

EFSA Panel on Food Contact Material, Enzymes, Flavourings and Processing Aids (CEF); Scientific Opinion on Flavouring Group Evaluation 73, Revision 1: Consideration of alicyclic primary alcohols, aldehydes, acids and related esters evaluated by JECFA (59th meeting) structurally related to primary saturated or unsaturated alicyclic alcohol, aldehyde, and esters evaluated by EFSA in FGE.12Rev2 (2011)

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SCIENTIFIC OPINION

Scientific Opinion on Flavouring Group Evaluation 73, Revision 1 (FGE.73Rev1):

Consideration of alicyclic primary alcohols, aldehydes, acids and related esters evaluated by JECFA (59th meeting) structurally related to primary saturated or unsaturated alicyclic alcohol, aldehyde, and esters evaluated by EFSA in FGE.12Rev2 (2011)¹

EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (CEF)^{2,3}

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ABSTRACT

The Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids of the European Food Safety Authority was requested to consider evaluations of flavouring substances assessed since 2000 by the Joint FAO/WHO Expert Committee on Food Additives (the JECFA), and to decide whether further evaluation is necessary, as laid down in Commission Regulation (EC) No 1565/2000. The present consideration concerns a group of 16 alicyclic primary alcohols, aldehydes, acids and related esters evaluated by the JECFA at the 59th meeting in 2002. The revision is made due to consideration of one additional substance compared to the previous version. The substances were evaluated through a stepwise approach that integrates information on structure-activity relationships, intake from current uses, toxicological threshold of concern, and available data on metabolism and toxicity. The Panel agrees with the application of the Procedure as performed by the JECFA for all 16

1 On request from the European Commission, Question No EFSA-Q-2010-01249, adopted on 22 March 2012.

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substances [FL-no: 02.114, 02.141, 05.098, 05.104, 05.112, 05.119, 05.123, 08.034, 08.060, 08.067, 09.028, 09.289, 09.488, 09.534, 09.536 and 09.615], considered in this FGE and agrees with the JECFA conclusion, “No safety concern at estimated levels of intake as flavouring substances” based on the MSDI approach. Besides the safety assessment of these flavouring substances, the specifications for the materials of commerce have also been considered and for all 16 substances, the information is adequate.

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KEY WORDS

Alicyclic, primary, alcohols, aldehyde, esters, JECFA, 59th meeting, FGE.12, FGE.73.

SUMMARY

The Scientific Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (the Panel) was asked to give scientific advice to the Commission on the implications for human health of chemically defined flavouring substances used in or on foodstuffs in the Member States. In particular, the Panel was requested to consider the Joint FAO/WHO Expert Committee on Food Additives (the JECFA) evaluations of flavouring substances assessed since 2000, and to decide whether no further evaluation is necessary, as laid down in Commission Regulation (EC) No 1565/2000. These flavouring substances are listed in the Register, which was adopted by Commission Decision 1999/217/EC and its consecutive amendments.

The revision is made due to consideration of one additional substance, 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104], compared to the previous version of FGE.73. This substance has been evaluated in FGE.209 due to structural concern for genotoxicity, and has been cleared from this concern and thus may be evaluated through the Procedure.

The present consideration therefore concerns 16 alicyclic primary alcohols, aldehydes, acids and related esters evaluated by the JECFA (59th meeting) and will be considered in relation to the European Food Safety Authority (EFSA) evaluation of nine primary saturated or unsaturated alicyclic alcohol, aldehyde and esters evaluated in the Flavouring Group Evaluation 12, Revision 2 (FGE.12Rev2).

A further 10 substances were evaluated by the JECFA in this group, one substance is not in the Register [mixture of 2-methyl-5-(2,3-dimethyltricyclo[2.2.1.0(2.6)]hept-3-yl)pent-2-en-1-ol and 2-methyl-5-(2-methyl-3-methylenebicyclo[2.2.1]hept-2-yl)pent-2-en-1-ol] (JECFA no: 984) and nine substances are alpha,beta-unsaturated aldehydes or precursors for such [FL-no: 02.060, 02.091, 05.106, 05.117, 05.121, 09.034, 09.272, 09.278 and 09.302]. The genotoxicity properties of these nine substances were considered together with other alpha,beta-unsaturated aldehydes and ketones in FGE.208 for which it was concluded that additional data were required.

The Panel agrees with the application of the Procedure as performed by the JECFA for the 16 substances considered in this FGE.

For all 16 substances evaluated through the Procedure use levels are needed to calculate the modified Theoretical Added Maximum Daily Intake (mTAMDI) in order to identify those flavouring substances that need more refined exposure assessment and to finalise the evaluation.

In order to determine whether the conclusion for the 16 JECFA evaluated substances can be applied to the materials of commerce, it is necessary to consider the available specifications. Adequate specifications including complete purity criteria and identity tests are available for all 16 JECFA evaluated substances.

Thus, for all 16 substances [FL-no: 02.114, 02.141, 05.098, 05.104, 05.112, 05.119, 05.123, 08.034, 08.060, 08.067, 09.028, 09.289, 09.488, 09.534, 09.536 and 09.615] the Panel agrees with the JECFA conclusion “No safety concern at estimated levels of intake as flavouring substances” based on the MSDI approach.

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BACKGROUND

Regulation (EC) No 2232/96 of the European Parliament and the Council (EC, 1996a) lays down a Procedure for the establishment of a list of flavouring substances, the use of which will be authorised to the exclusion of all other substances in the EU. In application of that Regulation, a Register of flavouring substances used in or on foodstuffs in the Member States was adopted by Commission Decision 1999/217/EC (EC, 1999a), as last amended by Commission Decision 2009/163/EC (EC, 2009a). Each flavouring substance is attributed a FLAVIS-number (FL-number) and all substances are divided into 34 chemical groups. Substances within a group should have some metabolic and biological behaviour in common.

Substances which are listed in the Register are to be evaluated according to the evaluation programme laid down in Commission Regulation (EC) No 1565/2000 (EC, 2000a), which is broadly based on the Opinion of the Scientific Committee on Food (SCF, 1999a).

Commission Regulation (EC) No 1565/2000 lays down that substances that are contained in the Register and will be classified in the future by the Joint FAO/WHO Expert Committee on Food Additives (the JECFA) so as to present no safety concern at current levels of intake will be considered by the European Food Safety Authority (EFSA), who may then decide that no further evaluation is necessary.

In the period 2000 – 2008, during its 55th, 57th, 59th, 61st, 63rd, 65th, 68th and 69th meetings, the JECFA evaluated about 1000 substances, which are in the EU Register.

TERMS OF REFERENCE

The European Food Safety Authority (EFSA) is requested to consider the Joint FAO/WHO Expert Committee on Food Additives (the JECFA) evaluations of flavouring substances assessed since 2000, and to decide whether no further evaluation is necessary, as laid down in Commission Regulation (EC) No 1565/2000 (EC, 2000a). These flavouring substances are listed in the Register, which was adopted by Commission Decision 1999/217/EC (EC, 1999a) and its consecutive amendments.

