Safety and efficacy of a feed additive consisting of *Lactiplantibacillus plantarum* (formerly *Lactobacillus plantarum*) IMI 507026 for all animal species (ALL-TECHNOLOGY (IRELAND) LIMITED [Alltech Ireland])

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 Fašmon Durjava, Maryline Koubé, Marta López-Alonso, Secundino López Puente, Francesca Marcon, Baltasar Mayo, Alena Pechová, Mariana Petkova, Fernando Ramos, Yolanda Sanz, Roberto Edoardo Villa, Ruud Woutersen, Pier Sandro Cocconcelli, Boet Glandorf, Miguel Prieto Maradona, Maria Saarela, Rosella Brozzi, Jaume Galobart, Matteo Innocenti and Joana Revez

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 *Lactiplantibacillus plantarum* (formerly *Lactobacillus plantarum*) IMI 507026 as a technological additive for all animal species. The additive is intended to improve the production of silage at a proposed application rate of $1 \times 10^9$ colony forming units (CFU)/kg fresh material. The bacterial species *L. plantarum* is considered by EFSA to be suitable for the qualified presumption of safety approach. As the identity of the strain has been established and no antimicrobial resistance determinants of concern were detected, the use of the strain as a silage additive is considered safe for livestock species, for consumers and for the environment. In the absence of data, the FEEDAP Panel cannot conclude on the potential of the additive to be a skin/eye irritant or a skin sensitiser. Given the proteinaceous nature of the active agent, the additive should be considered a respiratory sensitiser. The additive at the proposed application rate of $1 \times 10^9$ CFU/kg fresh material has the potential to improve the fermentation of the silages from easy to moderately difficult to ensile forages.

© 2021 European Food Safety Authority. *EFSA Journal* published by John Wiley and Sons Ltd on behalf of European Food Safety Authority.

**Keywords:** technological additive, silage additive, *Lactiplantibacillus plantarum* IMI 507026, safety, efficacy, QPS

**Requestor:** European Commission

**Question number:** EFSA-Q-2020-00695

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

**Legal notice:** Relevant information or parts of this scientific output have been blackened in accordance with the confidentiality requests formulated by the applicant pending a decision thereon by the European Commission. The full output has been shared with the European Commission, EU Member States and the applicant. The blackening will be subject to review once the decision on the confidentiality requests is adopted by the European Commission.

**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](https://ess.efsa.europa.eu/doi/doiweb/doisearch).

**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, Cocconcelli PS, Glandorf B, Maradona MP, Saarela M, Brozzi R, Galobart J, Innocenti M and Reve J, 2021. Scientific Opinion on the safety and efficacy of a feed additive consisting of *Lactiplantibacillus plantarum* (formerly *Lactobacillus plantarum*) IMI 507026 for all animal species (ALL-TECHNOLOGY (IRELAND) LIMITED [Alltech Ireland]). EFSA Journal 2021;19(7):6703, 10 pp. [https://doi.org/10.2903/j.efsa.2021.6703](https://doi.org/10.2903/j.efsa.2021.6703)

**ISSN:** 1831-4732

© 2021 European Food Safety Authority. *EFSA Journal* published by John Wiley and Sons Ltd on behalf of European Food Safety Authority.

This is an open access article under the terms of the Creative Commons Attribution-NoDerivs License, which permits use and distribution in any medium, provided the original work is properly cited and no modifications or adaptations are made.

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

Abstract................................................................................................................................................... 1
1. Introduction................................................................................................................................4
1.1. Background and Terms of Reference as provided by the requestor.................................................. 4
1.2. Additional information.................................................................................................................. 4
2. Data and methodologies.............................................................................................................. 4
2.1. Data........................................................................................................................................... 4
2.2. Methodologies............................................................................................................................. 4
3. Assessment................................................................................................................................. 5
3.1. Characterisation .......................................................................................................................... 5
3.1.1. Characterisation of the active agent.............................................................................................. 5
3.1.2. Characterisation of the additive.................................................................................................... 5
3.1.3. Stability ...................................................................................................................................... 6
3.1.4. Conditions of use ........................................................................................................................ 6
3.2. Safety ........................................................................................................................................ 6
3.2.1. Safety for the target species, consumer and the environment......................................................... 6
3.2.2. Safety for user ............................................................................................................................ 6
3.2.2.1. Conclusions on safety for user...................................................................................................... 6
3.3. Efficacy ..................................................................................................................................... 6
3.3.1. Conclusions on efficacy ................................................................................................................ 8
4. Conclusions................................................................................................................................... 8
5. Documentation as provided to EFSA/Chronology............................................................................ 8
References............................................................................................................................................... 8
Abbreviations ........................................................................................................................................... 9
Annex A – Executive Summary of the Evaluation Report of the European Union Reference Laboratory for Feed Additives on the Method(s) of Analysis for Lactobacillus plantarum IMI 507026 ........................................... 10
1. Introduction

