SCIENTIFIC OPINION

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Safety of potassium polyaspartate (A-5D K/SD) for use as a stabiliser in wine

EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS)

Abstract

Potassium polyaspartate (A-5D K/SD) is proposed for use as a stabiliser in wine, with a maximum use level of 300 mg/L and typical levels in the range of 100-200 mg/L. The data provided in support of the current application were in accordance with the Tier 1 requirement of the Guidance for submission for food additive evaluations issued by the ANS Panel in 2012. In the *in vitro* tests provided by the applicant, potassium polyaspartate (A-5D K/SD) showed minimal proteolytic digestion and no absorption of the intact compound. Potassium polyaspartate (A-5D K/SD) tested negative in a bacterial reverse mutation assay performed in accordance with OECD TG 471 and in an *in vitro* mammalian cell micronucleus test performed in accordance with OECD TG 487. From a 90-day oral toxicity study in rats performed in accordance with OECD TG 408, a no observed adverse effect level (NOAEL) was set at 1,000 mg/kg bw per day, the highest dose tested. The Panel considered these data as fulfilling the requirements for the evaluation of the new food additive and did not request additional testing for chronic toxicity and carcinogenicity, nor for reprotoxicity and developmental toxicity. Exposure estimates to potassium polyaspartate (A-5D K/SD) from its proposed use were calculated for both typical and maximum use levels. In the worst case scenario of high-level intakes of potassium polyaspartate (A-5D K/SD) when used at the maximum proposed use level of 300 mg/L, the maximum estimated intake would be 1.8 mg/kg bw per day in the elderly and 1.4 mg/kg bw per day in adults, resulting in a margin of safety of approximately 550. The Panel concluded that there was no safety concern from the proposed use and use levels of potassium polyaspartate (A-5D K/SD).

Keywords: food additive, wine stabiliser, potassium polyaspartate, A-5D K/SD

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Summary

Following a request from the European Commission, in accordance with Regulation (EC) No 1331/2008 establishing a common authorisation procedure for food additives, food enzymes and food flavourings, the Panel on Food Additives and Nutrient Sources added to Food (ANS) was asked to deliver a scientific opinion on the safety of the proposed use of potassium polyaspartate (A-5D K/SD) as a stabiliser in wine. According to the applicant potassium polyaspartate (A-5D K/SD) is proposed for use as a stabiliser against tartrate crystal precipitation (anti-scaling additive) in wine (red, rosé and white wine) at a maximum level (ML) of use of 300 mg/L.

According to the applicant, potassium polyaspartate (A-5D K/SD) is proposed for use as a stabiliser against tartrate crystal precipitation in wine (red, rosé and white wine) at the typical use level of 100-200 mg/L and an ML of 300 mg/L, depending on the level of instability of the wine to be treated. The food category for which an authorisation is sought with the current application is 14.2 Alcoholic beverages, including alcohol-free and low-alcohol counterparts.

The Panel based its opinion on a dossier submitted by the applicant and on the additional clarification provided upon request from the European Food Safety Authority (EFSA) during the assessment process. The tiered approach to toxicological testing described in the 2012 ANS Panel 'Guidance for submission for food additive evaluations' was followed by the applicant and the minimal biological and toxicological dataset applicable to all compounds was submitted for evaluation by the Panel. Following the evaluation of the Tier 1 toxicological studies described below, the Panel did not request additional testing to be conducted.

The EFSA Comprehensive European Food Consumption Database was used to estimate the dietary exposure. Dietary exposure to potassium polyaspartate (A-5D K/SD) from its use as a food additive was estimated combining the food consumption data available within the EFSA Comprehensive European Food Consumption Database with the proposed MLs and the proposed typical use levels provided by the applicant. Different exposure scenarios were calculated. Uncertainties on the exposure assessment were identified and discussed with regard to their impact on the final exposure calculation.

Potassium polyaspartate (A-5D K/SD) is the potassium salt of polyaspartic acid, produced from L-aspartic acid and potassium hydroxide. The applicant has proposed specifications of 98% purity for the material. The Panel considered that the analytical information provided on five batches of the proposed food additive, produced independently according to the method of manufacture, showed that the additive can be consistently manufactured within its proposed specifications.

According to the applicant, the presence of potassium polyaspartate (A-5D K/SD) in red or white wine can be determined and quantified by calculating the difference in aspartic acid content before and after complete sample hydrolysis to aspartic acid monomer.

The applicant has provided information on the stability of the food additive potassium polyaspartate (A-5D K/SD) at different storage conditions, as well as in water and in wine.

The applicant submitted results from two *in vitro* tests aimed at assessing gastrointestinal digestibility and intestinal absorption of potassium polyaspartate (A-5D K/SD): a sequential proteolytic attack with pepsin (porcine) and pancreatin (porcine), and an absorption study in human colon adenocarcinoma Caco-2 cells *in vitro*. The results from these studies showed that proteolytic digestion of potassium polyaspartate (A-5D K/SD) was minimal and that no absorption of intact A-5D K/SD was observed *in vitro*.

The genotoxic potential of potassium polyaspartate (A-5D K/SD) was investigated in a bacterial reverse mutation assay and using an *in vitro* mammalian cell micronucleus test. No genotoxic effect was observed in either of these two standard regulatory studies carried out in recognised testing facilities according to the relevant guideline and Good Laboratory Practice (GLP) compliance and reported in accordance with the relevant guideline.

Data from two toxicity studies performed in rats were submitted as part of the dossier, a 14-day range-finding study performed to collect information of target organs and to appropriate dosing, and a 90-day subchronic toxicity study. The 90-day study was performed in accordance with the OECD Test

Guideline 408, modified to include assessment of additional parameters to allow for the identification of chemicals with the potential to cause neurotoxic, immunological or reproductive organ effects or endocrine-mediated effects. In this study, potassium polyaspartate (A-5D K/SD) was administered daily via gavage to groups of Wistar rats (10 animals per sex per dose), at doses of 250, 500 and 1,000 mg/kg bw per day.

Based on the findings of this study, the Panel considered that a no observed adverse effect level (NOAEL) of potassium polyaspartate (A-5D K/SD) was to be set at 1,000 mg/kg bw per day, the highest dose tested, and also that there were no triggers for additional toxicological testing.

The mean dietary exposure from the proposed use level of 200 mg/L ranged from 0.01 to 0.2 mg/kg bw per day in adults up to 0.04 to 0.4 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.0 in adults and from 0.3 to 1.2 mg/kg bw per day in the elderly.

At the proposed maximum level of 300 mg/L, the mean dietary exposure ranged from 0.02 to 0.4 mg/kg bw per day in adults up to 0.05 to 0.6 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.4 in adults and from 0.4 to 1.8 mg/kg bw per day in the elderly.

In consideration of the proposed use of potassium polyaspartate (A-5D K/SD) as a food additive, limited to wine, the Panel considered it appropriate to consider dietary exposure only in adults and in the elderly. The Panel acknowledged that data from the younger age groups (i.e. infants, toddlers, children and adolescents) showed some levels of intake from wine or other alcoholic consumption. Consumption of alcoholic beverages is not appropriate for these age groups, and these exposure estimates, which are very low, are most likely a result of the indirect consumption of alcoholic beverages (ranging from < 0.001 up to 0.10 mg/kg bw per day) as recipe ingredients of composite foods. Therefore, the Panel considered that these were not relevant for the current risk assessment.

Based on the NOAEL of the 90-day study and these exposure estimates, the Panel considered that there would be an adequate margin of safety from the proposed use and use levels (approximately 550 for the high-level elderly consumers at the proposed ML of 300 mg/L).

The Panel considered that the estimated margin of safety of 550 was higher than the value of 200 which could be derived for the uncertainty factor based on the default safety factors for toxicokinetics and toxicodynamics and extrapolation from a 90-day study to chronic exposure when using a 90-day study to derive an acceptable daily intake (ADI) (EFSA SC, 2012).

