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Safety and efficacy of a feed additive consisting of a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) for all poultry species (Biomin GmbH)

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Abstract

Following a request from the European Commission, EFSA was asked to deliver a scientific opinion on the safety and efficacy of a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) for all poultry species. The additive is authorised for use in feed for chickens for fattening, chickens reared for laying and minor poultry species reared to the point of lay. The safety and efficacy of the additive for those species have been previously evaluated by the EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP Panel). The current application is for an extension of use of the additive for all poultry species under the same conditions of use (recommended level ranging between 65 and 105 mg/kg complete feed). The FEEDAP Panel concluded that the additive Biomin® DC-P is safe for poultry species for fattening or reared for laying, but in the absence of adequate data cannot conclude on the safety for laying/breeding birds. The use of Biomin® DC-P in feed for all poultry species under the proposed conditions of use was considered safe for consumers and the environment. The FEEDAP Panel considered that exposure of users by inhalation is unlikely. In the absence of data, the Panel could not conclude on the effects of Biomin® DC-P on skin and eyes. Biomin® DC-P has the potential to be efficacious in poultry species for fattening or reared for laying when incorporated into feed at a minimum concentration of 65 mg/kg complete feed. In the absence of sufficient data, the Panel could not conclude on the efficacy for laying hens or for other poultry species for laying/breeding.

Keywords: zootechnical additives, other zootechnical additives, Biomin® DC-P, carvacrol, safety, efficacy, all poultry species

Requestor: European Commission

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Table of contents

1. Introduction

1.1. Background and Terms of Reference

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

The European Commission received a request from Biomin GmbH[2] for the authorisation of the additive consisting of a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) when used as a feed additive for all poultry species (category: zootechnical additives; functional group: other zootechnical additives).

According to Article 7(1) of Regulation (EC) No 1831/2003, the Commission forwarded the application to the European Food Safety Authority (EFSA) as an application under Article 4(1) (authorisation of a feed additive or new use of a feed additive). The particulars and documents in support of the application were considered valid by EFSA as of 8 March 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 preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P), when used under the proposed conditions of use (see Section 3.2.1).

1.2. Additional information

EFSA issued one opinion on the safety and efficacy of this product when used in feed for chickens for fattening, chickens reared for laying and minor poultry species reared to the point of lay (EFSA FEEDAP Panel, 2019a). The additive is currently authorised for use in those species and categories (4d20).[3]

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of a technical dossier[4] in support of the authorisation request for the use of a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) as a feed additive.

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.

EFSA has verified the European Union Reference Laboratory (EURL) report as it relates to the methods used for the control of the active substances in animal feed. The Executive Summary of the EURL report can be found in Annex A.[5]

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) is in line with the principles laid down in Regulation (EC) No 429/2008[6] and the relevant guidance documents: Guidance on studies concerning the safety of use of the additive for users/workers (EFSA FEEDAP Panel, 2012),

[1] Regulation (EC) No 1831/2003 of the European Parliament and of the council of 22 September 2003 on the additives for use in animal nutrition. OJ L 268, 18.10.2003, p. 29.
[2] Erber Campus 1, 3131, Getzersdorf, Austria.
[3] COMMISSION IMPLEMENTING REGULATION (EU) 2020/996 of 9 July 2020 concerning the authorisation of the preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol as a feed additive for chickens for fattening, chickens reared for laying and minor poultry species reared for laying (holder of authorisation Biomin GmbH) OJ L 221 10.07.2020 p. 87.
[4] FEED dossier reference: FAD-2020-0070.
[5] The full report is available on the EU science HUB webpage: https://joint-research-centre.ec.europa.eu/system/files/2021-07/finrep_fad-2020-0070-biomin-dc-p.pdf
[6] Commission Regulation (EC) No 429/2008 of 25 April 2008 on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the preparation and the presentation of applications and the assessment and the authorisation of feed additives. OJ L 133, 22.5.2008, p. 1.

Guidance on the identity, characterisation and conditions of use of feed additives (EFSA FEEEDAP Panel, 2017a), Guidance on the assessment of the safety of feed additives for the target species (EFSA FEEDAP Panel, 2017b), Guidance on the assessment of the safety of feed additives for the consumer (EFSA FEEDAP Panel, 2017c), Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018) and Guidance on the assessment of the safety of feed additives for the environment (EFSA FEEDAP Panel, 2019b).

3. Assessment

The additive, hereinafter referred as Biomin® DC-P, is a preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol intended to be used as a zootechnical feed additive (functional group: other zootechnical additives) for all poultry species. The additive is currently authorised for use in feed for chickens for fattening, chickens reared for laying and minor poultry species reared to the point of lay.

3.1. Characterisation

3.1.1. Characterisation of the additive

The additive is a blend of five individual compounds (carvacrol, thymol, D-carvone, methyl salicylate and L-menthol) encapsulated with a hydrogenated vegetable oil.[7] Amorphous silica is added as a carrier. The typical additive composition as it is specified in the authorisation[8] is shown in Table 1.

