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Assessment of the application for renewal of authorisation of endo-1,4- β -xylanase produced by *Aspergillus niger* CBS 109.713 and endo-1,4- β -glucanase produced by *Aspergillus niger* DSM 18404 for poultry species, ornamental birds and weaned piglets, from BASF SE

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Abstract

Natugrain[®] TS/TS L is the trade name of the feed additive under assessment and contains endo-1,4- β -xylanase and endo-1,4- β -glucanase produced by genetically modified strains of *Aspergillus niger*. The product is currently authorised for use as a feed additive for poultry species, ornamental birds, weaned piglets and pigs for fattening. This scientific opinion concerns the renewal of the authorisation of this additive for poultry species, ornamental birds and weaned piglets. The applicant provided evidence that the additive in the market complies with the conditions of the authorisation. According to the information provided by the applicant, no new evidence has been identified that would make the FEEDAP Panel reconsider the previous conclusions regarding the safety for the target species, consumer, user and environment under the authorised conditions of use. The additive is safe for the target species for which the renewal of the authorisation is requested, the consumers and the environment. The additive is a potential skin and a respiratory sensitiser. The present application for renewal of the authorisation did not include a proposal for amending or supplementing the conditions of the original authorisation that would have an impact on the efficacy of the additive. Therefore, there was no need for assessing the efficacy of the additive in the context of the renewal of the authorisation.

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Table of contents

Abstract.....	1
1. Introduction.....	4
1.1. Background and Terms of Reference.....	4
1.2. Additional information.....	4
2. Data and methodologies.....	5
2.1. Data.....	5
2.2. Methodologies.....	5
3. Assessment.....	5
3.1. Characterisation of the additive.....	5
3.1.1. Shelf-life of the additive.....	7
3.1.2. Conditions of use.....	7
3.2. Safety.....	7
3.3. Efficacy.....	8
3.4. Post-market monitoring.....	8
4. Conclusions.....	8
5. Documentation as provided to EFSA/Chronology.....	8
References.....	9
Abbreviations.....	9

1. Introduction

1.1. Background and Terms of Reference

Regulation (EC) No 1831/2003¹ establishes the rules governing the Community authorisation of additives for use in animal nutrition. In particular, Article 14(1) of that Regulation lays down that an application for renewal shall be sent to the Commission at the latest one year before the expiry date of the authorisation.

The European Commission received a request from BASF SE² for renewal of the authorisation of the additive that contains endo-1,4- β -xylanase produced by *Aspergillus niger* CBS 109.713 and endo-1,4- β -glucanase produced by *A. niger* DSM 18404, when used as a feed additive for poultry species, ornamental birds and weaned piglets (category: zootechnical additive; functional group: digestibility enhancers).

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 14(1) (renewal of the authorisation). 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 30 July 2018.

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 Natugrain[®] TS/TS L (endo-1,4- β -xylanase and endo-1,4- β -glucanase), when used under the proposed conditions of use (see Section 3.1.2).

1.2. Additional information

The feed additive contains endo-1,4- β -xylanase and endo-1,4- β -glucanase, produced by two genetically modified strains of *A. niger* (CBS 109.713 and DSM 18404, respectively) and its trade name is Natugrain[®] TS/TS L. The product is available in solid (Natugrain[®] TS) and liquid (Natugrain[®] TS L) forms. EFSA issued an opinion on the safety and efficacy of this additive when used in piglets (weaned), chickens for fattening, laying hens, turkeys for fattening and ducks for fattening, and including the assessment of the safety for the consumer, the user and the environment, as well as the safety aspects of the genetic modification (EFSA, 2008). Further opinions on the use of this product have been issued by EFSA: opinion on the use in other avian species (EFSA FEEDAP Panel, 2011), pigs for fattening (EFSA FEEDAP Panel, 2013a), modification on the terms of the authorisation in relation to the efficacy in laying hens (EFSA FEEDAP Panel, 2014) and in chickens reared for laying and for minor poultry species for laying (EFSA FEEDAP Panel, 2016), and lately on the use of the additive in sows (EFSA FEEDAP Panel, 2020).

