EFSA CEF Panel (EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids), 2014. Scientific Opinion on Flavouring Group Evaluation 74, Revision 3 (FGE.74Rev3): Consideration of Simple Aliphatic Sulphides and Thiols evaluated by the JECFA (53rd and 61st meeting) Structurally related to Aliphatic and Alicyclic Mono-, Di-, Tri-, and Polysulphides with or wi thout Additional Oxygenated Functional Groups from Chemical Group 20 evaluated by EFSA in FGE.08Rev5 (2012)

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SCIENTIFIC OPINION

Scientific Opinion on Flavouring Group Evaluation 74, Revision 3 (FGE.74Rev3): Consideration of Simple Aliphatic Sulphides and Thiols evaluated by the JECFA (53rd and 61st meeting) Structurally related to Aliphatic and Alicyclic Mono-, Di-, Tri-, and Polysulphides with or without Additional Oxygenated Functional Groups from Chemical Group 20 evaluated by EFSA in FGE.08Rev5 (2012)[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 19 simple aliphatic sulphides and thiols evaluated by the JECFA at the 53rd meeting in 1999 and the 61st meeting in 2003. 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. For nine substances [FL-no: 12.088, 12.179, 12.198, 12.212, 12.238, 12.239, 12.255, 12.257 and 12.291] considered in this FGE, the Panel concluded that they would pose "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 for the substances evaluated through the Procedure and for all nine substances, the information is adequate. Thus, the Panel concluded that nine substances [FL-no: 12.088, 12.179, 12.198, 12.212, 12.238, 12.239, 12.255, 12.257 and 12.291] do not give rise to safety concern at their levels of dietary intake, estimated on the basis of the MSDI approach. For 10 candidate substances in FGE.74Rev3 [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155, 12.169, 12.241 and 12.280] evaluated through the Procedure, the Panel concluded that additional toxicity data are required.

[1] On request from the European Commission, Question No EFSA-Q-2013-00410 to -00411 and EFSA-Q-2013-00866 to -00873, adopted on 21 May 2014.

[2] Panel members: Ulla Beckman Sundh, Mona-Lise Binderup, Claudia Bolognesi, Leon Brimer, Laurence Castle, Alessandro Di Domenico, Karl-Heinz Engel, Roland Franz, Nathalie Gontard, Rainer Gürtler, Trine Husøy, Klaus-Dieter Jany, Martine Kolf-Clauw, Wim Mennes, Maria Rosaria Milana, Iona Pratt †, Kettil Svensson, Maria de Fátima Tavares Poças, Fidel Toldra and Detlef Wölfle. Correspondence: fip@efsa.europa.eu

[3] Acknowledgement: The Panel wishes to thank the members of the Working Group on Flavourings: Ulla Beckman Sundh, Leon Brimer, Angelo Carere, Karl-Heinz Engel, Henrik Frandsen, Rainer Gürtler, Frances Hill, Trine Husøy, Wim Mennes, Gerard Mulder and Harriet Wallin for the preparatory work on this scientific opinion and the hearing experts: Vibe Beltoft, Pia Lund and Karin Nørby and EFSA staff: Annamaria Rossi and Kim Rygaard Nielsen for the support provided to this scientific opinion.

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KEY WORDS

safety, flavourings, sulphides, thiols, JECFA 53rd meeting, 61st meeting, FGE.74

SUMMARY

Following a request from the European Commission, the EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (CEF Panel) was asked to deliver a scientific opinion 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 CEF 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 JECFA has evaluated a group of 12 simple aliphatic sulphides and thiols at the 61st meeting and seven trisulphides and one monosulphide in a group of simple aliphatic and aromatic sulphides and thiols at the 53rd meeting. One of the substances evaluated by the JECFA at its 61st meeting is not in the Register (spiro[2,4-dithia-1-methyl-8-oxabicyclo(3.3.0)octane-3,3'-(1'-oxa-2'-methyl)-cyclo-pentane], JECFA-no: 1296). Accordingly, this consideration will deal with 19 JECFA evaluated substances.

This revision is made due to additional genotoxicity data have been submitted for 4-mercapto-4-methyl-2-pentanone [FL-no: 12.169]. In addition, for seven substances [FL-no: 12.009, 12.020, 12.045, 12.169, 12.238, 12.239 and 12.291] new data on specifications have been included. Industry also submitted additional information intended to support the derivation of a NOAEL for substances in subgroup VI (tri- and poly-sulphides).

The Panel concluded that the 19 substances in the JECFA flavouring group of simple aliphatic sulphides and thiols are structurally related to the group of aliphatic and alicyclic mono-, di-, and polysulphides with or without additional oxygenated functional groups evaluated by EFSA in the Flavouring Group Evaluation 08, Revision 5 (FGE.08Rev5).

The results of the additional genotoxicity study on [FL no: 12.169] alleviate the concern for genotoxicity of two substances [FL no: 12.169 and 12.241] and therefore these two substances can be evaluated through the Procedure. For the remaining 17 substances in FGE.74Rev2 the Panel considered already before, that the genotoxicity data available do not preclude evaluating these substances through the Procedure.

The Panel agrees with the application of the Procedure as performed by the JECFA for five of the 19 aliphatic sulphides and thiols [FL-no: 12.238, 12.239, 12.255, 12.257 and 12.291], and concluded, similar the JECFA that these would not a pose safety concern at the current levels of exposure based on the MSDI approach. For three substances [FL- no: 12.179, 12.198 and 12.212] the Panel reached the same conclusion, but used a NOAEL from a different study as the one used by the JECFA.

For diallyl sulphide [FL-no: 12.088] the JECFA evaluated this substance at step B5 to be of no safety concern as the estimated intake in the USA is 0.4 µg/*capita*/day, which is below 1.5 µg/day. The Panel does not make use of this threshold of 1.5 µg per person per day. However, the Panel decided that this substance could be allocated to subgroup I, for which a supporting substance [FL-no: 12.006] provides a NOAEL. Based on the intake estimate (MSDI) for diallyl sulphide [FL-no: 12.088] and this NOAEL an adequate margin of safety of 4.3×10^6 could be calculated.

