Adopted: 12 December 2023

DOI: 10.2903/j.efsa.2024.8563

efsa JOURNAL

SCIENTIFIC OPINION

Follow-up of the re-evaluation of quillaia extract (E 999) as a food additive and safety of the proposed extension of uses

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Abstract

Quillaia extract (E 999) was re-evaluated in 2019 by the EFSA Panel on Food Additives and Flavourings (FAF). EFSA derived an acceptable daily intake (ADI) of 3 mg saponins/kg bw per day for E 999. Following a European Commission call for data to submit data to fill the data gaps, the present follow-up opinion assesses data provided by interested business operators (IBOs) to support an amendment of the EU specifications for E 999. Additionally, this opinion deals with the assessment of the proposed extension of use for E 999 in food supplements supplied in a solid and liquid form, excluding food supplements for infants and young children and, as a carrier in botanical nutrients. The Panel concluded that the proposed extension of use, if authorised, could result in an exceedance of the ADI at the maximum of the ranges of the mean for children, adolescents and the elderly, and for all populations at the 95th percentile. An additional proposed extension of use for E 999 to be used as a carrier for glazing agents on entire fresh fruits and vegetables has been received. Since no information on the proposed use levels of E 999 on a saponins content basis has been provided by this applicant, the Panel was not able to evaluate the safety of this extension of use. Considering the technical data submitted, the Panel recommended some modifications of the existing EU specifications for E 999, mainly to lower the limits for lead, mercury and arsenic and to include a maximum limit for cadmium and for calcium oxalate. The Panel also recommended that the limits would be expressed on a saponins basis. The Panel proposed to revise the definition of E 999 to better describe the composition in a qualitative way.

KEYWORDS

CAS No 68990-67-0, E 999, food additive, Quillaia extract, Quillaja extract

CONTENTS

SUMMARY

Quillaia extract (E 999) was re-evaluated in 2019 by the Panel on Food Additives and Flavourings (FAF Panel) which concluded that any toxicity associated with quillaia extract (E 999) is due to its constituent saponins and, therefore, established an ADI of 3 mg saponins/kg bw per day for quillaia extract (E 999) based on the no observed adverse effect level (NOAEL) of 1500 mg quillaia extract/kg bw per day and the conservative assumption that it contained 20% saponins, and by applying an uncertainty factor of 100. In addition, the Panel made a list of recommendations to amend the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012.

The data gaps and uncertainties identified by the Panel required a follow-up by the European Commission by means of a subsequent call for additional data. The present opinion deals with the assessment of the data provided by Interested Business Operators (IBOs) in response to this call. Additionally, it deals with the assessment of the proposed extension of use for quillaia extract (E 999) in food supplements supplied in a solid form, excluding food supplements for infants and young children (FC 17.1) and in food supplements supplied in a liquid form, excluding food supplements for infants and young children (FC 17.2), according to Annex II of Regulation (EU) No 1333/2008, and as a carrier in botanical nutrients according to Annex III of Regulation (EU) No 1333/2008, for use in flavoured drinks (FC 14.1.4) and food supplements (FC 17.1 and 17.2). An additional proposed extension of use for quillaia extract (E 999) to be used as a carrier for glazing agents on entire fresh fruits and vegetables according to Annex II to Regulation (EC) No 1333/2008 has been received. Since no information on the proposed use levels of E 999 on a saponins content basis has been provided by this applicant, the Panel was not able to evaluate the safety of this extension of use.

At the time of the re-evaluation of E 999 in 2019, the Panel had recommended revising the maximum use levels for quillaia extract (E 999) established in Regulation (EC) No 1333/2008 to be expressed on saponin content, but this has not been implemented, yet. In order to estimate the current maximum permitted level (MPL), previously calculated as anhydrous extract, on a saponins basis, the maximum reported content of saponins in E 999 of 83.41% was considered. The Panel noted that had the reported content of saponins in E 999 been used, this could have resulted in a ~10% lower estimate of exposure to E 999 expressed on a saponins basis.

Quillaia extract (E 999) is currently authorised in two food categories according to Regulation (EU) No 1333/2008. The exposure to E 999 was estimated based on the currently authorised uses and the MPLs, in the regulatory maximum level exposure assessment scenario, or considering the reported use levels at the time of the re-evaluation for the refined exposure scenarios. Thus, different exposure scenarios have been calculated. The Panel considered that since the food additive is used in FC 14.1.4 Flavoured drinks, the brand-loyal scenario is the most relevant for the risk assessment of E 999. Using the refined brand-loyal exposure assessment scenario, the highest exposure levels for the mean and 95th percentile among the different population groups were considered, i.e. 0.3 and 0.9 mg saponins/kg bw per day, respectively, for toddlers.

Additional exposure scenarios, 'food supplements consumers only scenarios', have been performed considering the proposed extension of uses. These scenarios have been calculated using the MPLs or the typical use levels for the authorised food categories, respectively, and the proposed use levels for the food supplements. The Panel noted that these scenarios considered that all food supplements contain E 999 and that all flavoured drinks are assumed to have added botanical nutrients containing quillaia extract (E 999), resulting in an overestimation of the dietary exposure. The calculated 'food supplements consumers only exposure assessment scenarios' considering the proposed extension of use of quillaia extract (E 999) resulted in an exceedance of the ADI at the maximum of the ranges at the mean for children, adolescents and the elderly, and for all populations at the 95th percentile.

In the re-evaluation of E 999 in 2019, it was noted that existing EU specifications for E 999 do not describe any range for saponins content in the food additive. The Joint FAO/WHO Expert Committee on Food Additives (JECFA) specifications, however, differentiate two types of quillaia extracts, Type 1 and Type 2, containing a different percentage of saponins (Type 1: 20%–26% on the dried basis and Type 2: 65%–90% on the dried basis). The Panel considered, at that time, that similar differentiation of the extracts of quillaia should be presented in the EU specifications, including the percentage range for saponins, polyphenols (including tannins), protein, polysaccharides including fibre, reducing sugars, a maximum limit for calcium oxalate as well as microbiological specifications.

Based on the new data submitted in response to the European Commission call for data, regarding impurities, and considering that the maximum authorised use levels are proposed to be expressed on a saponins basis, the Panel considered that it is not needed to differentiate the extracts of quillaia with different saponins content. However, a minimum content for the functional component (not less than 20% on the dried basis) is recommended to be included in the EU specifications.

In response to the European Commisssion's call for data, analytical data on levels of toxic elements (arsenic, lead, cadmium, mercury) in commercial samples of E 999 were provided by one IBO and respective limit values were proposed. The Panel noted that the occurrence data on toxic elements are substantially lower than the current limits in the EU specifications. The Panel performed a risk assessment considering three different scenarios: (I) at the current limits in the EU specification calculated on a saponins basis; (II) at the limits proposed by one IBO for the toxic elements in E 999 Type 1, expressed on a saponins basis; and (III) at the rounded up highest measured level of the toxic elements in E 999 Type 1 or, in the absence of any measured value(s), the highest limit of quantification (LOQ) expressed on a saponins basis. The potential exposure to these impurities from the use of E 999 was compared against the available health-based guidance values (HBGV) and reference points (RP). For arsenic, in all scenarios, the lower end of the range of the calculated MOE values was insufficient, i.e. below the target value of 1000. For the other toxic elements (cadmium, mercury, lead), exposure to them due to the use

of the food additive does not give rise to safety concerns. The Panel recommended the maximum limits for lead, mercury and arsenic to be lowered on the basis of the information provided by the IBO. The Panel noted that in the data submitted, cadmium was present in some of the samples, and therefore, considered that a maximum limit for cadmium should be included in the specifications. The Panel also recommended that the limits would be expressed on a saponins basis.

Analytical data on the content of polyphenols (including tannins), protein, polysaccharides including fibre and reducing sugars were submitted. Differences among the results of the content of the different components have been observed by the Panel, but different methodology has been applied for each of the components between the two manufacturers. As a result, the Panel was not able to propose quantitative values (percentage ranges) that can define the composition. However, the data reported do not indicate any concern regarding the presence of these components in the food additive E 999. Thus, the Panel proposed to revise the definition of E 999 to better describe the composition in a qualitative way, only.

With respect to calcium oxalate, in order to avoid excessive contribution to the oxalate burden of the body, which could result in the formation of oxalate kidney stones, a maximum limit could be included in the specifications of the food additive E 999. Considering the proposed lowest technologically achievable level for oxalic acid of 0.08% on the dried basis for the quillaia extract, and the worst case of an extract containing 20% saponins, a limit for calcium oxalate of 0.6% expressed on a saponins basis is recommended.

Microbiological criteria are proposed to be included on the basis of the information provided. The Panel considered that the microbiological criteria for quillaia extract Type 2 proposed by an IBO at the time of the re-evaluation of 2019, cover quillaia extract Type 1 to be used as a food additive, according to the data submitted by one manufacturer.

The Panel concluded that the technical data provided by one IBO support an amendment of the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012.

1 | INTRODUCTION

The re-evaluation of quillaia extract (E 999) as a food additive was completed by EFSA in 2019 (EFSA FAF Panel, 2019). The EFSA Panel on Food Additives and Flavourings (FAF Panel) issued several recommendations to amend the specifications of the food additive E 999 in Commission Regulation (EU) No 231/2012.[1]

The data gaps and uncertainties identified by the FAF Panel required a follow-up by the European Commission by means of a call for additional data.[2]

The present opinion deals with the assessment of the data provided by interested parties in support of an amendment of the EU specifications for quillaia extract (E 999).

Additionally, the present opinion deals with the assessment of the proposed extension of use for quillaia extract (E 999) in food supplements supplied in a solid and liquid form, excluding food supplements for infants and young children, according to Annex II of Regulation (EU) No 1333/2008, and in botanical nutrients according to Annex III of Regulation (EU) No 1333/2008, for use in flavoured drinks and food supplements.

An additional proposed extension of use for quillaia extract (E 999) to be used as a carrier for glazing agents on entire fresh fruits and vegetables according to Annex II to Regulation (EC) No 1333/2008 has been received.

1.1 | BACKGROUND AND TERMS OF REFERENCE AS PROVIDED BY THE REQUESTOR

1.1.1 | Background

The use of food additives is regulated under the European Parliament and Council Regulation (EC) No 1333/2008 on food additives.[3] Only food additives that are included in the Union list, in particular in Annex II to that Regulation, may be placed on the market and used in foods under the conditions of use specified therein. Moreover, food additives shall comply with the specifications as referred to in Article 14 of that Regulation and laid down in Commission Regulation (EU) No 231/2012.

1.1.1.1 | *Follow-up of the re-evaluation of quillaia extract (E 999) as a food additive (EFSA-Q-2022-00541)*

Quillaia extract (E 999) is authorised for use as a food additive in the Union. Since quillaia extract (E 999) was permitted in the Union before 20 January 2009, it belongs to the group of food additives which are subject to a new risk assessment by the European Food Safety Authority (EFSA), according to Commission Regulation (EU) No 257/2010,[4] and in line with the provisions of Regulation (EC) No 1333/2008.

EFSA completed the re-evaluation of quillaia extract (E 999) as a food additive and safety of the proposed extension of use and published a scientific opinion on 6 March 2019.[5] In that opinion, EFSA derived an acceptable daily intake (ADI) for quillaia extract (E 999) of 3 mg saponins/kg bw per day. None of the exposure estimates for the different population groups of the refined brand-loyal scenario exceeded the ADI. However, EFSA made recommendations concerning the specifications for E 999.

Therefore, the European Commission published on 15 December 2020 a call for data[6] requesting business operators to submit technical data needed to address issues identified by EFSA in the re-evaluation of the safety of quillaia extract (E 999) as a food additive.

In October 2021, interested business operators completed the submission of data in reply to the call for data.

Consequently, the European Commission has decided to consult EFSA on this matter.

1.1.1.2 | *Extension of use for quillaia extract (E 999) (EFSA-Q-2022-00475; EFSA-Q-2022-00476)*

Two applications have been received for a modification of the conditions of use of the authorised food additive quillaia extract (E 999). The first one for the use in solid and liquid food supplements and nutrients, and the second for the use as a carrier for glazing agents applied on entire fresh fruits and vegetables.

1.1.2 | Terms of reference

1.1.2.1 | *Follow-up of the re-evaluation of quillaia extract (E 999) as a food additive*

In accordance with Article 29(1)(a) of Regulation (EC) No 178/2002,[7] the European Commission requests the European Food Safety Authority (EFSA) to provide a scientific opinion and confirm that the technical data provided by interested business

[1]Commission Regulation (EU) No 231/2012: https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=celex%3A32012R0231.

[2]Available online: https://food.ec.europa.eu/system/files/2020-12/fs_food-improvement-agents_reeval_call_20201215_e999_data.pdf.

[3]Council Regulation (EC) No 1333/2008: https://eur-lex.europa.eu/legal-content/en/TXT/?uri=CELEX%3A32008R1333.

[4]Commission Regulation (EU) No 257/2010: https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=celex%3A32010R0257.

[5]Re-evaluation of Quillaia extract (E 999) as a food additive and safety of the proposed extension of use: https://efsa.onlinelibrary.wiley.com/doi/full/10.2903/j.efsa.2019.5622.

[6]https://food.ec.europa.eu/system/files/2020-12/fs_food-improvement-agents_reeval_call_20201215_e999_data.pdf.

[7]Council Regulation (EC) No 178/2002: https://eur-lex.europa.eu/eli/reg/2002/178/oj L 31,1.2.2002, p. 1.

operators adequately support an amendment of the specifications of the food additive quillaia extract (E 999) in line with the recommendations made by EFSA during the re-evaluation of the safety of this food additive.

