Report of the Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) on a request for initial assessment for marketing of synthetic hydroxytyrosol under Regulation (EC) No 258/97 concerning novel foods and novel food ingredients

Section of Food Safety and Nutrition

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Abstract

The company Seprox Biotech S.L. has applied for authorisation to market synthetic hydroxytyrosol in the European Union, for addition, as an ingredient, to oils, fats and fruit and vegetable juices. This ingredient has no history of use in the European Union prior to 1997 and therefore comes within the scope of Regulation (EC) No 258/97 concerning novel foods and novel food ingredients.

The Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) is of the view that, on the basis of the information provided, there is no indication that the consumption of synthetic hydroxytyrosol under the conditions proposed by the applicant (involving its addition, as an ingredient, to oils and fats to be consumed uncooked and to fruit and vegetable juices) can have adverse effects on health. The Committee therefore concludes that the novel food presented by Seprox Biotech S.L. for assessment fulfils the criteria for acceptance laid down in Regulation (EC) No 258/97 concerning novel foods and novel food ingredients.

Key words

Hydroxytyrosol, novel food, novel food ingredients.

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1. Assessment of the novel food

Introduction

The company Seprox Biotech S.L. has applied for authorisation to market synthetic hydroxytyrosol in the European Union, for addition, as an ingredient, to fats and oils to be consumed uncooked and to fruit and vegetable juices.

According to the applicant, synthetic hydroxytyrosol has no history of use in the European Union prior to 1997 and therefore comes within the scope of Regulation (EC) No 258/97 concerning novel foods and novel food ingredients (EU, 1997a).

The applicant declares that the novel food belongs to Class 1 'Pure chemicals or simple mixtures from non-GM sources', Subclass 1 'the source of the novel food has a history of food use in the Community' pursuant to Commission Recommendation 97/618/EC of 29 July 1997. The report presented by the applicant therefore follows the guidelines laid down for category 1.1, covering sections I, II, III, IX, X, XI, XII and XIII set out in Table II of the said Recommendation (EU, 1997b).

The proposed novel ingredient has a high purity of above 99 % and is produced by chemical synthesis and physical processes, from which hydroxytyrosol is obtained in the form of a viscous, yellowish liquid.

Comments

The Scientific Committee does not agree with the applicant's categorisation of the product. It takes the view that as a new product obtained by synthesis, it should be categorised under Class 1, Subclass 2 (the source of the novel food has no history of food use in the Community). However, this does not significantly alter the documentation to be provided, the only difference being that information from previous human exposure to the novel food or its source therefore need not be provided. The ability to assess the documentation submitted is thus unaffected.

I. Specification of the novel food

Seprox Biotech's synthetic hydroxytyrosol (3,4-dihydroxyphenylethanol) (CAS No 10597-60-1) is a viscous, yellowish liquid (moisture content 4 %). The applicant states that the solvents used in the synthesis of the hydroxytyrosol are authorised for food use in the European Union or in the United States of America or are at low and safe residual levels.

The specifications determined by the applicant are set out in Table 1.

Table 1. Specifications of synthetic hydroxytyrosol

Parameter	**Criterion**	**Method of testing**
Description	Viscous yellowish liquid	Visual
Odour	Characteristic	Organoleptic
Taste	Slightly bitter	Organoleptic
Water solubility	Miscible with water	Saturation
Moisture	< 4 %	Thermogravimetry
pH	3.5-4.5	1 M aqueous solution
Hydroxytyrosol content	> 95.0 %	Calculated as 100 % - other components
Chromatographic purity	> 99.0 %	280 nm HPLC
Hydroxytyrosol acetate	< 0.3 %	HPLC
Other	< 0.1 %	HPLC
Heavy Metals		
Lead	< 0.03 ppm	ICP-MS
Cadmium	< 0.01 ppm	ICP-MS
Mercury	< 0.01 ppm	ICP-MS
Solvent residues		
Ethyl acetate	< 25.0 ppm	Headspace GC/MS
Isopropanol	< 2.50 ppm	Headspace GC/MS
Methanol	< 0.01 ppm	Headspace GC/MS
Tetrahydrofuran	< 0.01 ppm	Headspace GC/MS

In order to demonstrate compliance with the specifications, results from the analysis of six batches manufactured non-consecutively during the course of 2012 are provided in the dossier.