The evaluation programme was finalised at the end of 2009.

After the finalisation of the evaluation programme, in their letter of the 7th September 2010, the Commission requested EFSA, based on additional submitted data on genotoxicity, to carry out re-evaluation of the flavouring substance 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] and depending on the outcome to proceed to the evaluation of this flavouring substance through the Procedure, also according to Commission Regulation (EC) No 1565/2000 (EC, 2000a).

ASSESSMENT

The approach used by EFSA for safety evaluation of flavouring substances is referred to in Commission Regulation (EC) No 1565/2000 (EC, 2000a), hereafter named the “EFSA Procedure”. This Procedure is based on the Opinion of the Scientific Committee on Food (SCF, 1999a), which has been derived from the evaluation procedure developed by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1995; JECFA, 1996a; JECFA, 1997a; JECFA, 1999b), hereafter named the “JECFA Procedure”. The Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (the Panel) compares the JECFA evaluation of structurally related substances with the result of a corresponding EFSA evaluation, focussing on specifications, intake estimations and toxicity data, especially genotoxicity data. The evaluations by EFSA will conclude whether the flavouring substances are of no safety concern at their estimated levels of intake, whether additional data are required or whether certain substances should not be put through the EFSA Procedure.

The following issues are of special importance.

Intake

In its evaluation, the Panel as a default uses the “Maximised Survey-derived Daily Intake” (MSDI) approach to estimate the *per capita* intakes of the flavouring substances in Europe.

In its evaluation, the JECFA includes intake estimates based on the MSDI approach derived from both European and USA production figures. The highest of the two MSDI figures is used in the evaluation by the JECFA. It is noted that in several cases, only the MSDI figures from the USA were available, meaning that certain flavouring substances have been evaluated by the JECFA only on the basis of these figures. For Register substances for which this is the case the Panel will need EU production figures in order to finalise the evaluation.

When the Panel examined the information provided by the European Flavour Industry on the use levels in various foods, it appeared obvious that the MSDI approach in a number of cases would grossly underestimate the intake by regular consumers of products flavoured at the use level reported by the Industry, especially in those cases where the annual production values were reported to be small. In consequence, the Panel had reservations about the data on use and use levels provided and the intake estimates obtained by the MSDI approach. It is noted that the JECFA, at its 65th meeting considered “how to improve the identification and assessment of flavouring agents, for which the MSDI estimates may be substantially lower than the dietary exposures that would be estimated from the anticipated average use levels in foods” (JECFA, 2006c).

In the absence of more accurate information that would enable the Panel to make a more realistic estimate of the intakes of the flavouring substances, the Panel has decided also to perform an estimate of the daily intakes per person using a “modified Theoretical Added Maximum Daily Intake” (mTAMDI) approach based on the normal use levels reported by Industry.

As information on use levels for the flavouring substances has not been requested by the JECFA or has not otherwise been provided to the Panel, it is not possible to estimate the daily intakes using the mTAMDI approach for the substances evaluated by the JECFA. The Panel will need information on use levels in order to finalise the evaluation.

Threshold of 1.5 Microgram/Person/Day (Step B5) Used by the JECFA

The JECFA uses the threshold of concern of 1.5 microgram/person/day as part of the evaluation procedure:

“The Committee noted that this value was based on a risk analysis of known carcinogens which involved several conservative assumptions. The use of this value was supported by additional information on developmental toxicity, neurotoxicity and immunotoxicity. In the judgement of the Committee, flavouring substances for which insufficient data are available for them to be evaluated using earlier steps in the Procedure, but for which the intake would not exceed 1.5 microgram per person per day would not be expected to present a safety concern. The Committee recommended that the Procedure for the Safety Evaluation of Flavouring Agents used at the forty-sixth meeting be amended to include the last step on the right-hand side of the original Procedure (“Do the condition of use result in an intake greater than 1.5 microgram per day?”) (JECFA, 1999b).

In line with the Opinion expressed by the Scientific Committee on Food (SCF, 1999), the Panel does not make use of this threshold of 1.5 microgram per person per day.

Genotoxicity

As reflected in the Opinion of SCF (SCF, 1999a), the Panel has in its evaluation focussed on a possible genotoxic potential of the flavouring substances or of structurally related substances. Generally, substances for which the Panel has concluded that there is an indication of genotoxic potential *in vitro*, will not be evaluated using the EFSA Procedure until further genotoxicity data are provided. Substances for which a genotoxic potential *in vivo* has been concluded, will not be evaluated through the Procedure.

Specifications

Regarding specifications, the evaluation by the Panel could lead to a different opinion than that of JECFA, since the Panel requests information on e.g. isomerism.

Structural Relationship

In the consideration of the JECFA evaluated substances, the Panel will examine the structural relationship and metabolism features of the substances within the flavouring group and compare this with the corresponding FGE.

HISTORY OF THE EVALUATION OF THE SUBSTANCES IN THE PRESENT FGE

In FGE.73, which contains a group of 15 alicyclic primary alcohols, aldehydes, acids and related esters, the Panel considered that for nine substances [FL-no: 02.114, 02.141, 05.098, 05.112, 08.067, 09.289, 09.488, 09.534 and 09.615] additional data were needed (no European production volumes available, preventing them to be evaluated using the Procedure, and/or missing data on isomerism/composition). For the remaining six of the 15 JECFA evaluated substances [FL-no: 05.119, 05.123, 08.034, 08.060, 09.028 and 09.536] the Panel agreed with the JECFA conclusion “no safety concern at estimated levels of intake as flavouring substances” based on the MSDI approach.

FGE	Opinion adopted by EFSA	Link	No. of candidate substances
FGE.73	6 March 2008	http://www.efsa.europa.eu/en/efsajournal/pub/868.htm	15
FGE.73Rev1	22 March 2012		16

The present revision of FGE.73, FGE.73Rev1, includes the consideration of one additional substance [FL-no: 05.104]. The substance is a cyclic aldehyde with the conjugated alpha,beta-double bond incorporated in the ring system and was originally allocated to FGE.209 (EFSA, 2011d) in which the Panel concluded that the data available ruled out the concern for genotoxicity and thus concluded that this substance could be evaluated through the Procedure. The information concerning genotoxicity of this substance is described in Section 3.3. A search in open literature for the new substance did not provide any further data on toxicity or metabolism.

Since the publication of FGE.73, the EU production volume has been provided for three substances, [FL-no: 02.141, 09.488 and 09.534] for which the evaluation could not be finalised in the previous version of this FGE, due to lack of these data. Based on the newly submitted EU production volume the substances have already been evaluated in FGE.96⁴ (EFSA, 2011a) (Concerning FGE.73: “For the three substances the Panel concluded at step A3 that these substances would be of no safety concern at their estimated level of intake based on the MSDI approach”), but for the sake of completion, the information has been included in the present revision of FGE.73 as well.