1.1.2.2 | *Extension of use for quillaia extract (E 999)*

The European Commission requests the EFSA to provide a scientific opinion on the safety of the proposed extension of use for quillaia extract (E 999) in accordance with Regulation (EC) No 1331/2008 establishing a common authorisation procedure for food additives, food enzymes and food flavourings.[8]

1.2 | Summary of the EFSA re-evaluation of quillaia extract (E 999)

Quillaia extract (E 999) was re-evaluated by the Panel which concluded that any toxicity associated with quillaia extract (E 999) is due to its constituent saponins and, therefore, established an ADI of 3 mg saponins/kg bw per day for quillaia extract (E 999) based on the no observed adverse effect level (NOAEL) of 1500 mg quillaia extract/kg bw per day and the conservative assumption that it contained 20% saponins, and by applying an uncertainty factor of 100 (EFSA FAF Panel, 2019). In addition, exposure estimates for the different population groups of the brand-loyal scenario did not exceed the ADI of 3 mg saponins/kg bw per day at the reported use levels.

The Panel recommended that the European Commission considers:

- Revising the EU specifications for quillaia extract (E 999) in order to differentiate extracts of quillaia according to the saponins content (including a description of the principle of the method of analysis to quantify the content of saponins in line with the JECFA specifications), i.e. Type 1 and Type 2.[9]
- Revising the EU specifications to include the percentage range for polyphenols (including tannins), protein, polysaccharides including fibre, reducing sugars, a maximum limit for calcium oxalate as well as microbiological specifications.
- Lowering the current limits for toxic elements (arsenic, lead and mercury) in the EU specifications for quillaia extract (E 999) in order to ensure that the food additive will not be a significant source of exposure to these toxic elements in food.
- Revising the maximum use levels for quillaia extract (E 999) established in Regulation (EC) No 1333/2008 to be expressed on saponin content.

2 | DATA AND METHODOLOGIES

2.1 | Data

The Panel based its assessment on:

- Information submitted in response to the public call for data issued by the European Commission (Documentation provided to EFSA n. 1) and additional information submitted during the assessment process by one IBO, representing two manufacturers, in response to a follow-up request from EFSA (Documentation provided to EFSA n. 2);
- Food consumption data from the EFSA Comprehensive European Food Consumption Database (Comprehensive Database), which were used to estimate the dietary exposure to quillaia extract (E 999);
- Use levels reported at the time of the re-evaluation (EFSA FAF Panel, 2019) to estimate the dietary exposure to quillaia extract (E 999);
- Information from Mintel's Global New Products Database (GNPD) to identify the use of quillaia extract (E 999) in beverage products and food supplements. Mintel's GNPD is an online database that contains the compulsory ingredient information present on the label of numerous products.

In addition, two dossiers in support of the application for the extension of use of E 999 as a food additive (Documentation provided to EFSA n. 3, 4) and additional information submitted during the assessment process (Documentation provided to EFSA n. 5) were provided to EFSA and have been evaluated.

2.2 | Methodologies

This opinion was formulated following the principles described in the EFSA Guidance on transparency with regard to scientific aspects of risk assessment (EFSA Scientific Committee, 2009) and following the relevant existing guidance documents from the EFSA Scientific Committee.

[8]Council Regulation (EC) No 1333/2008: https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A32008R1331&qid=1689345562212.

[9]Quillaia extract Type 1 and Type 2 are differentiated on the basis of their saponins content: Type 1 (20%–26% on the dried basis) and Type 2 (65%–90% on the dried basis).

The FAF Panel assessed the safety of the proposed extension of use of the food additive quillaia extract (E 999) in line with the EFSA Guidance for submission for food additive evaluations in 2012 (EFSA ANS Panel, 2012).

Dietary exposure to quillaia extract (E 999) from its use as a food additive was estimated combining the food consumption data available within the Comprehensive Database with maximum permitted levels according to Annex II to Regulation (EC) No. 1333/2008, reported use levels submitted to EFSA (EFSA FAF Panel, 2019), and the proposed use levels indicated in the extension of use. The exposure was estimated according to different exposure scenarios (EFSA ANS Panel, 2017). Uncertainties in the exposure assessment were identified and discussed (Section 3.5.4).

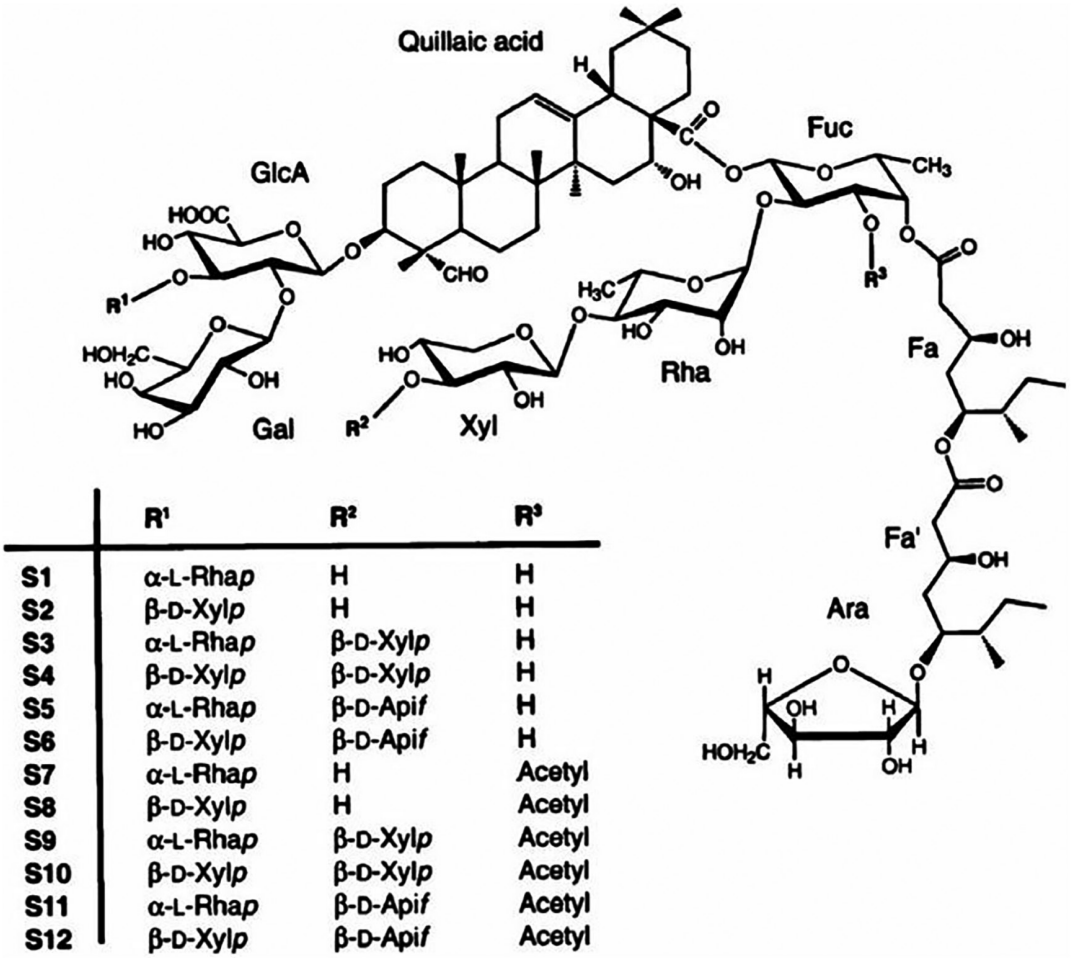
3 | ASSESSMENT

3.1 | Identity and specifications of E 999

According to Commission Regulation (EU) No 231/2012, quillaia extract (E 999) is obtained by aqueous extraction of *Quillaia saponaria* Molina, or other Quillaia species, trees of the family Rosaceae. It contains a number of triterpenoid saponins consisting of glycosides of quillaic acid. According to the EFSA re-evaluation of quillaia extract (E 999) (EFSA FAF Panel, 2019), sugars – including glucose, galactose, arabinose, xylose and rhamnose – are also present, along with polyphenols including tannins, calcium oxalate and other minor components, such as protein and polysaccharides.

As mentioned in the EFSA re-evaluation of quillaia extract (E 999) (EFSA FAF Panel, 2019), no chemical name, EC/EINECS Number, chemical formula or molecular weight is provided in the EU specifications. *Quillaia saponaria* extract is registered with EC No 273-620-4 and CAS No 68990-67-0 according to the EC Inventory. The same CAS number is cited for quillaia extract Type 1 and Type 2 in the JECFA specifications (JECFA, 2006, 2014).

The general structural formulae of quillaia saponins is shown in Figure 1.



	R¹	R²	R³
S1	α-L-Rha*p*	H	H
S2	β-D-Xyl*p*	H	H
S3	α-L-Rha*p*	β-D-Xyl*p*	H
S4	β-D-Xyl*p*	β-D-Xyl*p*	H
S5	α-L-Rha*p*	β-D-Api*f*	H
S6	β-D-Xyl*p*	β-D-Api*f*	H
S7	α-L-Rha*p*	H	Acetyl
S8	β-D-Xyl*p*	H	Acetyl
S9	α-L-Rha*p*	β-D-Xyl*p*	Acetyl
S10	β-D-Xyl*p*	β-D-Xyl*p*	Acetyl
S11	α-L-Rha*p*	β-D-Api*f*	Acetyl
S12	β-D-Xyl*p*	β-D-Api*f*	Acetyl

FIGURE 1 General structure of quillaia saponins (quillaic acid as aglycone (sapogenin) component) (Güçlü-Ustündağ & Mazza, 2007). Apif, apiofuranose; Ara, D-arabinose; Gal, D-galactose; GlcA, D-glucoronic acid; Rha, D-rhamnose; Rhap, D-rhamnopyranose; Xyl, D-xylose; Xylp, D-xylopyranose.

Specifications for quillaia extract (E 999) have been defined in the Commission Regulation (EU) No 231/2012 and by JECFA (2006, 2014) as described in Table 1.

TABLE 1 Specifications for quillaia extract (E 999) according to Commission Regulation (EU) No 231/2012 and JECFA (2006, 2014).

	Commission Regulation (EU) No 231/2012	JECFA (2006) Type 1 [INS No 999(i)]	JECFA (2014) Type 2 [INS No 999(ii)]
Definition	Quillaia extract is obtained by aqueous extraction of *Quillaia saponaria* Molina, or other *Quillaia* species, trees of the family Rosaceae. It contains a number of triterpenoids saponins consisting of glycosides of quillaic acid. Some sugars including glucose, galactose, arabinose, xylose, and rhamnose are also present, along with tannin, calcium oxalate and other minor components	Quillaia extract (Type 1) is obtained by aqueous extraction of the milled inner bark or of the wood of pruned stems and branches of *Quillaja saponaria* Molina (family Rosaceae). It contains triterpenoid saponins (quillaia saponins, QS) consisting predominantly of glycosides of quillaic acid. Polyphenols and tannins are major components and some sugars and calcium oxalate will be present Quillaia extract (Type 1) is available commercially as liquid product or as spray-dried powder that may contain carriers such as lactose, maltitol or maltodextrin. The liquid product is usually preserved with sodium benzoate or ethanol	Quillaia extract (Type 2) is obtained either by chromatographic separation or ultrafiltration of the aqueous extraction of the milled inner bark or of the wood of pruned stems and branches of *Quillaja saponaria* Molina (family Rosaceae). It contains triterpenoid saponins (quillaia saponins, QS) consisting predominantly of glycosides of quillaic acid. Polyphenols and tannins are minor components. Some sugars and calcium oxalate will also be present Quillaia extract (Type 2) is available commercially as a liquid product or as a spray-dried powder that may contain carriers such as lactose, maltitol or maltodextrin. The liquid product is usually preserved with sodium benzoate or ethanol
CAS Number	-	68990-67-0	
Formula weight	-	Monomeric saponins range from ca. 1800 to ca. 2300, consistent with a triterpene with 8–10 monosaccharide residues	
Assay			
Saponin content	-	Not less than 20% and not more than 26% on the dried basis	Not less than 65% and not more than 90% on the dried basis
Description	Quillaia extract in the powder form is light brown with a pink tinge. It is also available as an aqueous solution	Red-brownish liquid or light brown powder with a pink tinge	Light red-brownish liquid or powder
Identification			
pH	Between 3.7 and 5.5 (4% solution)[a]	3.7–5.5 (4% solution)	
Solubility	-	Very soluble in water, insoluble in ethanol, acetone, methanol and butanol	
Foam	-	Dissolve 0.5 g of powder extract in 9.5 g of water or 1 mL of liquid extract in 9 mL of water. Add 1 mL of this mixture to 350 mL of water in a 1000-mL graduated cylinder. Cover the cylinder, vigorously shake it 30 times and allow settling. Record the foam level (mL) after 30 min. Typical values are 150 mL of foam	Dissolve 0.5 g of the powder form in 9.5 mL of water or 1 mL of the liquid form in 9 mL of water. Add 1 mL of this solution to 350 mL of water in a 1000-mL graduated cylinder. Cover the cylinder, vigorously shake it 30 times and allow settling. Record the foam volume (mL) after 30 min. Typical volumes are about 260 mL
Chromatography	-	The retention time of major peak of the sample corresponds to the major saponin peak (QS-18) of the standard	
Colour and turbidity	-	Powder form only: Dissolve 0.5 g in 9.5 g of water. The solution is not turbid. Determine the absorbance of the solution against water at 520 nm. The absorbance is less than 1.2	Powder form only: Dissolve 0.5 g in 9.5 mL of water. The solution shall not be turbid. Determine the absorbance of the solution against water at 520 nm. The absorbance shall be less than 0.7
Purity			
Water	Not more than 6.0% (Karl Fischer method) (powder form only)	Powder form: not more than 6% (Karl Fischer Method)	
Loss on drying	-	Liquid form: 50%–80% (2 g, 105°, 5 h)	Liquid form: 50%–90% (2 g, 105°, 5 h)
Ash	-	Not more than 14% on a dried basis (use 1.0 g for powder samples; for liquid samples, use the residue from loss on drying)	Not more than 5% on a dried basis (use 1.0 g for powder samples; for liquid samples, use the residue from loss on drying)
Tannins	-	Not more than 8% on a dried basis	

(Continues)

TABLE 1 (Continued)

	Commission Regulation (EU) No 231/2012	JECFA (2006) Type 1 [INS No 999(i)]	JECFA (2014) Type 2 [INS No 999(ii)]
Arsenic	Not more than 2 mg/kg	-	
Lead	Not more than 2 mg/kg	Not more than 2 mg/kg	
Mercury	Not more than 1 mg/kg	-	

[a]According to recital (33) of Regulation (EC) 231/2012, the current specification relating to the pH range should be adjusted in order to bring it in line with JECFA.