The Panel noted that the NOAEL of 1,000 mg/kg bw per day was the highest dose tested and that the exposure used for this comparison is a conservative estimate because the applicant indicated that for most wines typical levels of 100 to 200 mg/L of potassium polyaspartate (A-5D K/SD) would be sufficient. The Panel considers that, because of the possible uncertainties, the margin of safety estimated above is likely to be lower than the actual margin of safety.

Based on 4% breakdown of aspartic acid, the Panel estimated that the maximal amount of aspartic acid released would be 0.04 mg/kg bw per day at the typical use level and 0.07 mg/kg bw per day at the proposed ML. This intake could be compared with the estimates of dietary intake of aspartic acid estimates of mean and high-level exposure to aspartate ions from the diet (9.1 and 13 g/day, respectively, which is equivalent to approximately 130 and 186 mg/kg bw per day). The Panel considered that this additive use would increase dietary exposures by less than 0.05% at the proposed ML which it considered negligible.

The Panel concluded that there was no safety concern from the proposed use and use levels of potassium polyaspartate (A-5D K/SD) as a stabiliser in wine.

The Panel recommended that specifications for potassium polyaspartate (A-5D K/SD) should be revised in order to define limits for the possible presence of toxic elements and to clarify the identity of the other significant impurities.

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1. Introduction

The present scientific opinion deals with the evaluation of the safety of potassium polyaspartate (A-5D K/SD) for use as a stabiliser in wine.

1.1. Background and Terms of Reference as provided by the European Commission

1.1.1. Background

The use of food additives is regulated under the European Parliament and Council Regulation (EC) No 1333/2008[1] on food additives. 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.

An application has been introduced for the authorisation of the use of potassium polyaspartate (A-5D K/SD) as a stabiliser in wine (Food Category 14.2.2 of part E of Annex II to Regulation (EC) No 1333/2008) at the maximum level (ML) of use of 300 mg/L.

1.1.2. Terms of Reference

The European Commission requests the European Food Safety Authority to provide a scientific opinion on the safety of the proposed use of potassium polyaspartate (A-5D K/SD) as a stabiliser in wine in accordance with Regulation (EC) No 1331/2008[2] establishing a common authorisation procedure for food additives, food enzymes and food flavourings.

1.2. Information on existing evaluations and authorisations

1.2.1. Authorisation for experimental trials in accordance with Article 4 of Commission Regulation (EC) No 606/2009

Potassium polyaspartate (A-5D K/SD) has been authorised for experimental trials on tartaric stabilisation in white wine, rosé wine and red wine, in accordance with Art 4 of Commission Regulation (EC) No 606/2009[3], in Italy[4] and in Spain[5]. The same request was submitted to the relevant competent authority of France on 22 December 2014 and, at the time of submission of the dossier, it was still pending authorisation[6].

These authorisations have been granted/requested after the submission of physico-chemical, toxicokinetic, immunological and (geno)toxicological data available for A-5D K/SD and provided with the dossier submitted in support of the current application.

1.2.2. Authorisation and evaluations for polyaspartic acid and its sodium salt

Polyaspartic acid is not listed by the US National Toxicology Program (NTP) or the International Agency for Research on Cancer (IARC), nor is it regulated as a carcinogen by the US Occupational Safety & Health Administration (OSHA) (Documentation provided to EFSA n.1).

Authorisations and evaluations are available for the sodium salt of polyaspartic acid (CAS 34345-47-6), the polyaminoacid containing sodium instead of potassium. The sodium salt of polyaspartic acid is

[1] Regulation (EC) No 1333/2008 of the European Parliament and of the Council of 16 December 2008 on food additives. OJ L 354, 31.12.2008, pp. 16–33.
[2] Regulation (EC) No 1331/2008 of the European Parliament and of the Council of 16 December 2008 establishing a common authorisation procedure for food additives, food enzymes and food flavourings. OJ L 354, 31.12.2008, pp. 1–6.
[3] Commission Regulation (EC) No 606/2009 of 10 July 2009 laying down detailed rules for implementing Council Regulation (EC) No 479/2008 as regards the categories of grapevine products, oenological practices and the applicable restrictions. OJ L 193, 24.7.2009, p. 1–59
[4] Italian Ministry of Agricultural, Food and Forestry Policies – MIPAAF Protocol N. 0007017, 15 December 2014
[5] Department of Agriculture, Food and Environment, Aragon Government, authorisation issued on 4 September 2014
[6] General Directorate for Competition Policy, Consumer Affairs and Fraud Control – DGCCRF dossier n 4C/2014/12/9108

authorised for use in USA and Australia as a food contact substance, as a dispersant for fillers and an anti-scale additive in sugar processing; and as a water treatment agent used as a scale inhibitor in cooling tower and boiler water applications, with properties of non-phosphor, non-nitrogen, non-pollution and complete biodegradation.

2. Data and Methodologies

2.1. Data

The applicant has submitted a dossier in support of its application for the authorisation of potassium polyaspartate (A-5D K/SD) as a new food additive for the proposed use in food category 14.2 Alcoholic beverages, including alcohol-free and low-alcohol counterparts (Documentation provided to EFSA n.1).

Additional clarification was sought from the applicant during the assessment process (Documentation provided to EFSA n.2).

The EFSA Comprehensive European Food Consumption Database (Comprehensive Database[7]) was used to estimate the dietary exposure.

2.2. Methodologies

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

The current 'Guidance for submission for food additive evaluations' (EFSA ANS Panel, 2012) has been followed by the Panel for the evaluation of the application for authorisation of the new food additive potassium polyaspartate (A-5D K/SD).

The tiered approach to toxicological testing described in the Guidance above was followed by the applicant and the minimal dataset applicable to all compounds was submitted by the applicant for evaluation by the Panel. Following the evaluation of the Tier 1 toxicological studies described below, the Panel did not request additional testing to be conducted.

Dietary exposure to potassium polyaspartate (A-5D K/SD) from its use as a food additive was estimated combining the food consumption data available within the EFSA Comprehensive European Food Consumption Database with the proposed MLs and the proposed typical use levels provided by the applicant. Different scenarios were used to calculate exposure (see Section 3.2.2.). Uncertainties on the exposure assessment were identified and discussed with regard to their impact on the final exposure calculation (see Section 3.2.3.).

3. Assessment

3.1. Technical data

3.1.1. Identity of the substance

The applicant has provided the following information with respect to the identity of the food additive.

Chemical name:	L-aspartic acid, homopolymer, potassium salt
CAS No.:	64723-18-8
EINECS No.:	Not available

[7] Available online: http://www.efsa.europa.eu/en/datexfoodcdb/datexfooddb.htm

Synonyms:	Potassium polyaspartate; A-5D K/SD; A-5D K / SD; A-5D K SD; A-5DK/SD; A-5DK; KPA
Trade name:	Not yet assigned
Chemical formula:	$[C_4H_4NO_3K]_n$
Number average molecular weight:	1,100 g/mol
Weight average molecular weight:	5,300 g/mol

The structural formula of potassium polyaspartate (A-5D K/SD) is shown in Figure 1.

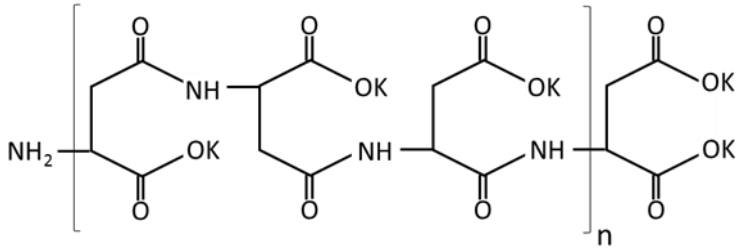


Figure 1: Structural formula of potassium polyaspartate (A-5D K/SD)

The applicant has submitted spectral data obtained with three different methods: Fourier transform infrared spectroscopy (FT-IR), proton magnetic resonance (H-NMR) and ultraviolet-visible spectrophotometry (UV-vis) for the potassium polyaspartate polymer and L-aspartic acid which demonstrated the expected spectral characteristics by each method.

With respect to particle size and distribution, the applicant has submitted data obtained by means of sieving (shaking time 5 min) showing that the majority of the test material (57.36% in weight) was greater than 125 µm; 24.27% was in the range 55–125 µm; 17.66% was in the range 45–75 µm and only 0.48% was lower than 45 µm. On the basis of the data provided, the Panel concluded that the possible nanofraction of the material was extremely small.