Table 1: Typical composition of Biomin® DC-P

Ingredient (CAS number)	Content (mg/g additive)
Carvacrol (499–75–2)	120–160
Thymol (89–83–8)	1–3
D-Carvone (2244–16–8)	3–6
Methyl salicylate (119–36–8)	10–35
L-Menthol (2216–51–8)	30–55
Amorphous silica (68611–44–9)	Maximum 100
Hydrogenated vegetable oil	Maximum 700

The information relating to the characterisation of the additive and its manufacturing process has been assessed by the FEEDAP Panel in a previous opinion (EFSA FEEDAP Panel, 2019a) and is considered to apply to the present assessment.

3.1.2. Conditions of use

The additive is intended to be used in feed for all poultry species at a minimum recommended level of 65 mg/kg complete feed and a maximum recommended level of 105 mg/kg complete feed. The additive is intended to be incorporated directly to feed or via premixture.

3.2. Safety

The safety of the product for the user and environment was evaluated in the context of the previous opinion (EFSA FEEDAP Panel, 2019a). The FEEDAP Panel concluded that the use of the additive under the conditions of use proposed would raise no concerns for the environment. Concerning the safety for the users, the FEEDAP Panel considered that exposure of Biomin® DC-P by inhalation is unlikely. In the absence of data, the Panel could not conclude on the effects on skin and eyes.

The applicant did not provide any new data in the current application and the FEEDAP Panel is not aware of new information that would lead to reconsider the previous conclusions on the safety for the

[7] [redacted]

[8] COMMISSION IMPLEMENTING REGULATION (EU) 2020/996 of 9 July 2020 concerning the authorisation of the preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol as a feed additive for chickens for fattening, chickens reared for laying and minor poultry species reared for laying (holder of authorisation Biomin GmbH) OJ L 221 10.07.2020 p. 87.

environment and users. In addition, the proposed extension of use to the new species/categories would not introduce risks not already considered in the previous assessment. Therefore, the FEEDAP Panel considers that the conclusions of the previous opinion regarding the safety for the environment and the users still apply to the present assessment.

3.2.1. Safety for the target species

In a previous opinion, the Panel evaluated a tolerance study in chickens for fattening (EFSA FEEDAP Panel, 2019a). Based on that study, the Panel concluded that the additive is safe for chickens for fattening at the maximum recommended level of 105 mg/kg complete feed; this conclusion was extended to chickens reared for laying and extrapolated to minor poultry species to the point of lay. The Panel considers that the conclusions can also be extrapolated to other physiologically related poultry species, namely turkeys.

The applicant submitted a new tolerance study in laying hens, including data on the deposition of the active compounds in the eggs, to support the safety for the target species.

3.2.1.1. Tolerance study in laying hens

A total of 600 20-week-old hens (ISA Brown) were distributed in 60 cages of 10 hens each and allocated to four dietary treatments (24 replicates (cages) for control diet and 12 replicates per treatment).[9] A basal diet based on maize and soyabean meal was either not supplemented (control) or supplemented with Biomin® DC-P to provide 105 (1×maximum recommended level), 525 (5×) and 1,050 (10×) mg/kg feed. The 5× and 10× doses could not be confirmed analytically, reaching 68% and 50% of the intended additive concentration in the final feed, respectively.[10] Water and feed (in mash form) were offered *ad libitum* for 56 days. Mortality and general health were monitored throughout the study. Individual body weight was recorded at the beginning and at the end of the trial. Feed intake, laying performance and egg quality parameters were recorded at days 28 and 56. At the end of the study, eggs were collected for the analysis of the active substances to evaluate possible residues (see Section 3.2.2.2) and blood samples from 12 hens per dietary treatment were taken and the animals killed. Haematological and biochemical parameters[11] were analysed, and the weight of liver, kidneys, lungs and heart, as well as the length of jejunum and ileum were recorded.

The maximum intended overdose level applied in the study, which could not be demonstrated analytically, was of 10× the maximum recommended level. Therefore, the battery of observations included in the tolerance study provided showed limitations that prevent the Panel to perform a complete assessment of the safety of the additive for laying hens: several relevant blood parameters were not measured (prothrombin time, fibrinogen, magnesium, cholesterol, acute phase protein, amylase and creatine kinase) and a complete necropsy was not performed at the end of the study. Therefore, in the absence of an adequate tolerance study, the FEEDAP Panel cannot conclude on the safety of the additive for laying hens.

3.2.1.2. Conclusions on the safety for the target species

The FEEDAP Panel concludes that the additive Biomin® DC-P is safe for poultry species for fattening or reared for laying/breeding. In the absence of adequate data, the FEEDAP Panel cannot conclude on the safety for laying/breeding birds.