In addition to the parameters stipulated in the specifications, the test reports provide information on boron (five batches), sodium, potassium, calcium, chromium, nickel, iron, nitrates, nitrites, chlorides, sulphates and acetone content.

The analyses of hydroxytyrosol, hydroxytyrosol acetate and moisture content were performed by Seprox Biotech. All the other analyses were carried out by an external laboratory (Laboratorio Químico-Microbiológico S.A.).

The method for the determination of hydroxytyrosol by HPLC/UV was validated by Harlan Laboratories S.A. This laboratory is accredited by the Spanish National Accreditation Body (Entidad Nacional de Acreditación, ENAC) in accordance with the standard UNE-EN ISO/IEC 17025 for the performance of veterinary clinical analysis (accreditation number 80/LE179) and in accordance with Good Laboratory Practice for the performance of phytosanitary product studies in the area of laboratory toxicity testing (26/BPL030) and studies on chemical substances in the areas of laboratory toxicity and toxin kinetics testing (26/BPL037). After validation by Harlan Laboratories S.A., the applicant carried out a study of several parameters connected with the

determination of hydroxytyrosol (linear range, detection and quantification limits, reproducibility and stability).

Laboratorio Químico-Microbiológico S.A. for its part, is accredited by ENAC for the performance of various physico-chemical tests on agri-food products in accordance with the standard UNE-EN ISO/IEC 17025. The analyses carried out by this laboratory do not fall within its accreditation, so information has been provided about the precision and uncertainty of the methods used.

Analysis for inorganic compounds

The results of the analysis of six batches of hydroxytyrosol for various inorganic compounds are set out in Table 2.

Table 2. Results of analysis for various inorganic compounds in six batches of synthetic hydroxytyrosol

Inorganic compounds (mg/kg)	Mean (mg/kg) ± standard deviation	Inorganic compounds (mg/kg)	Mean (mg/kg) ± standard deviation
Mercury	< 0.01	Sodium	433.13 ± 57.59
Lead	0.03 ± 0.01	Potassium	431.72 ± 295.92
Nickel	0.04 ± 0.03	Calcium	119.45 ± 61.04
Cadmium	< 0.01	Chlorides	508.13 ± 310.64
Chromium	0.08 ± 0.06	Sulphates	219.35 ± 117.99
Iron	2.05 ± 1.38	Nitrites	2.05 ± 3.70
Boron (5 batches)	7.44 ± 3.39	Nitrates	10.7 ± 10.8

Analysis for organic compounds

The applicant provides data from analyses carried out using HPLC-MS/MS on four batches for the following four organic impurities: 3-methoxy-4-hydroxyphenylglycol, homovanillyl alcohol, isohomovanillyl alcohol and hydroxytyrosol acetate. The sum of all of these is stated to be below 0.1 %.

Analysis for solvent residues

The applicant provides data from six batches of hydroxytyrosol on the level of residues of the five solvents potentially present (Table 3).

Table 3. Results of analysis for different solvent residues in six batches of synthetic hydroxytyrosol	
Solvents (mg/kg)	**Mean (mg/kg) ± standard deviation**
Ethyl acetate	9.63 ± 8.84
Acetone	1.85[1]
Isopropyl alcohol	1.17 ± 0.74
Methanol	1.93[1]
Tetrahydrofuran	< 0.01

[1]Only one positive reading.

Comments

In the Scientific Committee's view, the novel food to be marketed is properly identified by the nuclear magnetic resonance (NMR), chromatographic and composition analysis studies submitted and the information on the presence of other organic and inorganic compounds is sufficient.

II. Effects of the production process applied to the novel food

Description of the production process

The applicant produces hydroxytyrosol by chemical and enzymatic synthesis. The details provided in the dossier are considered by the applicant to be confidential.

Production control

The applicant declares that quality controls have been put in place to optimise the production process and verify that the solvents and reagents used comply with the corresponding food law.

The solvents used are eliminated by evaporation in water, which is the solvent with the highest boiling point out of those used.