⁴ Scientific Opinion on Flavouring Group Evaluation 96 (FGE.96), addendum to FGE. 51, 52, 53, 54, 56, 58, 61, 62, 63, 64, 68, 69, 70, 71, 73, 76, 77, 79, 80, 83, 84, 85 and 87: Consideration of 88 flavouring substances considered by EFSA for which EU production volumes / anticipated production volumes have been submitted on request by DG SANCO).

Finally, new information on the stereoisomeric composition has been provided for six substances [FL-no: 02.114, 02.141, 05.098, 08.067, 09.289 and 09.615] and for one substance [FL-no: 05.112] further information on the composition has been submitted, since the previous version of FGE.73 (EFFA, 2010a).

1. Presentation of the Substances in the JECFA Flavouring Group

1.1. Description

1.1.1. JECFA Status

The JECFA has at the 59th meeting evaluated a group of 26 flavouring substances consisting of alicyclic primary alcohols, aldehydes, acids and related esters (JECFA, 2003a).

1.1.2. EFSA Considerations

One of the 26 JECFA evaluated substances is not in the Register [Mixture of 2-methyl-5-(2,3-dimethyltricyclo[2.2.1.0(2,6)]hept-3-yl)pent-2-en-1-ol and 2-methyl-5-(2-methyl-3-methylenebicyclo[2.2.1]hept-2-yl)pent-2-en-1-ol] (JECFA-no: 984).

Ten substances [FL-no: 02.060, 02.091, 05.104, 05.106, 05.117, 05.121, 09.034, 09.272, 09.278 and 09.302] are alpha,beta-unsaturated aldehydes or may be metabolised to alpha,beta-unsaturated aldehydes and have been considered together with other alpha,beta-unsaturated aldehydes and ketones. One of these alpha,beta-unsaturated substances [FL-no: 05.104] has been considered with respect to genotoxicity in FGE.209 (EFSA, 2011d), and the Panel concluded that the data available ruled out the concern for genotoxicity and thus concluded that this substance can be evaluated through the Procedure in this revision of FGE.73. The genotoxic properties of the remaining nine of these 10 alpha,beta-unsaturated carbonyl substances were considered together with other alpha,beta-unsaturated aldehydes and ketones in FGE.208 (EFSA, 2008b) for which it was concluded that additional data were required for all nine substances.

This consideration will therefore deal with 16 JECFA evaluated substances. The Panel concluded that the 16 substances in the JECFA flavouring group of alicyclic primary alcohols, aldehydes, acids and related esters are structurally related to the group of primary saturated or unsaturated alicyclic alcohol, aldehyde and esters evaluated by EFSA in the Flavouring Group Evaluation 12, Revision 2 (FGE.12Rev2).

1.2. Isomers

1.2.1. Status

Eight substances in the group of the JECFA evaluated alicyclic primary alcohols, aldehydes, acids and related esters have one or more chiral centres [FL-no: 02.114, 02.141, 05.098, 05.119, 05.123, 08.067, 09.289 and 09.615].

1.2.2. EFSA Considerations

In FGE.73, information was lacking about stereoisomerism for six of these eight substances [FL-no: 02.114, 02.141, 05.098, 08.067, 09.289 and 09.615]. After publication of FGE.73, Industry has specified the stereoisomeric composition (EFFA, 2010a).

For the two stereoisomeric substances [FL-no: 05.119 and 05.123], the CAS register number (CASrn) is considered to specify the stereoisomeric composition (Table 1).

1.3. Specifications

1.3.1. JECFA Status

The JECFA specifications are available for all 16 substances (JECFA, 2002d). See Table 1.

1.3.2. EFSA Considerations

The available specifications are considered adequate for all 16 substances (See Section 1.2).

2. Intake Estimations

2.1. JECFA Status

For all 16 substances evaluated through the JECFA Procedure intake data are available for the EU, see Table 3.1.

2.2. EFSA Considerations

Tonnage data are available for the EU allowing calculation of the intake estimates (MSDI). The Panel noted that since no use levels were submitted no mTAMDI values can be calculated.

3. Genotoxicity Data

3.1. Genotoxicity Studies - Text Taken⁵ from the JECFA (JECFA, 2003a)

No data on genotoxicity were available for the JECFA-evaluated substances. As these substances are rapidly metabolised *in vivo* to compounds of lower toxicological potential, the Committee concluded that the monocyclic and bicyclic terpenes with alkyl ring substituents and containing an alcohol, aldehyde or carboxylic acid group would have little genotoxic potential *in vivo*.

3.2. Genotoxicity Studies - Text Taken⁶ from EFSA FGE.12Rev2 (EFSA, 2010x)

There are no studies available on genotoxicity neither for the nine candidate substances nor for the 15 supporting substances. The genotoxic potential of this group of flavouring substances can therefore not be assessed properly. However, this does not preclude evaluation of the candidate substances in the present group using the Procedure.

3.3. Genotoxicity Studies - Text Taken⁷ from EFSA FGE.209 (EFSA, 2011d)

The Industry has submitted data concerning genotoxicity studies for 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] (safranal), which is the only substance considered in FGE.209.

In Vitro Data

In vitro genotoxicity assays have been performed on the alpha,beta-unsaturated aldehyde safranal [FL-no: 05.104].

Bacterial Reverse Mutation Assay

⁵ The text is taken verbatim from the indicated reference source, but text related to substances not included in the present FGE has been removed.

⁶ The text is taken verbatim from the indicated reference source.

⁷ The text is taken verbatim from the indicated reference source.

Safranal has been tested for its ability to induce gene mutations in the bacterial reverse mutation assay according to OECD guideline 471 (Bevers, 2010b) (for details see Table 2.4). The concentrations used in the different experiments were based on concentrations observed to give toxic effects in previous experiments. Positive and negative controls were included in all experiments according to current guidelines.

There were some increases in revertant numbers in TA102 in the absence and presence of S9 in the first experiment, but these were of insufficient magnitude to be considered as evidence of mutagenicity, they were not concentration-related, and were not reproducible in the other experiments. In all other strains there was no evidence of mutagenic activity either in the absence or presence of S9 in any of the experiments.

It is concluded that under the test conditions applied safranal did not induce gene mutations in bacteria.

Micronucleus Assays

Safranal was evaluated in an *in vitro* micronucleus assay in human peripheral blood lymphocytes for its ability to induce chromosomal damage or aneuploidy in the presence and absence of S9 (Whitwell, 2010c). The maximum soluble concentration of 1250 µg/ml was selected as the maximum concentration for the cytotoxicity range finder test. The concentrations in the main tests were based on toxicity shown in this range finding study (for details see Table 2.4).

At the highest concentration used in the 3+21 hour treatment in the presence of S9, a small statistical increase in the frequency of micronucleated binucleate cells (MNBN) was observed, but this was set against a low mean concurrent vehicle control response. This concentration induced 62 % cytotoxicity, and there was no statistically significant increase in MNBN at the next lowest concentration, which induced 42 % cytotoxicity. Therefore, this isolated increase was not considered to be of biological importance. Outside of this isolated observation at a high level of toxicity, no evidence of chromosomal damage or aneuploidy was observed in terms of any increase in the frequency of MNBN in the presence or absence of S9.