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

Regulation (EC) No 1831/2003¹ 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 ALL-TECHNOLOGY (IRELAND) LIMITED [Alltech Ireland]² for the authorisation of the product Lactiplantibacillus plantarum (formerly Lactobacillus plantarum) IMI 507026, when used as a feed additive for all animal species (category: technological additives; functional group: silage 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 7 January 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 Lactiplantibacillus plantarum IMI 507026, when used under the proposed conditions of use (see Section 3.1.4).

1.2. Additional information

The additive is a preparation containing viable cells of Lactiplantibacillus plantarum (formerly Lactobacillus plantarum) IMI 507026. 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 dossier³ in support of the authorisation request for the use of Lactiplantibacillus plantarum IMI 507026 as a feed additive.

EFSA has verified the European Union Reference Laboratory (EURL) report as it relates to the methods used for the control of the active agent in animal feed. The Executive Summary of the EURL report can be found in Annex A.⁴

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of Lactiplantibacillus plantarum IMI 507026 is in line with the principles laid down in Regulation (EC) No 429/2008⁵ and the relevant guidance documents: Guidance on the characterisation of microorganisms used as feed additives or as production organisms (EFSA FEEDAP Panel, 2018a), Guidance on the identity, characterisation and conditions of use of feed 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 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, 2019) and Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018b).

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 ALL-TECHNOLOGY (IRELAND) LIMITED [Alltech Ireland], Sarney, Summerhill Rd., A86X006 Dunboyne, Co. Meath, Ireland.
³ FEED dossier reference: FAD-2020-0078.
⁴ The full report is available on the EURL website: https://ec.europa.eu/jrc/sites/jrcsh/files/finrep-fad-2020-0075-0078-0079-0080-lactobacilli.pdf
⁵ 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.
3. Assessment
The product under assessment is a preparation of viable cells of *Lactiplantibacillus plantarum* IMI 507026 intended for use as a technological additive (functional group: silage additives) in easy and moderately difficult to ensile forages for all animal species.

3.1. Characterisation

3.1.1. Characterisation of the active agent

The active agent was isolated from maize silage. It is deposited in the Centre for Agriculture and Bioscience International culture collection, formerly International Mycological Institute, CABI-IMI Culture collection, with the accession number IMI 507026. It has not been genetically modified.

Taxonomical identification was confirmed with alignment-free genome distance estimation with Mash using MinHash and OrthoANI to calculate *in silico* average nucleotide identity (ANI) based on the whole genome sequence (WGS). Results showed that *L. plantarum* JDM1 was the closest matching NCBI RefSeq genome with a Mash distance of 0.00016 and an OrthoANI value of 99.98%. In addition, the strain *L. plantarum* IMI 507026 has a calculated OrthoANI value of 99.15% with the type strain of this species, *L. plantarum* ATCC 14917T.

The bacterial strain was tested for antibiotic susceptibility using a broth microdilution method. The battery of antibiotics used included those recommended by EFSA (EFSA FEEDAP Panel, 2018a). All the minimum inhibitory concentration values were equal or fell below the corresponding cut-off values for *L. plantarum*. Therefore, the strain is considered to be susceptible to all the relevant antibiotics.

The whole genome sequence of the strain was searched for antibiotic resistance genes using the ABRicate tool with thresholds of 70% for both identity and coverage, at nucleotide and protein level. The databases used were ARG-ANNOT, ecOH, MEGARes, NCBI Bacterial Antimicrobial Resistance Reference Gene Database and ResFinder. No hits of concern were identified.

3.1.2. Characterisation of the additive

The inoculum of the active agent is prepared to guarantee a minimum concentration of active agent of $1 \times 10^{10}$ CFU/g of additive.

Analysis of five batches showed viable lactic acid bacteria counts exceeding the specifications (mean: $1.0 \times 10^{12}$ CFU/g, range: $8.2 - 12 \times 10^{10}$ CFU/g).