3.1.2. Specifications

The specifications for potassium polyaspartate (A-5D K/SD) as proposed by the applicant are presented in Table 1.

The applicant has provided reference FT-IR and H-NMR spectra.

The Panel noted there were no proposed specifications for toxic elements and that the identity of the other significant impurities was not clearly explained.

The Panel also noted that, based on the data presented in Table 1, the nominal purity of the substance is 98% w/w on dry matter and therefore the overall impurities should be lower than 2%, in contrast to what is reported in the proposed specifications.

Table 1: Specifications for potassium polyaspartate (A-5D K/SD) as proposed by the applicant (Documentation provided to EFSA n.2)

Parameter	Proposed specification	
Description:	Light brown powder without odour	
Identification:		
pH	7.5–8.5 (40% aqueous solution)	
Solubility	Water	> 1,000 g/L
	Xylene	< 5.0 g/L
	Dichloromethane	< 5.0 g/L
	Methanol	< 5.0 g/L
	Acetone	< 5.0 g/L
	Ethylacetate	< 5.0 g/L
	n-Heptane	< 5.0 g/L
Purity:		
Assay	Not less than 98.0% w/w on dry matter	
Degree of substitutions	Not less than 91.5% w/w on dry matter	
KOH	Not more than 2.0% w/w on dry matter	
Loss on drying	Not more than 11.0% w/w	
Aspartic acid	Not more than 1.0% w/w	
Other significant impurities	Not more than 0.1% w/w	

Certificate of analysis of five production batches

Five typical production batches of potassium polyaspartate (A-5D K/SD) from the production plant were analysed for their content of active substance and impurities. The batches were selected to cover a manufacturing period ranging from 2012 to 2015. The certificates of analysis were submitted by the applicant as part of the dossier (Documentation provided to EFSA n.1) and as additional information (Documentation provided to EFSA n.2). Information on the batches tested is reported in Table 2.

Table 2: Results of analysis of five production batches of potassium polyaspartate (A-5D K/SD)

Batch No.	KHKS 040412	KHKS-072512-1	KHKS-070214-1	KHKS-060414-1	KDK-110515-1
Assay (% w/w)	99	101	100	100	101
Degree of substitution (% w/w)	94	96	94	96	98
Loss of drying (% w/w)	8.4	8.3	5.5	5.4	4.9
Aspartic acid (% w/w)	0.4	0.4	0.5	0.5	0.4

The Panel considered that the analytical information provided on five batches of the proposed food additive, produced independently according to the method of manufacture, showed that the additive can be consistently manufactured within its proposed specifications.

3.1.3. Manufacturing process

The applicant has provided information on the manufacturing process used for the production of the food additive potassium polyaspartate (A-5DK SD).

Potassium polyaspartate (A-5DK SD) is produced from L-aspartic acid according to the reaction reported in Figure 2.

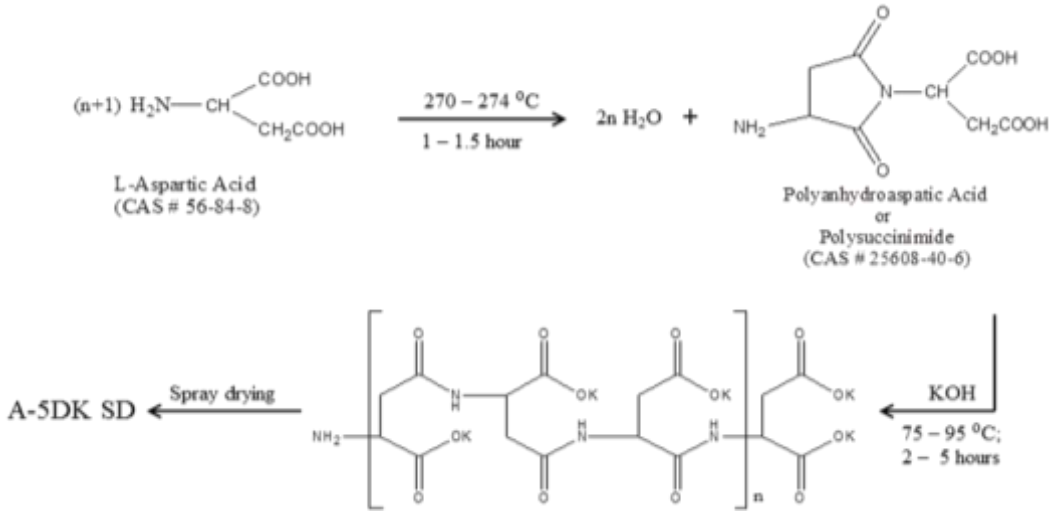


Figure 2: Manufacturing process for potassium polyaspartate (A-5DK SD) (Documentation provided to EFSA n.1).

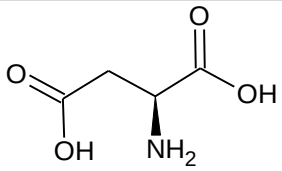
The thermic process transforms the aspartic acid in polysuccinimide that is insoluble. Polysuccinimide is then treated with potassium hydroxide under controlled conditions, thus allowing the opening of the ring and polymerisation of the units. Through this process, a 40% solution of potassium polyaspartate at pH 8.3 is obtained.

The last step of the production of the preparation of potassium polyaspartate (A-5D K/SD) is the spray drying phase, which results in a light tan powder at 92–95% dry matter.

Information on the two starting materials

The applicant has provided the following information on the two starting materials, L-aspartic acid monomer and caustic potash liquid 45%, used for the manufacturing of potassium polyaspartate (A-5D K/SD). The information provided is reported in Table 3.

Table 3: Information on the two starting materials used in the manufacture of potassium polyaspartate (A-5D K/SD)

	Starting materials	
Name:	L-Aspartic acid monomer	Caustic potash liquid 45%
IUPAC name:	L-Aspartic acid	Potassium hydroxide
CAS No.:	56-84-8	1310-58-3
EC No.:	200-291-6	215-181-3
Chemical formula:	$C_4H_7NO_4$	KOH
Structural formula:		KOH
Molecular weight:	133.10 g/mol	56.11 g/mol
Purity	≥98.5%	45–47%

Analysis of residual KOH cannot be performed directly because it is not possible to differentiate between K linked to potassium polyaspartate (A-5D K/SD) or KOH. Based on the proposed specification of 98% potassium polyaspartate (A-5D K/SD) in the dry material, residual KOH cannot be greater than 2%. As the pH of wine ranges between 3 and 3.8, any residual KOH would be neutralised during the production and storage of wine.

3.1.4. Methods of analysis in food

The applicant has submitted information on the single-laboratory validated method used for the determination of potassium polyaspartate (A-5D K/SD) in red or white wine. According to the method proposed, the quantification of the food additive is achieved by calculating the difference in aspartic acid content before and after complete sample hydrolysis to aspartic acid monomer.

The determination of aspartic acid liberated into wine after acid hydrolysis is performed by high-performance liquid chromatography using an external standard and fluorescence detector. The recovery was between 73% and 119% for different samples of red and white wine, which was within the acceptance criteria of 70–120%. The limit of detection (LoD) and limit of quantification of the method were 0.7 and 2.1 mg/mL respectively.

The applicant has also submitted information on the single-laboratory validated method used for the determination of potassium polyaspartate (A-5D K/SD) in water (e.g. the vehicle used for the toxicological studies). In this case, a UV spectrophotometric method based on the addition of a cationic polymer to a buffered solution of potassium polyaspartate (A-5D K/SD) was used. Turbidity was developed and absorbance of the solution was measured at 420 nm. The mean recovery was found to be 93.24%.

3.1.5. Stability of the substance, and reaction and fate in food

The applicant has provided information on the stability of the food additive potassium polyaspartate (A-5D K/SD) at different storage conditions, as well as in water and in wine (Documentation provided to EFSA n.1).