It is concluded that under the conditions of this study, safranal did not induce micronuclei in cultured human lymphocytes.

In Vivo Data

Based on the *in vitro* data available no *in vivo* data are needed.

Discussion of Mutagenicity/Genotoxicity Data

2,6,6-Trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] was tested for all three genetic endpoints: gene mutations, structural and numerical chromosomal aberrations. The substance did not induce gene mutations in bacteria and was not clastogenic and/or aneugenic in mammalian cells *in vitro*.

For validation and study results see Table 2.1.

Conclusion on Genotoxicity and Carcinogenicity

The *in vitro* genotoxicity data on 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] do not indicate genotoxic potential. 2,6,6-Trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] will then be evaluated through the Procedure in FGE.73Rev1.

3.4. EFSA Considerations

The present revision of FGE.73, Revision 1, contains 16 substances, one substance [FL-no: 05.104] has been added. This substance has a structural alert for genotoxicity, but this concern has been alleviated as shown in FGE.209. Therefore, this substance can also be evaluated through the Procedure. No genotoxicity data are available for the remaining 15 JECFA evaluated substances. However, this will not preclude the evaluation of these substances using the Procedure, and the Panel agreed with the JECFA that these 15 substances can be evaluated using the Procedure.

4. Application of the Procedure

4.1. Application of the Procedure to 16 Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters by the JECFA (JECFA, 2003a)

According to the JECFA all 16 substances belong to structural class I using the decision tree approach presented by Cramer et al. (Cramer et al., 1978).

The JECFA concluded for 15 of the alicyclic primary alcohols, aldehydes, acids and related esters at step A3 in the JECFA Procedure – i.e. the substances are expected to be metabolised to innocuous products (step 2) and the intakes for all substances are below the thresholds for their structural class I (step A3).

The JECFA concluded for 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] (safranal) at step B4 in the JECFA Procedure – i.e. the substance cannot be expected to be metabolised to innocuous products (step 2) and an adequate NOAEL exists to provide a margin of safety (step B4). This evaluation was reached by the following procedure: Step B3. The daily per capita intake of the monocyclic substance with two endocyclic double-bonds evaluated at this step, 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104], was below the threshold for daily human intake of compounds of structural class I, and its evaluation therefore proceeded to step B4.

Step B4. As the agent evaluated at this step, 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] (safranal), is structurally related to perillyl alcohol [FL-no: 02.060], data on the toxicity of perillyl alcohol were used to evaluate its safety. Perillyl alcohol given by intragastric gavage changed the weights of several organs in female rats when given at 400 mg/kg bw per day, but not at 120 mg/kg bw per day, in a 90-day study; changes in organ weights were not reported in male rats. Doses of 40, 120 and 400 mg/kg bw per day produced hyperexcitability and salivation, which the authors considered may have been due to its irritating properties (National Cancer Institute, 1996). A daily dose of 120 mg/kg bw was well tolerated by dogs in a 90-day study (National Cancer Institute, 1996). The daily intake of 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] (safranal) is 0.058 microg/kg bw in Europe and 0.001 microg/kg bw in the USA. The margin of safety between these intakes and 120 mg/kg bw per day is > 2000000. The compound also shares structural similarities with alpha-ionone and beta-ionone [FL-no: 07.007] and [FL-no: 07.008], which were evaluated by the Committee at its fifty-first meeting (JECFA, 2000a). The NOELs for these compounds were 10 mg/kg bw per day in a 90-day study in rats, providing a margin of safety of about 200000. Therefore, 2,6,6-trimethylcyclohexa-1,3-diene-1-carbaldehyde [FL-no: 05.104] (safranal) would not be a safety concern.

In conclusion, the JECFA evaluated all 16 substances as to be of no safety concern at the estimated levels of intake as flavouring substances based on the MSDI approach.

The evaluations of the 16 substances are summarised in Table 3.1: Summary of Safety Evaluation of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2003a).

4.2. Application of the Procedure to Nine Primary Saturated or Unsaturated Alicyclic Alcohol, Aldehyde, and Esters by EFSA in FGE.12Rev2 (EFSA, 2010x)

Nine candidate substances were evaluated in FGE.12Rev2. All nine substances were classified into structural class I, using the decision tree approach presented by Cramer et al. (Cramer et al., 1978).

It was anticipated that all nine substances will be metabolised to innocuous products at the estimated levels of intake and accordingly proceed via the A-side of the Procedure. The estimated daily *per capita* intakes of the nine substances range from 0.011 to 43 microgram, which is below the threshold of concern of 1800 microgram/person/day for structural class I.

The Panel concluded all substances in FGE.12Rev2 at step A3 as to be of no safety concern at the estimated levels of intake as flavouring substances based on the MSDI approach.

The stepwise evaluations of the nine substances are summarised in Table 3.2: Summary of Safety Evaluation Applying the Procedure (EFSA, 2010x).

4.3. EFSA Considerations

The Panel agrees with the application of the Procedure as performed by the JECFA for the 16 substances in the group of alicyclic primary alcohols, aldehydes, acids and related esters.

The Panel noted that one substance [FL-no: 05.123] has a terminal double bond. Although theoretically, the double bond may be oxidised to give reactive epoxides, it is expected that for this substance, the metabolism via this pathway is negligible. The terminal double bond is present in a molecule that has an aldehyde function at the end distal from the double bond. The aldehyde function is expected to be readily attacked by oxidation processes, ultimately yielding unsaturated carboxylic acids. Biochemical attack of these carboxylic acids via e.g. beta-oxidation or conjugation with glucuronic acid is expected to be much more efficient and rapid than microsomal oxidation.

5. Conclusion

This consideration deals with 16 flavouring substances, which belong to a group of 26 alicyclic primary alcohols, aldehydes, acids and related esters evaluated by the JECFA of at the 59th meeting in 2002. One substance is not in the Register [Mixture of 2-methyl-5-(2,3-dimethyltricyclo[2.2.1.0(2,6)]hept-3-yl)pent-2-en-1-ol and 2-methyl-5-(2-methyl-3-methylenebicyclo[2.2.1]hept-2-yl)pent-2-en-1-ol] (JECFA-no: 984). Ten substances [FL-no: 02.060, 02.091, 05.104, 05.106, 05.117, 05.121, 09.034, 09.272, 09.278 and 09.302] are alpha,beta-unsaturated aldehydes or may be metabolised to alpha,beta-unsaturated aldehydes. The genotoxic properties of nine of these 10 substances were considered together with other alpha,beta-unsaturated aldehydes and ketones in FGE.208 for which it was concluded that additional data were required. The remaining alpha,beta-unsaturated substance [FL-no: 05.104] has been considered with respect to genotoxicity in FGE.209 (EFSA, 2011d), and the Panel concluded that the data available ruled out the concern for genotoxicity and thus concluded that this substance can be evaluated through the Procedure in this revision of FGE.73. No genotoxicity data are available for the remaining 15 JECFA evaluated substances. However, this will not preclude the evaluation of these substances using the Procedure.