Residues of organic and inorganic salts are eliminated by extractions with solvents after the reactions in which these salts are involved.

Stability of the finished product

Seprox Biotech recommends storing the novel ingredient in the dark, at 4 °C, and protected from oxidising atmospheres.

A study is provided on the stability of five batches of synthetic hydroxytyrosol kept in these conditions for 20 or 23 months and the stability of one batch kept for 8 months at ambient temperature in three different sets of conditions: darkness and protective atmosphere, light and protective atmosphere, and light and open vial.

In the open vial only, some change in colour was observed at 4 months, as a result of oxidation. Moisture content also increased, owing to the hygroscopic nature of the product, and purity was slightly reduced.

In all cases, the percentage of hydroxytyrosol was above 99 % purity. The applicant has not established an expiry date for the synthetic product.

Stability of the product when added to foodstuffs

A study is provided on the product's stability in grape and pineapple, apple, orange, tomato, and mixed vegetable juices and in a grape and pineapple juice containing 400 mg/kg of vitamin C and added iron (40 mg/kg of $Fe_2(SO_4)_3$). A study is also provided on sunflower, soybean, refined olive and virgin olive oils.

In the case of the juices, the studies were conducted at 40 °C and 90 °C, with the exception of the juice with added iron where temperatures of 40 °C and 60 °C were used, and three different concentrations of hydroxytyrosol (15, 30 and 50 mg/kg) were studied. Due to its hydrophilic nature, hydroxytyrosol is easily dissolved in all the aqueous matrices (juices) studied. The length of the studies varied from 3-4 hours up to 11 weeks, depending on the type of juice, the temperature applied and the concentration of hydroxytyrosol added. The studies at 60 °C and 90 °C were carried out in open vials.

All juice and oil samples were analysed by HPLC using a standard curve, in each case in an aqueous solution prepared for the particular matrix. To ensure the validity of the extrapolation, a control analysis was carried out using a matrix to which three different concentrations of hydroxytyrosol had been added: low (50 % of the chosen concentration), medium (same as chosen concentration) and high (150 % of the chosen concentration).

In the case of the oils, the studies were conducted at 40 °C, 90 °C and 180 °C, for three different concentrations of hydroxytyrosol (100, 300 and 500 mg/kg). The applicant indicates that the solubility of hydroxytyrosol in these matrices is limited to 2.5 g/kg. The length of the studies varied from 1 hour up to 12 months, depending on the type of oil, the temperature applied and the concentration of hydroxytyrosol added.

Two previously-published HPLC methods were used to quantify the hydroxytyrosol in the oils. In the first month, one method of analysis (Carrasco-Pancorbo et al., 2007) was used; from the fifth month, another method (Romero and Brenes, 2012) was used, in order to achieve better extraction of hydroxytyrosol.

According to the dossier submitted, none of the results obtained for any of the juices showed significant loss of hydroxytyrosol in the conditions studied: 40 °C (8 weeks) and 90 °C (3-4 hours).

The results obtained for oils subjected to 40 °C show that the percentage of the hydroxytyrosol originally added that is still present in the oils after 12 months is approximately 85 % in sunflower oil, 70 % in olive oil and less than 40 % in soybean oil.

At 90 °C, losses of at least 62 % of the hydroxytyrosol originally added (500 mg/kg) were observed when three samples of extra virgin olive oil were subjected to this temperature for 4 days.

For oils subjected to 180 °C for 60 minutes, losses of hydroxytyrosol of more than 93 % of the amount measured at 120 °C (5 minutes before reaching 180 °C) were observed for refined olive oil, extra virgin olive oil and soybean oil; losses at 20 minutes exceeded 50 %.

For sunflower oil under the same conditions, losses of 54 % and 26 % were observed at 60 minutes and 20 minutes respectively.

The applicant therefore considers that oils enriched with hydroxytyrosol are suitable for consumption in uncooked form, but does not recommend their use at high temperatures (normal

cooking temperatures), due to the loss of hydroxytyrosol that occurs and the deterioration of the organoleptic properties of the oils.

Comments

In the view of the Committee, the production process has been adequately described. The enzyme used is authorised in another EU Member State.