For the two tertiary thiols [FL-no: 12.169 and 12.241] the Panel did not agree with the JECFA that appropriate studies were available for deriving NOAELs, and accordingly the Panel concluded that additional data are required for these two substances.

For the eight tri- and polysulphides [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155 and 12.280], the Panel concluded that the additional information submitted by the Industry was

insufficient to evaluate the safety of the tri- and polysulphides. Therefore there is still a need for a 90-day study for these substances.

For one substance use levels have been provided by the Industry. The mTAMDI figure calculated for the substances [FL-no: 12.291] is below the threshold of concern for structural class I. For the remaining 18 substances use levels must be provided. These are needed to calculate the mTAMDIs in order to identify those flavouring substances that need a more refined exposure assessment and to finalise the evaluation.

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

Thus, for 10 candidate substances in FGE.74Rev3 [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155, 12.169, 12.241 and 12.280] evaluated through the Procedure, the Panel concluded that additional toxicity data are required.

For the remaining nine JECFA evaluated simple aliphatic sulphides and thiols [FL-no: 12.088, 12.179, 12.198, 12.212, 12.238, 12.239, 12.255, 12.257 and 12.291] 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 AS PROVIDED BY THE EUROPEAN COMMISSION

The use of flavourings is regulated under Regulation (EC) No 1334/2008 of the European Parliament and Council of 16 December 2008[4] on flavourings and certain food ingredients with flavouring properties for use in and on foods. On the basis of Article 9(a) of this Regulation, an evaluation and approval are required for flavouring substances.

The Union list of flavourings and source materials was established by Commission Implementing Regulation (EC) No 872/2012[5]. The list contains flavouring substances for which the scientific evaluation should be completed in accordance with Commission Regulation (EC) No 1565/2000[6].

On 24 November 2011, the EFSA Panel on Food Contact Material, Enzymes, Flavouring and Processing Aids (CEF) adopted an opinion on FGE.74 Revision 2: consideration of simple aliphatic sulphides and thiols evaluated by the JECFA (53rd and 61st meeting) structurally related to aliphatic and alicyclic mono-, di-, tri- and polysulphides with and without additional oxygenated functional groups from chemical group 20 evaluated by EFSA in FGE.08 Rev3 (2011).

In this opinion EFSA has considered the Joint FAO/WHO Expert Committee on Food Additives (the JECFA) evaluation of 19 simple aliphatic sulphides and thiols with and without an additional oxygenated functional group evaluated in the flavouring group evaluation 74 (FGE.74) and its latest revision. EFSA concluded in its opinion, contrary to the JECFA, that the two tertiary thiols [FL-no: 12.169 and 12.241] should not be evaluated using the Procedure due to the concern for genotoxicity and , therefore, additional data was requested.

In addition, in the same opinion, for eight tri- and polysulphides [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155 and 12.280], the Panel did not agree with the JECFA that appropiate studies were available for deriving NOAELs, and accordingly additional data are required for these eight substances. For two of these substances [FL-no: 12.045 and 12.155] the JECFA evaluation is only based on MSDI values derived from production figures from the USA. EU production figures are needed in order to finalise the evaluation of these substances.

Subsequently, these substances were included in the Union List with a Footnote 1, 2 or 4**.**

The requested information on the representative material, 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169], was submitted by the European Flavour Association and forwarded by the Commission to EFSA on 26 April 2013. This information is intended to cover the re-evaluation of this substance and of 2-mercapto-2-methylpentan-1-ol [FL-no: 12.241] from FGE.74.

On 8 and 11 July 2013, the applicant submitted additional relevant data on these polysulphydes, represented by methyl propyl trisulfide [FL-no: 12.020].

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

The European Commission requests the European Food Safety Authority (EFSA) to evaluate this new information and, depending on the outcome, proceed to the full evaluation on these flavouring substances in accordance with Commission Regulation (EC) N° 1565/2000**.**

[4] Regulation (EC) No 1334/2008 of the European Parliament and of the Council of 16 December 2008 on flavourings and certain food ingredients with flavouring properties for use in and on foods and amending Council Regulation (EEC) No 1601/91, Regulations (EC) No 2232/96 and (EC) No 110/2008 and Directive 2000/13/EC. OJ L 354, 31.12.2008, p. 34-50.

[5] Commission implementing Regulation (EU) No 872/2012 of 1 October 2012 adopting the list of flavouring substances provided for by Regulation (EC) No 2232/96 of the European Parliament and of the Council, introducing it in Annex I to Regulation (EC) No 1334/2008 of the European Parliament and of the Council and repealing Commission Regulation (EC) No 1565/2000 and Commission Decision 1999/217/EC. OJ L 267, 2.10.2012, p, 1-161.

[6] 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. OJ L 180, 19.7.2000, p. 8-16.

ASSESSMENT

The approach used by EFSA for safety evaluation of flavouring substances is referred to in Commission Regulation (EC) No 1565/2000, hereafter named the "EFSA Procedure". This Procedure is based on the Opinion of the Scientific Committee on Food (SCF, 1999), which has been derived from the evaluation procedure developed by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1995; JECFA, 1996; JECFA, 1997; JECFA, 1999a), 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 evaluated 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, 2006).

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 (µg)/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 µg 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 µg per day?")" (JECFA, 1999a).

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 µg per person per day.

Genotoxicity

As reflected in the Opinion of SCF (SCF, 1999), 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.

1. History of the Evaluation of the Substances in the Present FGE

At its 61st meeting the JECFA evaluated a group of 12 flavouring substances consisting of simple aliphatic sulphides and thiols. One substance was not in the Register. The remaining 11 flavouring substances have originally been considered by EFSA in the FGE.74 (EFSA, 2008). The Panel concluded that for two substances [FL-no: 12.169 and 12.241], the Procedure should not be applied until adequate genotoxicity data become available and for three substances [FL-no: 12.179, 12.198 and 12.212] additional toxicity data were required.