Dry matter was determined for two samples from the same batch stored for 8 weeks at 40 and 25°C, respectively. No significant variation in the content of dry matter occurred during storage in either of the two conditions. The dry matter content of two batches of A-5D K/SD did not significantly change (3% difference) after storage at ambient temperature for 2 years.

A validated spectroscopic method was used for the determination of the test article in water maintained at room temperature (27 ± 9°C) for 24 h. The two concentrations tested, approximately 6 and 100 mg/mL, showed small percentage changes compared to their initial concentration (–1.43% and –2.42%, respectively).

The applicant has submitted data from several experiments conducted in different varieties of red and white wines mainly intended to assess the efficacy of the food additive in preventing tartaric crystallisation and maintaining colour stability in red wines.

3.1.6. Proposed use and use levels

The proposed use of potassium polyaspartate (A-5D K/SD) is as a stabiliser against tartrate crystal precipitation (anti-scaling additive) in wine (red, rosé and white wine) at the typical use level of 100–200 mg/L. The applicant suggested a maximum level (ML) of use of 300 mg/L. The levels of potassium polyaspartate (A-5D K/SD) to be used depend on the level of instability of the wine to be treated. The applicant claimed that level of 100 mg/L is sufficient to obtain a complete inhibition of tartrate crystal formation during storage; however, in case of wines with high levels of tartrate instability, higher doses may be needed up to an ML of 300 mg/L.

The food category for which an authorisation is sought with the current application is 14.2 Alcoholic beverages, including alcohol-free and low-alcohol counterparts.

3.2. Exposure data

3.2.1. Food consumption data used for 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, 2011a). New consumption surveys recently[8] added to the Comprehensive database were also taken into account in this assessment[9].

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 as a result of under-reporting and/or misreporting of the consumption amounts by subjects. Nevertheless, the EFSA Comprehensive Database represents the best available source of food consumption data across Europe at present.

Food consumption data used for the exposure assessment were from the population groups: infants, toddlers, children, adolescents, adults and the elderly. For the present assessment, food consumption data were available from 33 different dietary surveys carried out in 19 European countries (Table 4).

Table 4: Population groups considered for the exposure estimates of potassium polyaspartate (A-5D K/SD)

Population	Age range	Countries with food consumption surveys covering more than 1 day
Infants	From 4 months up to and including 11 months of age	Bulgaria, Denmark, Finland, Germany, Italy, UK
Toddlers	From 12 months up to and including 35 months of age	Belgium, Bulgaria, Denmark, Finland, Germany, Italy, the Netherlands, Spain, UK
Children [(a)]	From 36 months up to and including 9 years of age	Austria, Belgium, Bulgaria, Czech Republic, Denmark, Finland, France, Germany, Greece, Italy, Latvia, Netherlands, Spain, Sweden, UK
Adolescents	From 10 years up to and including 17 years of age	Austria, Belgium, Cyprus, Czech Republic, Denmark, Finland, France, Germany, Italy, Latvia, Spain, Sweden, UK
Adults	From 18 years up to and including 64 years of age	Austria, Belgium, Czech Republic, Denmark, Finland, France, Germany, Hungary, Ireland, Italy, Latvia, Netherlands, Romania, Spain, Sweden, UK
The elderly [(a)]	From 65 years of age and older	Austria, Belgium, Denmark, Finland, France, Germany, Hungary, Ireland, Italy, Romania, Sweden, UK

(a): The terms 'children' and 'the elderly' correspond, respectively, to 'other children' and the merging of 'elderly' and 'very elderly' in the Guidance of EFSA on the 'Use of the EFSA Comprehensive European Food Consumption Database in Exposure Assessment' (EFSA, 2011a).

Consumption records were codified according to the FoodEx classification system (EFSA, 2011b). Nomenclature from the FoodEx classification system has been linked to the Food Classification System (FCS) as presented in Annex II of Regulation (EC) No 1333/2008, part D, to perform exposure estimates. In practice, FoodEx food codes were matched to the FCS food categories.

Food categories selected for the exposure assessment of potassium polyaspartate (A-5D K/SD)

The food category in which the use of potassium polyaspartate (A-5D K/SD) is proposed for use was selected from the nomenclature of the EFSA Comprehensive Database (FoodEx classification system), at the most detailed level possible (up to FoodEx Level 3) (EFSA, 2011b).

Overall, all wine or fortified wine food categories available in the FoodEx nomenclature were included in the exposure assessment as shown in Table 5.

[8] Available online: http://www.efsa.europa.eu/en/press/news/150428.htm
[9] Available online: http://www.efsa.europa.eu/en/datexfoodcdb/datexfooddb.htm

Table 5: Food categories available in the FoodEx nomenclature and considered for the exposure estimates of potassium polyaspartate (A-5D K/SD)

FoodEx Level 2	FoodEx Level 3	FoodEx Category No.
Wine	Wine (undefined)	A.14.02
	Wine, white	A.14.02.001
	Wine, white, sparkling	A.14.02.002
	Wine, red	A.14.02.003
	Wine, red, sparkling	A.14.02.004
Fortified and liqueur wines (e.g. vermouth, sherry, Madeira)	Fortified and liqueur wines (undefined)	A.14.03
	Vermouth	A.14.03.001
	Sherry	A.14.03.002

3.2.2. Exposure to potassium polyaspartate (A-5D K/SD) from its proposed use as a food additive

Estimate of exposure based on the Food Additives Intake Model (FAIM) template

The applicant has provided an estimate of exposure to potassium polyaspartate (A-5D K/SD) based on the output obtained using the FAIM model at the proposed ML of 300 mg/L and at the proposed typical use level of 200 mg/L in food category 14.2 Alcoholic beverages, including alcohol-free and low-alcohol counterparts (Documentation provided to EFSA n.1).

The Panel decided not to use the estimate of exposure generated from the FAIM tool and provided by the applicant because aggregation of the data resulted in an overestimate of the exposure.

The Panel therefore decided to perform a more refined assessment, limited to the consideration of the food categories described in Table 4.

Refined exposure assessment scenario

The exposure scenarios resulting from the refined assessment are presented in Table 6. Detailed results by population groups and survey are presented in Appendix A.

Table 6: Estimated exposure to potassium polyaspartate (A-5D K/SD) from its proposed use as a food additive: at the proposed use levels and at the proposed MLs

Estimated exposure (mg/kg bw per day)	Infants (4–11 months)	Toddlers (12–35 months)	Children (3–9 years)	Adolescents (10–17 years)	Adults (18–64 years)	The elderly (≥65 years)
Proposed typical use level: 200 mg/L						
Mean	<0.001	0–0.005	0–0.01	0–0.01	0.01–0.2	0.04–0.4
High level	<0.001	0–0.002	0–0.08	0–0.08	0–1.0	0.3–1.2
Proposed maximum Level: 300 mg/L						
Mean	<0.001	0–0.007	0–0.02	0–0.02	0.02–0.4	0.05–0.6
High level	<0.001	0–0.004	0–0.1	0–0.1	0–1.4	0.4–1.8

In consideration of the proposed use of potassium polyaspartate (A-5D K/SD) as a food additive, limited to wine, the Panel considered it appropriate to consider dietary exposure only in adults and in the elderly. The Panel acknowledged that data from the younger age groups (i.e. infants, toddlers, children and adolescents) showed some levels of intake from wine or other alcoholic consumption. Consumption of alcoholic beverages is not appropriate for these age groups, and these exposure estimates, which are very low, are most likely a result of the indirect consumption of alcoholic

beverages (ranging from < 0.001 up to 0.1 mg/kg bw per day) as recipe ingredients of composite foods. Therefore, the Panel considered that these were not relevant for the current risk assessment.

The mean dietary exposure from the proposed typical use level of 200 mg/L ranged from 0.01 to 0.2 mg/kg bw per day in adults up to 0.04 to 0.4 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.0 in adults and from 0.3 to 1.2 mg/kg bw per day in the elderly

At the proposed ML of 300 mg/L, the mean dietary exposure ranged from 0.02 to 0.4 mg/kg bw per day in adults up to 0.05 to 0.6 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.4 in the adults and from 0.4 to 1.8 mg/kg bw per day in the elderly

Main food categories contributing to exposure to potassium polyaspartate (A-5D K/SD) using the proposed ML and typical level of use

Within the food categories selected to perform the current exposure assessment, the main categories contributing to exposure to potassium polyaspartate (A-5D K/SD) using both levels of 200 mg/L and 300 mg/L proposed by the applicant are presented in Table 7.