Given the lack of stability of the novel ingredient when added to oils and subjected to various heat treatments, consumers should be advised that oils and fats containing it are only to be consumed uncooked, as indicated by the applicant.

The applicant must provide users with information on its stability in the foods to which it is intended to be added.

III. History of the organism used as the source of the food

The applicant indicates that it produces hydroxytyrosol using a well-established chemical synthesis process and that there is no history of use of the novel food in the form of synthetic hydroxytyrosol, despite natural hydroxytyrosol being present in virgin olive oil and olives, among other dietary sources.

Comments

The Scientific Committee considers the novel ingredient to be produced by a process of chemical synthesis and not to have a proven history of consumption as a food in the European Union prior to 1997.

IX. Anticipated intake/extent of use of the novel food

The applicant is seeking authorisation for the use of hydroxytyrosol in concentrations of 5 mg per portion (FSA, 2008) of the proposed foods, i.e. 5 mg/10 ml (0.5 mg/ml) of fat or oil for consumption uncooked and 5 mg/200 ml (0.025 mg/ml) of fruit or vegetable juice. Foods for breastfeeding women and children under 36 months of age are expressly excluded.

With the help of the food consumption database of the European Food Safety Authority (EFSA, 2011a), estimated intake has been calculated for children, teenagers, adults and old people in various countries (Belgium, Denmark, France, Germany, Italy, Spain, Czech Republic and Sweden). The greatest intake of hydroxytyrosol by adult consumers in the extreme case of the aggregate of 95th percentiles would occur in Germany (64.8 mg/day); for children, it would occur in the Czech Republic (40.5 mg/day).

Comments

Figures for average and maximum intakes of hydroxytyrosol have been obtained using EFSA data on habitual food consumption only for consumers. Data on intake of fats and oils and of vegetable and fruit juices was used. The worst case scenario, i.e. the maximum theoretical consumption, would be calculated as the sum of the intake from oils and fats and the intake from fruit and

vegetable juices, assuming 95th percentile consumption of these foods. Portions are taken to be 10 ml in the case of fats and oils and 200 ml for vegetable and fruit juices. On the basis of this data and of the proposed amount to be included in each portion (5 mg), average and maximum intake are calculated.

The Scientific Committee considers the estimates of intake made by the applicant to be appropriate.

X. Information from previous human exposure to the novel food or its source

Seprox Biotech's hydroxytyrosol is obtained by chemical synthesis and, as such, has not previously been available commercially.

The applicant points out that naturally-occurring hydroxytyrosol is found in many foods and plants, is used as a food supplement, and has been the subject of several studies on humans. In the dossier, the applicant indicates an estimated intake of hydroxytyrosol and other phenolic compounds occurring naturally in olives of 20-40 mg/day. This estimate is based on an average consumption of 20 medium-sized olives a day and a hydroxytyrosol content of between 1 mg and 2 mg per olive. This intake would not only be of hydroxytyrosol but also of oleuropein and other polyphenols in olives, and consumption is expected to be higher than this in Mediterranean countries where both olive oil and olives are commonly consumed.

Various studies are cited on the effectiveness in humans of the polyphenols present in olive oil, and on the hydroxytyrosol and other polyphenol content of olives.

The nuclear magnetic resonance (NMR) study provided shows that the compound obtained by synthesis has the same structure as naturally-occurring hydroxytyrosol.

Comments

The Scientific Committee considers that the novel ingredient is produced by a process of chemical synthesis and that there is no previous human exposure, although we can say from the aforementioned NMR study that the results for synthetic hydroxytyrosol could be extrapolated from those for naturally-occurring hydroxytyrosol in relation to human exposure. However, we must point out that exposure to naturally-occurring hydroxytyrosol never occurs in isolated form and/or in the chemical form obtained by synthesis, as it is always accompanied, both in virgin olive oil and in olives, by other polyphenols (for example, tyrosol), and is present in different chemical species (for example, oleuropein).

XI. Nutritional information on the novel food

The applicant states that the aim is not to replace olive oil, olives or other sources of hydroxytyrosol with the novel ingredient and that the proposed intake would not be nutritionally disadvantageous to the consumer.