The additive is currently authorised for poultry species and ornamental birds,^{3,4} as well as for weaned piglets⁵ and pigs for fattening.⁶ The applicant requested for the renewal of the authorisation of the product as a feed additive for poultry species, ornamental birds and weaned piglets.

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 BASF SE, G-ENA/MR 68623/Lampertheim, Germany.

³ Commission Regulation (EC) No 271/2009 of 2 April 2009 concerning the authorisation of a preparation of endo-1,4-beta-xylanase and endo-1,4-beta-glucanase as a feed additive for weaned piglets, chickens for fattening, laying hens, turkeys for fattening and ducks for fattening (holder of the authorisation BASF SE). OJ L 91, 2.4.2009, p. 5. Amended by Commission Implementing Regulation (EU) No 1070/2014 of 10 October 2013 amending Regulation (EC) No 271/2009 as regards the minimum content of the preparation of endo-1,4-beta-xylanase produced by *Aspergillus niger* (CBS 109.713) and endo-1,4-beta-glucanase produced by *Aspergillus niger* (DSM 18404) as a feed additive for laying hens (holder of authorisation BASF SE) OJ L 295, 11.10.2014, p. 49.

⁴ Commission Implementing Regulation (EU) No 1068/2011 of 21 October 2011 concerning the authorisation of an enzyme preparation of endo-1,4-beta-xylanase produced by *Aspergillus niger* (CBS 109.713) and endo-1,4-beta-glucanase produced by *Aspergillus niger* (DSM 18404) as a feed additive for chickens reared for laying, turkeys for breeding purposes, turkeys reared for breeding, other minor avian species (other than ducks for fattening) and ornamental birds (holder of authorisation BASF SE). OJ L 277, 22.10.2011, p. 11. Amended by Commission Implementing Regulation (EU) 2017/950 of 2 June 2017 amending Implementing Regulation (EU) No 1068/2011 as regards the minimum content of the preparation of endo-1,4-beta-xylanase produced by *Aspergillus niger* (CBS 109.713) and endo-1,4-beta-glucanase produced by *Aspergillus niger* (DSM 18404) as a feed additive for chickens reared for laying and all avian species for laying (holder of authorisation BASF SE) OJ L 143, 3.6.2017, p. 5.

⁵ Commission Regulation (EC) No 271/2009 of 2 April 2009 concerning the authorisation of a preparation of endo-1,4-beta-xylanase and endo-1,4-beta-glucanase as a feed additive for weaned piglets, chickens for fattening, laying hens, turkeys for fattening and ducks for fattening (holder of the authorisation BASF SE). OJ L 91, 2.4.2009, p. 5.

⁶ Commission Implementing Regulation (EU) No 1404/2013 of 20 December 2013 concerning the authorisation of a preparation of endo-1,4-beta-xylanase produced by *Aspergillus niger* (CBS 109.713) and endo-1,4-beta-glucanase produced by *Aspergillus niger* (DSM 18404) as a feed additive for pigs for fattening (holder of authorisation BASF SE) OJ L 349, 21.12.2013, p. 88.

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of a technical dossier⁷ in support of the authorisation request for the use of Natugrain[®] TS/TS L (endo-1,4- β -xylanase and endo-1,4- β -glucanase) as a feed additive.

The European Union Reference Laboratory (EURL) considered that the conclusions and recommendations reached in the previous assessment are valid and applicable for the current application.⁸

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety of Natugrain[®] TS/TS L (endo-1,4- β -xylanase and endo-1,4- β -glucanase) is in line with the principles laid down in Regulation (EC) No 429/2008⁹ and the relevant guidance documents: Guidance on the renewal of the authorisation of feed additives (EFSA FEEDAP Panel, 2013b) and Guidance on the characterisation of microorganisms used as feed additives or as production organisms (EFSA FEEDAP Panel, 2018).

3. Assessment

The additive under assessment contains endo-1,4- β -xylanase produced by *A. niger* CBS 109.713 and endo-1,4- β -glucanase produced by *A. niger* DSM 18404. In the text of this opinion the additive will be referred to as Natugrain[®] TS/TS L. This feed additive is authorised for use as a zootechnical additive in poultry species, ornamental birds, piglets and pigs for fattening. This opinion deals with the renewal of its authorisation as a zootechnical additive (functional group of digestibility enhancers) for poultry species, ornamental birds and weaned piglets.