3.2.2. Safety for the consumer

3.2.2.1. Toxicological studies

The FEEDAP Panel assessed in its previous opinion on Biomin® DC-P genotoxicity studies (bacterial reverse mutation test and a mammalian chromosome aberration test) and a sub-chronic oral toxicity study (90-day toxicity study) performed with a concentrated preparation of the active compounds of Biomin® DC-C, which includes all the active compounds of Biomin® DC-P at a concentration ca. 3–4

[9] Technical dossier/Section III/Annex 01.
[10] Technical dossier/Section III/Annex 03.
[11] Technical dossier/Section III/Annex 01: Haematology parameters analysed were total erythrocyte count, total leucocyte count, haemoglobin concentration, haematocrit, mean cell volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), platelet count and differential leucocyte count. Blood chemistry consisted of analysis of concentrations of glucose, urea, total protein, albumin, albumin/globulin ratio, creatinine, total bilirubin, alkaline phosphate activity (ALP), alanine aminotransferase activity (ALT), lactate dehydrogenase activity (LDH), aspartate aminotransferase activity (AST), gamma glutamyl transferase (GGT), calcium, phosphorus, chloride, sodium and potassium.

times higher than in the additive plus linalool (EFSA FEEDAP Panel, 2019a,c). Based on the genotoxicity studies and considering that all the identified components of the additive (D-carvone, carvacrol, thymol, methyl salicylate and L-menthol) were already assessed and authorised for use in food and feed as flavouring compounds, the Panel concluded that the additive under assessment was not genotoxic. The 90-day study was not further considered as it did not comply with the requirements of the relevant Organisation for Economic Co-operation and Development (OECD) Testing Guideline (TG).

In the current dossier, to support the lack of data for the assessment of the clastogenic and aneugenic potential of the additive, the applicant submitted a new *in vivo* mammalian micronucleus test in accordance with OECD TG 474 and following Good Laboratory Practices (GLP) performed in peripheral blood reticulocytes of NMRI mice. The Panel noted that the test item used was the same concentrated mixture of the active compounds from Biomin® DC-C used in the abovementioned toxicological tests. Based on the results of a dose-finding test showing mild toxicity signs at 2,000 mg/kg body weight (bw), animals were treated twice by oral gavage, 2 h apart, at 500, 1,000 and 2,000 mg/kg bw and sacrificed 44 and 68 h after last administration. Flow cytometric analysis showed a dose-related, statistically significant increase in the percentage of immature reticulocytes (also called polychromatic erythrocytes, PCE) induced by the mixture of active compounds of Biomin® DC-C, confirming the exposure of the target tissue. No increase in the frequency of micronuclei was observed in treated animals compared to the negative control group. Based on the results of this study, the Panel concluded that the test item did not induce structural and numerical chromosome aberrations *in vivo* when tested up to 2,000 mg/kg bw.

The FEEDAP Panel considered that the existing genotoxicity studies performed with a concentrated preparation of the active compounds of Biomin® DC-C are relevant for the assessment of the additive currently under assessment and, therefore, concluded that Biomin® DC-P is not genotoxic.

3.2.2.2. Residue study

In the previous opinion, the Panel concluded that the use of Biomin® DC-P in feed for chickens for fattening and reared for laying and minor avian species under the proposed conditions of use is safe for the consumers (EFSA FEEDAP Panel, 2019a). The applicant provided new data in laying hens based on the analyses of the active compounds' residues (L-menthol, D-carvone, methyl salicylate, thymol and carvacrol) in egg samples taken from the tolerance study (see Section 3.2.1.1) performed with laying hens.[12]

Six eggs from each of the twelve replicates (cages) were collected in 3 consecutive days (2 eggs/cage/day from control, 105 and 525 mg/kg groups). Yolks and albumens were separated and samples coming from the same cage were pooled to form 12 independent yolk and albumen samples from each treatment. The marker residues (L-menthol, D-carvone, methyl salicylate, thymol and carvacrol) were analysed by gas chromatography-tandem mass spectrometry (GC-MS/MS) with a limit of quantification (LOQ) of 20 μg/kg for each substance. Data showed non-normal distribution (Shapiro-Wilk test). Therefore, statistical comparison between treatments were done by Wilcoxon Rank-Sum-Test. Significance level was set at 0.05.

Regardless of the use level tested, no residue above the LOQ (20 μg/kg) was found for any of the analytes in the yolk samples, and in the albumen for D-carvone and thymol. In albumen, L-menthol and carvacrol were only detected in 2 out of the 12 replicates in the group supplemented with the maximum recommended level of Biomin® DC-P (105 mg/kg complete feed), with a maximum content of 23.5 and 23.9 μg/kg, respectively. In contrast, methyl salicylate was detected in all samples of the 105 mg/kg group, with an average content of 30.0 (range from 23.6 to 43.9) μg/kg.