Various studies are cited regarding the effectiveness of hydroxytyrosol and other polyphenols in olive oil, including health claims endorsed by the EFSA for the polyphenols in olive oil (EFSA,

2011b). Further studies –both on animals and humans– are cited concerning the absorption, distribution, metabolism and excretion of phenolic compounds including hydroxytyrosol.

Seproch Biotech provides a study of the pharmacokinetics of 3,4-dihydroxyphenylacetic acid (DOPAC), hydroxytyrosol and hydroxytyrosol acetate. The study was conducted by Vivotecnia Research S.L. in accordance with the principles of Good Laboratory Practice laid down in Directive 2004/10/EC (EU, 2004) and Spanish law (Royal Decree No 1369/2000) (BOE, 2000).

The experiment involved the administration by gavage of single oral doses of 1 mg and 5 mg of hydroxytyrosol per kg to Sprague Dawley rats of both sexes. At both concentrations, significant differences were observed in the maximum concentration (C_{max}) of each analyte measured in the plasma following administration of a single dose, with DOPAC exhibiting the highest C_{max}, followed by hydroxytyrosol acetate.

The three compounds showed rapid absorption (T_{max}: 0.5-1 hour), with the exception of DOPAC, which had a T_{max} of 2 hours for males treated with 1 and 5 mg/kg and females treated with 5 mg/kg.

Concerning bioavailability, DOPAC was the analyte with the highest $AUC_{(0-24)}$ after each test, with the highest values being observed in the case of females treated with 5 mg/kg of hydroxytyrosol acetate.

The parameters calculated for hydroxytyrosol (C_{max}, T_{max} and AUC) correspond to a limited number of samples obtained with quantifiable concentrations.

The results obtained suggest that both hydroxytyrosol and hydroxytyrosol acetate are quickly converted to DOPAC after oral administration.

Comments

The health claim approved in Regulation (EU) No 432/2012 (EU, 2012) concerning polyphenols in olive oil does not apply to synthetic hydroxytyrosol. The addition of the novel ingredient to fats and oils does not make them equivalent to olive oil in terms of their nutritional or health properties. If any such claim is to be made, it must first be authorised in accordance with the procedure laid down in Regulation (EU) No 1924/2006 (EU, 2006).

Taking into account the above, the Scientific Committee is of the view that the addition of this novel ingredient to fats, oils and fruit and vegetable juices under the conditions proposed by the applicant does not pose any nutritional disadvantage.

XII. Microbiological information on the novel food

The applicant considers the risk of bacterial proliferation to be very low, considering both the nature and purity of the hydroxytyrosol and the process by which it is produced. Attention is also drawn to the antimicrobial properties of hydroxytyrosol. The applicant has provided information on counts of all aerobic microbes combined, *E. coli*, *Enterobacteriaceae*, and moulds and yeasts, with a negative result being returned in each case.

Comments

The Scientific Committee considers the information provided to be adequate.

XIII. Toxicological information on the novel food

1. Toxicity and genotoxicity studies on hydroxytyrosol

Seproch Biotech has provided studies on sub-chronic toxicity and mutagenicity (Ames test and *in vitro* human lymphocyte chromosomal aberration test) for the synthetic hydroxytyrosol under assessment.

The sub-chronic toxicity study (OECD No 408) was performed by Harlan Laboratories S.A. in accordance with the principles of Good Laboratory Practice and the requirements and recommendations set out in various European guides and directives. The study has been published (Auñon-Calles et al., 2013a).

Test conditions were as follows: doses of 0 (control), 5, 50 and 500 mg of hydroxytyrosol per kg of body weight per day were administered orally by gavage to Wistar Hannover rats of both sexes for a period of 13 weeks, with a 4-week recovery period.

After 91-92 days of treatment, four groups of 10 animals of each sex were killed in order to carry out toxicity tests. Two additional groups of five animals of each sex were given a 4-week recovery period in order to assess the progression or reversibility of certain changes observed in the tests.

In the study, samples from each group were analysed during the first and eighth weeks of treatment in order to determine hydroxytyrosol content, and results of ophthalmoscopic examinations, clinical signs, food consumption and body weight were periodically recorded during the acclimatisation, treatment and recovery periods.