3.1. Characterisation of the additive

The additive is presented in two different formulations, solid (Natugrain[®] TS) and liquid (Natugrain[®] TS L). The information submitted confirms that the manufacturing process is the same as the one described in the first assessment of the product (EFSA, 2008) and the applicant stated that no antimicrobial substances are used during the process.

The two formulations ensure a minimum activity level of 5,600 (xylanase) TXU¹⁰ /g and 2,500 (glucanase) TGU¹¹ /g.

Three batches of Natugrain[®] TS and three of Natugrain[®] TS L were analysed for chemical and microbiological contamination.¹⁴

⁷ FEED dossier reference: FAD-2018-0022.

⁸ The full report is available on the EURL website: <https://ec.europa.eu/jrc/en/eurl/feed-additives/evaluation-reports/fad-2010-0034?search&form-return>

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 Thermostable xylanase unit (TXU) is the amount of enzyme which liberates 5 micromole reducing sugars (xylose equivalents) from wheat arabinoxylan per minute at pH 3.5 and 40°C.

¹¹ Thermostable glucanase unit (TGU) is the amount of enzyme which liberates 1 micromole reducing sugars (glucose equivalents) from barley β -glucan per minute pH 3.5 and 40°C.

¹² Technical dossier/Section II/Annex II.3 and Supplementary information July 2019/Annexes 5 and 6.

¹³ Technical dossier/Section II/Annex II.3.

¹⁴ Technical dossier/Section II/Annex II.116a and b.

Further analytical data for mycotoxins and other secondary metabolites were provided for three batches of the two enzyme concentrates and of the liquid formulation, which showed contents all below the limits of detection (LODs).¹⁵

¹⁶

¹⁴

The two enzymes are produced by fermentation. The xylanase is produced with a genetically modified strain of *A. niger* which is deposited at the Centraalbureau voor Schimmelcultures (CBS) with the accession number CBS 109.713.¹⁷ The glucanase is produced with a genetically modified strain of *A. niger* which is deposited at the Deutsch Sammlung von Microorganismen und Zellkulturen (DSMZ) with the accession number DSM 18404.¹⁸

¹⁹

The assessment of the genetic modifications of the two production strains was done in a previous opinion (EFSA, 2008), and the FEEDAP and GMO Panels concluded that the genetic modifications do not raise any safety concern. The FEEDAP Panel is not aware of any further genetic modifications applied to the production strains and therefore considers that the conclusions drawn in the previous assessment are still valid.

The presence of viable cells and of DNA of the production strains was analysed

²⁰ No cells of the production strains were detected in any of the samples.

The presence of recombinant DNA of the two production strains was analysed

DNA was not detected in the samples, while positive controls performed as expected.

¹⁵ Technical dossier/Supplementary information July 2019/Annex 7. The analysis included (limit of detection): alternariol methyl ether (< 10 µg/kg), alternariol (< 10 µg/kg), aflatoxins (B1, B2, G1 and G2) (< 10 µg/kg), 3-acetyldeoxynivalenol (< 50 µg/kg), deoxynivalenol (< 100 µg/kg), diacetoxyscirpenol (< 50 µg/kg), fumonisin B1 (< 5 µg/kg), HT-2 Toxin (< 20 µg/kg), ochratoxin A (< 1 µg/kg), sterigmatocystin (< 1 µg/kg), T-2 toxin (< 20 µg/kg), zearalenol (< 50 µg/kg), zearalenone (< 5 µg/kg), fumonisin B2 (< 0.5 µg/kg), cyclopiazonic acid (< 25 µg/kg), roquefortine C (< 10 µg/kg), citrinin (< 10 µg/kg), mycophenolic acid (< 25 µg/kg), penitrem A (< 50 µg/kg) and penicillic acid (< 50 µg/kg).

¹⁶ Technical dossier/Section II/Annexes II.39 and II.40.

¹⁷ Technical dossier/Section II/Annex 115a.

¹⁸ Technical dossier/Section II/Annex 115b.

¹⁹ Technical dossier/Supplementary information June 2019/Annexes 1 and 3 for CBS 109.713 and Annexes 2 and 4 for DSM 18404.