The revisions of the existing EU specifications proposed by the Panel are provided under Section 3.6.

3.2 | Technical data submitted

3.2.1 | Content of saponins

According to JECFA (2006, 2014), two types of quillaia extracts exist containing different percentages of saponins, i.e. Type 1 (20%–26% on the dried basis) and Type 2 (65%–90% on the dried basis). No specification limits are provided in the EU specifications of E 999 for the content of saponins.

One IBO, representing two manufacturers, provided analytical data on the content of saponins, on Type 1 and Type 2 quillaia extracts, in liquid and powder formulations (Documentation provided to EFSA n. 1, 2).

For the quillaia extract Type 1, one manufacturer provided data on nine samples of a liquid formulation with saponins ranging from 23.2% to 27.0% on the dried basis. Another manufacturer provided data on four samples of a liquid formulation with saponins ranging from 10.2% to 12.4% on extract basis, and two samples of a powder formulation with saponins being 16.2% and 17.6% on extract basis. The Panel calculated the equivalence of the saponins content expressed on extract basis to the saponins content expressed on the dried basis, considering the reported moisture content of two samples. The sample of a liquid formulation with saponins content of 10.2% on extract basis is equivalent to 19.6% on the dried basis, while the sample of a powder formulation with saponins content of 17.6% on extract basis is equivalent to 18.1% on the dried basis. No information on moisture content was provided for the rest of the samples in order to calculate the saponins content on the dried basis (Documentation provided to EFSA n. 1, 2).

For the quillaia extract Type 2, one manufacturer provided data on nine samples of a liquid formulation with saponins ranging from 67.3% to 73.6% on the dried basis. Another manufacturer provided data on four samples of a liquid formulation with saponins ranging from 66.7% to 83.4% on the dried basis, and six samples of a powder formulation with saponins ranging from 65.2% to 82.5% on the dried basis (Documentation provided to EFSA n. 1, 2).

The analytical method used to analyse saponins in the quillaia extracts was reverse-phase high-performance liquid chromatography (RP-HPLC) with an ultraviolet (UV) detector at 210 nm, following the method described by San Martín and Briones (2000) (Documentation provided to EFSA n. 2).

3.2.2 | Toxic elements

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of arsenic, lead and mercury in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for arsenic, lead and mercury and cadmium in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

Information on the content of toxic elements (arsenic, cadmium, lead and mercury) on Type 1 and Type 2 quillaia extracts was provided by one IBO representing two manufacturers, in liquid and powder formulations (Documentation provided to EFSA n. 1, 2). One of the manufacturers analysed these four toxic elements in the food additive with inductively coupled plasma mass spectrometry (ICP-MS) (AOAC 993.14 method), while the other one analysed the four toxic elements in the food additive with atomic absorption spectrophotometry (AAS) according to AOAC 986.15 for arsenic, AOAC 999.10 for cadmium and lead, and with an in-house validated method for mercury. Analytical data were expressed as mg/kg of quillaia extract.

Specifically, for the quillaia extract Type 1, one manufacturer provided data on six samples of a liquid formulation with arsenic ranging from 0.21 to 0.48 mg/kg, cadmium from 0.015 to 0.037 mg/kg, lead from 0.44 to 1.1 mg/kg and mercury being lower than the limit of detection (LOD) of 0.003 mg/kg in all samples. Another manufacturer, provided data on two samples of a liquid formulation with arsenic being in one sample 0.065 mg/kg and in the other lower than the limit of quantification (LOQ) of 0.0002 mg/kg, cadmium being lower than the LOQs in both samples (being 0.01 mg/kg and 0.001 mg/kg, respectively), lead being in one sample 0.24 mg/kg and in the other lower than the LOQ of 0.01 mg/kg and mercury being in one sample 0.036 mg/kg and in the other lower than the LOQ of 0.001 mg/kg. Data on three samples of a powder

formulation were also provided with arsenic ranging from below the LOQ of 0.0002 to 0.30 mg/kg, cadmium ranging from below the LOQ of 0.001 to 0.047 mg/kg, lead ranging from below the LOQ of 0.01 to 0.30 mg/kg and mercury being below the LOQs in all samples (0.001, 0.005 and 0.01 mg/kg, respectively) (Documentation provided to EFSA n. 1).

For the quillaia extract Type 2, one manufacturer provided data on five samples of a liquid formulation with arsenic ranging from 0.16 to 0.39 mg/kg, cadmium from 0.007 to 0.012 mg/kg, lead from 0.28 to 0.61 mg/kg and mercury being lower than the LOD of 0.003 mg/kg in all samples. Another manufacturer provided data on two samples of a liquid formulation and three samples of a powder formulation with arsenic being below the LOD of 0.0002 mg/kg, cadmium being below the LOD of 0.001 mg/kg, lead being below the LOD of 0.01 mg/kg and mercury being below the LOD of 0.001 mg/kg in all samples (Documentation provided to EFSA n. 1). A summary of the analytical data is presented in Table 2.

TABLE 2 Summary of the analytical data reported by one IBO on toxic elements expressed as mg/kg of quillaia extract (Documentation provided to EFSA n. 1).

Quillaia extract type	Type 1			Type 2		
Formulation	Liquid		Powder	Liquid		Powder
Manufacturer	Manufacturer 1	Manufacturer 2	Manufacturer 2	Manufacturer 1	Manufacturer 2	Manufacturer 2
Number of samples	6	2	3	5	2	3
Arsenic (mg/kg of quillaia extract)	0.21–0.48	< 0.0002 (LOQ) and 0.065	< 0.0002 (LOQ)–0.30	0.16–0.39	< 0.0002 (LOD)	< 0.0002 (LOD)
Cadmium (mg/kg of quillaia extract)	0.015–0.037	< 0.001 (LOQ) and < 0.01 (LOQ)	< 0.001 (LOQ)–0.047	0.007–0.012	< 0.001 (LOD)	< 0.001 (LOD)
Lead (mg/kg of quillaia extract)	0.44–1.1	< 0.01 (LOQ) and 0.24	< 0.01 (LOQ)–0.30	0.28–0.61	< 0.01 (LOD)	< 0.01 (LOD)
Mercury (mg/kg of quillaia extract)	< 0.003 (LOD)	< 0.001 (LOQ) and 0.036	< 0.001 (LOQ)–< 0.01 (LOQ)	< 0.003 (LOD)	< 0.001 (LOD)	< 0.001 (LOD)

The Panel noted that the LOQs and LODs of each toxic element differ substantially depending on the laboratory that conducted the analysis and the time period when the elements were analysed.

The levels of arsenic, lead and mercury analysed were all well below the limits as defined in the Commission Regulation (EU) No 231/2012, of 2, 2 and 1 mg/kg of quillaia extract, respectively. No limit for cadmium is included in the EU specifications of quillaia extract (E 999).

Considering the analytical data on toxic elements provided by the two manufacturers, the IBO proposed the lowest technologically achievable levels for arsenic, cadmium, lead and mercury, for Type 1 and Type 2 quillaia extracts as shown in Table 3. These levels cover both liquid and powder formulations. These levels were derived by the average values plus six standard deviations (average + 6SD) of the results of the analysis.

TABLE 3 Lowest technologically achievable levels of the toxic elements Pb, Hg, Cd and As in commercial Type 1 and 2 quillaia extracts on quillaia extract basis, as proposed by one IBO (Documentation provided to EFSA n. 1).

Quillaia extract type	Lead (mg/kg of extract)	Mercury (mg/kg of extract)	Cadmium (mg/kg of extract)	Arsenic (mg/kg of extract)
Type 1	2.0	0.10	0.13	1.0
Type 2	2.0	0.003	0.021	1.0

Since the FAF Panel recommended at the time of the re-evaluation to revise the maximum use levels for quillaia extract (E 999) established in Regulation (EC) No 1333/2008 to be expressed on saponins content (EFSA FAF Panel, 2019), analytical data on the toxic elements, expressed as mg/kg of saponins, in the food additive were requested.

Upon EFSA's request, new analytical data on the toxic elements, expressed as mg/kg of saponins, were provided (Documentation provided to EFSA n. 2). The manufacturers analysed for each sample the content of saponins and toxic elements as mg/kg of quillaia extract and used the analytical results to calculate the corresponding concentration of toxic elements expressed as mg/kg of saponins.

For the quillaia extract Type 1, one manufacturer provided data on five samples of a liquid formulation with a content of saponins ranging from 11.3% to 12.0% on extract basis. The levels of arsenic ranged from 2.93 to 5.50 mg/kg of saponins, for cadmium were below the LOQs (LOQs from 0.42 to 0.44 mg/kg of saponins), for lead ranged from 1.23 to 1.75 mg/kg of saponins and for mercury were below the LOQs (LOQs from 0.42 to 0.44 mg/kg of saponins). Another manufacturer provided data on three samples of a liquid formulation with saponins ranging from 11.5% to 12.4% on extract basis. The levels of arsenic ranged from 0.89 to 1.05 mg/kg of saponins, for cadmium were below the LOD (LODs from 0.40 to 0.43 mg/kg of saponins), for lead were below the LOD (LODs from 1.61 to 1.74 mg/kg of saponins) and for mercury were below the

LOD (LODs from 0.40 to 0.43 mg/kg of saponins). The IBO also provided data on one sample of a powder formulation with saponins being 16.2% on extract basis, arsenic being 0.68 mg/kg of saponins, cadmium being below the reporting limit of 0.06 mg/kg of saponins, lead being 2.16 mg/kg of saponins and mercury being below the reporting limit of 0.43 mg/kg of saponins. The Panel noted that no information on the LOQ and LOD was provided for this sample (Documentation provided to EFSA n. 2).

For the quillaia extract Type 2, one manufacturer provided data on five samples of a liquid formulation with saponins ranging from 14.9% to 15.9% in the liquid preparation. The levels of arsenic ranged from 0.88 to 1.79 mg/kg of saponins, for cadmium were below the LOQ (LOQs from 0.32 to 0.34 mg/kg of saponins), for lead ranged from below the LOQ of 0.64 to 1.20 mg/kg of saponins and for mercury were below the LOQ (LOQs from 0.32 to 0.34 mg/kg of saponins). Another manufacturer provided data on two samples of a liquid formulation, with saponins being 15.2% and 18.0% in the liquid preparation. The levels for arsenic were below the LOD of 0.001 mg/kg of saponins in both samples, for cadmium were below the LOD (LODs of 0.006 and 0.007 mg/kg of saponins), for lead were below the LOD (LODs of 0.056 and 0.066 mg/kg of saponins) and for mercury were below the LOD (LODs of 0.006 and 0.007 mg/kg of saponins). The manufacturer also provided data on two samples of a powder formulation with saponins being 74.0% and 82.5%. The levels for arsenic were below the LOD (LODs of 0.0002 and 0.0003 mg/kg of saponins), for cadmium were below the LOD of 0.001 mg/kg of saponins in both samples, for lead were below the LOD (LODs of 0.012 and 0.014 mg/kg of saponins), and for mercury were below the LOD of 0.001 mg/kg of saponins in both samples (Documentation provided to EFSA n. 2). A summary of the analytical data is presented in Table 4.

TABLE 4 Summary of the analytical data reported by one IBO on toxic elements expressed as mg/kg of saponins (Documentation provided to EFSA n. 2).

Quillaia extract type	Type 1			Type 2		
Formulation	Liquid		Powder	Liquid		Powder
Manufacturer	Manufacturer 1	Manufacturer 2	Manufacturer 2	Manufacturer 1	Manufacturer 2	Manufacturer 2
Number of samples	5	3	1	5	2	2
Arsenic (mg/kg of saponins)	2.93–5.50	0.89–1.05	0.68	0.88–1.79	< 0.001 (LOD)	< 0.0002 (LOD) and < 0.0003 (LOD)
Cadmium (mg/kg of saponins)	< 0.42 (LOQ)–< 0.44 (LOQ)	< 0.40 (LOD)–< 0.43 (LOD)	< 0.06 (reporting limit)	< 0.32 (LOQ)–< 0.34 (LOQ)	< 0.006 (LOD) and < 0.007 (LOD)	< 0.001 (LOD)
Lead (mg/kg of saponins)	1.23–1.75	< 1.61 (LOD)–< 1.74 (LOD)	2.16	<0.64 (LOQ)–1.20	< 0.056 (LOD) and < 0.066 (LOD)	< 0.012 (LOD) and < 0.014 (LOD)
Mercury (mg/kg of saponins)	< 0.42 (LOQ)–< 0.44 (LOQ)	< 0.40 (LOD)–< 0.43 (LOD)	< 0.43 (reporting limit)	< 0.32 (LOQ)–< 0.34 (LOQ)	< 0.006 (LOD) and < 0.007 (LOD)	< 0.001 (LOD)

The Panel noted that the LOQs and LODs of each toxic element differ substantially depending on the laboratory that conducted the analysis and the time period when those parameters were analysed.

