the intestinal microbial mass and excreted as such. The use of amino acids in water for drinking, when given in addition to complete diets with a well-balanced amino acid profile, would disturb the nitrogen balance and increase nitrogen excretion via urine. The use of the additive in animal nutrition would not lead to any localised increase in the concentration of l-valine or its metabolites in the environment.

3.2.2. Conclusions on the safety for the target species, consumer and the environment

The FEEDAP Panel concludes that L-valine produced using *C. glutamicum* CGMCC 7.366 is safe for the target species when supplemented in appropriate amounts to the diet according to the nutritional needs of the target species. The FEEDAP Panel has concerns on the use of amino acids in water for drinking for hygienic reasons, and due to the risk of imbalances when administered simultaneously via feed.

The use of L-valine produced using *C. glutamicum* CGMCC 7.366 in animal nutrition is considered safe for the consumer and for the environment.

3.2.3. Safety for the user

The additive under assessment is a powder with a dusting potential up to 0.3 g/m^3 and containing a fraction of inhalable particles (< 100 μm diameter) up to 64%. Therefore, inhalation exposure of users is possible.

No specific data on inhalation toxicity, skin/eye irritation or skin sensitisation were provided for the additive under application.

In the absence of data, the FEEDAP Panel cannot conclude on the potential of the additive to be toxic by inhalation, irritant to skin or eyes, or on its potential to be a dermal sensitiser.

3.3. Efficacy

Efficacy studies are not required for amino acids that occur naturally in plant and animal proteins. The nutritional role of the amino acid L-valine is well established in the scientific literature. The product L-valine produced by fermentation using *C. glutamicum* CGMCC 7.366 is regarded as an efficacious source of the essential amino acid L-valine for non-ruminant nutrition. The Panel indicated in a previous opinion (EFSA FEEDAP Panel, 2013) that ruminal degradation would reduce the delivery of the amino acid to the abomasum, and that protective measures should be considered.

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[32] and Good Manufacturing Practice.

4. Conclusions

The FEEDAP Panel concludes that L-valine produced using *C. glutamicum* CGMCC 7.366 is safe for the target species when supplemented in appropriate amounts to the diet according to the nutritional needs of the target species. The FEEDAP Panel has concerns on the use of amino acids in water for drinking for hygienic reasons, and due to the risk of imbalances when administered simultaneously via feed.

The use of L-valine produced using *C. glutamicum* CGMCC 7.366 in animal nutrition is considered safe for the consumer and for the environment.

The FEEDAP Panel cannot conclude on the potential of L-valine produced using *C. glutamicum* CGMCC 7.366 to be toxic by inhalation, irritant to the skin or eyes, or a dermal sensitiser due to the lack of data.

The additive L-valine produced by fermentation using *C. glutamicum* CGMCC 7.366 is regarded as an efficacious source of the essential amino acid L-valine for non-ruminant nutrition. For the supplemental L-valine to be as efficacious in ruminants as in non-ruminant species, it requires protection against degradation in the rumen.

5. Documentation as provided to EFSA/Chronology

Date	Event
06/05/2020	Reception mandate from the European Commission
08/05/2020	Dossier received by EFSA. L-valine for all animal species. Submitted by Ningxia Eppen Biotech Co., Ltd. represented in EU by Welding GmbH & Co. KG.
22/06/2020	Application validated by EFSA – Start of the scientific assessment
23/07/2020	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 of the additive*
20/08/2020	Reception of supplementary information from the applicant - Scientific assessment re-started
07/09/2020	Reception of the Evaluation report of the European Union Reference Laboratory for Feed Additives
11/09/2020	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 of the additive*
23/09/2020	Comments received from Member States
23/11/2020	Reception of supplementary information from the applicant - Scientific assessment re-started
17/03/2021	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

[32] 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.

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Abbreviations

CAS	Chemical Abstracts Service
CFU	colony forming unit
CGMCC	China General Microbiological Culture Collection Center
CIR	Cosmetic Ingredient Review
CV	coefficient of variation
DM	dry matter
EINECS	European Inventory of Existing Commercial Chemical Substances
EURL	European Union Reference Laboratory
FCC	Food Chemical Codex
FEEDAP	EFSA Panel on Additives and Products or Substances used in Animal Feed
IEC-VIS	ion exchange chromatography coupled with post-column derivatisation and photometric detection
IUPAC	International Union of Pure and Applied Chemistry
LOD	limit of detection
LOQ	limit of quantification

MIC	minimum inhibitory concentration
PCB	polychlorinated biphenyl
PCDD/F	polychlorinated dibenzodioxin/dibenzofuran
RH	relative humidity
RSDr	Relative standard deviation for repeatability
RSDR	Relative standard deviation for reproducibility
WGS	whole genome sequence

Annex A – Executive Summary of the Evaluation Report of the European Union Reference Laboratory for Feed Additives on the Method(s) of Analysis for L-valine produced by fermentation with *Corynebacterium glutamicum* CGMCC 7.366

In the current application an authorisation is sought under Article 4(1) for L-valine produced by fermentation with *Corynebacterium glutamicum* CGMCC 7.366, under the category/functional groups 3 (c) 'nutritional additives'/'amino acids, their salts and analogues', according to Annex I of Regulation (EC) No 1831/2003. Specifically, the authorisation is sought for all animal species.

According to the Applicant, L-valine has a minimum purity (mass fraction) of 98%. The feed additive is intended to be mixed either into premixtures, incorporated through complementary feed or added directly to feedingstuffs or water for drinking. However, the Applicant did not propose any minimum or maximum content of L-valine in feedingstuffs.

For the characterisation of the feed additive, the EURL found the "L-valine monograph" of the Food Chemical Codex (FCC), where different tests (including an optical rotation) are used for the identification of L-valine.

For the quantification of valine in the feed additive, premixtures, feedingstuffs and water the Applicant submitted the ring-trial validated European Union (EU) method based on ion-exchange chromatography coupled to post-column derivatisation and photometric detection (IEC-VIS). The method does not distinguish between the salts of amino acids and cannot differentiate between enantiomers. The following performance characteristics were reported for the quantification of total valine in feed: a relative standard deviation for repeatability (RSDr) ranging from 1.7 to 3.8% and a relative standard deviation for reproducibility (RSDR) ranging from 8.8 to 16.1%. In addition, in the frame of the batch-to-batch analysis and for the stability studies of valine in the feed additive and water, the Applicant presented acceptable experimental data when applying the above mentioned EU method.

In the frame of this authorisation the EURL recommends for official control (i) the "L-valine monograph" of the Food Chemical Codex (FCC) for the identification of L-valine in the feed additive, and (ii) the ring-trial validated European Union method based on IEC-VIS for the quantification of valine in the feed additive, premixtures, feedingstuffs and water.

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, as last amended by Regulation (EU) 2015/1761) is not considered necessary.