A total of four batches were analysed for microbiological contamination and mycotoxins, heavy metals and arsenic concentrations. Regarding the specifications for the microbiological purity, limits are set for total coliforms (1,000 CFU/g), β-glucuronidase-positive *Escherichia coli* (100 CFU/g), coagulase-positive staphylococci (including *Staphylococcus aureus*) (10 CFU/g), Salmonella spp. (no detection in 25 g), *Listeria monocytogenes* (no detection in 25 g), *Clostridium perfringens* (100 CFU/g), anaerobic sulphite reducers (100 CFU/g), yeasts and filamentous fungi (1,000 CFU/g). Analysis of four batches of the additive showed compliance with these limits. The same batches were tested for the following mycotoxins which were not detected: aflatoxins (B1, B2, G1, and G2), deoxynivalenol, zearalenone, ochratoxin A, fumonisins B1+B2, HT-2 toxin and T-2 toxin. Average values of the analyses of the heavy metals and arsenic were: 0.042 mg Pb/kg (range 0.024–0.060), 0.007 mg Hg/kg (range 0–0.011) and 0.052 mg As/kg (range 0–0.077). Cadmium was not detected. The levels of the detected impurities do not raise concerns.

The additive is a powder with an average density of 1,259 kg/m³ (range: 1,257–1,262 kg/m³) and an average bulk density of 363 kg/m³ (range: 359–365 kg/m³). The dusting potential of the additive was measured in three batches (Stauber–Heubach) and showed a mean value of 5.57 g/m³ air (range: 6.0–7.0 g/m³ air).

---

6 Technical dossier/Section II/Annex II 2.4.
7 Technical dossier/Section II/Annex II 2.3.
8 Technical dossier/Section II/Annex II 2.8.
9 Technical dossier/Section II/Annex II 1.2.
10 Technical dossier/Section II/Annex II 1.3.
11 Technical dossier/Section II/Annex II 1.3 with the following Limits of Detection: aflatoxins (B1, B2, G1, and G2): 1 μg/kg, deoxynivalenol 20 μg/kg, zearalenone (10 μg/kg), ochratoxin A (0.50 μg/kg), fumonisins B1 and B2 (10 μg/kg), HT-2 toxin (5.0 μg/kg), T-2 toxin (2.5 μg/kg), Pb (0.0017 mg/kg), Hg (0.0017 mg/kg), Cd (0.0017 mg/kg) and As (0.0067 mg/kg).
4.81–6.17 g/m³). The same three batches were tested for particle size distribution by laser diffraction. Results showed that ~37% of the additive consists of particles with diameters below 100 μm, 28% below 50 μm and 10% below 10 μm.12

3.1.3. Stability

Four batches of the additive were tested for shelf-life by storing in sealed aluminium foil bags at 4°C for 3 months13 and 25°C/60% relative humidity (RH) for 2 months.14 Negligible losses (< 0.5 log of the initial value) were observed under the aforementioned conditions.

The stability in water was studied by suspending 1 g of the additive (one batch) in 1 L of water and then storing for 48 h at 4°C and 20°C. Negligible losses were observed for both temperatures tested, with < 0.5 log of the initial value.15

3.1.4. Conditions of use

The additive is intended for use in easy and moderately difficult to ensile forages at a proposed minimum inclusion level of 1 × 10⁹ CFU/kg fresh material for all animal species.16 It is to be applied as such or as an aqueous suspension.

3.2. Safety

3.2.1. Safety for the target species, consumer and the environment

The species L. plantarum is considered by EFSA to be suitable for the Qualified Presumption of Safety (QPS) approach to safety assessment (EFSA, 2007, EFSA BIOHAZ Panel, 2020). This approach requires the identity of the strain to be conclusively established and evidence that the strain lacks acquired determinants for resistance to antibiotics of human and veterinary importance. In the view of the FEEDAP Panel, the identity of the strain was established as L. plantarum and the antibiotic resistance qualification has been met. Consequently, L. plantarum IMI 507026 is presumed safe for the target species, consumers and the environment.

3.2.2. Safety for user

No studies were submitted regarding the effects of the additive to the respiratory tract, skin or eyes.

The dusting potential reported is high (5.57 g/m³), thus exposure by inhalation is possible. Owing to the proteinaceous nature of the active agent, the additive should be considered a respiratory sensitiser.