The FEEDAP Panel has considered the residues in eggs from the study above and of muscle and skin + fat from the previous opinion on the same additive (EFSA FEEDAP Panel, 2019a) to assess the chronic consumer exposure for L-menthol and D-carvone for which an acceptable daily intake (ADI) is established: 4 mg L-menthol/kg bw (WHO, 1999) and 60 mg D-carvone/kg bw (EFSA Scientific Committee, 2014). For this assessment, the European food consumption data of different age classes from EFSA's Comprehensive European Food Consumption Database was used as detailed in the Guidance on the assessment of the safety of feed additives for the consumer (EFSA FEEDAP Panel, 2017a). Table 2 shows the input data used for the calculation by the Panel, based on the highest analysed value and/or the LOQ for L-menthol and D-carvone in eggs, muscle and skin + fat.

[12] Technical dossier/Section III/Annex 08.

Table 2: Input data on concentration (mg/kg) of L-menthol and D-carvone in eggs of hens, breast meat and skin+fat of chickens for fattening used for the consumer exposure assessment[13]

Compound	Eggs(1)	Meat(2)	Skin + Fat
L-Menthol	0.022	0.030	0.050
D-Carvone	0.020	0.127	0.315

(1): Calculated by default as 66% albumen and 34% yolk (w/w).
(2): Calculated by default as 90% muscle and 10% skin + fat (w/w).

The results of the chronic dietary exposure to menthol and carvone are summarized in Table 3, while the detailed results are given in Appendix A.

Table 3: Chronic dietary exposure of consumers to L-menthol and D-carvone total residues based on residue data in chicken tissues and hen eggs - Summary statistics across European dietary surveys

Active substance	Population class	Maximum HRP(1) (mg/kg bw per day)	% ADI
L-Menthol	Infants	0.00023	0.006
	Toddlers	0.00027	0.007
	Other children	0.00023	0.006
	Adolescents	0.00014	0.004
	Adults	0.00008	0.002
	Elderly	0.00008	0.002
	Very elderly	0.00007	0.002
D-Carvone	Infants	0.00086	0.001
	Toddlers	0.00100	0.002
	Other children	0.00082	0.001
	Adolescents	0.00058	<0.001
	Adults	0.00030	<0.001
	Elderly	0.00027	<0.001
	Very elderly	0.00026	<0.001

(1): Highest Reliable Percentile = the highest percentile that is considered statistically robust for combinations of dietary survey, age class and possible raw primary commodity, considering that a minimum of 5, 12, 30 and 61 observations are respectively required to derive 50th, 75th, 90th and 95th percentile estimates.

As it can be seen in Table 3, for all population groups, the chronic exposure to L-menthol and D-carvone was at least 4–5 orders of magnitude lower than the corresponding ADI. The Panel notes that no data on residues of L-menthol and D-carvone in liver were available. However, considering the low contribution to the ADI of the residues in other tissues and products, it is not expected that any potential residue from liver would significantly increase the consumer exposure. For the rest of the components of the additive, an ADI is not available and, thus, the procedure used for L-menthol and D-carvone cannot be applied. However, the FEEDAP Panel notes that those components (carvacrol, thymol and methyl salicylate) – and the rest of the components of the additive: D-carvone and L-menthol – have been assessed and considered safe for use as flavouring compounds, and they are currently authorised for food[14] and feed[15] uses.

[13] Samples taken from the hens and chickens used in the tolerance studies from the current and previous Biomin DC-P opinion (EFSA FEEDAP Panel, 2019a).
[14] Commission Implementing Regulation (EU) No 872/2012 of 1 October 2012 adopting the list of flavouring substances provided for by Regulation (EC) No 2232/96 of the European Parliament and of the Council, introducing it in Annex I to Regulation (EC) No 1334/2008 of the European Parliament and of the Council and repealing Commission Regulation (EC) No 1565/2000 and Commission Decision 1999/217/EC. OJ L 267, 2.10.2012, p. 1.
[15] European Union Register of Feed Additives pursuant to Regulation (EC) No 1831/2003. Available online: https://ec.europa.eu/food/sites/food/files/safety/docs/animal-feed-eu-reg-comm_register_feed_additives_1831-03.pdf

3.2.2.3. Conclusions on safety for the consumer

The FEEDAP Panel concludes that the use of Biomin® DC-P in feed for all poultry species under the proposed conditions of use is considered safe for consumers.

3.3. Efficacy

The efficacy of Biomin® DC-P for chickens for fattening was evaluated by the Panel in a previous opinion (EFSA FEEDAP Panel, 2019a). The Panel concluded that Biomin® DC-P has potential to increase the growth performance of chickens for fattening when incorporated into feed at a minimum concentration of 65 mg/kg complete feed. The conclusion was extended to chickens reared for laying and extrapolated to minor poultry species reared up to the point of lay at the same minimum level. The Panel considers that the conclusions can also be extrapolated to other physiologically related poultry species, namely turkeys for fattening/reared for breeding.