After EFSA's request to propose limits for toxic elements on the basis of saponins content of the commercial products, one IBO indicated the preference to retain the specifications limits of toxic elements expressed as mg/kg of quillaia extract. Nevertheless, the IBO proposed limits for the toxic elements arsenic, cadmium, lead and mercury expressed on a saponins basis, for Type 1 and Type 2 quillaia extracts. These limits were derived by the average values plus six standard deviations (average+6SD) of the results of the analysis (Table 5).

TABLE 5 Proposed limits for the toxic elements Pb, Hg, Cd and As on a saponins basis in commercial Type 1 and 2 quillaia extracts (Documentation provided to EFSA n. 2).

Quillaia extract type	Lead (mg/kg of saponins)	Mercury (mg/kg of saponins)	Cadmium (mg/kg of saponins)	Arsenic (mg/kg of saponins)
Type 1	10.0	1.0	2.0	10.0
Type 2	5.0	1.0	1.0	5.0

3.2.3 | Polyphenols (including tannins)

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of polyphenols (including tannins) in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for polyphenols (including tannins) in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

One IBO representing two manufacturers provided analytical data on the content of polyphenols (including tannins), on Type 1 and Type 2 quillaia extracts, in liquid and powder formulations (Documentation provided to EFSA n. 1). The results were expressed, by the IBO, on the dried basis (w/w), calculated considering the moisture content of the analysed samples.

Specifically, for the quillaia extract Type 1, one manufacturer provided data on four samples of a liquid formulation with total polyphenols content ranging from 2.0% to 3.7% on the dried basis and tannins ranging from 0.8% to 1.5% on the dried basis. Another manufacturer provided data on one sample of a liquid formulation with total polyphenols content (expressed as tannic acid equivalents) being 5.2% on the dried basis, and on two samples of a powder formulation with total polyphenols content (expressed as tannic acid equivalents) being 4.5% and 9.5% on the dried basis.

For the quillaia extract Type 2, one manufacturer provided data on four samples of a liquid formulation with total polyphenols content ranging from 2.9% to 4.2% on the dried basis and tannins content ranging from 0.9% to 1.4% on the dried basis. Another manufacturer provided data on three samples of a liquid formulation with total polyphenols content (expressed as tannic acid equivalents) ranging from 0.5% to 1.0%, and on four samples of a powder formulation with total polyphenols content (expressed as tannic acid equivalents) ranging from 1.7% and 5.8% on the dried basis.

The IBO proposed a lowest technologically achievable level for polyphenols (including tannins) of 27.1% on the dried basis for the quillaia extract Type 1 and 17.4% on the dried basis for the quillaia extract Type 2, derived by the results of the analyses considering the average values plus six standard deviations of polyphenols content on the dried basis.

The Panel noted that spectrophotometric analytical methods have been used by the two different manufacturers, i.e. the Folin–Ciocalteu method and an internal method.

3.2.4 | Protein

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of protein in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for protein in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

One IBO representing two manufacturers provided analytical data on the content of protein, on Type 1 and Type 2 quillaia extracts, only in liquid formulations (Documentation provided to EFSA n. 1). The results were expressed, by the IBO, on the dried basis (w/w), calculated considering the moisture content of the analysed samples.

For the quillaia extract Type 1, one manufacturer provided data on four samples of a liquid formulation with protein content being up to 0.1% on the dried basis. Another manufacturer provided data on two samples of a liquid formulation with protein content being 2.9% and 4.5% on the dried basis.

For the quillaia extract Type 2, one manufacturer provided data on four samples of a liquid formulation with protein content being up to 0.3% on the dried basis. Another manufacturer provided data on two samples of a liquid formulation with protein content being 3.1% and 2.2% on the dried basis.

The IBO proposed a lowest technologically achievable level for protein content of 10.3% on the dried basis for the quillaia extract Type 1 and 6.4% on the dried basis for the quillaia extract Type 2, derived by the results of the analyses considering the average values plus six standard deviations of protein content on the dried basis.

The Panel noted that the content of protein in the analysed samples from the first manufacturer was lower than in the samples from the second manufacturer. The analytical methods used by the manufacturers are the Folin–Ciocalteu spectrophotometric method, or the same method as modified by Lowry.

3.2.5 | Polysaccharides (including fibre)

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of polysaccharides (including fibre) in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for polysaccharides (including fibre) in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

Regarding the content of polysaccharides, one IBO representing two manufacturers provided analytical data on Type 1 and Type 2 quillaia extracts, in only liquid formulations (Documentation provided to EFSA n. 1). The results were expressed, by the IBO, on the dried basis (w/w), calculated considering the moisture content of the analysed samples.

Specifically, for the quillaia extract Type 1, one manufacturer provided data on four samples of a liquid formulation with polysaccharides content ranging from 5.7% to 9.2% on the dried basis. However, the other manufacturer provided data on the content of carbohydrates, in two samples of a liquid formulation, being 78.0% and 84.6% on the dried basis.

For the quillaia extract Type 2, one manufacturer provided data on four samples of a liquid formulation with polysaccharides content ranging from 0.9% to 1.4% on the dried basis. The other manufacturer provided data on the content of carbohydrates, in two samples of a liquid formulation, being 85.3% and 89.5% on the dried basis.

The IBO proposed a lowest technologically achievable level for polysaccharides of 17.39% on the dried basis for the quillaia extract Type 1 and 2.35% on the dried basis for the quillaia extract Type 2, derived by the results of the analyses, by the one of the manufacturers, considering the average values plus six standard deviations of polysaccharides on the dried basis.

The Panel noted that that the data provided by the two manufacturers differ, since the parameters were analysed with different analytical methods. Specifically, the second manufacturer likely overestimated the carbohydrate content, since it was calculated gravimetrically by difference, including the saponins content. The analytical method of the other manufacturer was a modified phenol-sulfuric method.

3.2.6 | Reducing sugars

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of reducing sugars in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for reducing sugars in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

Regarding the content of reducing sugars, one IBO representing two manufacturers provided analytical data on Type 1 and Type 2 quillaia extracts, in only liquid formulations (Documentation provided to EFSA n. 1). The results were expressed, by the IBO, on the dried basis (w/w), calculated considering the moisture content of the analysed samples.

Specifically, for the quillaia extract Type 1, one manufacturer provided data on three samples of a liquid formulation with reducing sugars content ranging from 21.8% to 40.7% on the dried basis. Another manufacturer provided data on two samples of a liquid formulation with reducing sugars content being 22.8% and 28.9% on the dried basis.

For the quillaia extract Type 2, one manufacturer provided data on three samples of a liquid formulation with reducing sugars content ranging from 12.6% to 21.2% on the dried basis. Another manufacturer provided data on two samples of a liquid formulation with reducing sugars being 9.3% and 13.6% on the dried basis.

The IBO proposed a lowest technologically achievable level for reducing sugars of 60% on the dried basis for the quillaia extract Type 1 and 30% on the dried basis for the quillaia extract Type 2, derived by the results of the analyses considering the average values plus three standard deviations of reducing sugars on the dried basis.

The Panel noted that there is an approximate twofold reduction between the reducing sugars content from quillaia extract Type 1 to Type 2 for each of the two manufacturers. One of the manufacturers reported using AOAC 906.03 Munson–Walker method for the analysis, while the other manufacturer mentioned only an internal method without further information.

3.2.7 | Calcium oxalate

The following was requested in the European Commission call for data:

- Analytical data, if possible supported by certificate of analysis, on current levels of calcium oxalate in commercial samples of the food additive (Type 1 and Type 2 extracts);
- The lowest technologically achievable level for calcium oxalate in order to adequately propose maximum limits in the specifications for Type 1 and Type 2 extracts of E 999.

One IBO representing two manufacturers provided analytical data on the content of calcium oxalate, on Type 1 and Type 2 quillaia extracts, in liquid and powder formulations (Documentation provided to EFSA n. 1). The results were calculated, by the IBO, from the results of the analysis of oxalic acid content, considering the moisture content and expressed on the dried basis (w/w).

Specifically, for the quillaia extract Type 1, one manufacturer provided data on five samples of a liquid formulation with calcium oxalate content being in all samples below the LOD of 0.0014% on the dried basis (calculated from the LOD of 0.001% of oxalic acid). Another manufacturer provided data on one sample of a liquid formulation with calcium oxalate content being 0.0548% on the dried basis (corresponding to 0.2802% on a saponins basis as calculated by the Panel

considering the reported saponins content), and two samples of a powder formulation for calcium oxalate being 0.0295% and 0.0294% on the dried basis.

For the quillaia extract Type 2, one manufacturer provided data on three samples of a liquid formulation with calcium oxalate content being below the LOD of 0.0028% on the dried basis (calculated from the LOD of 0.002% of oxalic acid) in all samples. Another manufacturer provided data on three samples of a liquid formulation with calcium oxalate content ranging from 0.0365% to 0.0379% on the dried basis, and on four samples of a powder formulation with calcium oxalate ranging from 0.0294% to 0.0303% on the dried basis.

The Panel noted that oxalic acid was analysed with different analytical methods between the two manufacturers, meaning by HPLC-UV or with ion chromatography with a conductimetry detector.

The IBO proposed a lowest technologically achievable level not for calcium oxalate, but for oxalic acid of 0.08% on the dried basis for the quillaia extract Type 1 and 0.04% on the dried basis for the quillaia extract Type 2 derived by the results of the analyses considering the average values plus six standard deviations of oxalic acid on the dried basis.

The Panel noted that the maximum proposed level of oxalic acid of 0.08% corresponds to 0.114% calcium oxalate on the dried basis in the quillaia extract. Considering a minimum content of 20% saponins (worst case) in the quillaia extract to be used as a food additive, the corresponding maximum content of calcium oxalate would be 0.6% on a saponins basis.

3.2.8 | Microbiological parameters

Because of the botanical origin, quillaia extract can be subject to microbiological contamination. At the time of the re-evaluation (EFSA FAF Panel, 2019), IBOs had proposed microbiological specifications (Table 6). The Panel recommended the inclusion of microbiological parameters in the EU specifications for E 999 as proposed in Table 6, including also specifications for the absence of *Escherichia coli*, *Salmonella* and *Staphylococcus aureus*.

TABLE 6 Microbiological specification for Quillaia extract (Types 1 and 2) as proposed by interested parties at the time of the re-evaluation (EFSA FAF Panel, 2019).

Parameter	Type 1 limit (CFU/g)	Type 2 limit (CFU/g)
Aerobic plate count	< 5000	< 100
Yeast	< 100	< 10
Mould	< 100	< 10

Abbreviation: CFU, colony forming unit.

One IBO provided analytical data on behalf of one of the manufacturers on microbiological parameters (aerobic plate count, yeasts, moulds, coliforms, Enterobacteriaceae, Salmonella, *E. coli*, *S. aureus*, *Candida albicans*, *Pseudomonas aeruginosa*, Gram-negative bacteria) in 17 samples for liquid and powder formulations of quillaia extract Type 1 and Type 2 (Documentation provided to EFSA n. 1).

For the quillaia extract Type 1, one manufacturer provided analytical data on three samples of a liquid formulation and two samples of a powder formulation with aerobic plate count being below 100 CFU/g, yeasts being below 10 CFU/g and moulds being below 10 CFU/g in all samples, respectively. In addition, the two samples of a powder formulation were also analysed for coliforms (below 100 CFU/g in both samples), and Enterobacteriaceae (below 100 CFU/g in both samples) and one of the samples of a powder formulation was analysed for Salmonella (not detected in 25 g) (Documentation provided to EFSA n. 1).

For the quillaia extract Type 2, one manufacturer provided analytical data on five samples of a liquid formulation and six samples of a powder formulation with aerobic plate count being below 100 CFU/g, yeasts being below 10 CFU/g and moulds being below 10 CFU/g in all samples, respectively. In addition, the three samples of a liquid formulation were also analysed for *E. coli* (negative in 1 g), *S. aureus* (negative in 1 g), *C. albicans* (negative in 1 g) and two of the samples of a liquid formulation were analysed for *P. aeruginosa* (negative per g). Two samples of a liquid formulation were analysed for coliforms (not detected in 1 g) and for Salmonella (not detected in 25 g), and one sample of a liquid formulation was analysed for *E. coli* (negative in 1 g), *S. aureus* (negative in 1 g), *C. albicans* (negative in 1 g), *P. aeruginosa* (negative in 1 g) and Gram-negative bacteria (negative in 1 g) (Documentation provided to EFSA n. 1).

The Panel noted that the analytical results provided for the quillaia extract Type 1 meet the specifications of the quillaia extract Type 2 proposed at the time of the EFSA re-evaluation of the quillaia extract (E 999) (Table 6). Therefore, the Panel considered that the proposed microbiological specifications for quillaia extract Type 2, cover quillaia extract Type 1 to be used a food additive, according to the data submitted.

3.3 | Authorised use and use levels

Maximum levels of quillaia extract (E 999) have been defined in Annex II to Regulation (EC) No 1333/2008 on food additives, as amended. In this document, these levels are named maximum permitted levels (MPLs).

Currently, quillaia extract (E 999) is an authorised food additive in the EU at an MPL of 200 mg/L, calculated as anhydrous extract, in food category (FC) 14.1.4. Flavoured drinks and in FC 14.2.3 Cider and perry (excluding *cidre bouché*) (Table 7).