The Panel concluded that the 16 substances are structurally related to the group of nine primary saturated or unsaturated alicyclic alcohol, aldehyde and esters evaluated by EFSA in the Flavouring Group Evaluation 12, Revision 2 (FGE.12Rev2).

The Panel agrees with the application of the Procedure as performed by the JECFA for the 16 substances considered in this FGE.

For all 16 substances evaluated through the Procedure use levels are needed to calculate the mTAMDI in order to identify those flavouring substances that need more refined exposure assessment and to finalise the evaluation.

In order to determine whether the conclusion for the 16 JECFA evaluated substances can be applied to the materials of commerce, it is necessary to consider the available specifications. Adequate specifications including complete purity criteria and identity tests are available for all 16 JECFA evaluated substances.

Thus, for all 16 substances [FL-no: 02.114, 02.141, 05.098, 05.104, 05.112, 05.119, 05.123, 08.034, 08.060, 08.067, 09.028, 09.289, 09.488, 09.534, 09.536 and 09.615] the Panel agrees with the JECFA conclusion “No safety concern at estimated levels of intake as flavouring substances” based on the MSDI approach.

TABLE 1: SPECIFICATION SUMMARY

Table 1: Specification Summary of the Substances in the JECFA Flavouring Group of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2002d)

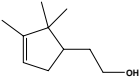
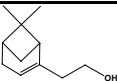
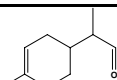


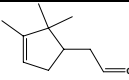

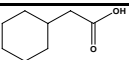
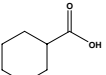
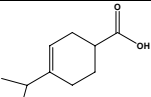
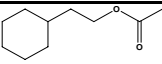
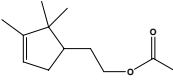
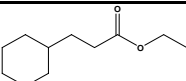
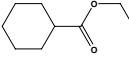
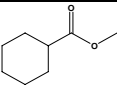
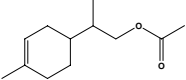
FL-no JECFA- no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility 1) Solubility in ethanol 2)	Boiling point, °C 3) Melting point, °C ID test Assay minimum	Refrac. Index 4) Spec.gravity 5)	EFSA comments
02.114 970	2-(2,2,3-Trimethylcyclopent-3-enyl)ethan-1-ol		3741 1901-38-8	Liquid C ₁₀ H ₁₈ O 154.25	Slightly soluble Miscible	74 (0.8 hPa) NMR 96 %	1.470-1.478 0.882-0.894 (20°)	Racemate (EFFA, 2010a). Synonym (+/-)-campholene alcohol (EFFA, 2010a).
02.141 986	2-(6,6-Dimethylbicyclo[3.1.1]hept-2-en-2-yl)ethan-1-ol		3938 128-50-7	Liquid C ₁₁ H ₁₈ O 166.26	Insoluble Miscible	230 IR NMR 95 %	1.490-1.500 0.965-0.973	Racemate (EFFA, 2010a).
05.098 971	p-Menth-1-en-9-al		3178 10347 29548-14-9	Liquid C ₁₀ H ₁₆ O 152.23	Insoluble Miscible	95 (13 hPa) NMR 99 %	1.458-1.466 0.904-0.916 (20°)	Racemate (EFFA, 2010a).
05.104 977	2,6,6-Trimethylcyclohexa-1,3-diene-1-carbaldehyde		3389 10383 116-26-7	Liquid C ₁₀ H ₁₄ O 150.22	Insoluble Miscible	70 (1 hPa) NMR 96 %	1.525-1.533 0.968-0.980 (20°)	
05.112 978	2,6,6-Trimethylcyclohex-1-en-1-acetaldehyde		3474 10338 472-66-2	Liquid C ₁₁ H ₁₈ O 166.26	Insoluble Miscible	58 (0.5 hPa) IR NMR 92 %	1.480-1.487 0.873-0.885 (20°)	Min. assay (92 %) secondary components β-cyclocitral (2-3 %), β-ionone (0.5-1 %), methyl β-homocyclogeranate (2-4 %), ethyl β-homocyclogeranate (0.6-1 %) (EFFA, 2010a).
05.119 967	2,2,3-Trimethylcyclopent-3-en-1-yl acetaldehyde		3592 10325 4501-58-0	Liquid C ₁₀ H ₁₆ O 152.23	Insoluble Miscible	75 (137 hPa) NMR 99 %	1.462-1.469 0.918-0.924	CASrn in Register refers to (R)-isomer. Register name to be changed to (1R) 2,2,3-Trimethylcyclopent-3-en-1-yl acetaldehyde.
05.123 968	5-Isopropenyl-2-methylcyclopentanecarboxaldehyde		3645 55253-28-6	Liquid C ₁₀ H ₁₆ O 152.23	Insoluble Miscible	80 (14 hPa) IR 95 %	1.501-1.508 0.940-0.952 (20°)	CASrn in Register refers to (1R,2R,5S)-isomer. Register name to be changed to (1R,2R,5S) 5-Isopropenyl-2-methylcyclopentanecarboxaldehyde.

Table 1: Specification Summary of the Substances in the JECFA Flavouring Group of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2002d)

FL-no JECFA- no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility 1) Solubility in ethanol 2)	Boiling point, °C 3) Melting point, °C ID test Assay minimum	Refrac. Index 4) Spec.gravity 5)	EFSA comments
08.034 965	Cyclohexylacetic acid		2347 34 5292-21-7	Solid C ₈ H ₁₄ O ₂ 142.20	Slightly soluble Miscible	242 28-33 NMR 98 %	1.459-1.467 1.001-1.009	
08.060 961	Cyclohexanecarboxylic acid		3531 11911 98-89-5	Solid C ₇ H ₁₂ O ₂ 128.17	Slightly soluble Miscible	232-233 28-32 IR NMR 98 %	1.516-1.520 1.029-1.037	
08.067 976	1,2,5,6-Tetrahydrocuminic acid		3731 71298-42-5	Solid C ₁₀ H ₁₆ O ₂ 168.24	Slightly soluble Soluble	n.a. 61 NMR 95 %	n.a. n.a.	Racemate (EFFA, 2010a).
09.028 964	2-Cyclohexylethyl acetate		2348 218 21722-83-8	Liquid C ₁₀ H ₁₈ O ₂ 170.25	Insoluble Miscible	211 (996 hPa) NMR 98 %	1.442-1.450 0.945-0.948	
09.289 969	alpha-Campholene acetate		3657 36789-59-0	Liquid C ₁₂ H ₂₀ O ₂ 196.29	Insoluble Miscible	96 (7 hPa) IR NMR 98 %	1.453-1.460 0.943-0.949	Commercial product (<i>S</i>)-enantiomer (EFFA, 2010a). Synonym (-)-campholenyl acetate (EFFA, 2010a). Register name to be changed to (-)-campholenyl acetate or (<i>S</i>)-campholenyl acetate.
09.488 966	Ethyl cyclohexanepropionate		2431 2095 10094-36-7	Liquid C ₁₁ H ₂₀ O ₂ 184.28	Insoluble Miscible	91 (10 hPa) NMR 98 %	1.444-1.452 0.926-0.932	
09.534 963	Ethyl cyclohexanecarboxylate		3544 11916 3289-28-9	Liquid C ₉ H ₁₆ O ₂ 156.22	Insoluble Miscible	82 (16 hPa) IR NMR 99 %	1.447-1.454 0.966-0.978 (20°)	
09.536 962	Methyl cyclohexanecarboxylate		3568 11920 4630-82-4	Liquid C ₈ H ₁₄ O ₂ 142.19	Insoluble Miscible	183 IR NMR 98 %	1.439-1.447 0.990-0.999	
09.615 972	p-Menth-1-en-9-yl acetate		3566 10748 28839-13-6	Liquid C ₁₂ H ₂₀ O ₂ 196.28	Insoluble Miscible	228-232 NMR 97 %	1.441-1.448 0.931-0.937	Racemate (EFFA, 2010a).