Table 7: Main food categories contributing to exposure to potassium polyaspartate (A-5D K/SD) using both level of 200 mg/L and 300 mg/L for the proposed uses (% min–max) and the number of surveys ≥5% contribution (*n*) in which each food category contributes

FoodEX Level 3	Infants	Toddlers	Children	Adolescents	Adults	The elderly
	Range of % contribution to the total exposure (number of surveys)[(a)]					
Wine (undefined)	100 (1)	90.2–100 (2)	7.6–78.8 (2)	12.4–100 (10)	6.1–85.5 (13)	5.8–97.9(8)
Wine, white	97.7–100 (3)	11.7–82.5 (4)	13.7–100 (14)	15.3–92.4 (13)	5.0–59.6 (17)	6.3–65.1 (13)
Wine, white, sparkling	–	–		7.0–100 (5)	8.3–22.9 (2)	–
Wine, red	–	9.8–100 (6)	5.8–72.2 (13)	5.4–94.5 (14)	6.8–77.8 (16)	21.5–81.7 (13)
Fortified and liqueur wines (undefined)	–	–	11.1 (1)	7.2–23.3 (2)	–	5.8 (1)
Vermouth	–	19.2 (1)	16.6–64.8 (2)	9.4 (1)	–	–
Sherry	–	–	10.8–17.5 (2)	17.1 (1)	–	5.7 (1)

–: no information available
(a): The total number of surveys may be greater than the total number of countries listed in Table 3 because some countries submitted more than one survey for a specific population.

3.2.3. Uncertainty analysis

Uncertainties in the exposure assessment of potassium polyaspartate (A-5D K/SD) for its proposed use as a food additive have been discussed 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 are summarised in Table 8.

Overall, the Panel considered that the uncertainties identified would, in general, result in an overestimation of exposure to potassium polyaspartate (A-5D K/SD) as a food additive in European countries for the maximum level exposure scenario.

Table 8: Qualitative evaluation of influence of uncertainties on the dietary exposure estimate

Sources of uncertainties	Direction [(a)]
Consumption data: different methodologies/representativeness/under-reporting/misreporting/no portion size standard	+/–
Use of data from food consumption survey of a few days to estimate long-term (chronic) exposure for high percentiles (95th percentile)	+
Correspondence of reported use levels and analytical data to the food items in the EFSA Comprehensive Food Consumption Database: uncertainties regarding which types of food the levels refer to	+/–
Maximum and typical proposed use level: levels considered applicable for all items within the entire food category	+/-
Maximum level exposure assessment scenario: all food categories at the maximum proposed use level	+
Uncertainty in possible national differences in use levels of food categories	+/–

(a): +, uncertainty with potential to cause over-estimation of exposure; –, uncertainty with potential to cause underestimation of exposure.

3.3. Biological and toxicological data

3.3.1. Absorption, distribution, metabolism and excretion

Tier 1 absorption studies and *in vitro* gastrointestinal digestion

The applicant submitted results from two *in vitro* tests aimed at assessing gastrointestinal digestibility and intestinal absorption of potassium polyaspartate (A-5D K/SD):

- Sequential proteolytic attack with pepsin (porcine) and pancreatin (porcine).
- Absorption in human colon adenocarcinoma Caco-2 cells *in vitro*.

There was very limited breakdown to aspartic acid (less than 4%) in the simulated gastric digestion following incubation with porcine pepsin and pancreatin. However, any aspartic acid released and subsequently absorbed would go into the amino acid pool. The majority of the material remained as polyaspartate following simulated gastric digestion *in vitro*. Based on 4% breakdown of aspartic acid, the Panel estimated that the maximal amount of aspartic acid released would be 0.04 mg/kg bw per day at the typical use level and 0.07 mg/kg bw per day at the proposed ML. This intake could be compared with the estimates of dietary intake of aspartic acid estimates of mean and high-level exposure to aspartate ions from the diet (9.1 and 13 g/day, respectively, which is equivalent to approximately 130 and 186 mg/kg bw per day) (EFSA, 2008). The Panel considered that this additive use would increase dietary exposures by less than 0.05% at the proposed ML which it considered negligible. Based on a hypothetical 100% breakdown of potassium polyaspartate (A-5D K/SD), the Panel noted that corresponding intake of aspartic acid would correspond to the maximal intake levels of 1.2 and 1.8 mg/kg bw per day at the proposed typical and ML, respectively. Under this assumption, the Panel noted that intake of aspartic acid from the proposed use of potassium polyaspartate (A-5D K/SD) would correspond to less than 1.5% at the proposed ML.

There was very limited (less than 0.07% based on the LoD), if any, absorption of intact polyaspartate into either the cell layer or the receiving fluid (below the LoD of 0.7 mg/L in the basolateral fluid) in the Caco-2 cell absorption model at concentrations of 1 mg/mL *in vitro*. There was good correlation between the duplicate samples at each of the time points studied. All the administered material remained in the apical fluid. The Panel considered that there was negligible absorption of polyaspartate in this model.

The Panel noted that the Caco-2 cells model used was not specified with respect to the expression of transporters and metabolising enzymes.

3.3.2. Genotoxicity

Tier 1 genotoxicity testing

The applicant has submitted data from two *in vitro* genotoxicity tests in accordance with the recommendations of the EFSA Scientific Committee on genotoxicity testing (EFSA Scientific Committee, 2011). The basic test battery performed consisted of:

- A bacterial reverse mutation assay (OECD TG 471).
- An *in vitro* mammalian cell micronucleus test (OECD TG 487).

These were standard regulatory studies carried out in recognised testing facilities according to the relevant guideline and GLP compliance and were reported in accordance with the relevant guideline.

Potassium polyaspartate (A-5D K/SD) was evaluated in an Ames/Salmonella pre-incubation assay to determine its ability to induce reverse mutation at selected histidine loci in five tester strains of *Salmonella typhimurium* (TA1535, TA97a, TA98, TA100 and TA102) in the presence and absence of a metabolic activation system (S9). The tester strains were exposed to the test article in triplicate cultures at doses of 5000, 1500, 500, 150 and 50 µg/plate, both in the presence and absence of a metabolic activation system (S9). Dimethyl sulfoxide was used as a vehicle. The exposed bacteria were plated onto minimal glucose agar medium supplemented with L-histidine. The plates were incubated at 37ºC for 68–69 hours after which the histidine revertant colonies were counted and their frequency was compared with that in the vehicle control group. Concurrent positive control groups were also included in the experiment, as specified by the Test Guideline.

Results of this test indicated that the frequencies of histidine revertant colonies at all concentrations of potassium polyaspartate (A-5D K/SD) in the strains TA1535, TA97a, TA98, TA100 and TA102, with and without the presence of a metabolic activation system, were comparable to those observed in the vehicle control group, as per the criteria employed for evaluation of mutagenic potential. Concurrent positive controls demonstrated sensitivity of the assay both in the presence and absence of metabolic activation. Plate counts for the spontaneous histidine revertant colonies in the vehicle control groups were found to be within the frequency ranges expected from the laboratory historical control data.

The Panel considered that potassium polyaspartate (A-5D K/SD) was not mutagenic in *Salmonella* typhimurium strains TA1535, TA97a, TA98, TA100 and TA102.