The Panel noted that, at the time of the re-evaluation of E 999 (EFSA FAF Panel, 2019), it was recommended revising the maximum use levels for quillaia extract (E 999) established in Regulation (EC) No 1333/2008 to be expressed on saponin content, and this has not been implemented. In order to calculate the MPL expressed on a saponins basis, and in line with the proposal from the IBO (Documentation provided to EFSA n. 2), the maximum content of saponins in E 999 of 83.41% was used, resulting in an MPL for E 999 of 167 mg/L or mg/kg expressed on a saponins basis. The Panel noted that this is the worst case, but it is in line with the proposed use levels on a saponins basis for the proposed extension of uses (Tables 8 and 9). If the reported average saponin content of 73.48% had been used, an MPL of approximately 150 mg/L or mg/kg expressed on a saponins basis could have been considered, resulting in an ~10% lower estimate of exposure to E 999 expressed on a saponins basis.

TABLE 7 MPLs of quillaia extract (E 999) in foods according to Annex II to Regulation (EC) No 1333/2008.

Food category number	Food category name	Restrictions/ exception	MPL calculated as anhydrous extract[a] (mg/L or mg/kg as appropriate)	MPL on a saponins basis[b] (mg/L or mg/kg as appropriate)
14.1.4	Flavoured drinks		200	167
14.2.3	Cider and perry	Excluding *cidre bouché*	200	167

Abbreviation: MPL, maximum permitted level.

[a]Terminology as in Regulation No 1333/2008, dried basis is the terminology used in this document.

[b]Rounded value calculated by the Panel, considering the maximum saponins content of 83.41% on the dried basis (for the Type 2 quillaia extract) (Documentation provided to EFSA n. 1, 2).

At the time of the re-evaluation, an extension of use for quillaia extract (E 999) to be included in Annex III Part 4 'Food additives including carriers in food flavourings' of Regulation (EC) No 1333/2008 was evaluated. It was noted that exposure estimates considering such an extension of use were almost identical to those considering only the food categories in which quillaia extract (E 999) is authorised (EFSA FAF Panel, 2019). Since then, the authorised uses and use levels of E 999 have not been modified in Regulation (EC) No 1333/2008 and, therefore, the proposed extension of uses evaluated in 2019 was not considered in the current assessment.

3.4 | Proposed extension of uses

3.4.1 | Proposed extension of uses in solid and liquid food supplements and in botanical nutrients

The current opinion considers the proposed inclusion of quillaia extract (E 999) in annex II to Regulation (EC) No 1333/2008 on food additives, for its use as a food additive in FC 17.1 'Food supplements supplied in a solid form, excluding food supplement for infants and young children' and FC 17.2 'Food supplements supplied in a liquid form, excluding food supplement for infants and young children' (Table 8).

TABLE 8 Proposed uses and use levels of quillaia extract (E 999) in food categories according to Annex II to Regulation (EC) No 1333/2008 (Documentation provided to EFSA n. 4).

		Proposed use levels calculated as dried basis (mg/L or mg/kg as appropriate)		Proposed use levels on a saponins basis (mg/L or mg/kg as appropriate)	
Food category number	Food category name	Normal use level of quillaia extract in the final food	Maximum use level of quillaia extract in the final food	Normal use level of quillaia extract in the final food[a]	Maximum use level of quillaia extract in the final food[b]
17.1	Food supplements supplied in a solid form, excl. food supplement for infants and young children	20,000	20,000	14,696	16,682
17.2	Food supplements supplied in a liquid form, excl. food supplement for infants and young children	20,000	20,000	14,696	16,682

[a]As proposed by the applicant considering the average saponins content of 73.48% on the dried basis (for the Type 2 quillaia extract).

[b]As proposed by the applicant considering the maximum saponins content of 83.41% on the dried basis (for the Type 2 quillaia extract).

The applicant also proposed to use quillaia extract (E 999) as a food additive, including as a carrier, in botanical nutrients, according to Annex III, part 5, section A of Regulation (EC) No 1333/2008, to be used in flavoured drinks and food supplements in solid and liquid form (excluding food supplement for infants and young children) (Table 9).

TABLE 9 Proposed uses and use levels of quillaia extract (E 999) as a carrier in botanical nutrient preparations and in the final food according to Annex III to Regulation (EC) No 1333/2008 (Documentation provided to EFSA n. 4).

		Use levels on an anhydrous basis (mg/L or mg/kg as appropriate)			Use levels on a saponins basis (mg/L or mg/kg as appropriate)		
Food category number	Food category name	Normal use levels of quillaia extract in the botanical nutrient preparation	Maximum use levels as dried basis of quillaia extract in the botanical nutrient preparation	Maximum use levels as dried basis of quillaia extract in the final food	Normal use levels of quillaia extract on a saponins basis in the botanical nutrient preparation[a]	Maximum use levels of quillaia extract on a saponins basis in the botanical nutrient preparation[b]	Maximum use levels of quillaia extract on a saponins basis in the final food[b]
14.1.4	Flavoured drinks	20,000	200,000	200	14,696	166,820	166.8
17.1	Food supplements supplied in a solid form, excl. food supplement for infants and young children	20,000	200,000	20,000	14,696	166,820	16,682
17.2	Food supplements supplied in a liquid form, excl. food supplement for infants and young children	20,000	200,000	20,000	14,696	166,820	16,682

[a]As proposed by the applicant considering the average saponins content of 73.48% on the dried basis (for the Type 2 quillaia extract).
[b]As proposed by the applicant considering the maximum saponins content of 83.41% on the dried basis (for the Type 2 quillaia extract).

The proposed level in final foods (Table 9) includes the authorised use in FC 14.1.4 'Flavoured drinks' or the proposed extension of use in FC 17.1 and 17.2, respectively, 'Food supplements supplied in a solid form, excluding food supplements for infants and young children' and 'Food supplements supplied in a liquid form, excluding food supplements for infants and young children' according to Annex II and the proposed use as a food additive, including as a carrier, in botanical nutrients to be used in the same FCs according to Annex III. Therefore, the exposure estimates calculated considering these categories present the exposure taking into account the use according to both annexes.

3.4.2 | Proposed extension of uses to be used as a carrier for glazing agents on entire fresh fruits and vegetables

An additional proposed extension of use for quillaia extract (E 999) to be used as a carrier for glazing agents on entire fresh fruits and vegetables according to Annex II to Regulation (EC) No 1333/2008 was requested to be evaluated (Documentation provided to EFSA n. 5).

EFSA requested information on the proposed use levels of E 999 on a saponins content basis. Since no information was provided by the applicant, the Panel was not able to evaluate the safety of this extension of use.

3.5 | Exposure assessment

A new exposure assessment to quillaia extract (E 999) has been performed to evaluate the new extension of uses requested for solid and liquid food supplements and botanical nutrients.

3.5.1 | Exposure data

Reported use levels or data on analytical levels of quillaia extract (E 999)

Data on the occurrence of quillaia extract (E 999) in food were collected at the time of its re-evaluation by the FAF Panel by means of a call for data launched in 2016.[10] In response to this call, four use levels were submitted to EFSA by the industry (EFSA FAF Panel, 2019).

No analytical results were made available by the Member States.

For the current exposure assessment, the Panel considered the data collected during the EFSA call for data in 2016–2017. Levels used for estimating the dietary exposure assessment of quillaia extract (E 999) are presented in Appendix A.

Summarised data extracted from the Mintel's Global New Products Database

Mintel's Global New Products Database (GNPD) is an online database which monitors new introductions of packaged goods in the market worldwide. It contains information of over 4.2 million food and beverage products of which more than 1,300,000 are or have been available on the European food market. Mintel started covering EU's food markets in 1996, currently having 24 of its 27 member countries and Norway presented in the Mintel GNPD.[11]

For this Scientific Opinion, Mintel's GNPD[12] was used for checking the labelling of food and beverage products and food supplements for quillaia extract (E 999) within the EU's food market as the database contains the required ingredient information on the label.

According to Mintel's GNPD, quillaia extract (E 999) was labelled on 20 products between January 2018 and November 2023. These products belong to 'carbonated soft drinks' ($n = 12$), 'beer' ($n = 5$), 'energy drinks' ($n = 2$) and 'ready-to-drink iced tea' ($n = 1$).

Appendix B lists the percentages of the food products labelled to contain quillaia extract (E 999) out of the total number of food products per food subcategory according to Mintel's GNPD food classification. The percentages ranged from 0.1% to 0.2% with an average percentage of foods labelled to contain quillaia extract (E 999) of 0.1%.

Food consumption data used for the exposure assessment

EFSA Comprehensive European Food Consumption Database

Since 2010, the EFSA Comprehensive European Food Consumption Database (Comprehensive Database) has been populated with national data on food consumption at a detailed level. Competent authorities in the European countries provide EFSA with data on the level of food consumption by the individual consumer from the most recent national dietary survey in their country (cf. Guidance of EFSA on the 'Use of the EFSA Comprehensive European Food Consumption Database in Exposure Assessment', EFSA, 2011). The version of the Comprehensive database taken into account in this assessment was the one available in November 2022.[13]

The food consumption data gathered by EFSA were collected by different methodologies, and thus, direct country-to-country comparisons should be interpreted with caution. Depending on the food category and the level of detail used for exposure calculations, uncertainties could be introduced owing to possible subjects' underreporting and/or misreporting of the consumption amounts. Nevertheless, the EFSA Comprehensive Database includes the currently best available food consumption data across Europe.

Food consumption data from infants, toddlers, children, adolescents, adults and the elderly were used in the exposure assessment. For the present assessment, food consumption data were available from 41 different dietary surveys carried out in 22 Member States (Table 10). Not all Member States provided consumption information for all population groups, and in some cases, the same country provided food consumption data from more than one consumption survey. In most cases, when, for one country and age class, different dietary surveys were available, only the most recent was used. However, when two national surveys from the same country gave a better coverage of the age range than using only the most recent one, both surveys were kept.

TABLE 10 Population groups considered for the exposure estimates of quillaia extract (E 999).

Population	Age range	EU Member States with food consumption surveys covering more than 1 day
Infants	From more than 12 weeks up to and including 11 months of age	Bulgaria, Cyprus, Denmark, Estonia, Finland, France, Germany, Italy, Latvia, Portugal, Slovenia
Toddlers[a]	From 12 months up to and including 35 months of age	Belgium, Bulgaria, Cyprus, Denmark, Estonia, Finland, France, Germany, Hungary, Italy, Latvia, Netherlands, Portugal, Slovenia, Spain

[10] https://www.efsa.europa.eu/en/data/call/160524.

[11] Missing Cyprus, Luxembourg, and Malta.

[12] http://www.gnpd.com/sinatra/home/ accessed on 22/11/2023.

[13] https://www.efsa.europa.eu/en/data-report/food-consumption-data.

TABLE 10 (Continued)

Population	Age range	EU Member States with food consumption surveys covering more than 1 day
Children[b]	From 36 months up to and including 9 years of age	Austria, Belgium, Bulgaria, Cyprus, Czechia, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Italy, Latvia, Netherlands, Portugal, Spain, Sweden
Adolescents	From 10 years up to and including 17 years of age	Austria, Belgium, Cyprus, Czechia, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Italy, Latvia, Netherlands, Portugal, Romania, Slovenia, Spain, Sweden
Adults	From 18 years up to and including 64 years of age	Austria, Belgium, Croatia, Cyprus, Czechia, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Ireland, Italy, Latvia, Netherlands, Portugal, Romania, Slovenia, Spain, Sweden
The elderly[b]	From 65 years of age and older	Austria, Belgium, Cyprus, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Ireland, Italy, Latvia, Netherlands, Portugal, Romania, Slovenia, Spain, Sweden

[a]The term 'toddlers' in the Comprehensive Database (EFSA, 2011) corresponds to 'young children' in Regulations (EC) No 1333/2008 and (EU) No 609/2013.
[b]The terms 'children' and 'the elderly' correspond, respectively, to 'other children' and the merge of 'elderly' and 'very elderly' in Comprehensive Database (EFSA, 2011).

Since 2018, all consumption records in the Comprehensive Database are codified according to the FoodEx2 classification system (EFSA, 2015). Nomenclature from the FoodEx2 classification system has been linked to the food categorisation system of Annex II to Regulation (EC) No 1333/2008, part D, to perform exposure assessments of food additives. In practice, the FoodEx2 food codes were matched to the food categories.

Food categories considered for the exposure assessment of quillaia extract (E 999)

Food categories for which concentration data of quillaia extract (E 999) were provided were selected from the nomenclature of the EFSA Comprehensive Database (FoodEx2 classification system), at the most detailed level possible (up to FoodEx2 Level 7) (EFSA, 2015).

- All flavoured drinks were selected for the FC 14.1.4. This includes the proposed use according to Annex III of Regulation (EU) No 1333/2008.
- All cider and perry were selected for the FC 14.2.3 as the exclusion of '*cidre bouché*' could not be considered.
- All food supplements were also considered for the FC 17, in the extension of use scenario, as the applicant requested that both liquid and solid food supplements to be able to contain quillaia extract (E 999) at the same use level. This includes the proposed use according to Annex III of Regulation (EU) No 1333/2008.

Concentration levels of quillaia (E 999) used in the exposure assessment scenarios (mg/L or mg/kg as appropriate) are indicated in Appendix C.