²⁰ Technical dossier/Section II/Annex II.37a and II.37b and supplementary information June 2019 – general answer and annexes 8–13.

²¹ Technical dossier/Supplementary information June 2019/Annexes 14 and 15 and Supplementary information March 2020.

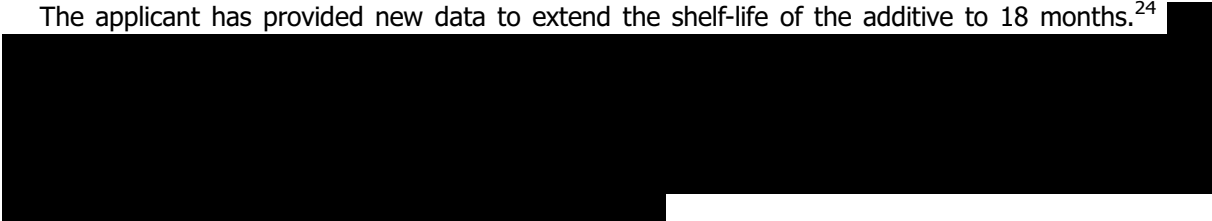


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3.1.1. Shelf-life of the additive

The applicant has provided new data to extend the shelf-life of the additive to 18 months.²⁴



3.1.2. Conditions of use


The additive is currently authorised for use in feed for chickens for fattening, ducks for fattening, laying hens, minor poultry species for fattening (other than ducks) and ornamental birds, chickens reared for laying and all minor poultry species for laying at a recommended level of 280–840 TXU and 125–375 TGU/kg complete feed. The additive is also authorised in feed for turkeys for fattening and turkeys for breeding purposes, turkeys reared for breeding and weaned piglets at 560–840 TXU and 250–375 TGU/kg complete feed. The applicant proposes to maintain the same conditions of use.

Under other provisions of the authorisation, it is stated that 'for safety: breathing protection, glasses and gloves shall be used during handling'.

3.2. Safety

Safety aspects regarding the use of this additive in feed including the safety of the genetic modification of the production strain, the safety for the target species, consumers, users and the environment have been previously assessed (EFSA, 2008). The Panel concluded that the additive is safe for the target species at the recommended levels, that there are no concerns for the consumers and that no risks for the environment are expected. The Panel also concluded that the additive in both forms is not irritant to eyes or skin, but it is a potential skin and respiratory sensitiser.

In line with the requirements established in the EFSA guidance on the renewal (EFSA FEEDAP Panel, 2013b), the applicant performed two literature searches to provide evidence that in the light of the current knowledge the additive remains safe under the approved conditions for target species, consumers, users and the environment.



²² Technical dossier/Section II/Annex II.112 and 113.

²³ Technical dossier/Section II/Annex II.114.

²⁴ Technical dossier/Section II/Annex II.46 and II.47.

²⁵ Technical dossier/Section III, Supplementary information July 2019/Annex 16 and Supplementary information July 2020/Annex 1, 2 and 3 and related annexes.

²⁶ Technical dossier/Supplementary information July 2020/Annex 3 to 23 for Glucanase/Annex 20.

²⁷ Technical dossier/Supplementary information July 2020/Annex 9.

██████████ The outcome of these two publications is in line with the conclusion from the Panel that the additive is a potential skin sensitiser and a respiratory sensitiser.

In view of the above and the fact that the manufacturing and composition of the additive have not been modified since the original authorisation, the FEEDAP Panel concludes that the additive remains safe for the target animals, consumers and the environment. The Panel confirms its previous conclusions that the additive in both forms is not irritant to eyes or skin but is a potential skin sensitiser and a respiratory sensitiser.

3.3. Efficacy

The present application for renewal of the authorisation does not include a proposal for amending or supplementing the conditions of the original authorisation that would have an impact on the efficacy of the additive. Therefore, there is no need for assessing the efficacy of the additive in the context of the renewal of the authorisation.

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 Regulation²⁸ and Good Manufacturing Practice.

4. Conclusions

The Panel concludes that the additive currently in the market complies with the existing conditions of authorisation.