Functional observation batteries and measurements of locomotor activity and grip strength were also carried out during the thirteenth week of treatment and at the end of the recovery period, and blood samples were taken for haematological, biochemical and urinary analysis.

The applicant states that no signs of adverse effects or mortality were observed during the study. The salivation observed in several of the animals treated is attributed to the bitter taste of hydroxytyrosol and/or the physical characteristics of the formulation (slightly oily and thick).

Small changes in body weight were observed in male and female rats dosed with 500 mg/kg body weight per day. It is stated that all significant changes in haematology and chemical parameters are considered to be chance changes/biological variations and not adverse effects of the treatment as these changes were not observed in both sexes, correlative changes in other clinical parameters were not observed, they are of small magnitude, they were not observed at all doses, or they were not associated with microscopic changes in the related organs.

The changes observed in the weight of some organs after the recovery period are considered to be due to chance because they do not depend on the dose and because of the absence of correlated histopathological and chemical changes.

On the basis of the results obtained, the dose of 500 mg/kg body weight per day has been set as the No Observed Adverse Effect Level (NOAEL).

The Ames test (OECD No 471) was carried out by Vivotecnia Research S.L. in accordance with the principles of Good Laboratory Practice. This study has been published (Auñon-Calles et al., 2013b).

The test was conducted using strains TA98, TA100, TA1535 and TA1537 of *Salmonella typhimurium* and strain WP2 (pKM101) of *Escherichia coli*, with and without metabolic activation, using five concentrations in a range of 0.06 to 5 µl/plate. In order to set test doses, a preliminary cytotoxicity study was carried out on one of the strains, with negative results at a concentration of 50.0 µl/ml.

The result was negative in all the strains and at all the concentrations used, indicating that the synthetic hydroxytyrosol being studied does not induce mutations and can be considered to be non-mutagenic on the basis of the tests.

The *in vitro* human lymphocyte chromosomal aberration test (OECD No 473) was carried out by Harlan Laboratories S.A. in accordance with the principles of Good Laboratory Practice. This study has been published (Auñon-Calles et al., 2013b).

The test was performed with and without metabolic activation, using ten concentrations in a range of 10.0 to 1 542 µl/ml. In order to set test doses, a preliminary cytotoxicity study was carried out.

Clastogenic effects were only observed at concentrations of 10 mM, which are very unlikely to be reached following oral ingestion.

Various studies on toxicity, genotoxicity, reproductive toxicity and teratogenicity, carried out mainly with olive pulp extracts, are also cited.

The information available on the toxicity of synthetic hydroxytyrosol is limited, as only two papers have been published on the subject (Auñón et al., 2013a,b). However, the NMR study provided by the applicant supports the identity of the naturally-occurring and synthetic molecules. A sub-chronic NOAEL of 500 mg/kg body weight per day has been suggested for synthetic hydroxytyrosol (Auñón et al., 2013a), although this is actually just the highest dose used in the tests, so it could be even higher. At these levels, no relevant toxic effects have been found. The test was performed on rats and is, as indicated, the only existing test. It would be desirable to have information from other species which could be more sensitive.

In vitro tests (Auñón et al., 2013b) have shown that hydroxytyrosol is neither genotoxic nor mutagenic at much higher concentrations than are expected following its ingestion. Clastogenic effects have been observed at concentrations (10 mM) very difficult to reach by oral ingestion. The possibility of adverse effects from prolonged exposure to hydroxytyrosol and/or its metabolites is not ruled out (Auñón et al., 2013b), although the information available (Auñón et al., 2013a) apparently does not point to this.