In the first revision of Flavouring Group Evaluation 74 (FGE.74Rev1) there was a reassessment of four candidate substances due to sub-grouping of the substances based on the type of sulphur-containing functional groups. This is in accordance with what has been done in FGE.08Rev1 (EFSA CEF Panel, 2010a) and in FGE.91 (EFSA CEF Panel, 2010b), which also consider substances with sulphur-containing functional groups. The candidate substances in FGE.74Rev1 that have been reassessed due to this are [FL-no: 12.179, 12.198, 12.212 and 12.280]. The outcome of the evaluation is explained in Section 6.3. Furthermore, the FGE.74Rev1 included the assessment of seven additional substances [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074 and 12.155] evaluated by the JECFA at the 53rd meeting in 1999. The reason for the inclusion of these seven substances is explained in Section 2.1.2.

In the second revision of FGE.74, FGE.74Rev2, one candidate substance has been added, diallyl sulphide [FL-no: 12.088]. This substance has been evaluated by the JECFA at the 53rd meeting in 1999. The reason for the inclusion of this substance is explained in Section 2.1.2. For four substances [FL-no: 12.009, 12.020, 12.045 and 12.169] additional information on specifications received after publication of FGE.74Rev1 has been included.

FGE	Opinion adopted	Link	No. of substances
FGE.74	January 2008	http://www.efsa.europa.eu/en/efsajournal/pub/987.htm	11
FGE.74Rev1	September 2010	http://www.efsa.europa.eu/en/efsajournal/pub/1842.htm	18
FGE.74Rev2	November 2011	http://www.efsa.europa.eu/en/efsajournal/pub/2458.htm	19
FGE.74Rev3			19

The present revision of FGE.74 (FGE.74Rev3) concerns the re-consideration of two JECFA-evaluated substances [FL-no: 12.169 and 12.241]. For these two substances, the Panel concluded previously (FGE.74) that the Procedure should not be applied until adequate genotoxicity data would be available.

For 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169] additional genotoxicity data (bacterial reverse mutation assay) have been submitted (Mc Garry, 2012). This substance [FL-no: 12.169] from FGE.74Rev3 is considered a representative for tertiary monothiols in the Union List, i.e. [FL-no: 12.241] in this FGE, [FL-no: 12.304] in FGE.08Rev5 and [FL-no: 12.038, 12.085, 12.137, 12.138, 12.145, 12.252 and 12.259] in FGE.91.

Additional data for the tri- and polysulphides have been submitted by the Industry (IOFI, 2013). The data include argument for "structure activity relationship" between disulphides and trisulphides, a 90-day inhalation study on dimethyl disulphide and two 90-day studies by Morgareidge and Oser (Morgareidge and Oser, 1970a; Morgareidge and Oser, 1970b).

Since the publication of FGE.74Rev2, information on European production figures has been provided by EFFA for four substances: [FL-no: 12.045, 12.155, 12.169 and 12.241] (IOFI, 2012; EFFA, 2013). For four substances [FL-no: 12.009, 12.020, 12.045 and 12.169] information has been provided and for three substances [12.238, 12.239 and 12.291] additional information on stereoisomerism has been submitted (EFFA, 2013; EFFA, 2014). These data are also included in the present revision.

2. Presentation of the Substances in the JECFA Flavouring Group

2.1. Description

2.1.1. JECFA Status

The JECFA has evaluated a group of 12 flavouring substances consisting of simple aliphatic sulphides and thiols at the 61st meeting (JECFA, 2004a; JECFA, 2004b).

The JECFA has at the 53rd meeting (JECFA, 2000), before 2000, evaluated a group of 137 flavouring substances consisting of simple aliphatic and aromatic sulphides and thiols with and without an additional oxygenated functional group.

2.1.2. EFSA Considerations

This FGE deals with 19 JECFA evaluated substances. Eleven substances from the 61st meeting, 2003, and eight substances from the 53rd meeting, 1999, because:

- Of the 12 simple aliphatic sulphides and thiols evaluated by the JECFA at the 61st meeting one is not in the Register (spiro[2,4-dithia-1-methyl-8-oxabicyclo(3.3.0)octane-3,3'-(1'-oxa-2'-methyl)-cyclopentane], JECFA-no: 1296). From the 61st JECFA meeting, 11 substances remain to be evaluated by EFSA.

- Of the 137 simple aliphatic and aromatic sulphides and thiols with and without an additional oxygenated functional group evaluated by the JECFA at the 53rd meeting, seven are acyclic polysulphides [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074 and 12.155]. These seven substances were evaluated by the JECFA before the year 2000 and have been used as supporting substances in FGE.08 and following revisions. For flavouring substances evaluated by the JECFA before 2000 it is laid down in Commission Regulation (EC) No 1565/2000 (EC, 2000) that if they are considered acceptable at the current estimated intake by the JECFA and comply with the general use criteria, they could be included in the list of authorised substances without undergoing a separate evaluation for the time being. In the FGE.08Rev1 (EFSA CEF Panel, 2010a), it was recognised that tri- and polysulphides may form reactive metabolites and accordingly in FGE.74Rev1, the Panel decided to reconsider these seven polysulphides previously evaluated by the JECFA (see Comment on Subgroup VI (Acyclic tri- and polysulphides), below). Further, for diallyl sulphide [FL-no: 12.088], which the JECFA evaluated at step B5, no NOAEL exists to provide a margin of safety. However, as the estimated intake in the USA of 0.4 µg/capita/day is below the threshold of concern of 1.5 µg/person/day the JECFA Committee noted that intakes below this value would not be expected to present a safety concern. 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 µg/person/day. From the 53rd JECFA meeting eight substances remain to be evaluated by EFSA. In addition, in FGE.08Rev1 the genotoxicity issues that were noted for candidate tertiary thiols are obviously also of relevance for two candidate JECFA-evaluated tertiary thiols [FL nos: 12.169 and 12.241] in this consideration.