- 1) Solubility in water, if not otherwise stated.
- 2) Solubility in 95 % ethanol, if not otherwise stated.
- 3) At 1013.25 hPa, if not otherwise stated.
- 4) At 20°C, if not otherwise stated.

- 5) At 25°C, if not otherwise stated.

TABLE 2: GENOTOXICITY DATA

Table 2.1: GENOTOXICITY (*in vitro*) EFSA / FGE.209 (EFSA, 2011d)

FL-no	Chemical Name	Test System <i>in vitro</i>	Test Object	Concentrations of Substance and Test Conditions	Result	Reference	Comments
[05.104]	2,6,6-Trimethylcyclohexa-1,3-diene-1-carbaldehyde	Reverse Mutation	<i>S. typhimurium</i> TA98, TA100, TA1535, TA1537 and TA102	1.6, 8, 40, 200, 1000, 5000 µg/plate	Negative ⁴	(Beevers, 2010b)	Valid study. First experiment: Standard plate ± S9. Toxicity was observed in all strains with and without S9 at 5000 µg/plate and in TA1537 and TA102 with S9 at 1000 µg/plate.
			<i>S. typhimurium</i> TA98, TA100, TA1535, TA1537 and TA102	125, 250, 500, 1000, 2000, 5000 µg/plate	Negative ⁴	(Beevers, 2010b)	Valid study. Second experiment: Standard plate without S9. Toxicity was observed at 2000 µg/plate and above.
			<i>S. typhimurium</i> TA98, TA100, TA1535	62.5, 125, 250, 500, 1000, 2000, 5000 µg/plate	Negative ⁴	(Beevers, 2010b)	Valid study. Second experiment with S9 and preincubation: Toxicity was observed at 500 µg/plate and above.
			<i>S. typhimurium</i> TA1537 and TA102	62.5, 125, 250, 500, 1000, 2000 µg/plate	Negative ⁴	(Beevers, 2010b)	Valid study. Second experiment with S9 and preincubation: Toxicity was observed at 500 µg/plate and above.
			<i>S. typhimurium</i> TA98, TA100, TA1535, TA1537 and TA102	15.625, 31.25, 62.5, 125, 250, 500 µg/plate	Negative ⁴	(Beevers, 2010b)	Valid study. Third experiment with S9 and preincubation: Toxicity was observed at 250 µg/plate and above.
		Micronucleus induction	Human peripheral blood lymphocytes	0, 40, 60, 90 µg/ml ¹ 0, 80, 100, 120, 140 µg/ml ² 0, 4, 8, 12 µg/ml ³	Negative ⁵	(Whitwell, 2010c)	Valid study.

- 1) 3 hours treatment 21 hours recovery without S9.
- 2) 3 hours treatment 21 hours recovery with S9.
- 3) 24 hours treatment no recovery without S9.
- 4) The assays were performed according to OECD guideline 471 and in compliance with GLP.
- 5) This assay is performed in accordance with OECD 487.

TABLE 3: SUMMARY OF SAFETY EVALUATIONS

Table 3.1: Summary of Safety Evaluation of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2003a)

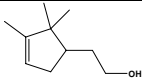
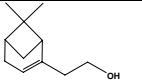
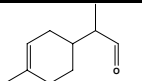
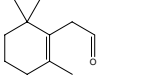
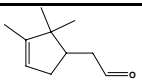
FL-no JECFA-no	EU Register name	Structural formula	EU MSDI 1) US MSDI ($\mu\text{g/capita/day}$)	Class 2) Evaluation procedure path 3)	Outcome on the named compound [4) or 5)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOAEL, genotoxicity)	EFSA conclusion on the material of commerce
02.114 970	2-(2,2,3-Trimethylcyclopent-3-enyl)ethan-1-ol		0.012 ND	Class 1 A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
02.141 986	2-(6,6-Dimethylbicyclo[3.1.1]hept-2-en-2-yl)ethan-1-ol		33 0.01	Class 1 A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
05.098 971	p-Menth-1-en-9-al		0.12 ND	Class 1 A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
05.112 978	2,6,6-Trimethylcyclohex-1-en-1-yl acetaldehyde		0.24 2	Class 1 A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	According to JECFA: Min. assay value is "92 %". Secondary components β -cyclocitral (2-3 %), β -ionone (0.5-1 %), methyl β -homocyclogeranate (2-4 %), ethyl β -homocyclogeranate (0.6-1 %) (EFFA, 2010a). No safety concern at the estimated level of intake based on the MSDI approach.
05.119 967	2,2,3-Trimethylcyclopent-3-en-1-yl acetaldehyde		5.0 ND	Class 1 A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	CASrn in Register refers to (R)-isomer. Register name to be changed to (1R) 2,2,3-Trimethylcyclopent-3-en-1-yl acetaldehyde. No safety concern at the estimated level of intake based on the MSDI approach.

Table 3.1: Summary of Safety Evaluation of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2003a)


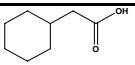
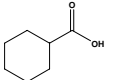
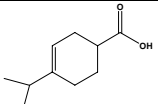
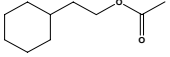
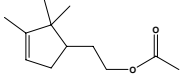
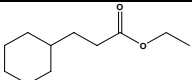
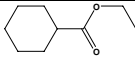
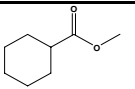
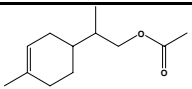

FL-no JECFA-no	EU Register name	Structural formula	EU MSDI 1) US MSDI ($\mu\text{g}/\text{capita}/\text{day}$)	Class 2) Evaluation procedure path 3)	Outcome on the named compound [4) or 5)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOEL, genotoxicity)	EFSA conclusion on the material of commerce
05.123 968	5-Isopropenyl-2-methylcyclopentanecarboxaldehyde		0.012 ND	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	CASrn in Register refers to (1 <i>R</i> ,2 <i>R</i> ,5 <i>S</i>)-isomer Register name to be changed to (1 <i>R</i> ,2 <i>R</i> ,5 <i>S</i>) 5-Isopropenyl-2-methylcyclopentanecarboxaldehyde. No safety concern at the estimated level of intake based on the MSDI approach.
08.034 965	Cyclohexylacetic acid		0.12 0.4	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake as flavouring substance based on the MSDI approach.
08.060 961	Cyclohexanecarboxylic acid		0.061 4	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake as flavouring substance based on the MSDI approach.
08.067 976	1,2,5,6-Tetrahydrocuminic acid		0.012 ND	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
09.028 964	2-Cyclohexylethyl acetate		0.97 ND	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake as flavouring substance based on the MSDI approach.
09.289 969	alpha-Campholene acetate		0.061 ND	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	Register name to be changed to (-)-campholenyl acetate or (S)-campholenyl acetate. No safety concern at the estimated level of intake based on the MSDI approach.
09.488 966	Ethyl cyclohexanepropionate		0.12 0.1	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.