3.3.1. Efficacy for laying hens

A total of 3 long-term efficacy trials with laying hens were submitted. However, one of the studies was not further considered as no information on the initial and final body weight of the hens was provided. The other two studies shared a common design (Table 4) and the main results are reported in Table 5.

Table 4: Trial design and use level of the efficacy trials performed in laying hens

Trial	Total n° of animals (animals × replicate) Replicates × treatment	Breed start age (duration)	Composition feed (form)	Groups (mg/kg feed)	
				Intended	Analysed
1[16]	600 (10) 30	Tetra SL 22 weeks (84 days)	Maize, soyabean meal and wheat (mash)	0 65	– 52
2[17]	600 (10) 30	Tetra SL 22 weeks (84 days)	Maize, soyabean meal and wheat (mash)	0 65	– 52

In both trials, 22-week-old laying hens were distributed in cages and allocated to two different treatments. The animals were fed either a non-supplemented diet (control) or a diet containing Biomin® DC-P at the minimum recommended use level of 65 mg/kg feed (52 mg/kg analytically confirmed for both studies) for 84 days. The health and mortality were monitored throughout the study and the initial and final body weight, feed intake and egg weight were recorded. Average daily feed intake, egg mass per hen and day and the feed to egg mass ratio were calculated. The data were analysed with t-test, using the cage as experimental unit. Significance level was set at 0.05.

Table 5: Effects of Biomin® DC-P on the performance of laying hens

Trial	Groups	Initial body weight	Final body weight	Average daily feed intake	Laying rate	Daily egg mass per hen	Egg weight	Feed to egg mass ratio
	(mg/kg feed)	(g)	(g)	(g)	(%)	(g/hen per day)	(g)	
1	0	1,784	2,038	117^{a}	93.9^{b}	55.2^{b}	58.8^{b}	2.12^{a}
	65	1,782	2,037	116^{b}	94.8^{a}	55.8^{a}	58.9^{a}	2.08^{b}
2	0	1,790	2,040	122^{a}	93.3^{b}	54.8^{b}	58.6^{b}	2.23^{a}
	65	1,792	2,027	121^{b}	93.8^{a}	55.2^{a}	58.9^{a}	2.18^{b}

^{a,b}: Mean values within a trial and within a column with a different superscript are significantly different $p < 0.05$.

No hens died during the experiment. The inclusion of the minimum use level of Biomin® DC-P in both trials resulted in significantly higher laying rate and average egg mass, which, together with a reduction of the feed intake, was translated into an improved feed to egg mass ratio.

[16] Technical dossier/Section IV/Annex 06.
[17] Technical dossier/Section IV/Annex 09.

3.3.2. Conclusions on efficacy

Biomin® DC-P has the potential to be efficacious in poultry species for fattening or reared for laying/breeding when incorporated into feed at a minimum concentration of 65 mg/kg complete feed.

Positive effects of the supplementation of the hen's diets with Biomin® DC-P at the minimum recommended use level on the performance of the hens were observed in two studies. In the absence of a third positive study, the Panel cannot conclude on the efficacy for laying hens or for other poultry species for laying/breeding.

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[18] and Good Manufacturing Practice.

4. Conclusions

The FEEDAP Panel concludes that the additive Biomin® DC-P is safe for poultry species for fattening or reared for laying/breeding, but in the absence of adequate data, cannot conclude on the safety for laying/breeding birds.

The use of Biomin® DC-P in feed for all poultry species under the proposed conditions of use is considered safe for consumers of animal products and for the environment.

The FEEDAP Panel considers that exposure of users by inhalation is unlikely. In the absence of data, the Panel cannot conclude on the effects of Biomin® DC-P on skin and eyes.

Biomin® DC-P has the potential to be efficacious in poultry species for fattening or reared for laying/breeding when incorporated into feed at a minimum concentration of 65 mg/kg complete feed. In the absence of sufficient data, the Panel cannot conclude on the efficacy for laying hens or for other poultry species for laying/breeding.

5. Documentation provided to EFSA/Chronology

Date	Event
30/09/2020	Dossier received by EFSA. FAD-2020-0070 Biomin® DC-P for all poultry species. Submitted by Biomin GmbH.
17/12/2020	Reception mandate from the European Commission
08/03/2021	Application validated by EFSA – Start of the scientific assessment
01/06/2021	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. *Issues: characterisation of the additive/ safety for the target species/user safety/efficacy*
09/06/2021	Comments received from Member States
14/06/2021	Reception of the Evaluation report of the European Union Reference Laboratory for Feed Additives
12/11/2021	Reception of supplementary information from the applicant - Scientific assessment re-started
18/01/2022	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. *Issues: toxicological studies*
22/03/2022	Reception of supplementary information from the applicant - Scientific assessment re-started
29/06/2022	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