Once an active agent has been authorised as a silage additive, different formulations can be placed on the market with reference to that authorisation. For this specific product, the excipients used in the preparation of the final formulation are not expected to introduce additional risks.

3.2.2.1. Conclusions on safety for user

The additive should be considered a respiratory sensitiser. No conclusions can be drawn on the potential of the additive to cause skin and eye irritation or skin sensitisation.

3.3. Efficacy

Five laboratory studies were conducted with forages representing materials easy to ensile (study 1)17 and moderately difficult to ensile (studies 2,17 3,17 418 and 517) as specified by Regulation (EC) No 429/2008 (Table 1). All the studies included a control and a group in which L. plantarum IMI 507026 was applied to the forage at a concentration of 1 × 10⁹ CFU/kg of fresh forage. Analytical confirmation of the counts of the two batches of the additive used for the studies were provided. An
aqueous suspension of the additive was prepared and then sprayed onto the forage prior to ensiling. In the control silos, the same volume of water was added, but without the additive. In studies 1-3 and 5, the forage was ensiled for 90 days in mini-silos (five replicates per treatment) with a capacity of 1.75 L with the potential to vent gas. In study 4, the forage was ensiled for 100 days in mini-silos (four replicates per treatment) with a capacity of 20 L and a device to vent gas. All experiments were conducted at 20 ± 1°C.

After 90 days (or 100 days for study 4), the silos were opened and the contents were analysed for dry matter (DM), pH, lactic, acetic and propionic acids and ethanol concentrations, and ammonia. Aerobic stability was assessed at the end of each experiment by taking samples from each silo and exposing to air with continuous monitoring of temperature. A rise of 3°C above room temperature was considered as indicator of silage deterioration, and the time at which that rise was observed was taken as a measure of the aerobic stability of treated and control silages. A minimum increase of stability of the treated silage of two days compared to that shown by the untreated control is considered as evidence of aerobic stability.

Data were analysed using the non-parametric Wilcoxon Signed-Rank test (studies 1, 2, 3 and 5) or Mann-Whitney test (study 4) and significance declared at p < 0.05. Results are shown in Table 2.

The addition of the additive resulted in a significant reduction of the dry matter loss and ammonia-N production and increased lactic acid concentration in four out of the five studies. Regarding the aerobic stability, only in study 4 a positive outcome was observed but the difference was below the 48 h threshold.
3.3.1. Conclusions on efficacy

The use of *L. plantarum* IMI 507026 at the proposed inclusion rate has the potential to improve the production of silage with easy and moderately difficult to ensile materials by enhancing the preservation of nutrients.

4. Conclusions

Based on the QPS approach to safety assessment, *Lactiplantibacillus plantarum* IMI 507026 is presumed safe for the target species, consumers and the environment.

The additive should be considered a respiratory sensitiser. No conclusions can be drawn on the eye and skin irritancy, or skin sensitisation potential of the additive.

*L. plantarum* IMI 507026 at a concentration of $1 \times 10^9$ CFU/kg plant material has the potential to improve the preservation of nutrients in silage prepared with easy and moderately difficult to ensile material.

5. Documentation as provided to EFSA/Chronology

| Date       | Event                                                                 |
|------------|-----------------------------------------------------------------------|
| 14/10/2020 | Dossier received by EFSA. *L. plantarum* IMI 507026. Submitted by ALL-TECHNOLOGY (IRELAND) LIMITED [Alltech Ireland] |
| 20/10/2020 | Reception mandate from the European Commission                         |
| 08/01/2021 | Application validated by EFSA – Start of the scientific assessment     |
| 11/02/2021 | 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 and efficacy* |
| 08/03/2021 | Reception of the Evaluation report of the European Union Reference Laboratory for Feed Additives |
| 07/04/2021 | Reception of supplementary information from the applicant - Scientific assessment re-started |
| 09/04/2021 | Comments received from Member States                                  |
| 23/06/2021 | Opinion adopted by the FEEDAP Panel. End of the Scientific assessment  |

References

EFSA (European Food Safety Authority), 2007. Opinion of the Scientific Committee on a request from EFSA on the introduction of a Qualified Presumption of Safety (QPS) approach for assessment of selected microorganisms referred to EFSA. EFSA Journal 2007;5(12):587, 16 pp. https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2007.587