Table 3.1: Summary of Safety Evaluation of Alicyclic Primary Alcohols, Aldehydes, Acids and Related Esters (JECFA, 2003a)

FL-no JECFA-no	EU Register name	Structural formula	EU MSDI 1) US MSDI ($\mu\text{g}/\text{capita}/\text{day}$)	Class 2) Evaluation procedure path 3)	Outcome on the named compound [4) or 5)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOEL, genotoxicity)	EFSA conclusion on the material of commerce
09.534 963	Ethyl cyclohexanecarboxylate		0.24 0.1	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
09.536 962	Methyl cyclohexanecarboxylate		0.073 0.01	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake as flavouring substance based on the MSDI approach.
09.615 972	p-Menth-1-en-9-yl acetate		0.85 ND	Class I A3: Intake below threshold	4)	No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.
05.104 977	2,6,6-Trimethylcyclohexa-1,3-diene-1-carbaldehyde		3.5 0.07	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	4)	Evaluated in FGE.209, genotoxicity concern could be ruled out (EFSA, 2011). No safety concern at the estimated level of intake based on the MSDI approach.	No safety concern at the estimated level of intake based on the MSDI approach.

1) EU MSDI: Amount added to food as flavour in (kg / year) x 10E9 / (0.1 x population in Europe (= 375 x 10E6) x 0.6 x 365) = $\mu\text{g}/\text{capita}/\text{day}$.

2) Thresholds of concern: Class I = 1800 $\mu\text{g}/\text{person}/\text{day}$, Class II = 540 $\mu\text{g}/\text{person}/\text{day}$, Class III = 90 $\mu\text{g}/\text{person}/\text{day}$.

3) Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.

4) No safety concern based on intake calculated by the MSDI approach of the named compound.

5) Data must be available on the substance or closely related substances to perform a safety evaluation.

ND) Not Determined.

Table 3.2: Summary of Safety Evaluation Applying the Procedure (based on intakes calculated by the MSDI approach) (EFSA / FGE.12Rev2)

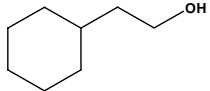
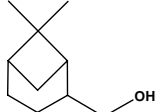
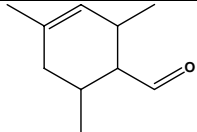
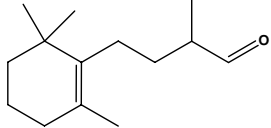
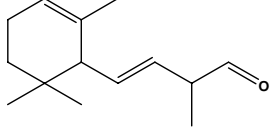
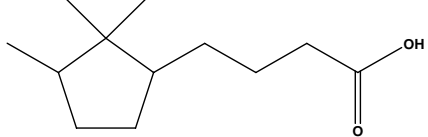
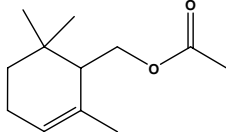
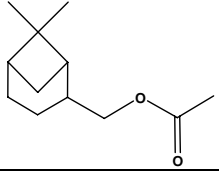
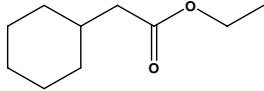
FL-no	EU Register name	Structural formula	MSDI 1) ($\mu\text{g}/\text{capita}/\text{day}$)	Class 2) Evaluation procedure path 3)	Outcome on the named compound [4) or 5]	Outcome on the material of commerce [6), 7), or 8)]	Evaluation remarks
02.134	2-Cyclohexylethan-1-ol		0.011	Class I A3: Intake below threshold	4)	6)	
02.186	Myrtanol		0.37	Class I A3: Intake below threshold	4)	7)	
05.157	Isocyclocitral		0.011	Class I A3: Intake below threshold	4)	7)	
05.183	4-(2,6,6-Trimethylcyclohexenyl)-2-methylbutanal		0.012	Class I A3: Intake below threshold	4)	6)	
05.198	alpha-Methyl ional		0.011	Class I A3: Intake below threshold	4)	7)	
08.135	4-(2,2,3-Trimethylcyclopentyl)butanoic acid		43	Class I A3: Intake below threshold	4)	6)	
09.342	Cyclogeranyl acetate		0.24	Class I A3: Intake below threshold	4)	6)	

Table 3.2: Summary of Safety Evaluation Applying the Procedure (based on intakes calculated by the MSDI approach) (EFSA / FGE.12Rev2)

FL-no	EU Register name	Structural formula	MSDI 1) ($\mu\text{g}/\text{capita}/\text{day}$)	Class 2) Evaluation procedure path 3)	Outcome on the named compound [4) or 5]	Outcome on the material of commerce [6), 7), or 8)]	Evaluation remarks
09.670	Myrtanyl acetate		0.58	Class I A3: Intake below threshold	4)	7)	
09.829	Ethyl cyclohexyl acetate		0.61	Class I A3: Intake below threshold	4)	6)	

1) EU MSDI: Amount added to food as flavour in (kg / year) x 10E9 / (0.1 x population in Europe (= 375 x 10E6) x 0.6 x 365) = $\mu\text{g}/\text{capita}/\text{day}$.

2) Thresholds of concern: Class I = 1800 $\mu\text{g}/\text{person}/\text{day}$, Class II = 540 $\mu\text{g}/\text{person}/\text{day}$, Class III = 90 $\mu\text{g}/\text{person}/\text{day}$.

3) Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.

4) No safety concern based on intake calculated by the MSDI approach of the named compound.

5) Data must be available on the substance or closely related substances to perform a safety evaluation.

6) No safety concern at estimated level of intake of the material of commerce meeting the specification of Table 1 (based on intake calculated by the MSDI approach).

7) Tentatively regarded as presenting no safety concern (based on intake calculated by the MSDI approach) pending further information on the purity of the material of commerce and/or information on stereoisomerism.

8) No conclusion can be drawn due to lack of information on the purity of the material of commerce.