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Abbreviations

ADFI	average daily feed intake
ADI	acceptable daily intake
ALP	alkaline phosphate activity
ALT	alanine aminotransferase activity
AST	aspartate aminotransferase activity
BW	body weight
CAS	Chemical Abstracts Service
DM	dry matter
EC	European Commission
EURL	European Union Reference Laboratory
FEEDAP	EFSA Scientific Panel on Additives and Products or Substances used in Animal Feed
GC-MS/MS	gas chromatography-mass spectrometry/mass spectrometry
GGT	gamma glutamyl transferase
GLP	good laboratory practices

HRP	highest reliable percentile
LDH	lactate dehydrogenase activity
LOQ	limit of quantification
MCH	mean corpuscular haemoglobin
MCHC	mean corpuscular haemoglobin concentration
MCV	mean corpuscular volume
OECD	Organisation for Economic Co-operation and Development
PCE	polychromatic erythrocytes
TG	Test guidelines
WHO	World Health Organization

Appendix A – Detailed results on chronic exposure calculation Chronic dietary exposure per population class, country and survey (mg/kg bw per day) of consumers to L-menthol (Table A.1) and D-carvone (Table A.2)

Table A.1: Chronic dietary exposure per population class, country and survey (mg/kg bw per day) of consumers to L-menthol based on residue data

Population class	Survey's country	Number of subjects	HRP value	HRP description
Infants	Bulgaria	523	0.0020844786	95th
Infants	Germany	142	0.0003821946	95th
Infants	Denmark	799	0.0004675338	95th
Infants	Finland	427	0.0006973620	95th
Infants	Italy	9	0.0000000000	50th
Infants	United Kingdom(1)	1,251	0.0009328590	95th
Toddlers	Belgium	36	0.0010382479	90th
Toddlers	Bulgaria	428	0.0024357064	95th
Toddlers	Germany	348	0.0006838511	95th
Toddlers	Denmark	917	0.0005350626	95th
Toddlers	Spain	17	0.0011418147	75th
Toddlers	Finland	500	0.0011146343	95th
Toddlers	Italy	36	0.0009356891	90th
Toddlers	Netherlands	322	0.0011287518	95th
Toddlers	United Kingdom	1,314	0.0010838867	95th
Toddlers	United Kingdom	185	0.0010899219	95th
Other children	Austria	128	0.0009212682	95th
Other children	Belgium	625	0.0011893324	95th
Other children	Bulgaria	433	0.0020160104	95th
Other children	Germany	293	0.0007059453	95th
Other children	Germany	835	0.0007247079	95th
Other children	Denmark	298	0.0005945892	95th
Other children	Spain	399	0.0013405985	95th
Other children	Spain	156	0.0019013278	95th
Other children	Finland	750	0.0009430660	95th
Other children	France	482	0.0008461731	95th
Other children	Greece	838	0.0009499360	95th
Other children	Italy	193	0.0010039080	95th
Other children	Latvia	187	0.0010692526	95th
Other children	Netherlands	957	0.0008259735	95th
Other children	Netherlands	447	0.0010531076	95th
Other children	Sweden	1,473	0.0007864807	95th
Other children	Czechia	389	0.0019119224	95th
Other children	United Kingdom	651	0.0009542122	95th
Adolescents	Austria	237	0.0006222662	95th
Adolescents	Belgium	576	0.0005656936	95th
Adolescents	Cyprus	303	0.0005994523	95th
Adolescents	Germany	393	0.0005472404	95th
Adolescents	Germany	1,011	0.0004424992	95th
Adolescents	Denmark	377	0.0004544047	95th
Adolescents	Spain	651	0.0007764022	95th
Adolescents	Spain	209	0.0010379790	95th
Adolescents	Spain	86	0.0008458131	95th
Adolescents	Finland	306	0.0005733529	95th
Adolescents	France	973	0.0005170738	95th