The Panel concluded that the substances in the JECFA flavouring group of simple aliphatic sulphides and thiols are structurally related to the group of aliphatic and alicyclic mono-, di-, tri-, and polysulphides with or without additional oxygenated functional groups evaluated by EFSA in the Flavouring Group Evaluation 08, Revision 5 (FGE.08Rev5). Depending on the type of sulphur-containing functional groups, the substances in FGE.08Rev5 were subdivided into 11 subgroups:

I *Acyclic sulphides*

II *Cyclic sulphides*

III *Monothiols, including tertiary monothiols*

IV *Dithiols*

V *Acyclic and cyclic disulphides*

VI *Acyclic polysulphides*

VII *Mono-, di-, tri- and polysulphides with thioacetal structure*

VIII *Thioesters*

IX *Thioic acid*

X *Sulphoxides/sulphones and sulphonates*

XI *Cyclic thioketal fused with an oxolane ring.*

In the following part of this third revision of FGE.74 (FGE.74Rev3) there will be reference to the revision 5 of FGE.08 (FGE.08Rev5) (EFSA CEF Panel, 2012b). It is also revision 5 of FGE.08 that is used in the application of the Procedure by EFSA (Section 5.2 of this FGE.74Rev3).

The 19 JECFA evaluated substances in the present FGE will be considered in compliance with these EFSA defined subgroups.

Comment on Subgroup VI (Acyclic tri- and polysulphides)

During the evaluation of the candidate substances in the FGE.08Rev1 (EFSA CEF Panel, 2010a), it was recognised that tri- and polysulphides (subgroup VI) may form reactive metabolites through reaction with endogenous thiols forming a thiol and a hydropersulphide or perthiol. Compared to thiols, perthiols may be strong reducing agents, forming reactive products when exposed to oxidants. Based on the above information it was concluded that tri- and polysulphides could not be covered by No Observed Adverse Effect Levels (NOAELs) for disulphides, due to the formation of more reactive metabolites.

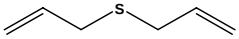
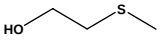
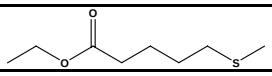
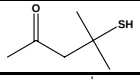
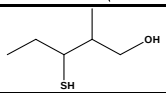
The Panel noted that in FGE.08Rev1 seven supporting substances are tri- or polysulphides [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074 and 12.155]. These substances were evaluated by JECFA before the year 2000[7] (accepted at step B4 based on NOAELs derived from studies with disulphides), and therefore at first not included in the consideration performed by EFSA on the JECFA evaluated substances in FGE.74.

Accordingly, the decision taken in FGE.08Rev1 has had an impact on the tri- and polysulphides in FGE.74 (one substance [FL-no: 12.280]) as well as those evaluated by the JECFA at its 53rd meeting, before 2000 (seven substances [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074 and 12.155]), which were therefore included in the first revision of FGE.74 (FGE.74Rev1) (EFSA CEF Panel, 2010c).

Distribution of the FGE.74Rev3 substances into subgroups

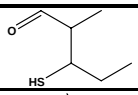
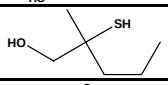
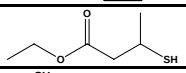
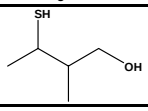
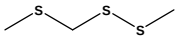
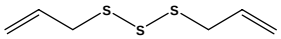
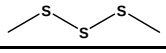
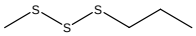
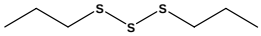
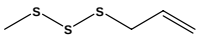
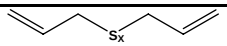
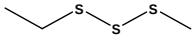
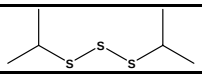
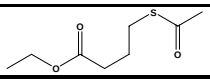
The 19 JECFA evaluated substances in this FGE have been assigned to five subgroups, in accordance with the subdivision in FGE.08Rev5. This subdivision is shown in Table 1 below.

Table 1: Allocation of the 19 JECFA evaluated Substances into Subgroups according to Subdivision in FGE.08Rev5

FL-no:	Register name	Structural formula
I	***Acyclic sulphides***	
12.088	Diallyl sulphide	
12.179	2-(Methylthio)ethan-1-ol	
12.212	Ethyl-5-(methylthio)valerate	
III	***Monothiols***	
12.169	2-Methyl-4-oxopentane-2-thiol	
12.238	3-Mercapto-2-methylpentan-1-ol	

[7] For flavouring substances evaluated by the JECFA before 2000 it is laid down in Commission Regulation (EC) 1565/2000 (EC, 2000a) that if they are considered acceptable at the current estimated intake by the JECFA and comply with the general use criteria, they could be included in the list of authorised substances without undergoing a separate evaluation for the time being.

Table 1: Allocation of the 19 JECFA evaluated Substances into Subgroups according to Subdivision in FGE.08Rev5

FL-no:	Register name	Structural formula
12.239	3-Mercapto-2-methylpentanal	
12.241	2-Mercapto-2-methylpentan-1-ol	
12.255	Ethyl 3-mercaptobutyrate	
12.291	3-Mercapto-2-methyl-1-butanol	
V	***Acyclic and cyclic disulphides***	
12.198	2,3,5-Trithiahexane	
VI	***Acyclic tri- and polysulphides***	
12.009	Diallyl trisulfide	
12.013	Dimethyl trisulfide	
12.020	Methyl propyl trisulfide	
12.023	Dipropyl trisulfide	
12.045	Methyl allyl trisulfide	
12.074	Diallyl polysulfides	 X=2,3,4 or 5
12.155	Methyl ethyl trisulfide	
12.280	Diisopropyl trisulphide	
VIII	***Thioesters***	
12.257	Ethyl 4-(acetylthio)butyrate	

2.2. Isomers

2.2.1. Status

Two substances have one chiral centre [FL-no: 12.241 and 12.255] and three substances have two chiral centres [FL-no: 12.238, 12.239 and 12.291] in the group of the JECFA evaluated sulphides and thiols.

2.2.2. EFSA Considerations

Adequate information on isomeric composition is available for all the substances in FGE.74Rev3. For the two stereoisomeric substances [FL-no: 12.241 and 12.255] with one chiral centre, the CAS register number (CASrn) is considered to cover the stereoisomeric composition as a racemate.

2.3. Specifications

2.3.1. *Status*

The JECFA specifications are available for all 19 substances (JECFA, 1999b; JECFA, 2003). See Table 4.

2.3.2. *EFSA Considerations*

The available specifications are considered adequate for all 19 substances. (see Section 2.2).

3. INTAKE ESTIMATION

3.1. Status

For all substances production volumes, based on which MSDI values can be calculated, are available for the EU, see Tables 3 and 10.