EFSA BIOHAZ Panel (EFSA Panel on Biological Hazards), Koutsoumanis K, Allende A, Alvarez-Ordóñez A, Bolton D, Bover-Cid S, Chemaly M, Davies R, De Cesare A, Hilbert F, Lindqvist R, Nauta M, Peixe L, Ru G, Simmons M, Skandamis P, Suffredini E, Cocconcelli PS, Fernández Escámez PS, Maradona MP, Querol A, Suarez JE, Sundh I, Vlak J, Barizzone F, Correia S and Herman L, 2020. Scientific Opinion on the update of the list of QPS-recommended biological agents intentionally added to food or feed as notified to EFSA (2017–2019). EFSA Journal 2020;18(2):5966, 56 pp. https://doi.org/10.2903/j.efsa.2020.5966

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

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Gropp J, Kolar B, Kouba M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Anguita M, Galobart J and Innocenti ML, 2017a. Guidance on the identity, characterisation and conditions of use of feed additives. EFSA Journal 2017a;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), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Gropp J, Kolar B, Kouba M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Anguita M, Galobart J, Innocenti ML and Martino L, 2017b. Guidance on the assessment of the safety of feed additives for the target species. EFSA Journal 2017b;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), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Gropp J, Kolar B, Koubka M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Glandorf B, Herman L, Kärenlampi S, Aguilera J, Anguita M, Brozzi R and Galobart J, 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), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Gropp J, Kolar B, Koubka M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Anguita M, Galobart J, Innocenti ML and Martino L, 2018b. Guidance on the assessment of the efficacy of feed additives. EFSA Journal 2018;16(5):5274, 25 pp. https://doi.org/10.2903/j.efsa.2018.5274

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Bampidis V, Bastos ML, Christensen H, Dusemund B, Kouba M, Kos Durjava M, López-Alonso M, López Puente S, Marcon F, Mayo B, Pechová A, Petkova M, Ramos F, Sanz Y, Villa RE, Woutersen R, Brock T, Knecht J, Kolar B, Beelen P, Padovani L, Tarrés-Call J, Vettori MV and Azimonti G, 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

Abbreviations

ANI average nucleotide identity
CFU colony forming unit
CV coefficient of variation
DM dry matter
EURL European Union Reference Laboratory
FEEDAP EFSA Panel on Additives and Products or Substances used in Animal Feed
MIC minimum inhibitory concentration
PFGE pulsed-field gel electrophoresis
QPS qualified presumption of safety
RH relative humidity
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 \textit{Lactobacillus plantarum} IMI 507026

In the current applications authorisations are sought under Article 4(1) for \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028 under the category/functional group 1(k) “technological additives”/ “silage additives”, according to Annex I of Regulation (EC) No 1831/2003. The authorisations are sought for the use of the feed additives for all animal species.

According to the Applicant, the feed additives contain as active substance viable cells of \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028, respectively. The feed additives are to be marketed as preparations containing a minimum content of $1 \times 10^{10}$ Colony Forming Units (CFU) of \textit{Lactobacillus rhamnosus} IMI 507023 or \textit{Lactobacillus plantarum} IMI 507026 or \textit{Lactobacillus plantarum} IMI 507027 or \textit{Lactobacillus plantarum} IMI 507028/g in the respective feed additive. The feed additives are intended to be used at a minimum dose of $1 \times 10^{6}$ CFU/kg fresh silage.

For the identification of \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028 the EURL recommends for official control Pulsed-Field Gel Electrophoresis (PFGE), a recognised methodology for the genetic identification of bacterial strains.

For the enumeration of \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028 in the feed additives the EURL recommends for official control the ring-trial validated spread plate method EN 15787.

Since the unambiguous determination of the content of \textit{Lactobacillus rhamnosus} IMI 507023 or \textit{Lactobacillus plantarum} IMI 507026 or \textit{Lactobacillus plantarum} IMI 507027 or \textit{Lactobacillus plantarum} IMI 507028 initially added to silage is not experimentally achievable, the EURL is not able to evaluate or recommend any method for official control for the determination \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028 in silage.

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.

---

19 The EURL produced a combined report for \textit{Lactobacillus rhamnosus} IMI 507023, \textit{Lactobacillus plantarum} IMI 507026, \textit{Lactobacillus plantarum} IMI 507027 and \textit{Lactobacillus plantarum} IMI 507028.

20 During the assessment, the applicant clarified that under the conditions of use, the minimum dose is $1 \times 10^{6}$ CFU/kg (Technical dossier/Response to EFSA - FAD-2020-0078_SIn_110221).