There is, however, a lot of information on the possible adverse effects of aqueous extracts of olive pulp. This information is brought together in detail in the GRAS (*Generally Recognized as Safe*) recognition by the American Food and Drug Administration (FDA, 2013). An acute NOAEL of 2 000 mg/kg body weight per day in mice and 1 000 mg/kg body weight per day in rats has been determined, with the value of 2 000 mg/kg body weight per day being considered more appropriate. In rats, a sub-chronic NOAEL of 2 000 mg/kg body weight per day has been found (equivalent to 72 mg hydroxytyrosol/kg body weight per day) and at these doses, no adverse reproductive effects nor any mutagenic effects have been observed. All that was observed were

slight changes (possible toxicity) at doses of 5 000 mg/kg body weight per day (equivalent to a dose of 350 g in a person weighing 70 kg), which are not considered to be of interest in any way. Furthermore, as is set out in the said document, there is a clear 'history of safe use' of olive oil and olives (with their high hydroxytyrosol content).

The above information led the FDA to grant GRAS status to aqueous extracts of olive pulp, some of which are marketed as so-called neutraceuticals.

The estimated intake of hydroxytyrosol as a result of its natural occurrence in table olives is 20-40 mg/day (similar to the intake that would result from the use of hydroxytyrosol as an ingredient in novel foods). In fact, the use of hydroxytyrosol anticipated by Seprox Biotech is 5 mg/portion in the chosen foods (fats and oils; vegetable/fruit juices). This implies a daily intake of 0.30 mg/kg body weight and 0.68 mg/kg body weight at average and 95th percentile consumption respectively, which translate into 18.1 mg/day and 40.6 mg/day for a person weighing 60 kg.

Although the information available on synthetic hydroxytyrosol is scarce, the said information confirms corresponding results obtained previously for olive pulp extracts, based on which there are considered to be no adverse effects. Experience of the use of olive oil and olives (which contain hydroxytyrosol as one of their polyphenolic components in quantities similar to, or greater than, those anticipated in the use of hydroxytyrosol as a 'novel food') likewise suggests that synthetic hydroxytyrosol is safe.

Comments

In light of all the information available, the Committee considers that there is no data to suggest possible adverse effects if hydroxytyrosol is used under the conditions proposed by the applicant. The information provided by Seprox Biotech on the safety of hydroxytyrosol is considered to be adequate from a toxicological viewpoint.

2. Toxicity of residues present in hydroxytyrosol

The possible toxicity of residues detected in synthetic hydroxytyrosol has been assessed using the data in tables 2 and 3. This was done taking into account the maximum daily hydroxytyrosol intake of 64.8 mg/day by adult consumers in an extreme case (sum of 95th percentiles), and 40.5 mg/day by children (figures provided by applicant). The amount of each residue contained in these daily intakes was calculated using the average value or the limit of detection (LOD) (Tables 2 and 3).

Calculations were performed using a weight of 60 kg for an adult and 18.3 kg for children of 0 to 9 years of age, in accordance with weight data from the World Health Organisation, which considers the median weight for children of 5 years of age to be 18.3 kg.

The daily intake of each residue considered was calculated taking into account the maximum daily intake of hydroxytyrosol (64.8 mg/day and 40.5 mg/day for adults and children respectively).

Comments

Based on the hydroxytyrosol intake figures, the estimated daily intake both for adults and for children represents a minimal percentage of acceptable daily intake (or equivalent). From a

toxicological viewpoint, the presence of residues in the concentrations considered, in principle, would therefore be of no significance.

3. Allergenicity studies

The applicant states that allergic reactions are not expected on the basis of the structure and nature of synthetic hydroxytyrosol. Information has been provided from the analysis of proteins in four batches of the product, with values of between 0.35 and 0.45 mg/ml being observed.

Comments

In the Scientific Committee's view, the only potential source of protein in the novel food is the enzyme used in the process, and since this enzyme is authorised for food use in Denmark and no references have been found to allergic reactions of food origin to lipases (EFSA, 2014a, 2014b, 2014c), it is unlikely that the novel food poses a risk of allergy.

Conclusions of the Scientific Committee

The Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) is of the view that, on the basis of the information provided, there is no indication that the consumption of synthetic hydroxytyrosol under the conditions proposed by the applicant (involving its addition, as an ingredient, to oils and fats to be consumed uncooked and to fruit and vegetable juices) can have adverse effects on health.

The Scientific Committee concludes that the novel food presented by Seprox Biotech S.L. for assessment fulfils the criteria for acceptance laid down in Regulation (EC) No 258/97 concerning novel foods and novel food ingredients.

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