3.2. EFSA Considerations

For one JECFA evaluated substance [FL-no: 12.291], normal and maximum use levels have been provided by the Flavour Industry in accordance with the Commission Regulation (EC) No 1565/2000 (Flavour Industry, 2008; EC, 2000) (see Table 2). Based on the normal use levels, the mTAMDI figure can be calculated (see Table 3). For calculation of mTAMDI figures, see e.g. FGE.03, Annex II (EFSA, 2004).

Table 2: Normal and Maximum Use Levels (mg/kg) available for the JECFA-evaluated substances in FGE.74Rev3

FL-no	Food Categories Normal use levels (mg/kg) Maximum use levels (mg/kg)																	
	01.0	**02.0**	**03.0**	**04.1**	**04.2**	**05.0**	**06.0**	**07.0**	**08.0**	**09.0**	**10.0**	**11.0**	**12.0**	**13.0**	**14.1**	**14.2**	**15.0**	**16.0**
12.291	-	0.1	-	0.01	-	-	-	0.1	0.1	-	-	-	0.1	-	-	-	0.1	0.1
	-	0.5	-	0.1	-	-	-	1	2	-	-	-	1	-	-	-	1	0.5

Table 3: Estimated Intakes Based on the MSDI- and the mTAMDI Approach

FL-no	EU Register name	MSDI – EU (µg/*capita*/day)	MSDI – USA (µg/*capita*/day)	mTAMDI (µg/person/day)	Structural class	Threshold of concern (µg/person/day)
12.013	Dimethyl trisulfide	1.1	0.02		Class I	1800
12.020	Methyl propyl trisulfide	0.21	0.1		Class I	1800
12.023	Dipropyl trisulfide	7.3	1		Class I	1800
12.155	Methyl ethyl trisulfide	0.012	1		Class I	1800
12.169	2-Methyl-4-oxopentane-2-thiol	0.69	0.02		Class I	1800
12.179	2-(Methylthio)ethan-1-ol	0.97	0.9		Class I	1800
12.198	2,3,5-Trithiahexane	0.024	0.04		Class I	1800
12.212	Ethyl-5-(methylthio)valerate	1.8	2		Class I	1800
12.238	3-Mercapto-2-methylpentan-1-ol	0.85	0.7		Class I	1800
12.239	3-Mercapto-2-methylpentanal	2.6	4		Class I	1800
12.241	2-Mercapto-2-methylpentan-1-ol	0.012	4		Class I	1800
12.255	Ethyl 3-mercaptobutyrate	3.4	4		Class I	1800
12.257	Ethyl 4-(acetylthio)-butyrate	3.4	4		Class I	1800
12.280	Diisopropyl trisulphide	0.24	0.007		Class I	1800
12.291	3-Mercapto-2-methyl-1-butanol	0.061	2	17	Class I	1800
12.009	Diallyl trisulfide	3.5	0.02		Class II	540
12.045	Methyl allyl trisulfide	0.012	0.9		Class II	540
12.074	Diallyl polysulfides	1.2	0.02		Class II	540
12.088	Diallyl sulfide	3.5	0.4		Class II	540

SUMMARY OF SPECIFICATION DATA

Table 4: Specification Summary of the Substances in the JECFA Flavouring Group (JECFA, 1999b; JECFA, 2003)

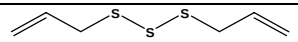
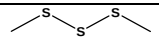
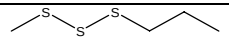
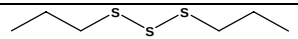
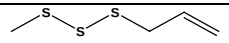
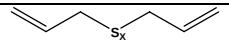
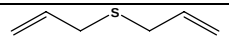
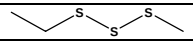
FL-no JECFA -no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility [(a)] Solubility in ethanol [(b)]	Boiling point, °C [(c)] Melting point, °C ID test Assay minimum	Refrac. Index [(d)] Spec.gravity [(e)]	EFSA comments
12.009 587	Diallyl trisulfide		3265 486 2050-87-5	Liquid $C_6H_{10}S_3$ 178.33	Insoluble Insoluble	112-120 (21hPa) IR 65 %	1.600-1.620 1.135-1.170	Min. Assay value 65 %, secondary components 20-25 % allyl disulfide; 5-7 % allylsulfide; 5-7 % allyl tetrasulfide (EFFA).
12.013 582	Dimethyl trisulfide		3275 539 3658-80-8	Liquid $C_2H_6S_3$ 126.26	Very slightly soluble Soluble	165-170 IR 97 %	1.595-1.605 1.195-1.210	
12.020 584	Methyl propyl trisulfide		3308 586 17619-36-2	Liquid $C_4H_{10}S_3$ 154.30	Very slightly soluble Soluble	52 (1.6 hPa) IR 45 %	1.558-1.570 1.095-1.101	Min. Assay value 45 %, secondary components 25 % dipropyl trisulfide, 12 % dipropyl disulfide, 14 % dimethyl disulfide, 3 % methyl propyl sulfide (EFFA).
12.023 585	Dipropyl trisulfide		3276 726 6028-61-1	Liquid $C_6H_{14}S_3$ 182.36	Almost insoluble Soluble	98 (5 hPa) IR 99 %	1.542-1.590 0.952	
12.045 586	Methyl allyl trisulfide		3253 11867 34135-85-8	Liquid $C_4H_8S_3$ 152.29	Very slightly soluble Soluble	47 (1 hPa) NMR 80 %	1.593-1.603 0.975-0.985	Min. Assay value 80 %, secondary components 10-12 % dimethyl trisulfide; 6-8 % allyl trisulfide (EFFA).
12.074 588	Diallyl polysulfides	 X=2,3,4 or 5	3533 11912 72869-75-1	Liquid $C_6H_{10}S_2$ 146.30	Insoluble Slightly soluble	68 (20 hPa) IR NMR 95 %	1.643-1.653 1.220 (20°)	
12.088 458	Diallyl sulfide		2042 11846 592-88-1	Liquid $C_6H_{10}S$ 114.21	Insoluble Sparingly soluble	138-139 IR 97 %	1.488-1.492 0.887-0.892	Solubility in ethanol (EFFA, 2011).
12.155	Methyl ethyl trisulfide		3861	Liquid	Very slightly	46-47 (5 hPa)	1.510-1.520	