Population class	Survey's country	Number of subjects	HRP value	HRP description
Adolescents	Italy	247	0.0004526719	95th
Adolescents	Latvia	453	0.0006617021	95th
Adolescents	Netherlands	1,142	0.0007808804	95th
Adolescents	Sweden	1,018	0.0005901800	95th
Adolescents	Czechia	298	0.0014051628	95th
Adolescents	United Kingdom	666	0.0007179633	95th
Adults	Austria	308	0.0007067125	95th
Adults	Belgium	1,292	0.0005383391	95th
Adults	Germany	10,419	0.0004346363	95th
Adults	Denmark	1,739	0.0003019610	95th
Adults	Spain	981	0.0007150283	95th
Adults	Spain	410	0.0007197629	95th
Adults	Finland	1,295	0.0005643747	95th
Adults	France	2,276	0.0004311251	95th
Adults	Hungary	1,074	0.0006386299	95th
Adults	Ireland	1,274	0.0007027243	95th
Adults	Italy	2,313	0.0003809462	95th
Adults	Latvia	1,271	0.0005833623	95th
Adults	Netherlands	2,055	0.0006400050	95th
Adults	Romania	1,254	0.0007232285	95th
Adults	Sweden	1,430	0.0006099221	95th
Adults	Czechia	1,666	0.0006881914	95th
Adults	United Kingdom	1,265	0.0005216291	95th
Elderly	Austria	67	0.0005605616	95th
Elderly	Belgium	511	0.0004362619	95th
Elderly	Germany	2,006	0.0003342854	95th
Elderly	Denmark	274	0.0002462935	95th
Elderly	Finland	413	0.0004644047	95th
Elderly	France	264	0.0003588130	95th
Elderly	Hungary	206	0.0004832871	95th
Elderly	Ireland	149	0.0005730619	95th
Elderly	Italy	289	0.0003685168	95th
Elderly	Netherlands	173	0.0004911107	95th
Elderly	Netherlands	289	0.0004152578	95th
Elderly	Romania	83	0.0006453365	95th
Elderly	Sweden	295	0.0005382478	95th
Elderly	United Kingdom	166	0.0004162738	95th
Very elderly	Austria	25	0.0001508615	75th
Very elderly	Belgium	704	0.0005028246	95th
Very elderly	Germany	490	0.0003623556	95th
Very elderly	Denmark	12	0.0001281694	75th
Very elderly	France	84	0.0004071186	95th
Very elderly	Hungary	80	0.0004687121	95th
Very elderly	Ireland	77	0.0005805875	95th
Very elderly	Italy	228	0.0003751192	95th
Very elderly	Netherlands	450	0.0004121388	95th
Very elderly	Romania	45	0.0006530012	90th
Very elderly	Sweden	72	0.0004376318	95th
Very elderly	United Kingdom	139	0.0003284572	95th

(1): Dietary data from the UK were included in FACE when the UK was a member of the European Union.

Table A.2: Chronic dietary exposure per population class, country and survey (mg/kg bw per day) of consumers to D-carvone based on residue data

Population class	Survey's country	Number of subjects	HRP value	HRP description
Infants	Bulgaria	523	0.0007253282	95th
Infants	Germany	142	0.0001307449	95th
Infants	Denmark	799	0.0001604675	95th
Infants	Finland	427	0.0002349959	95th
Infants	Italy	9	0.0000000000	50th
Infants	United Kingdom(1)	1,251	0.0003239569	95th
Toddlers	Belgium	36	0.0003553390	90th
Toddlers	Bulgaria	428	0.0008399293	95th
Toddlers	Germany	348	0.0002493247	95th
Toddlers	Denmark	917	0.0001903070	95th
Toddlers	Spain	17	0.0004606023	75th
Toddlers	Finland	500	0.0003837877	95th
Toddlers	Italy	36	0.0003249307	90th
Toddlers	Netherlands	322	0.0003804529	95th
Toddlers	United Kingdom	1,314	0.0003762606	95th
Toddlers	United Kingdom	185	0.0003754682	95th
Other children	Austria	128	0.0003276236	95th
Other children	Belgium	625	0.0004028917	95th
Other children	Bulgaria	433	0.0006898541	95th
Other children	Germany	293	0.0002465699	95th
Other children	Germany	835	0.0002603152	95th
Other children	Denmark	298	0.0002110713	95th
Other children	Spain	399	0.0004601920	95th
Other children	Spain	156	0.0006699533	95th
Other children	Finland	750	0.0003274705	95th
Other children	France	482	0.0003033864	95th
Other children	Greece	838	0.0003318328	95th
Other children	Italy	193	0.0003577633	95th
Other children	Latvia	187	0.0003632254	95th
Other children	Netherlands	957	0.0002942890	95th
Other children	Netherlands	447	0.0003550567	95th
Other children	Sweden	1,473	0.0002712973	95th
Other children	Czechia	389	0.0006511642	95th
Other children	United Kingdom	651	0.0003343055	95th
Adolescents	Austria	237	0.0002194632	95th
Adolescents	Belgium	576	0.0001972614	95th
Adolescents	Cyprus	303	0.0002041877	95th
Adolescents	Germany	393	0.0001981068	95th
Adolescents	Germany	1,011	0.0001540753	95th
Adolescents	Denmark	377	0.0001565004	95th
Adolescents	Spain	651	0.0002712899	95th
Adolescents	Spain	209	0.0003773982	95th
Adolescents	Spain	86	0.0002927712	95th
Adolescents	Finland	306	0.0001969166	95th
Adolescents	France	973	0.0001898801	95th
Adolescents	Italy	247	0.0001593830	95th
Adolescents	Latvia	453	0.0002324103	95th
Adolescents	Netherlands	1,142	0.0002677546	95th