3.5.2 | Exposure estimates to quillaia extract (E 999) based on the currently authorised uses

The Panel estimated the chronic dietary exposure to quillaia extract (E 999) for the following population groups: infants, toddlers, children, adolescents, adults and the elderly. The methodology to estimate dietary exposure to quillaia extract (E 999) expressed on a saponins basis for the different scenarios – regulatory maximum level exposure assessment scenario and refined exposure assessment scenarios (brand-loyal and non-brand-loyal) – presented in the current assessment are described in the approach for the refined exposure assessment of food additives under re-evaluation (EFSA ANS Panel, 2017).

Table 11 summarises the estimated exposure to quillaia extract (E 999), expressed on a saponins basis, from its use as a food additive in six population groups according to the different exposure scenarios. Detailed results per population group and survey are presented in Appendix D.

TABLE 11 Summary of estimated exposure to quillaia extract (E 999) expressed on a saponins basis, from its use as a food additive in the regulatory maximum level exposure assessment scenario and in the refined exposure scenarios, in six population groups.

	Infants (12 weeks–11 months)	Toddlers (12–35 months)	Children (3–9 years)	Adolescents (10–17 years)	Adults (18–64 years)	The elderly (≥ 65 years)
Regulatory maximum level exposure assessment scenario						
Mean	0–0.33	0.03–2.97	0.16–2.76	0.11–1.49	0.06–0.78	0.02–0.23
95th percentile	0–2.30	0.29–9.12	0.75–6.34	0.62–3.75	0.37–2.42	0.11–1.13
Refined estimated exposure assessment scenario						
Brand-loyal scenario						

(Continues)

TABLE 11 (Continued)

	Infants (12 weeks–11 months)	Toddlers (12–35 months)	Children (3–9 years)	Adolescents (10–17 years)	Adults (18–64 years)	The elderly (≥ 65 years)
Mean	0–0.03	< 0.01–0.29	0.02–0.27	0.01–0.15	< 0.01–0.11	< 0.01–0.02
95th percentile	0–0.23	0.03–0.91	0.08–0.62	0.06–0.37	0.04–0.39	0.01–0.16
Non-brand-loyal scenario						
Mean	0–0.03	< 0.01–0.25	0.01–0.23	0.01–0.13	0.005–0.08	< 0.01–0.02
95th percentile	0–0.20	0.02–0.78	0.06–0.54	0.05–0.32	0.03–0.29	0.01–0.10

In the regulatory maximum level exposure assessment scenario, mean exposure to quillaia extract (E 999) expressed on a saponins basis from the use of E 999 ranged from 0 mg/kg bw per day in infants to 3.0 mg/kg bw per day in toddlers. The 95th percentile of exposure to quillaia extract (E 999) expressed on a saponins basis ranged from 0 mg/kg bw per day in infants to 9.1 mg/kg bw per day in toddlers.

The results of the MPLs scenario slightly differ from the MPL results of the 2019 opinion as the dietary surveys considered in the current opinion are more recent and the saponins content used for the calculations is different (Section 3.3).

In the brand-loyal refined estimated exposure scenario, mean exposure to quillaia extract (E 999) expressed on a saponins basis from its use as a food additive ranged from 0 mg/kg bw per day in infants to 0.3 mg/kg bw per day in toddlers and children. The high exposure to quillaia extract (E 999) expressed on a saponins basis ranged from 0 mg/kg bw per day in infants to 0.9 mg/kg bw per day in toddlers. In the non-brand-loyal scenario, mean exposure to quillaia extract (E 999) expressed on a saponins basis from its use as a food additive ranged from 0 mg/kg bw per day in infants to 0.3 mg/kg bw per day in toddlers. The 95th percentile of exposure to quillaia extract (E 999) expressed on a saponins basis ranged from 0 mg/kg bw per day in infants to 0.8 mg/kg bw per day in toddlers.

The main contributing food category to the total mean exposure estimates for all population groups was flavoured drinks in all exposure scenarios (Appendix E).

3.5.3 | Dietary exposure to quillaia extract (E 999) including the proposed extension of uses in solid and liquid food supplements and botanical nutrients

A new exposure estimate was performed for the assessment of the request for extension of uses (Table 12). Considering the proposed request for extension of use in the food supplements and nutrients, the 'food supplements consumers only scenario' (EFSA ANS Panel, 2017) was performed (only the consumers of food supplements were considered while still considering their whole diet). This scenario considers the MPLs as defined for the already authorised food categories (i.e. FC 14.1.4 and FC 14.2.3) or the typical use levels from the reported use levels (Appendix A) at the time of the re-evaluation (EFSA FAF Panel, 2019), and the proposed maximum levels for the FCs 17.1 and 17.2 either for direct addition or considering the proposed use of the food additive, including as a carrier, in botanical nutrients.

TABLE 12 Summary of estimated exposure to quillaia extract (E 999) expressed on a saponins basis, from its use as a food additive in the 'food supplements consumers only scenario', in four population groups (minimum–maximum across the dietary surveys in mg saponins/kg bw per day).

	Children (3–9 years)	Adolescents (10–17 years)	Adults (18–64 years)	The elderly (≥ 65 years)
Food supplements consumer only scenario using the MPLs for the authorised food categories and the proposed uses for Annexes II and III				
Mean[a]	1.04–5.81	0.18–4.41	0.16–2.39	0.16–3.64
95th percentile[b]	2.54–11.12	2.65–14.71	2.43–11.40	1.43–6.14
Food supplements consumer only scenario using the typical use levels for the authorised food categories and the proposed uses for Annexes II and III				
Mean[a]	0.87–4.73	0.11–4.19	0.11–2.27	0.16–3.62
95th percentile[b]	2.35–10.49	1.70–14.24	1.51–11.38	1.22–6.14

[a]Mean calculated on a minimum of 5 consumers.

[b]p95 calculated on a minimum of 60 consumers.

In the 'food supplements consumers only scenario' using the MPLs for the authorised food categories and the proposed use levels for the food supplements, mean exposure to quillaia extract (E 999) expressed on a saponins basis, from its use as a food additive ranged from 0.2 mg/kg bw per day in adolescents, adults and the elderly to 5.8 mg/kg bw/day in children. The 95th percentile of exposure to quillaia extract (E 999) ranged from 1.4 mg/kg bw per day in the elderly to 14.7 mg/kg bw per day in adolescents.

In the 'food supplements consumers only scenario' using the typical use levels for the authorised food categories and the proposed use levels for the food supplements, mean exposure to quillaia extract (E 999) expressed on a saponins

basis, from its use as a food additive ranged from 0.1 mg/kg bw per day in adolescents and adults to 4.7 mg/kg bw/day in children. The 95th percentile of exposure to quillaia extract (E 999) ranged from 1.2 mg/kg bw per day in the elderly to 14.2 mg/kg bw per day in adolescents.

Quillaia extract is requested to be authorised in food supplements not intended to infants and young children, therefore for these populations, the regulatory scenario exposure results covered the extension of uses.

3.5.4 | Uncertainty analysis

Potential sources of uncertainty in the exposure assessment of quillaia extract (E 999) have been introduced above. In accordance with the guidance provided in the EFSA opinion related to uncertainties in dietary exposure assessment (EFSA, 2007), the following sources of uncertainties have been considered and summarised in Table 13.

TABLE 13 Qualitative evaluation of influence of uncertainties on the dietary exposure estimate.

Sources of uncertainties	Direction[a]
Consumption data: different methodologies/representativeness/underreporting/misreporting/no portion size standard	+/–
Methodology used to estimate high percentiles (95th) long-term (chronic) exposure based on data from food consumption surveys covering only a few days	+
Correspondence of reported use levels to the food items in the EFSA Comprehensive Database: uncertainties to which types of food the levels refer	+/–
Uncertainty in possible national differences in use levels of food categories	+/–
Concentration data	
Use levels considered applicable to all foods within the entire food category, whereas on average 0.1% of the foods, belonging to food categories with foods labelled with additive, was labelled with the additive	+
Food categories selected for the exposure assessment: inclusion of food category 14.2.3 without considering the restriction/exception of only '*cidre bouché*'	+
Regulatory maximum level exposure assessment scenario	
Exposure calculations based on the MPL according to Annex II to Regulation (EC) No 1333/2008	+
Refined exposure assessment scenarios	
Exposure calculations based on the maximum or mean levels (reported use from industries)	+/–

[a]+, uncertainty with potential to cause overestimation of exposure; –, uncertainty with potential to cause underestimation of exposure.

Quillaia extract (E 999) is currently authorised in two food categories. As use levels were reported by industries for both categories, all current authorisations were taken into account in the refined assessment [without taking into account the restriction for one of the two food categories (FC 14.2.3, Section 3.4.1). The Panel noted that information from the Mintel GNPD (Appendix B) indicated that no cider or perry were labelled with quillaia extract (E 999). In the 'Carbonated Soft Drinks' subcategory in Mintel (belonging to FC 14.1.4), only 0.2% ($n = 12$ products) were labelled with quillaia extract (E 999). In the assessment, all flavoured drinks are assumed to contain quillaia extract (E 999).

In the 'food supplements consumers only scenario', that was calculated to evaluate the request for extension of uses in the food supplements and as a carrier in botanical nutrients, food categories are considered at their MPLs/typical use levels or proposed maximum use levels. In addition, only the consumers of food supplements are considered, which for some food surveys were very few (Appendix F). Not considering the whole population avoids 'diluting' the exposure with non-consumers of food supplements but only reflects the potential exposure of these food supplements' consumers. It is also noted that these scenarios considered that all food supplements contain E 999 and that all flavoured drinks are assumed to have added botanical nutrients containing quillaia extract (E 999), resulting in an overestimation of the dietary exposure.

Overall, the Panel considered that the uncertainties summarised in Table 11 resulted in an overestimation of the exposure to quillaia extract (E 999) from its use as a food additive according to Annex II in both the MPL and refined exposure scenario. This was also true for the exposure scenario considering the requested extension of use of quillaia extract (E 999) as a food additive according to Annexes II and III.

3.6 | Proposed revision to existing EU specifications for quillaia extract (E 999)

The potential exposure to impurities from the use of the quillaia extract (E 999) can be calculated by assuming that the impurity is present in the food additive up to a limit value and then by calculation pro-rata to the estimates of exposure to the food additive itself.

With regard to the dietary exposure to the food additive, considering only the currently authorised uses, the Panel considered the exposure calculations for quillaia extract (E 999) as presented in Table 11.

The Panel considered the refined brand-loyal exposure assessment scenario covering the general population as the most appropriate and realistic scenario for risk assessment of the quillaia extract (E 999).

For the current assessment, the highest exposure levels for the mean and 95th percentile among the different population groups were considered, i.e. 0.3 and 0.9 mg saponins/kg bw per day, respectively, for toddlers (Table 11).

The level of the impurity in the food additive combined with the estimated or potential intakes of E 999, presented in Table 11, could result in an exposure which can be compared with the following reference points (RP) or health-based guidance values (HBGV) (Table 14) for the undesirable impurities present in E 999.

TABLE 14 Reference points/health-based guidance values for impurities and constituents potentially present in **quillaia extract (E 999).**

Impurity/constituent/ HBGV/RP (μg/kg bw)	Basis/reference
Lead (Pb)/0.5 ($BMDL_{01}$)	The reference point is based on a study demonstrating perturbation of intellectual development in children with the critical response size of 1 point reduction in IQ. The EFSA CONTAM Panel mentioned that a 1 point reduction in IQ is related to a 4.5% increase in the risk of failure to graduate from high school and that a 1 point reduction in IQ in children can be associated with a decrease of later productivity of about 2%. A risk cannot be excluded if the exposure exceeds the $BMDL_{01}$ (MOE lower than 1). EFSA CONTAM Panel (2010)
Mercury (Hg)/4 (TWI)	The HBGV was set using kidney weight changes in male rats as the pivotal effect. Based on the $BMDL_{10}$ of 0.06 mg/kg bw per day, expressed as mercury, and an uncertainty factor of 100 to account for inter and intra species differences, with conversion to a weekly basis and rounding to one significant figure, a TWI for inorganic mercury of 4 μg/kg bw per week, expressed as mercury was established. EFSA CONTAM Panel (2012)
Cadmium (Cd)/2.5 (TWI)	The derivation of the reference point is based on a meta-analysis to evaluate the dose–response relationship between selected urinary cadmium and urinary beta-2-microglobulin as the biomarker of tubular damage recognised as the most useful biomarker in relation to tubular effects. A group-based $BMDL_5$ of 4 μg Cd/g creatinine for humans was derived. A chemical specific adjustment factor of 3.9 was applied to account for human variability in urinary cadmium within each dose-subgroup in the analysis resulting in a reference point of 1.0 μg Cd per g creatinine. In order to remain below 1 μg Cd/g creatinine in urine in 95% of the population by age 50, the average daily dietary cadmium intake should not exceed 0.36 μg Cd/kg bw, corresponding to a weekly dietary intake of 2.5 μg Cd/kg bw. EFSA CONTAM Panel (2009a)
Arsenic (As)/0.3–8 ($BMDL_{01}$)	The reference point is based on a range of benchmark dose lower confidence limit ($BMDL_{01}$) values between 0.3 and 8 μg/kg bw per day identified for cancers of the lung, skin and bladder, as well as skin lesions. In general, the MOE should be at least 10,000 if the reference point is based on carcinogenicity in animal studies. However, as the BMDL for As is derived from human studies, an interspecies extrapolation factor (i.e. 10) is not needed, i.e. an MOE of 1000 would be sufficient. EFSA CONTAM Panel (2009b); EFSA Scientific Committee (2012)

Abbreviations: $BMDL_{01}$, benchmark dose (lower confidence limit); bw, body weight; HBGV, health-based guidance value; MOE, margin of exposure; RP, reference point; TWI, Tolerable Weekly Intake.