Table 4: Specification Summary of the Substances in the JECFA Flavouring Group (JECFA, 1999b; JECFA, 2003)

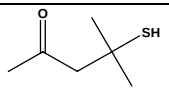
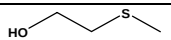
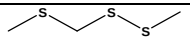
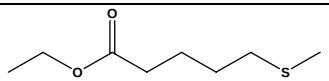
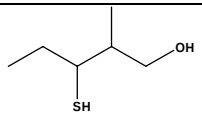
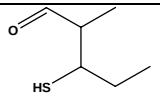
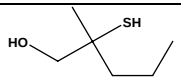
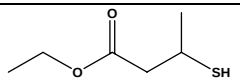
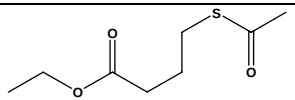
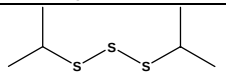
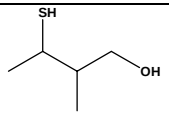
FL-no JECFA -no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility [(a)] Solubility in ethanol [(b)]	Boiling point, °C [(c)] Melting point, °C ID test Assay minimum	Refrac. Index [(d)] Spec.gravity [(e)]	EFSA comments
583				$C_3H_8S_3$	soluble		0.955-0.965	
			31499-71-5	140.28	Soluble	NMR 97 %		
12.169 1293	2-Methyl-4-oxopentane-2-thiol		3997 11500 19872-52-7	Liquid $C_6H_{12}OS$ 132.23	Soluble Very slightly soluble	47-49 (20 hPa) IR NMR MS 48 %	1.431-1.437 1.032-1.037	The Register name to be changed to 4-mercapto-4-methyl-2-pentanone. Min. assay value is 48 % and secondary component 4-methyl-3-penten-2-one [FL-no: 07.101] 48-50 % (EFFA); supplied as a 1 % solution in propylene glycol.
12.179 1297	2-(Methylthio)ethan-1-ol		4004 11545 5271-38-5	Liquid C_3H_8OS 92.16	Insoluble Soluble	169-171 IR NMR MS 98 %	1.490-1.498 1.055-1.065 (20º)	
12.198 1299	2,3,5-Trithiahexane		4021 42474-44-2	Liquid $C_3H_8S_3$ 140.30	Insoluble Soluble	56-58 (10 hPa) MS 95 %	1.436-1.444 1.157-1.163	
12.212 1298	Ethyl-5-(methylthio)valerate		3978 233665-98-0	Liquid $C_8H_{16}O_2S$ 176.27	Insoluble Soluble	227 IR NMR MS 96 %	1.460-1.464 0.993-1.003 (20º)	Register name to be changed to ethyl 5-(methylthio)valerate.
12.238 1291	3-Mercapto-2-methylpentan-1-ol		3996 227456-27-1	Liquid $C_6H_{14}OS$ 134.24	Slightly soluble Soluble	50 (0.7 hPa) IR NMR 99 %	1.480-1.490 0.985-0.995	Mixture of four diastereoisomers, each about 25 % (EFFA, 2014).
12.239 1292	3-Mercapto-2-methylpentanal		3994 227456-28-2	Liquid $C_6H_{12}OS$ 132.23	Insoluble Soluble	98-100 (13 hPa) IR 96 %	1.523-1.529 1.095-1.103	Mixture of four diastereoisomers, each about 25 % (EFFA, 2014).

Table 4: Specification Summary of the Substances in the JECFA Flavouring Group (JECFA, 1999b; JECFA, 2003)

FL-no JECFA -no	EU Register name	Structural formula	FEMA no CoE no CAS no	Phys.form Mol.formula Mol.weight	Solubility [(a)] Solubility in ethanol [(b)]	Boiling point, °C [(c)] Melting point, °C ID test Assay minimum	Refrac. Index [(d)] Spec.gravity [(e)]	EFSA comments
12.241 1290	2-Mercapto-2-methylpentan-1-ol		3995 258823-39-1	Liquid $C_6H_{14}OS$ 134.24	Slightly soluble Soluble	57-59 (0.8 hPa) IR NMR 99 %	1.476-1.483 0.968-0.974 (20º)	Racemate. CASrn is considered to cover the stereoisomeric composition as racemate.
12.255 1294	Ethyl 3-mercaptobutyrate		3977 156472-94-5	Liquid $C_6H_{12}O_2S$ 148.22	Insoluble Soluble	188 IR NMR MS 97 %	1.448-1.453 1.011-1.021 (20º)	Racemate. CASrn is considered to cover the stereoisomeric composition as racemate.
12.257 1295	Ethyl 4-(acetylthio)-butyrate		3974 104228-51-5	Liquid $C_8H_{14}O_3S$ 190.26	Insoluble Soluble	262 IR NMR MS 96 %	1.468-1.472 1.073-1.083 (20º)	
12.280 1300	Diisopropyl trisulphide		 5943-34-0	Liquid $C_6H_{14}S_3$ 182.40	Insoluble Soluble	107-108(13 hPa) NMR MS 95 %	1.441-1.445 1.134-1.140	
12.291 1289	3-Mercapto-2-methyl-1-butanol		3993 227456-33-9	Liquid $C_5H_{12}OS$ 120.21	Slightly soluble Freely soluble	98 (at 2.7 hPa) IR NMR MS 98 %	1.482-1.490 1.002-1.008	Mixture of four diastereoisomers, each about 25 % (EFFA, 2014).

(a): Solubility in water, if not otherwise stated.
(b): Solubility in 95 % ethanol, if not otherwise stated.
(c): At 1013.25 hPa, if not otherwise stated.
(d): At 20°C, if not otherwise stated.
(e): At 25°C, if not otherwise stated.

4. GENOTOXICITY DATA

4.1. Genotoxicity Studies – Text Taken[8] from the JECFA Report (JECFA, 2000; JECFA, 2004b)

Reverse mutation test was performed for diallyl sulphide [FL-no: 12.088] (0.004 - 0.44 µg/ml), using *S. typhimurium* strain TA100. No genotoxicity was observed (Eder et al., 1982)[9].