Population class	Survey's country	Number of subjects	HRP value	HRP description
Adolescents	Sweden	1,018	0.0002021040	95th
Adolescents	Czechia	298	0.0004859321	95th
Adolescents	United Kingdom	666	0.0002473109	95th
Adults	Austria	308	0.0002385432	95th
Adults	Belgium	1,292	0.0001843464	95th
Adults	Germany	10,419	0.0001506788	95th
Adults	Denmark	1,739	0.0001055213	95th
Adults	Spain	981	0.0002463967	95th
Adults	Spain	410	0.0002471020	95th
Adults	Finland	1,295	0.0001944203	95th
Adults	France	2,276	0.0001538804	95th
Adults	Hungary	1,074	0.0002228277	95th
Adults	Ireland	1,274	0.0002409213	95th
Adults	Italy	2,313	0.0001333074	95th
Adults	Latvia	1,271	0.0002025854	95th
Adults	Netherlands	2,055	0.0002194633	95th
Adults	Romania	1,254	0.0002526306	95th
Adults	Sweden	1,430	0.0002167512	95th
Adults	Czechia	1,666	0.0002341881	95th
Adults	United Kingdom	1,265	0.0001783861	95th
Elderly	Austria	67	0.0001889250	95th
Elderly	Belgium	511	0.0001508964	95th
Elderly	Germany	2,006	0.0001173208	95th
Elderly	Denmark	274	0.0000886698	95th
Elderly	Finland	413	0.0001617826	95th
Elderly	France	264	0.0001300537	95th
Elderly	Hungary	206	0.0001904336	95th
Elderly	Ireland	149	0.0001961810	95th
Elderly	Italy	289	0.0001286382	95th
Elderly	Netherlands	173	0.0001676834	95th
Elderly	Netherlands	289	0.0001412697	95th
Elderly	Romania	83	0.0002295353	95th
Elderly	Sweden	295	0.0001824187	95th
Elderly	United Kingdom	166	0.0001432642	95th
Very elderly	Austria	25	0.0000536653	75th
Very elderly	Belgium	704	0.0001762514	95th
Very elderly	Germany	490	0.0001251077	95th
Very elderly	Denmark	12	0.0000478329	75th
Very elderly	France	84	0.0001443068	95th
Very elderly	Hungary	80	0.0001667012	95th
Very elderly	Ireland	77	0.0001986284	95th
Very elderly	Italy	228	0.0001264070	95th
Very elderly	Netherlands	450	0.0001422361	95th
Very elderly	Romania	45	0.0002200472	90th
Very elderly	Sweden	72	0.0001549586	95th
Very elderly	United Kingdom	139	0.0001141547	95th

(1): Dietary data from the UK were included in FACE when the UK was a member of the European Union.

Annex A – Executive Summary of the Evaluation Report of the European Union Reference Laboratory for Feed Additives on the Method(s) of the Analysis for carvacrol, thymol, D-carvone, methyl salicylate and L-menthol

In the current application an authorisation is sought under Article 4(1) for a currently authorised preparation of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol (Biomin® DC-P) under the category/functional group (4d) "zootechnical additives"/"other zootechnical additives", according to the classification system of Annex I of Regulation (EC) No 1831/2003. Specifically, the authorisation is sought for the use of the feed additive for all poultry species. The feed additive is an off-white powder composed of a mixture of the following active substances: 12–16% (w/w) of carvacrol, 0.1–0.3% (w/w) of thymol, 0.3–0.6% (w/w) of D-carvone, 1.0–3.5% (w/w) of methyl salicylate and 3.0–5.5% (w/w) of L-menthol. In addition, Biomin® DC-P contains hydrogenated vegetable oil and silica as carriers.

The feed additive is intended to be incorporated through premixtures or directly into feedingstuffs at a recommended Biomin® DC-P content ranging from 65 to 105 mg/kg feedingstuffs.

For the quantification of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol in the feed additive the Applicant submitted a single-laboratory validated and further verified multianalyte method based on gas chromatography coupled to flame ionisation detection (GC-FID).

Based on the experimental evidence available the EURL recommends for the official control the GC-FID method for the quantification of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol in the feed additive.

Additionally, the Applicant submitted a single-laboratory validated and further verified multianalyte method based on gas chromatography coupled to mass spectrometry (GC-MS) for the quantification of carvacrol, thymol, D-carvone, methyl salicylate and L-menthol in premixtures and feedingstuffs. The EURL considers the GC-MS method proposed by the Applicant fit-for-purpose for the quantification of the total content of each active substances, namely carvacrol, thymol, D-carvone, methyl salicylate and L-menthol, in premixtures and feedingstuffs at the content levels investigated in the frame of the validation and verification studies.

However, as the accurate determination of the Biomin® DC-P content added to premixtures and feedingstuffs is not achievable experimentally, the EURL cannot evaluate or recommend any method for official control to determine Biomin® DC-P in premixtures and feedingstuffs.

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.