The risk assessment of the undesirable impurities helps inform whether there could be a possible health concern if these impurities would be present at the limit values in the food additive. The assessment is performed by calculating the MOE (margin of exposure) by dividing the reference point (e.g. BMDL) by the exposure estimate (Table 11), or by estimating the contribution of the use of E 999 to the HBGV (expressed as percentage of the HBGV).

3.6.1 | Toxic elements

The Panel noted that the occurrence data on toxic elements submitted by the IBO are substantially lower than the current limits in the EU specifications (Documentation provided to EFSA n. 1, 2).

Analytical results for arsenic, cadmium, lead and mercury in commercial samples of E 999 were reported by the IBO and are compiled in Section 3.2.2.

Taking into account the data submitted by the IBO on toxic elements (Section 3.2.2), the Panel performed the risk assessment that would result if these toxic elements were present in E 999, using three different scenarios:

I) at the current limits in the EU specification calculated on a saponins basis, considering a minimum content of 20%[14] saponins (i.e. 2, 2, and 1 mg/kg for arsenic, lead and mercury, respectively on the product basis would correspond to 10, 10, and 5 mg/kg on a saponins basis);
II) at the limits proposed by one IBO for the toxic elements in E 999 Type 1,[15] expressed on a saponins basis (Table 5); and
III) at the rounded up highest measured level of the toxic elements in E 999 Type 1 or, in the absence of any measured value(s), the highest LOQ expressed on a saponins basis.

The Panel emphasised that the choice of the maximum limit values as well as other considerations, such as on multiple sources of exposure to conclude on the maximum limits for toxic elements in the specifications, is in the remit of risk

[14]The current limits of toxic elements in the EU specification are expressed on quillaia extract E 999. In order to express the limits on a saponins basis, the Panel considered a minimum amount of saponins content of 20% (minimum content of saponins in quillaia extract Type 1), as a worst-case scenario. This is because to reach a certain amount of saponins in a food, a higher amount of quillaia extract Type 1 than Type 2 has to be added, contributing to a higher exposure to impurities present in the extract.
[15]Highest proposed limits, compared to the ones of quillaia extract Type 2 (Table 5).

management. The numbers used here are merely taken to support the risk assessment of these toxic elements as presented below.

The outcome of the risk assessment (Table 15) illustrates the health impact that could result if revised maximum limits for toxic elements were to be used.

TABLE 15 Risk assessment for toxic elements.

	Scenario I based on the current limits in the EU specifications calculated on a saponins basis, considering a minimum content of 20% saponins			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 10 mg/kg of saponins**	**% of the TWI for Hg at 5 mg/kg of saponins**	**-**	**MOE for As at 10 mg/kg of saponins**
0.3[a]	167	0.26	-	100–2667
0.9[b]	56	0.79	-	33–889
	Scenario II based on the limits proposed by one IBO for the toxic elements in E 999 Type 1, expressed on a saponins basis (Documentation provided to EFSA n. 2)			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 10 mg/kg of saponins**	**% of the TWI for Hg at 1 mg/kg of saponins**	**% of the TWI for Cd at 2 mg/kg of saponins**	**MOE for As at 10 mg/kg of saponins**
0.3[a]	167	0.05	0.2	100–2667
0.9[b]	56	0.16	0.5	33–889
	Scenario III based on the rounded up highest measured level of the toxic elements in E 999 Type 1 or, in the absence of any measured value(s), the highest LOQ expressed on a saponins basis (Documentation provided to EFSA n. 2)			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 3 mg/kg of saponins**	**% of the TWI for Hg at 1 mg/kg of saponins**	**% of the TWI for Cd at 1 mg/kg of saponins**	**MOE for As at 6 mg/kg of saponins**
0.3[a]	556	0.05	0.1	167–4444
0.9[b]	185	0.16	0.3	56–1481

[a]Highest exposure level among the different population groups (refined brand-loyal scenario–toddlers–mean) considering the currently authorised uses, see Table 11, Section 3.5.2.

[b]Highest exposure level among the different population groups (refined brand-loyal scenario–toddlers–95th percentile) considering the currently authorised uses, see Table 11, Section 3.5.2.

The resulting figures in Table 15 show that for arsenic, in all scenarios, the lower end of the range of the calculated MOE values was insufficient, i.e. below the target value of 1000. For the other toxic elements (cadmium, mercury, lead), exposure to them due to the use of the food additive does not give rise to safety concerns.

The Panel considered that the maximum limits in the EU specifications for toxic elements should be established based on actual levels in the commercial food additive. Taking into account the results of the estimation calculated by the Panel (Table 15) and the fact that the food additive is not the only potential dietary source of toxic elements, the Panel recommended the maximum limits for lead, mercury and arsenic to be lowered on the basis of the information provided by the IBO. The Panel noted that in the data submitted (Documentation provided to EFSA n. 1), cadmium was present in some of the samples (Section 3.2.2), and therefore, considered that a maximum limit for cadmium should be included in the specifications. The Panel also recommended that the limits would be expressed on a saponins basis. If the European Commission decides to revise the EU specifications, the estimates of toxic elements intake as above could be considered.

3.6.2 | Proposed revisions to the EU specifications

Based on the data available at the time of the re-evaluation, it had been proposed to revise the EU specifications for quillaia extract (E 999) in order to differentiate extracts of quillaia according to the saponins content (including a description of the principle of the method of analysis to quantify the content of saponins in line with the JECFA specifications), i.e. Type 1 and Type 2 (EFSA FAF Panel, 2019).

The Panel noted that the classification of quillaia extracts in different types (Type 1 and 2), depending on the content of saponins on the dried basis, does not reflect the commercial preparations (liquid and powder) of quillaia extracts as E 999. Commercial liquid preparations of quillaia extract (E 999) Type 2 (for which > 65% saponins on the dried basis is expected) contain 15%–18% saponins, while commercial powder preparations contain 74%–83% saponins.

Based on the new data submitted in response to the European Commission call for data, regarding impurities and considering that the maximum authorised use levels are proposed to be expressed on a saponins basis, the Panel considered that it is not needed to differentiate the extracts of quillaia with different saponins content. However, a minimum

content for the functional component (not less than 20% on the dried basis) is recommended to be included in the EU specifications.

In addition, based on the data available at the time of the re-evaluation, it had been proposed to revise the EU specifications for quillaia extract (E 999) in order to include the percentage range for polyphenols (including tannins), protein, polysaccharides including fibre and reducing sugars.

Analytical data on the content of polyphenols (including tannins), protein, polysaccharides including fibre and reducing sugars were submitted. Differences among the results of the content of the different components have been observed by the Panel, but different methodology has been applied for each of the components between the two manufacturers. As a result, the Panel was not able to propose quantitative values (percentage ranges) that can define the composition. However, the data reported do not indicate any concern regarding the presence of these components in the food additive E 999. Thus, the Panel proposed to revise the definition of E 999 to better describe the composition in a qualitative way, only (Table 16).

In order to avoid excessive contribution to the oxalate burden of the body, which could result in the formation of oxalate kidney stones, a maximum limit for calcium oxalate could be included in the specifications of the food additive E 999. Considering the proposed lowest technologically achievable level for oxalic acid of 0.08%[16] on the dried basis for the quillaia extract, and the worst case of an extract containing 20% saponins, a limit for calcium oxalate of 0.6% expressed on a saponins basis is recommended.

In addition, based on the new data submitted, the Panel considered that the microbiological criteria for quillaia extract Type 2 (Table 6) proposed by an IBO at the time of the re-evaluation (EFSA FAF Panel, 2019) cover quillaia extract Type 1 to be used as a food additive according to the data submitted by one manufacturer.

The CAS number 68990-67-0 reported in the JECFA specifications for both Type 1 and 2 quillaia extracts (JECFA, 2006, 2014) is also proposed to be included in the specifications.

Overall, based on the information provided by the IBO in response to the call for data (Documentation provided to EFSA n. 1, 2) and the above considerations, the Panel recommended the following revisions of the existing EU specifications for quillaia extract (E 999) as listed in Table 16. The Panel noted that the choice of amendment of the EU specifications is in the remit of risk management.

TABLE 16 Proposal for a revision of the existing EU Specifications for quillaia extract E 999.

	Commission regulation (EU) No 231/2012	Comment/justification for revision
Definition	Quillaia extract is obtained by aqueous extraction of *Quillaia saponaria* Molina, or other *Quillaia* species, trees of the family Rosaceae. It contains a number of triterpenoids saponins consisting of glycosides of quillaic acid. Some sugars including glucose, galactose, arabinose, xylose and rhamnose are also present, along with tannin, calcium oxalate and other minor components	Proposed revised definition: Quillaia extract is obtained by aqueous extraction of *Quillaia saponaria* Molina, or other *Quillaia* species, trees of the family Rosaceae. It contains a number of triterpenoids saponins consisting of glycosides of quillaic acid, polyphenols, carbohydrates in particular polysaccharides and reducing sugars, and to a minor extend proteins
CAS number	Not presently specified	68990-67-0
Assay		
Saponin content	Not presently specified	To include a minimum content for the functional component: Not less than 20% on the dried basis
Description	See Table 1	Unchanged
Identification		
pH	See Table 1	Unchanged
Purity		
Water	See Table 1	Unchanged
Arsenic	Not more than 2 mg/kg	Maximum limit to be expressed as mg/kg of saponins and lowered on the basis of the information provided and on the considerations of the Panel
Lead	Not more than 2 mg/kg	Maximum limit to be expressed as mg/kg of saponins and lowered on the basis of the information provided and on the considerations of the Panel
Mercury	Not more than 1 mg/kg	Maximum limit to be expressed as mg/kg of saponins and lowered on the basis of the information provided and on the considerations of the Panel

[16]Highest of the proposed lowest technologically achievable levels for quillaia extract (Type 1).

TABLE 16 (Continued)

	Commission regulation (EU) No 231/2012	Comment/justification for revision
Cadmium	Not presently specified	A maximum limit to be included in the specifications expressed as mg/kg of saponins on the basis of the information provided and on the considerations of the Panel
Calcium oxalate	Not presently specified	A maximum limit to be included in the specifications expressed as mg/kg of saponins on the basis of the information provided and on the considerations of the Panel
Microbiological criteria		
Aerobic plate count	Not presently specified	A maximum limit to be included in the specifications on the basis of the information provided and on the considerations of the Panel
Yeasts	Not presently specified	A maximum limit to be included in the specifications on the basis of the information provided and on the considerations of the Panel
Moulds	Not presently specified	A maximum limit to be included in the specifications on the basis of the information provided and on the considerations of the Panel
Escherichia coli	Not presently specified	To be absent, as recommended in the EFSA FAF Panel (2019) re-evaluation
Salmonella spp.	Not presently specified	To be absent, as recommended in the EFSA FAF Panel (2019) re-evaluation
Staphylococcus aureus	Not presently specified	To be absent, as recommended in the EFSA FAF Panel (2019) re-evaluation

3.6.3 | Impact of the proposed extension of use in the proposed revisions to the EU specifications

The potential exposure to toxic elements from the use of quillaia extract (E 999) considering the additional proposed extension of use described in Section 3.5.3 has been carried out similarly to what is presented in Section 3.6.1. In this context, the highest exposure levels in the 'food supplements consumers only scenario' for the mean and 95th percentile among the different population groups have been considered, i.e. 5.8 and 14.7 mg saponins/kg bw per day, respectively, for children and adolescents (Table 12).

The outcome of the risk assessment of the Panel is illustrated in Appendix G . When comparing the potential exposure to toxic elements (Appendix G) considering the exposure to E 999 including the proposed extension of uses (Section 3.4.1) with the RP/HBGV for the undesirable impurities presented in Table 12, the Panel noted that the entire calculated range of MOEs for arsenic (Table G1) was insufficient. The Panel noted that an amendment of the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012, as presented by the considerations made in Table 16 would still be recommended in case the proposed extension of use for E 999 would be authorised.

4 | DISCUSSION

The current assessment addresses the EFSA recommendations and data gaps indicated during the re-evaluation of quillaia extract (E 999) as a food additive (EFSA FAF Panel, 2019) to update its EU specifications (E 999) in Commission Regulation (EU) No 231/2012.

Additionally, the present opinion deals with the assessment of the proposed extension of use for quillaia extract (E 999) in food supplements supplied in a solid form, excluding food supplements for infants and young children (FC 17.1) and in food supplements supplied in a liquid form, excluding food supplements for infants and young children (FC 17.2), according to Annex II of Regulation (EU) No 1333/2008 (Table 8), and as a carrier in botanical nutrients according to Annex III of Regulation (EU) No 1333/2008, for use in flavoured drinks (FC 14.1.4) and food supplements (FC 17.1 and 17.2) (Table 9).

An additional proposed extension of use for quillaia extract (E 999) to be used as a carrier for glazing agents on entire fresh fruits and vegetables according to Annex II to Regulation (EC) No 1333/2008 has been received. EFSA requested information on the proposed use levels of E 999 on a saponins content basis. Since no reply to EFSA's request for additional data has been provided by this applicant, the Panel was not able to evaluate the safety of this extension of use.

At the time of the re-evaluation of E 999, it was recommended revising the maximum use levels for quillaia extract (E 999) established in Regulation (EC) No 1333/2008 to be expressed on saponin content, but this has not been implemented, yet. In order to estimate the current MPL, previously calculated as anhydrous extract, on a saponins basis, the maximum reported content of saponins in E 999 of 83.41% was considered (Tables 7–9). The Panel noted that had the reported content of saponins in E 999 been used, this could have resulted in a ~10% lower estimate of exposure to E 999 expressed on a saponins basis.