Groups of male ICR mice were given two doses 48 hours apart of a mixture containing diallyl sulphide [FL-no: 12.088], allyl disulphide (JECFA-no: 572), or diallyl trisulphide [FL-no: 12.009] in corn oil at doses of 10 or 20 mg/ml by gavage. The doses were estimated to provide 0.33 or 0.67 mmol/kg bw or 50 or 100 mg/kg bw on the basis of the composition of the mixture. No increase in the frequency of micronucleated polychromatic erythrocytes was seen in bone-marrow cells (Marks et al., 1992).

Erythro- and threo-3-mercapto-2-methylbutanol [FL-no: 12.291 (3-mercapto-2-methyl-1-butanol)] (50 – 5000 µg/plate) was evaluated for mutagenic activity in the modified Ames test with pre-incubation in the presence and absence of metabolic activation in *S. typhimurium* strains TA97, TA98, TA100, TA102 and TA1535. No genotoxic effects were observed (Gocke, 1997).

For a summary of *in vitro / in vivo* genotoxicity data considered by the JECFA, see Table 6.

4.2. Genotoxicity Studies – Text Taken[10] from EFSA FGE.08Rev5 (EFSA CEF Panel, 2012b)

In vitro / in vivo

Genotoxicity *in vitro* data are available for three candidate substances: di-(1-propenyl)-sulphide (mixture) [FL-no: 12.298] (subgroup I), 2-methylpropane-2-thiol [FL-no: 12.174] (subgroup III), dibutyl disulphide [FL-no: 12.111] (subgroup V). In addition studies are available on 11 supporting substances from subgroups I (1), III (4), V (4) and VIII (2).

In vivo data are available for three supporting substances from subgroups I (1), III (1) and V (1).

Subgroup I (Acyclic sulphides)

In vitro data are available for the candidate substance, di-(1-propenyl)-sulphide [FL-no: 12.298]; Ames test: *S. typhimurium* TA98, TA100, TA102, TA1535, TA1537, 1-100 µg/plate. Result was negative with and without metabolic activation (Stien, 2005).

For supporting substances, only data on diallyl sulphide [FL-no: 12.088] are available. Diallyl sulphide was negative in a limited bacterial reversion assay using one strain only (TA100) and provided equivocal results in an *in vitro* cytogenetic test in which increased incidences of cells with chromosomal aberrations and sister chromatid exchanges (SCEs), statistically significant but not dose related, were observed. *In vivo* diallyl sulphide was evaluated as negative in a micronucleus test in mouse bone marrow, which was, however, not designed to evaluate the genotoxicity of the substance itself as it was tested in a mixture. Overall the data available do not allow evaluation of the genotoxicity of the substances of this subgroup.

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

[9] *The Panel noted that the publication of Eder et al., 1982 is not the correct paper to quote from. It has not been possible for EFSA to identify the correct paper.*

[10] The text is taken from the indicated reference source, but text related to subgroups not included in the present FGE has been removed.

Subgroup III (Monothiols)

2-Methylpropane-2-thiol [FL-no: 12.174] is reported to be negative in an Ames test. It is reported to be positive in a mouse lymphoma assay without metabolic activation and negative in the test with metabolic activation, and it is reported to be negative in an *in vitro* SCE assay. However, these studies are reported only as summaries (Phillips Petroleum Company, 1990a). Some details are available for methods but not for the results. Although the validity of these studies cannot be fully evaluated, the positive result in the mouse lymphoma assay raises concern with respect to the potential for genotoxicity of this tertiary thiol and structurally related compounds, i.e. candidate substance 2-methylbutane-2-thiol [FL-no: 12.172] and ethyl-2-mercapto-2-methyl propanoate [FL-no: 12.304] and the five supporting substances [FL-no: 12.038, 12.085, 12.137, 12.138 and 12.145].

The *in vitro* data available for the other substances in this subgroup do not provide indication of concern for genotoxicity.

Subgroup V (Acyclic and Cyclic disulphides)

Dibutyl disulphide [FL-no: 12.111] is reported to be negative in a mouse lymphoma assay (Dooley et al., 1987). However, the study is reported only as an abstract, and thus, the validity cannot be evaluated.

Further data are available for the supporting substances diallyl disulfide [FL-no: 12.008], dimethyldisulphide [FL-no: 12.026], phenyl disulfide [FL-no: 12.043] and benzyl disulfide [FL-no: 12.081]. All substances were reported to be negative in the Ames test. In addition, diallyl disulphide was reported to be positive in a chromosomal aberration assay *in vitro*, with and without metabolic activation, and weakly positive in a SCE assay. However, the validity of these findings is doubtful as chromosomal aberrations were only increased in conditions associated with extensive (> 90 %) lethality, and because of the limitation of SCE in genotoxic hazard identification.

Subgroup VI (Acyclic tri- and polysulphides)

No genotoxicity information of sufficient quality is available.

Subgroup VIII (Thioesters)

The *in vitro* data available on supporting substances provide no indication of concern for genotoxicity.

Conclusion on genotoxicity

Most *in vitro* and *in vivo* studies are of limited or insufficient quality and provide only limited information.

The available data raise concern with respect to genotoxicity of three tertiary thiols [FL-no: 12.172, 12.174 and 12.304], included as candidate substances in subgroup III. Hydrolysis of the candidate substance 2,4,4-trimethyl-1,3-oxathiane [FL-no: 16.057], included in subgroup VII, leads to the formation of a tertiary thiol structurally related to the above-mentioned compounds. Therefore, there is also concern with respect to genotoxicity of this candidate substance. The Panel noted that in FGE.08 five of the supporting substances were tertiary thiols [FL-no: 12.038, 12.085, 12.137, 12.138 and 12.145] for which a concern for genotoxicity has been raised in the FGE.08Rev1. These supporting substances have been evaluated by the JECFA at the 53rd meeting (JECFA, 2000b; JECFA, 2000). These supporting substances have been considered by EFSA in FGE.91 (EFSA CEF Panel, 2010b).