The FEEDAP Panel confirms its previous conclusion that Natugrain[®] TS/TS L is safe for the target species/categories, consumers of products from animals fed the additive and the environment. The additive is to be considered a potential skin sensitiser and a respiratory sensitiser.

There is no need for assessing the efficacy of the additive in the context of the renewal of the authorisation.

5. Documentation as provided to EFSA/Chronology

Date	Event
04/05/2017	Dossier received by EFSA. Natugrain [®] TS, Natugrain [®] TS L (endo-1,4-beta-xylanase and endo-1,4-beta-glucanase) for piglets (weaned), laying hens, chickens for fattening, chickens reared for laying, turkeys for fattening, turkeys reared for breeding, turkeys for breeding purposes, ducks for fattening, all minor avian species for laying, minor poultry species for fattening (other than ducks for fattening) and ornamental birds. Submitted by BASF SE
18/05/2018	Reception of mandate from the European Commission
30/07/2018	Application validated by EFSA – Start of the scientific assessment
13/12/2018	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. <i>Issues: characterization and safety</i>
01/07/2019	Reception of supplementary information from the applicant - Scientific assessment re-started
18/08/2019	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. <i>Issues: characterization and safety</i>
30/03/2020	Reception of supplementary information from the applicant - Scientific assessment re-started
05/06/2020	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. <i>Issues: safety</i>
31/07/2020	Reception of supplementary information from the applicant - Scientific assessment re-started
18/11/2020	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

²⁸ Regulation (EC) No 1831/2003 of the European Parliament and of the Council of 22 October 2003 laying down requirements for feed hygiene. OJ L 35, 8.2.2005, p. 1.

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- EFSA (European Food Safety Authority), 2008. Scientific Opinion of the Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) and of the Panel on Genetically Modified Organisms (GMO) on the safety and efficacy of Natugrain® TS (endo-1,4- β -xylanase and endo-1,4- β -glucanase) as a feed additive for piglets (weaned), chickens for fattening, laying hens, turkeys for fattening and ducks. EFSA Journal 2008;6(12):914, 19 pp. <https://doi.org/10.2903/j.efsa.2008.914>
- EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2011. Scientific Opinion on the safety and efficacy of Natugrain® TS (endo-1,4-beta-xylanase and endo-1,4-beta-glucanase) as a feed additive for chickens reared for laying, turkeys for breeding purposes, turkeys reared for breeding, other minor avian species (other than ducks) and ornamental birds. EFSA Journal 2011;9(5):2172, 10 pp. <https://doi.org/10.2903/j.efsa.2011.2172>
- EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2013a. Scientific Opinion on the safety and efficacy of Natugrain® TS (endo-1,4-beta-xylanase and endo-1,4-beta-glucanase) as a feed additive for pigs for fattening. EFSA Journal 2013;11(7):3285, 10 pp. <https://doi.org/10.2903/j.efsa.2013.3285>
- EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2013b. Guidance on the renewal of the authorisation of feed additives. EFSA Journal 2013;11(10):3431, 8 pp. <https://doi.org/10.2903/j.efsa.2013.3431>
- EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2014. Scientific Opinion on the efficacy of Natugrain® TS/TS L (endo-1,4-beta-xylanase and endo-1,4-beta-glucanase) as a feed additive for laying hens. EFSA Journal 2014;12(6):3723, 9 pp. <https://doi.org/10.2903/j.efsa.2014.3723>
- EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), 2016. Scientific Opinion on the efficacy of Natugrain® TS/TS L (endo-1,4-beta-xylanase and endo-1,4-beta-glucanase) as a feed additive for chickens reared for laying and minor poultry species for laying. EFSA Journal 2016;14(11):4626, 7 pp. <https://doi.org/10.2903/j.efsa.2017.4626>
- 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, Glandorf B, Herman L, Kärenlampi S, Aguilera J, Anguita M, Brozzi R and Galobart J, 2018. 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>
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Abbreviations

CBS	Centraalbureau voor Schimmelcultures
DSMZ	Deutsch Sammlung von Microorganismen und Zellkulturen
EURL	European Union Reference Laboratory
FEEDAP	EFSA Panel on Additives and Products or Substances used in Animal Feed
LOD	limit of detection
TGU	thermostable glucanase unit
TXU	thermostable xylanase unit