Quillaia extract (E 999) is currently authorised in two food categories (Table 7). The exposure to this food additive was estimated based on the currently authorised uses and the MPLs in the regulatory maximum level exposure assessment

scenario or considering the reported use levels at the time of the re-evaluation in the refined exposure scenarios (Table 11). An additional exposure scenario, 'food supplements consumers only scenario' (Table 12), has been calculated considering the proposed extension of uses (Tables 8 and 9). Overall, the Panel considered that the calculated exposure to quillaia extract (E 999) was overestimated in all scenarios.

At the time of the re-evaluation (EFSA FAF Panel, 2019), it had been concluded that any toxicity associated with quillaia extract (E 999) is due to its constituent saponins and, therefore, established an ADI of 3 mg saponins/kg bw per day for quillaia extract (E 999) based on the NOAEL of 1500 mg quillaia extract/kg bw per day and the conservative assumption that it contained 20% saponins, and by applying an uncertainty factor of 100. Therefore, exposure to quillaia extract (E 999) was calculated on a saponins basis.

Different exposure scenarios have been calculated (Sections 3.5.2 and 3.5.3). The Panel considered that since the food additive is used in FC 14.1.4 Flavoured drinks, the brand-loyal scenario is the most relevant for the risk assessment of E 999. Using the refined brand-loyal exposure assessment scenario, exposure ranged between 0 mg saponins/kg bw per day at the mean up to 0.9 mg saponins/kg bw per day at the 95th percentile. None of the exposure estimates for the different population groups of the brand-loyal scenario exceeded the ADI of 3 mg saponins/kg bw per day.

Two 'food supplements consumers only scenarios' have been calculated using the MPLs or the typical use levels for the authorised food categories, respectively, and the proposed use levels for the food supplements (Table 12). The Panel noted that these scenarios considered that all food supplements contain E 999 and that all flavoured drinks are assumed to have added botanical nutrients containing quillaia extract (E 999), resulting in an overestimation of the dietary exposure.

The calculated 'food supplements consumers only exposure assessment scenarios' considering the proposed extension of use of quillaia extract (E 999) resulted in an exceedance of the ADI at the maximum of the ranges at the mean for children, adolescents and the elderly, and for all populations at the 95th percentile.

In the re-evaluation (EFSA FAF Panel, 2019), it was noted that existing EU specifications for E 999 do not describe any range for saponins content in the food additive. The JECFA specifications, however, differentiate two types of quillaia extracts, Type 1 and Type 2, containing a different percentage of saponins and other parameters. The Panel considered, at that time, that similar differentiation of the extracts of quillaia should be presented in the EU specifications, including the percentage range for saponins, polyphenols (including tannins), protein, polysaccharides including fibre, reducing sugars, a maximum limit for calcium oxalate as well as microbiological specifications.

Based on the new data submitted in response to the European Commission call for data, regarding impurities, and considering that the maximum authorised use levels are proposed to be expressed on a saponins basis, the Panel considered that it is not needed to differentiate the extracts of quillaia with different saponins content. However, a minimum content for the functional component (not less than 20% on the dried basis) is recommended to be included in the EU specifications.

In response to the European Commission call for data, analytical data on levels of toxic elements (arsenic, lead, cadmium, mercury) in commercial samples of E 999 were provided by one IBO and respective limit values were proposed. The Panel noted that the occurrence data on toxic elements are substantially lower than the current limits in the EU specifications. Proposed limits for the four toxic elements were provided by the IBO. The Panel performed a risk assessment considering three different scenarios: (I) At the current limits in the EU specification calculated on a saponins basis, considering a minimum content of 20% saponins (i.e. 2, 2 and 1 mg/kg for arsenic, lead and mercury, respectively, on the product basis would correspond to 10, 10 and 5 mg/kg on a saponins basis), (II) at the limits proposed by one IBO for the toxic elements in E 999 Type 1, expressed on a saponins basis (Table 5) and (III) at the rounded up highest measured level of the toxic elements in E 999 Type 1 or, in the absence of any measured value(s), the highest LOQ expressed on a saponins basis. The potential exposure to these impurities from the use of E 999 was compared against the available health-based guidance values (HBGV) and reference points (RP). For arsenic, in all scenarios, the lower end of the range of the calculated MOE values was insufficient, i.e. below the target value of 1000. For the other toxic elements (cadmium, mercury, lead), exposure to them due to the use of the food additive does not give rise to safety concerns. The Panel recommended the maximum limits for lead, mercury and arsenic to be lowered on the basis of the information provided by the IBO. The Panel noted that in the data submitted, cadmium was present in some of the samples (Section 3.2.2), and therefore, considered that a maximum limit for cadmium should be included in the specifications. The Panel also recommended that the limits would be expressed on a saponins basis.

Analytical data on the content of polyphenols (including tannins), protein, polysaccharides including fibre and reducing sugars were submitted. Differences among the results of the content of the different components have been observed by the Panel, but different methodology has been applied for each of the components between the two manufacturers. As a result, the Panel was not able to propose quantitative values (percentage ranges) that can define the composition. However, the data reported do not indicate any concern regarding the presence of these components in the food additive E 999. Thus, the Panel proposed to revise the definition of E 999 to better describe the composition in a qualitative way, only.

With respect to calcium oxalate, in order to avoid excessive contribution to the oxalate burden of the body, which could result in the formation of oxalate kidney stones, a maximum limit could be included in the specifications of the food additive E 999. Considering the proposed lowest technologically achievable level for oxalic acid of 0.08% on the dried basis for the quillaia extract, and the worst case of an extract containing 20% saponins, a limit for calcium oxalate of 0.6% expressed on a saponins basis is recommended.

Microbiological criteria are proposed to be included on the basis of the information provided. The Panel considered that the microbiological criteria for quillaia extract Type 2 (Table 6) proposed by an IBO at the time of the re-evaluation (EFSA FAF Panel, 2019), cover quillaia extract Type 1 to be used as a food additive, according to the data submitted by one manufacturer.

Overall, the Panel considered that the technical data provided, support an amendment of the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012 as presented by the recommendations made in Table 16.

The potential exposure to impurities from the use of E 999, considering the proposed extension of uses, has also been carried out. The Panel noted that an amendment of the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012, as presented by the considerations made in Table 16 would still be recommended in case the proposed extension of use for E 999 would be authorised.

5 | CONCLUSIONS

The Panel concluded that the technical data provided by one IBO support an amendment of the specifications for quillaia extract (E 999) laid down in Commission Regulation (EU) No 231/2012, as presented by the recommendations made in Table 16.

Additionally, the Panel concluded that the exposure estimates considering the proposed extension of use for E 999 in FC 17.1 Food supplements supplied in a solid form, excluding food supplements for infants and young children and in FC 17.2 Food supplements supplied in a liquid form, excluding food supplements for infants and young children, and as a carrier in botanical nutrients according to Annex III of Regulation (EU) No 1333/2008, for use in flavoured drinks and food supplements, if authorised, could result in an exceedance of the ADI at the maximum of the ranges of the mean for children, adolescents and the elderly, and for all populations at the 95th percentile.

6 | DOCUMENTATION AS PROVIDED TO EFSA

1. Intertek Health Sciences, Inc., 2021. Submission of data in response to the call for technical data on the permitted food additive quillaia extract (E 999). Submitted on 05 October 2021.
2. Intertek Health Sciences, Inc., 2023. Clarification on the data submitted in response to the call for technical data on the permitted food additive quillaia extract (E 999). Submitted on 26 April 2023.
3. Naturex SA, 2022. Technical dossier for the request on the extension of use: Modification of the condition of use of quillaia extract (E 999) in food supplements supplied in a solid form, excluding food supplements for infants and young children and food supplements supplied in a liquid form, excluding food supplements for infants and young children, and in botanical nutrients. Submitted on 22 July 2022.
4. Naturex SA, 2023. Clarification on the data submitted for the request on the extension of use: Modification of the condition of use of quillaia extract (E 999) in food supplements supplied in a solid form, excluding food supplements for infants and young children and food supplements supplied in a liquid form, excluding food supplements for infants and young children, and in botanical nutrients. Submitted on 23 February 2023.
5. Polynatural Holding SPA, 2023. Technical dossier for the request on the extension of use: Modification of the condition of use of quillaia extract (E 999) to be used as a carrier for glazing agents applied on entire fresh fruits and vegetables. Submitted on 22 July 2023.

ABBREVIATIONS

AAS	atomic absorption spectrophotometry
ADI	acceptable daily intake
ANS Panel EFSA	Panel on Food Additives and Nutrient Sources added to Food
BMDL	benchmark dose (lower confidence limit)
bw	body weight
CAS	Chemical Abstract Service
CFU	colony forming unit
FAF Panel	Panel on Food Additives and Flavourings
FAO/WHO	Food and Drug Organization/World Health Organization
FC	food category
HBGV	health-based guidance value
HPLC	high-performance liquid chromatography
IBO	interested business operator
ICP-MS	inductively coupled plasma-mass spectrometry
JECFA	Joint FAO/WHO Expert Committee on Food Additives
LOD	limit of detection
LOQ	limit of quantification
Mintel's GNPD	Mintel's Global New Products Database

MOE	margin of exposure
MPL	maximum permitted level
NOAEL	no observed adverse effect level
RP	reference point
RP-HPLC	reverse-phase high performance liquid chromatography
TWI	Tolerable Weekly Intake
UV	ultraviolet

ACKNOWLEDGEMENTS
The FAF Panel wishes to acknowledge all European competent institutions, Member State bodies and other organisations that provided data for this scientific output.

CONFLICT OF INTEREST
If you wish to access the declaration of interests of any expert contributing to an EFSA scientific assessment, please contact interestmanagement@efsa.europa.eu.

REQUESTOR
European Commission

QUESTION NUMBER(S)
EFSA-Q-2022-00541; EFSA-Q-2022-00475; EFSA-Q-2022-00476

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: EFSA FAF Panel (EFSA Panel on Food Additives and Flavourings), Younes, M., Aquilina, G., Castle, L., Degen, G., Engel, K.-H., Fowler, P., Frutos Fernandez, M. J., Gundert-Remy, U., Gürtler, R., Husøy, T., Manco, M., Mennes, W., Moldeus, P., Passamonti, S., Shah, R., Waalkens-Berendsen, I., Wright, M., Cheyns, K. … Fürst, P. (2024). Follow-up of the re-evaluation of quillaia extract (E 999) as a food additive and safety of the proposed extension of uses. *EFSA Journal*, *22*(2), e8563. https://doi.org/10.2903/j.efsa.2024.8563

APPENDICES

Appendix A: Summary of reported use levels (mg/kg or mg/L as appropriate) of quillaia extract (E 999) as anhydrous extract, provided by industry.

Appendix B: Number and percentage of food products labelled with quillaia extract (E 999) out of the total number of food products present in the Mintel GNPD per food subcategory between 2018 and 2023..

Appendix C: Concentration levels of quillaia extract (E 999) used in the exposure assessment scenarios (mg/L or mg/kg as appropriate).

Appendix D: Summary of total estimated exposure of E 999 from its use as a food additive for the regulatory maximum level exposure assessment scenario and the refined exposure assessment scenarios per population group and survey: mean and 95th percentile (mg/kg bw per day).

Appendix E: Main food categories contributing to exposure to E 999 using the regulatory maximum level exposure assessment scenario and the refined exposure assessment scenarios (> 5% to the total mean exposure).

Appendix F: Summary of total estimated exposure of E 999 from its use as a food additive for the food supplements consumers only exposure assessment scenario, using the MPLs or the use levels for the authorised FCs per population group and survey: mean and 95th percentile (mg/kg bw per day).

Appendices A–F can be found in the online version of this output (in the 'Supporting information' section).

Appendix G: Risk assessment to toxic elements considering the proposed extension of use of quillaia extract (E 999) in food supplements according to Annex II and as a carrier in botanical nutrients according to Annex III.

TABLE G1 Risk assessment to toxic elements considering the proposed extension of use of quillaia extract (E 999).

	Scenario I based on the current limits in the EU specifications calculated on a saponins basis, considering a minimum content of 20% saponins			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 10 mg/kg of saponins**	**% of the TWI for Hg at 5 mg/kg of saponins**	**-**	**MOE for As at 10 mg/kg of saponins**
5.8[a]	9	5.0	-	5–138
14.7[b]	3	12.9	-	2–54
	Scenario II based on the limits proposed by one IBO for the toxic elements in E 999 Type 1, expressed on a saponins basis (Documentation provided to EFSA n. 2)			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 10 mg/kg of saponins**	**% of the TWI for Hg at 1 mg/kg of saponins**	**% of the TWI for Cd at 2 mg/kg of saponins**	**MOE for As at 10 mg/kg of saponins**
5.8[a]	9	1.0	3.2	5–138
14.7[b]	3	2.6	8.2	2–54
	Scenario III based on the rounded up highest measured level of the toxic elements in E 999 Type 1 or, in the absence of any measured value(s), the highest LOQ expressed on a saponins basis (Documentation provided to EFSA n. 2)			
Exposure to E 999 (mg saponins/kg bw per day)	**MOE for Pb at 3 mg/kg of saponins**	**% of the TWI for Hg at 1 mg/kg of saponins**	**% of the TWI for Cd at 1 mg/kg of saponins**	**MOE for As at 6 mg/kg of saponins**
5.8[a]	29	1.0	1.6	9–230
14.7[b]	11	2.6	4.1	3–91

[a]Highest exposure level among the different population groups (food supplements consumers only scenario–children–toddlers–mean), see Table 12, Section 3.5.3.
[b]Highest exposure level among the different population groups (food supplements consumers only scenario–children–95th percentile), see Table 12, Section 3.5.3.