REFERENCES

- Beevers C, 2010b. Reverse mutation in five histidine-requiring strains of *Salmonella typhimurium*. Safranal. Covance Laboratories Ltd, England. Study no. 8200446. May 2010. Unpublished report submitted by EFFA to FLAVIS Secretariat.
- Cramer GM, Ford RA and Hall RL, 1978. Estimation of toxic hazard - a decision tree approach. *Food and Cosmetics Toxicology* 16(3), 255-276.
- EC, 1996a. Regulation No 2232/96 of the European Parliament and of the Council of 28 October 1996. *Official Journal of the European Communities* 23.11.1996, L 299, 1-4.
- EC, 1999a. Commission Decision 1999/217/EC of 23 February 1999 adopting a register of flavouring substances used in or on foodstuffs. *Official Journal of the European Communities* 27.3.1999, L 84, 1-137.
- EC, 2000a. Commission Regulation No 1565/2000 of 18 July 2000 laying down the measures necessary for the adoption of an evaluation programme in application of Regulation (EC) No 2232/96. *Official Journal of the European Communities* 19.7.2000, L 180, 8-16.
- EC, 2009a. Commission Decision 2009/163/EC of 26 February 2009 amending Decision 1999/217/EC as regards the Register of flavouring substances used in or on foodstuffs. *Official Journal of the European Union* 27.2.2009, L 55, 41.
- EFFA, 2010a. EFFA Letters to EFSA for clarification of specifications and isomerism for which data were requested in published FGEs.
- EFSA, 2005g. Opinion of the Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in contact with food on a request from the Commission related to Flavouring Group Evaluation 12 (FGE.12): Primary saturated or unsaturated alicyclic alcohol, aldehyde, and esters from chemical group 7 (Commission Regulation (EC) No 1565/2000 of 18 July 2000). Adopted on 23 February 2005. EFSA-Q-2003-155.
- EFSA, 2008b. Minutes of the 26th Plenary meeting of the Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in Contact with Food, Held in Parma on 27 - 29 November 2007. Parma, 7 January 2008. [Online]. Available: http://www.efsa.europa.eu/EFSA/Event_Meeting/afc_minutes_26thplen_en.pdf
- EFSA, 2010x. Opinion of the Scientific Panel on contact Materials, Enzymes, Flavourings and Processing Aids on a request from Commission related to Flavouring Group Evaluation 12, Revision 2 (FGE.12Rev2): Primary saturated or unsaturated alicyclic alcohol, aldehyde, acid, and esters from chemical group 7 (Commission Regulation (EC) No 1565/2000 of 18 July 2000). Adopted on 30 September 2010. EFSA-Q-2009-00578.
- EFSA, 2010al. Opinion of the Scientific Panel on contact Materials, Enzymes, Flavourings and Processing Aids on a request from Commission related to Flavouring Group Evaluation 96 (FGE.96), addendum to FGE. 51, 52, 53, 54, 56, 58, 61, 62, 63, 64, 68, 69, 70, 71, 73, 76, 77, 79, 80, 83, 84, 85 and 87: Consideration of 88 flavouring substances considered by EFSA for which EU production volumes / anticipated production volumes have been submitted on request by DG SANCO1 (Commission Regulation (EC) No 1565/2000 of 18 July 2000). Adopted on 25 November 2010. EFSA-Q-2010-01246.

- EFSA, 2011d. Opinion of the Scientific Panel on contact Materials, Enzymes, Flavourings and Processing Aids on a request from the Commission related to Flavouring Group Evaluation 209 (FGE.209): Consideration of genotoxicity data on one alpha,beta-unsaturated aldehyde from chemical subgroup 2.3 of FGE.19 by EFSA (Commission Regulation (EC) No 1565/2000 of 18 July 2000). Adopted on 04 February 2011. EFSA-Q-2010-01249.
- JECFA, 1995. Evaluation of certain food additives and contaminants. Forty-fourth Meeting of the Joint FAO/WHO Expert Committee on Food Additives. 14-23 February 1995. WHO Technical Report Series, no. 859. Geneva.
- JECFA, 1996a. Toxicological evaluation of certain food additives. The forty-fourth Meeting of the Joint FAO/WHO Expert Committee on Food Additives and contaminants. WHO Food Additives Series: 35. IPCS, WHO, Geneva.
- JECFA, 1997a. Evaluation of certain food additives and contaminants. Forty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives. Geneva, 6-15 February 1996. WHO Technical Report Series, no. 868. Geneva.
- JECFA, 1999b. Evaluation of certain food additives and contaminants. Forty-ninth report of the Joint FAO/WHO Expert Committee on Food Additives. Rome, 17-26 June 1997. WHO Technical Report Series, no. 884. Geneva.
- JECFA, 2000a. Evaluation of certain food additives. Fifty-first Meeting of the Joint FAO/WHO Expert Committee on Food Additives. Geneva, 9-18 June 1998. WHO Technical Report Series, no. 891. Geneva.
- JECFA, 2002d. Compendium of food additive specifications. Addendum 10. Joint FAO/WHO Expert Committee of Food Additives 59th session. Geneva, 4-13 June 2002. FAO Food and Nutrition paper 52 Add. 10.
- JECFA, 2003a. Safety evaluation of certain food additives. Fifty-ninth Meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Food Additives Series: 50. IPCS, WHO, Geneva.
- JECFA, 2006c. Joint FAO/WHO Expert Committee on Food Additives. Sixty-seventh Meeting. Rome, 20-29 June 2006, Summary and Conclusions. Issued 7 July 2006.
- National Cancer Institute, 1996. Clinical development plan: l-perillyl alcohol. Division of Cancer Prevention and Control, Chemoprevention Branch and Agent Development Committee. J. Cell. Biochem. 265, 137-148.
- SCF, 1999a. Opinion on a programme for the evaluation of flavouring substances (expressed on 2 December 1999). Scientific Committee on Food. SCF/CS/FLAV/TASK/11 Final 6/12/1999. Annex I the minutes of the 119th Plenary meeting. European Commission, Health & Consumer Protection Directorate-General.
- Whitwell J, 2010c. Induction of micronuclei in cultured human peripheral blood lymphocytes. Safranal. Covance Laboratories Ltd, England. Study no. 8222662. May 2010. Unpublished report submitted by EFA to FLAVIS Secretariat.

ABBREVIATIONS

BW	Body Weight
CAS	Chemical Abstract Service
CEF	Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids
CHO	Chinese Hamster Ovary (cells)
CoE	Council of Europe
DNA	Deoxyribonucleic Acid
EFSA	European Food Safety Authority
EPA	United States Environmental Protection Agency
EU	European Union
FAO	Food and Agriculture Organization of the United Nations
FEMA	Flavor and Extract Manufacturers Association
FGE	Flavouring Group Evaluation
FLAVIS (FL)	Flavour Information System (database)
GLP	Good Laboratory Practise
ID	Identity
Ip	Intraperitoneal
IR	Infrared spectroscopy
JECFA	The Joint FAO/WHO Expert Committee on Food Additives
MNBN	Micronucleated binucleate
MSDI	Maximised Survey-derived Daily Intake
mTAMDI	Modified Theoretical Added Maximum Daily Intake
NCE	Normochromatic Erythrocyte
No	Number
NOAEL	No Observed Adverse Effect Level
NTP	National Toxicology Program
OECD	The Organisation for Economic Co-operation and Development
PCE	Polychromatic Erythrocyte
SCE	Sister Chromatic Exchange

SCF Scientific Committee on Food
WHO World Health Organisation