Cultures of human peripheral blood lymphocytes were exposed to potassium polyaspartate (A-5D K/SD) dissolved in analytical grade water at concentrations of 5000, 1500 and 500 µg/mL (concentrations based upon preliminary solubility/precipitation and cytotoxicity studies) with and without a metabolic activation system (i.e. rat liver S9). Duplicate cultures were used at each concentration. In experiments No. 1 and No. 2, cells were exposed to the test article for 3 and 20 hours, respectively, without the supplementary metabolic activation system. In experiment No. 3, conducted with the supplementary metabolic activation system, the cells were exposed for 3 hours to the test article, 48 hours after the start of the culture. Cytochalasin B was added at 68 hours, whereas cell harvesting was performed 96 hours after the start of the culture. Positive, negative and vehicle controls, both with and without metabolic activation, were tested concurrently with the test article. Analytical grade water was used as the vehicle. Mitomycin C and vinblastine, known micronucleus forming agents, were employed as positive controls, at concentrations of 0.8 and 0.08 µg/mL, respectively, for the experiments without the metabolic activation system, whereas cyclophosphamide was employed at a concentration of 6.25 µg/mL for the experiment with the metabolic activation system. Each culture was harvested and slide preparations were made and stained with 5% Giemsa. Two thousand binucleated cells with well spread cytoplasm were evaluated microscopically for the presence of micronuclei, if any.

A comparison of the percentage incidence of micronucleated binucleated cells (BNCs) for each of the three experiments conducted with potassium polyaspartate (A-5D K/SD), either with or without the metabolic activation system, did not reveal any biologically significant or dose-related increase. Also, there was no incidence of a biologically significant increase in the percentage incidence of micronucleated BNCs at any of the concentration levels in the cultures treated with potassium polyaspartate (A-5D K/SD).

Sensitivity of the test system and activity of S9 mix were demonstrated in the positive control group. The positive controls (i.e. directly acting clastogen mitomycin C, directly acting aneugen vinblastine and indirectly acting clastogen cyclophosphamide) induced significant increase in frequencies of micronucleated binucleated cells over the concurrent controls, which validated the test method.

Assessment of the cytokinesis-block proliferation index made during the preliminary cytotoxicity test and the main study indicated that potassium polyaspartate (A-5D K/SD) exerted no cytotoxic effects on the cultured human lymphocytes at the test concentration of 5,000 µg/mL (3 hours of exposure and 20 hours of exposure).

The Panel considered that potassium polyaspartate (A-5D K/SD) did not induce chromosome breaks and/or gain or loss (i.e. it is not clastogenic and aneugenic) in cultured mammalian cells (i.e. human peripheral blood lymphocytes).

The Panel considered that, in line with its guidance 'In cases where all *in vitro* endpoints are clearly negative in adequately conducted tests, it can be concluded with reasonable certainty that the substance is not a genotoxic hazard' (EFSA ANS Panel, 2012).

3.3.3. Short-term and subchronic toxicity

Tier 1 toxicity testing – subchronic toxicity

The applicant has submitted data from two toxicity studies performed in rats:

- A 14-day range-finding study performed to collect information of target organs and to select appropriate doses for a 90-day study.
- A 90-day subchronic toxicity study (OECD TG 408), modified to include assessment of additional parameters described in the more recent guideline on repeated-dose 28-day oral toxicity study in rodents (OECD TG 407) to allow for the identification of chemicals with the potential to cause neurotoxic, immunological or reproductive organ effects or endocrine-mediated effects.

These were standard regulatory studies carried out in recognised testing facilities according to the relevant guideline and GLP compliance and were reported in accordance with the relevant guideline.

A repeated dose 14-day oral toxicity study of potassium polyaspartate (A-5D K/SD) in the Wistar rat was performed as a dose range finding study to assist in the design of the 90-day toxicity study. Based on these findings, the doses selected for the repeated dose 90-days oral toxicity study of potassium polyaspartate (A-5D K/SD) in the Wistar rat are 250, 500 and 1000 mg/kg bw.

The 90-day study performed involved daily oral administration via gavage of potassium polyaspartate (A-5D K/SD) to groups of Wistar rats 10 per sex per dose, at doses of 250, 500 and 1,000 mg/kg bw per day for 90 days to evaluate its toxicity and reversibility of toxicity, if any.

The findings of this study were:

- no mortality at any dose up to the dose of 1,000 mg/kg bw per day;
- no incidence of treatment related clinical abnormalities, and no ocular toxicity and neurotoxicity, at and up to the dose of 1,000 mg/kg bw per day;
- no adverse effect on body weight gain by the male and female rats treated at and up to the dose of 1,000 mg/kg bw per day;
- no effect on average daily food consumption by the male and female rats treated at and up to the dose of 1,000 mg/kg bw per day;
- no effect on the haematological parameters of male and female rats treated at and up to the dose of 1,000 mg/kg bw per day;
- no effect on the clinical chemistry and urinalysis parameters of male and female rats treated at and up to the dose of 1,000 mg/kg bw day;
- no significant alterations at the stage of the oestrous cycle at necropsy in females at and up to 1,000 mg/kg bw;

- no significant alterations in the absolute and relative organ weights;
- no treatment related gross and microscopic pathological alterations in the tissues of male and female rats treated at and up to the level of 1,000 mg/kg bw per day.

Based on the findings of this study, the authors report a no observed adverse effect level (NOAEL) of potassium polyaspartate (A-5D K/SD) in Wistar rats, following oral administration for 90 days, equal to or greater than 1,000 mg/kg bw. The Panel considered that the NOAEL was 1,000 mg/kg bw per day, the highest dose tested. The Panel acknowledged the choice of the top dose is related to the method of administration chosen (gavage), which was considered more relevant to the intended use of the food additive.

3.3.4. Chronic toxicity and carcinogenicity

In line with the current Guidance (EFSA ANS Panel, 2012), the data from the repeated dose 90-day oral toxicity study in rats conducted with potassium polyaspartate (A-5D K/SD) and from Tier 1 toxicokinetics did not trigger additional testing for chronic toxicity and carcinogenicity.

3.3.5. Reproductive and developmental toxicity

In line with the current Guidance (EFSA ANS Panel, 2012), the data from the repeated dose 90-day oral toxicity study in rats conducted with potassium polyaspartate (A-5D K/SD) and from Tier 1 toxicokinetics did not trigger additional testing for reproductive and developmental toxicity.

3.3.6. Hypersensitivity, allergenicity and food intolerance

Immunotoxicity

In the 90-day oral toxicity study performed with A-5D K/SD in rats, no effects have been observed in any of the parameters that may be indicative of an immunotoxic or immunomodulatory effect.

The potential stimulation of immuno cello has been assessed *in vitro* on pro-myelocytic human cell line THP-1, used as a surrogate of monocytes (Documentation provided to EFSA n.1). Prior to the study, the cytotoxicity of A-5D K/SD was assessed through the MTT test. The THP-1 cells were diluted to 10^6 cells/mL in RPMI 1640, supplemented with 10% heated-inactivated fetal calf serum (media) and cultured in 37°C in a 5% CO_2 incubator. Cytokine IL-8 release (with an enzyme-linked immunosorbent assay) and CD86 expression (with flow cytometric analysis) were then assessed. For IL-8 release, 1.0 x 10^6 cells were seeded in a 24-well plate. Cells were incubated for 24 h in the presence or absence of potassium polyaspartate (A-5D K/SD) at 2 mg/mL. Lipopolysaccharide from *Escherichia coli* serotype 0127:B8 10 ng/mL was used as a positive control.

The results showed that potassium polyaspartate (A-5D K/SD) did not induce any activation of the immune system (monitored parameters: up-regulation of CD86 and release of IL-8). Thus, the Panel considered that potassium polyaspartate (A-5D K/SD) has no immunotoxicity and no Tier 2 or 3 studies on immunotoxicity were necessary. In the positive control, a statistical significant increase in both CD86 expression and IL-8 release was observed.

Allergy

A literature search performed by the applicant in several scientific databases (PubMed, Medline, CAB Abstracts, Web of Sciences) revealed that no allergic reactions to the correspondent polymer of sodium salt have been observed.

Moreover, evaluation of the potential stimulation of immuno cells conducted on pro-myelocytic human cell lineTHP-1 showed that potassium polyaspartate (A-5D K/SD) did not induce any activation of immune response in this test system (Documentation provided to EFSA n.1).

Intolerance reactions

Up to now no inborn errors of metabolism have been reported for aspartic acid. Moreover, no intolerance reactions to the correspondent polymer of sodium salt have been observed. Thus,

potassium polyaspartate (A-5D K/SD) is not expected to cause intolerance reactions (Documentation provided to EFSA n.1).

3.3.7. Other studies

Neurotoxicity

On the 13th week of treatment of the 90-day oral toxicity study performed with potassium polyaspartate (A-5D K/SD) in rats, all animals were examined for assessment of sensory reactivity, assessment of grip strength and motor activity. These include the functional observational battery suggested by Moser (1989).

The neurological examinations (functional observations) conducted in the thirteenth week of the 90-day study did not reveal any remarkable and treatment related incidence of neurological abnormalities. Also, no findings indicative of a neurotoxic potential of potassium polyaspartate (A-5D K/SD) were encountered during these examinations. Thus, the Panel considered that potassium polyaspartate (A-5D K/SD) has no neurotoxicity and no Tier 2 or 3 studies were necessary.

Review of published literature

The published literature has been reviewed by the applicant to search for relevant references on polyaspartic acid. The methods used to identify relevant data, including the scope and criteria of literature searches, the database searched, the search strategy, language and time limitations, were submitted together with a compilation of the publication identified.

The majority of papers identified were on sodium polyaspartate or polyaspartic acid and were broadly similar to the studies submitted as part of the dossier.

Because of the presence of data for potassium polyaspartate, there was no need to use these as supporting evidence for read-across.

3.4. Discussion

According to the applicant, potassium polyaspartate (A-5D K/SD) is proposed for use as a stabiliser against tartrate crystal precipitation in wine (red, rosé and white wine) at the typical use level of 100–200 mg/L and a maximum level of 300 mg/L, depending on the level of instability of the wine to be treated. The food category for which an authorisation is sought with the current application is 14.2 Alcoholic beverages, including alcohol-free and low-alcohol counterparts.

Potassium polyaspartate (A-5D K/SD) is the potassium salt of polyaspartic acid, produced from L-aspartic acid and potassium hydroxide. The applicant has proposed specifications of 98% purity of the material.

According to the applicant, the presence of potassium polyaspartate (A-5D K/SD) in red or white wine can be determined and quantified by calculating the difference in aspartic acid content before and after complete sample hydrolysis to aspartic acid monomer.

The biological and toxicological data submitted were in accordance with the Tier 1 requirements of the 'Guidance for submission for food additive evaluations' (EFSA ANS Panel, 2012).

Based on these data the Panel considered that:

- Proteolytic digestion of A-5D K/SD was minimal.
- No absorption of intact A-5D K/SD was observed *in vitro*.
- Both required *in vitro* genotoxicity tests were negative.
- The NOAEL in the 90-day toxicity study was 1,000 mg/kg bw per day, the highest dose tested, and there were no triggers for additional toxicological testing.

In view of the Tier 1 results, the Panel considered that no Tier 2 or 3 testing was necessary.

The mean dietary exposure from the proposed use level of 200 mg/L ranged from 0.01 to 0.2 mg/kg bw per day in adults up to 0.04 to 0.4 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.0 in adults and from 0.3 to 1.2 mg/kg bw per day in the elderly.

At the proposed maximum level of 300 mg/L, the mean dietary exposure ranged from 0.02 to 0.4 mg/kg bw per day in adults up to 0.05 to 0.6 mg/kg bw per day in the elderly. The high-level intake ranged from 0 to 1.4 in adults and from 0.4 to 1.8 mg/kg bw per day in the elderly.

In consideration of the proposed use of potassium polyaspartate (A-5D K/SD) as a food additive, limited to wine, the Panel considered it appropriate to consider dietary exposure only in adults and in the elderly. The Panel acknowledged that data from the younger age groups (i.e. infants, toddlers, children and adolescents) showed some levels of intake from wine or other alcoholic consumption. Consumption of alcoholic beverages is not appropriate for these age groups, and these exposure estimates, which are very low, are most likely a result of the indirect consumption of alcoholic beverages (ranging from < 0.001 up to 0.1 mg/kg bw per day) as recipe ingredients of composite foods. Therefore, the Panel considered that these were not relevant for the current risk assessment.

Based on the NOAEL of the 90-day study and these exposure estimates, the Panel considered that there would be an adequate margin of safety from the proposed use and use levels (approximately 550 for the high-level elderly consumers at the proposed maximum level of 300 mg/L).

The Panel considered that the estimated margin of safety of 550 was higher than the value of 200 which could be derived for the uncertainty factor based on the default safety factors for toxicokinetics and toxicodynamics and extrapolation from a 90-day study to chronic exposure when using a 90-day study to derive an ADI (EFSA Scientific Committee, 2012).

The Panel noted that the NOAEL of 1,000 mg/kg bw per day was the highest dose tested and that the exposure used for this comparison is a conservative estimate because the applicant indicated that, for most wines, typical levels of 100–200 mg/L of potassium polyaspartate would be sufficient. The Panel considers that, because of the possible uncertainties, the margin of safety estimated above is likely to be lower than the actual margin of safety.

Based on 4% breakdown of aspartic acid, the Panel estimated that the maximal amount of aspartic acid released would be 0.04 mg/kg bw per day at the typical use level and 0.07 mg/kg bw per day at the proposed ML. This intake could be compared with the estimates of dietary intake of aspartic acid estimates of mean and high-level exposure to aspartate ions from the diet (9.1 and 13 g/day, respectively, which is equivalent to approximately 130 and 186 mg/kg bw per day) (EFSA, 2008). The Panel considered that this additive use would increase dietary exposures by less than 0.05% at the proposed ML which it considered negligible.

4. Conclusions

The Panel concluded that there was no safety concern from the proposed use and use levels of potassium polyaspartate (A-5D K/SD) as a stabiliser in wine.

5. Recommendations

The Panel recommended that specifications for potassium polyaspartate (A-5D K/SD) should be revised in order to define limits for the possible presence of toxic elements and to clarify the identity of the other significant impurities.

Documentation provided to EFSA

1. Application for registration of a new food additive. Summary dossier and annexes. February 2015. Submitted by ESSECO s.r.l.

2. Additional information. December 2015. Submitted by ESSECO s.r.l. in response to a request from EFSA. Updated in February 2016.

References

EFSA (European Food Safety Authority), 2007. Opinion of the Scientific Committee related to Uncertainties in Dietary Exposure Assessment. EFSA Journal, 2007;5(1):438, 54 pp. doi:10.2903/j.efsa.2007.438

EFSA (European Food Safety Authority), 2008. Scientific Opinion of the Panel on food additives, flavourings, processing aids and materials in contact with food (AFC) on a request from the Commission on magnesium aspartate, potassium aspartate, magnesium potassium aspartate, calcium aspartate, zinc aspartate, and copper aspartate added for nutritional purposes to food supplements. EFSA Journal 2008;6(12):883, 23 pp. doi: 10.2903/j.efsa.2008.883

EFSA (European Food Safety Authority), 2009. Guidance of the Scientific Committee on transparency in the scientific aspects of risk assessments carried out by EFSA. Part 2: General principles. EFSA Journal 2009;7(5):1051, 22 pp. doi:10.2903/j.efsa.2009.1051

EFSA (European Food Safety Authority), 2011a. Use of the EFSA Comprehensive European Food Consumption Database in Exposure Assessment. EFSA Journal 2011;9(3):2097, 34 pp. doi:10.2903/j.efsa.2011.2097

EFSA (European Food Safety Authority), 2011b. Evaluation of the FoodEx, the food classification system applied to the development of the EFSA Comprehensive European Food Consumption Database. EFSA Journal 2011;9(3):1970, 27 pp. doi:10.2903/j.efsa.2011.1970

EFSA ANS Panel (EFSA Panel on Food Additives and Nutrient Sources added to Food), 2012; Guidance for submission for food additive evaluations. EFSA Journal 2012;10(7):2760, 60 pp. doi:10.2903/j.efsa.2012.2760

EFSA Scientific Committee, 2011. Scientific opinion on genotoxicity testing strategies applicable to food and feed safety assessment. EFSA Journal 2011;9(9):2379, 66 pp. doi:10.2903/j.efsa.2011.2379

EFSA Scientific Committee, 2012. Guidance on selected default values to be used by the EFSA Scientific Committee, Scientific Panels and Units in the absence of actual measured data. EFSA Journal 2012;10(3):2579, 32 pp. doi:10.2903/j.efsa.2012.2579

Moser VC, 1989. Screening approaches for neurotoxicity–a functional observational battery. Journal of the American College of Toxicology. Volume 8, pp 85–93.

Abbreviations

ADI	acceptable daily intake
ANS Panel	EFSA Panel on Food Additives and Nutrient Sources added to Food
BNCs	micronucleated binucleated cells
bw	body weight
FAIM	Food Additives Intake Model
FCS	Food Classification System
FT-IR	Fourier transform infrared spectroscopy
H-NMR	proton magnetic resonance
IARC	International Agency for Research on Cancer
LoD	Limit of Detection
ML	maximum level
NOAEL	no observed adverse effect level
NTP	US National Toxicology Program
OSHA	US Occupational Safety & Health Administration
UV-vis	ultraviolet-visible spectrophotometry

Appendix A – Summary of total estimated exposure to potassium polyaspartate (A-5D K/SD) from its use as a stabiliser in wines for the maximum level scenario and the typical level scenario per population group and survey

	Number of subjects	Typical level scenario (200 mg/L)		Maximum level scenario (300 mg/L)	
		Mean	High level	Mean	High level
Infants					
Bulgaria (NUTRICHILD)	658	<0.001	0	<0.001	0
Germany (VELS)	159	<0.001	0	<0.001	0
Denmark (IAT 2006_07)	826	0	0	0	0
Finland (DIPP_2001_2009)	500	0	0	0	0
United Kingdom (DNSIYC_2011)	1366	0.001	0	0.001	0
Italy (INRAN_SCAI_2005_06)	12	<0.001		<0.001	
Toddlers					
Belgium (Regional_Flanders)	36	0.005		0.007	
Bulgaria (NUTRICHILD)	428	0.001	0	0.002	0
Germany (VELS)	348	0.001	0.002	0.001	0.004
Denmark (IAT 2006_07)	917	0	0	0	0
Spain (enKid)	17	0		0	
Finland (DIPP_2001_2009)	500	0	0	0	0
United Kingdom (NDNS-Rolling Programme Years 1–3)	185	0.001	0	0.002	0
United Kingdom (DNSIYC_2011)	1314	0.001	0	0.002	0
Italy (INRAN_SCAI_2005_06)	36	<0.001		<0.001	
Netherlands (VCP_kids)	322	0.001	0	0.001	0
Children					
Austria (ASNS_Children)	128	0.013	0.075	0.020	0.112
Belgium (Regional_Flanders)	625	<0.001	0	0.001	0
Bulgaria (NUTRICHILD)	433	0.001	0	0.001	0
Czech Republic (SISP04)	389	0.002	0	0.002	0
Germany (EsKiMo)	835	0.002	0.007	0.002	0.011
Germany (VELS)	293	0.001	0.004	0.001	0.006
Denmark (DANSDA 2005-08)	298	0.001	0	0.001	0
Spain (enKid)	156	0.001	0	0.001	0
Spain (NUT_INK05)	399	0	0	0	0
Finland (DIPP_2001_2009)	750	<0.001	0	<0.001	0
France (INCA2)	482	0.002	0.014	0.003	0.021
United Kingdom (NDNS-Rolling Programme Years 1–3)	651	0.001	0	0.001	0
Greece (Regional_Crete)	838	<0.001	0	0.001	0
Italy (INRAN_SCAI_2005_06)	193	0.003	0.001	0.005	0.002
Latvia (EFSA_TEST)	187	0	0	0	0
Netherlands (VCP_kids)	957	<0.001	0	<0.001	0
Netherlands (VCPBasis_AVL2007_2010)	447	0.001	0	0.001	0
Sweden (NFA)	1473	<0.001	0	<0.001	0

	Number of subjects	Typical level scenario (200 mg/L)		Maximum level scenario (300 mg/L)	
		Mean	High level	Mean	High level
Adolescents					
Austria (ASNS_Children)	237	0.005	0.033	0.007	0.049
Belgium (Diet_National_2004)	576	0.014	0.075	0.021	0.113
Cyprus (Childhealth)	303	0.001	0	0.001	0
Czech Republic (SISP04)	298	0.005	0	0.008	0
Germany (National_Nutrition_Survey_II)	1011	0.008	0	0.012	0
Germany (EsKiMo)	393	<0.001	0.002	0.001	0.002
Denmark (DANSDA 2005-08)	377	0.008	0.058	0.012	0.087
Spain (AESAN_FIAB)	86	0.012	0.049	0.017	0.074
Spain (enKid)	209	<0.001	0	<0.001	0
Spain (NUT_INK05)	651	0.011	0	0.016	0
Finland (NWSSP07_08)	306	<0.001	0	0.001	0
France (INCA2)	973	0.007	0.024	0.011	0.036
United Kingdom (NDNS-Rolling Programme Years 1–3)	666	0.006	0.008	0.010	0.012
Italy (INRAN_SCAI_2005_06)	247	0.005	0.001	0.007	0.002
Latvia (EFSA_TEST)	453	<0.001	0	<0.001	0
Netherlands (VCPBasis_AVL2007_2010)	1142	0.009	0	0.013	0
Sweden (NFA)	1018	0	0	0	0
Adults					
Austria (ASNS_Adults)	308	0.079	0.439	0.118	0.658
Belgium (Diet_National_2004)	1292	0.197	0.964	0.296	1.446
Czech Republic (SISP04)	1666	0.109	0.749	0.163	1.124
Germany (National_Nutrition_Survey_II)	10419	0.132	0.772	0.198	1.157
Denmark (DANSDA 2005-08)	1739	0.246	0.911	0.370	1.367
Spain (AESAN)	410	0.064	0.403	0.095	0.604
Spain (AESAN_FIAB)	981	0.108	0.578	0.163	0.866
Finland (FINDIET2012)	1295	0.040	0.283	0.060	0.424
France (INCA2)	2276	0.208	0.962	0.312	1.443
United Kingdom (NDNS-Rolling Programme Years 1–3)	1266	0.165	0.798	0.247	1.197
Hungary (National_Repr_Surv)	1074	0.054	0.417	0.081	0.625
Ireland (NANS_2012)	1274	0.139	0.747	0.209	1.121
Italy (INRAN_SCAI_2005_06)	2313	0.196	0.821	0.293	1.231
Latvia (EFSA_TEST)	1271	0.012	0	0.019	0
Netherlands (VCPBasis_AVL2007_2010)	2057	0.122	0.749	0.183	1.124
Romania (Dieta_Pilot_Adults)	1254	0.050	0.306	0.075	0.459
Sweden (Riksmaten 2010)	1430	0.140	0.625	0.210	0.938
The elderly					
Austria (ASNS_Adults)	92	0.150	0.625	0.225	0.938
Belgium (Diet_National_2004)	1215	0.190	0.913	0.285	1.370
Germany (National_Nutrition_Survey_II)	2496	0.142	0.781	0.212	1.172
Denmark (DANSDA 2005-08)	286	0.370	1.176	0.555	1.765

	Number of subjects	Typical level scenario (200 mg/L)		Maximum level scenario (300 mg/L)	
		Mean	High level	Mean	High level
Finland (FINDIET2012)	413	0.036	0.342	0.054	0.514
France (INCA2)	348	0.299	1.049	0.448	1.573
United Kingdom (NDNS-Rolling Programme Years 1–3)	305	0.117	0.705	0.175	1.057
Hungary (National_Repr_Surv)	286	0.070	0.519	0.105	0.779
Ireland (NANS_2012)	226	0.093	0.539	0.140	0.809
Italy (INRAN_SCAI_2005_06)	518	0.272	1.009	0.408	1.514
Netherlands (VCPBasis_AVL2007_2010)	173	0.211	0.929	0.316	1.394
Netherlands (VCP-Elderly)	739	0.164	0.716	0.247	1.074
Romania (Dieta_Pilot_Adults)	128	0.048	0.263	0.072	0.395
Sweden (Riksmaten 2010)	367	0.189	0.700	0.284	1.050