In addition, genotoxicity of the candidate substance methyl methanethiosulphonate [FL-no: 12.159], included in subgroup X, could not be assessed from the data available. However, due to the similarity

with methyl methanesulphonate, a direct acting mutagen and carcinogen, there is concern with respect to genotoxic potential of this candidate substance.

Therefore, the Panel decided that the Procedure could not be applied to the candidate substances [FL-no: 12.159, 12.172, 12.174, 12.304 and 16.057] until adequate *in vivo* genotoxicity data become available.

The other *in vitro/in vivo* genotoxicity data available, often from limited or poorly reported studies do not provide clear indication of concern for genotoxicity for the remaining candidate substances included in the present evaluation.

For a summary of *in vitro / in vivo* genotoxicity data considered by EFSA, see Tables 7 and 8 of this FGE.

4.3. New Genotoxicity Study on 2-Methyl-4-oxopentane-2-thiol [FL-no: 12.169]

In vitro

2-Methyl-4-oxopentane-2-thiol [FL-no: 12.169] was tested in *S. typhimurium* strains TA98, TA100, TA102, TA1535 and TA1537 in the presence or absence of S9-mix (Mc Garry, 2012). In the first experiment, the concentrations tested were 5.0, 15.8, 50.0, 158.1, 500.0, 1581 and 5000 µg/plate, and the plate incorporation method was used. No evidence of toxicity was observed in the absence or presence of S9-mix in any tester strains. In the second experiment, the concentrations were 156.3, 312.5, 625.0, 1250, 2500 and 5000 µg/plate of 2-methyl-4-oxopentane-2-thiol, and treatments in the presence of S9-mix used the pre-incubation method. Evidence of toxicity was observed through slight thinning of the background lawn and/or marked reduction in revertant numbers in all strains at 2500 and/or 5000 µg/plate in the presence of S9-mix and in TA1537 in the absence of S9-mix. Thus, the study design complied with current recommendations and an acceptable top concentration was achieved. There was no evidence of any mutagenic effect induced by 2-methyl-4-oxopentane-2-thiol in any of the strains, either in the absence or presence of S9-mix.

For a summary of the genotoxicity data on 2-methyl-4-oxopentane-2-thiol, see Table 9.

4.4. EFSA Considerations

Subgroup III includes the tertiary thiols for which a genotoxicity concern was established based on data from a limited gene mutation assay for candidate substances in FGE.08Rev1 [FL-no: 12.174] and additional genotoxicity data were requested for this group of substances. Since the publication of the latest revision of FGE.08, FGE.08Rev5 the Industry has submitted a new bacterial mutation assay for the tertiary thiol [FL-no: 12.169] included in FGE.74. This substance is considered by the Panel to be representative for the whole group of tertiary thiols (in FGE.08, FGE.74 and FGE.91). Based on the new genotoxicity data the Panel concluded that 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169] was not genotoxic in the assay and that 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169] and 2-mercapto-2-methylpentan-1-ol [FL-no: 12.241] do no longer give rise to concern with respect to gene mutations. Therefore, these two substances can be evaluated using the Procedure in the present FGE. The Panel noted that of the material of commerce for [FL no: 12.169], approximately half consists of the α,β-unsaturated carbonyl, 4-methyl-3-penten-2-one [FL-no: 07.101], for which concern for genotoxicity was ruled out in FGE.204 (EFSA CEF Panel, 2012a) and evaluated using the Procedure in FGE.63Rev2 (EFSA CEF Panel, 2013).

Although the available data are limited[11] the Panel considered that for the 19 substances in FGE.74Rev3 the genotoxicity data do not preclude evaluating these substances through the Procedure.

[11] The Panel noted that few days before the adoption of the Opinion EFFA provided a new in vivo micronucleus study on [FL-no:12.169] which will be considered in the next revision of this Opinion.

5. Considerations about Additional Data Submitted for the Tri- and Polysulphides in FGE.74

Based on the new data submitted for the tri- and polysulphides, the Panel concluded as follows:

In the earlier EFSA-evaluations of acyclic tri- and polysulphides (subgroup VI in FGE.08), the evaluation stopped at step B4 due to lack of NOAEL for a representative substance. The available 90-day feeding studies on dipropyltrisulphide and diallyltrisulphide by Morgareidge and Oser (Morgareidge and Oser, 1970a; Morgareidge and Oser, 1970b) were not considered good enough for derivation a NOAEL, and the reasons given was that the studies lacked information on the stability of the test substances, and that histopathology results were missing.

The shortcomings of the Morgareidge and Oser studies are (still) the following:

- no data on the stability of the test substance in feed is given.
- nearly all animals, including control animals, were affected by inflammatory changes in respiratory tract, and in other organs (mainly liver). These changes (probably caused by infections) prohibit adequate interpretation of the study results.
- the data on haematology, clinical chemistry and urine analysis (performed for eight animals in the test-substance groups respectively, and eight animals in the control group at weeks 6 and 12) are only shown as a mean for the three groups, and without any indication of variation between the individuals (e.g. no SD etc.).

Thus the studies are not adequate for derivation of a NOAEL.

The additional data submitted contains general information on biology and metabolism of polysulphides as well as toxicological data. The general information on metabolism and structure-activity relationships of sulphides is mainly in accordance with what is referred in FGE.08 and FGE.74.

Industry also submitted a discussion with the aim to show that NOAELs from toxicity studies for sulphides, disulphides and for tri- and polysulphides are of the same magnitude. In this discussion, route to route extrapolation was applied, but the technique to accomplish this has been presented as being inadequate (Rennen et al., 2004). Additionally, the oral long-term studies that are referred to are one-dose-level-only studies, and as such it is not appropriate to use the NOAELs derived from these studies for comparing the magnitudes with NOAELs from other studies. The discussion on magnitude of NOAELs is therefore not considered relevant. Therefore, the Panel concluded that the extrapolations made by the applicant cannot be used to support derivation of a NOAEL for the oral route.

Thus, the Panel concluded that the additional information as submitted by Industry (IOFI, 2013) is insufficient to evaluate the safety of the tri- and polysulphides. There is still need for a 90-day study from which a NOAEL can be derived with which the safety of these substances can be evaluated.

6. Application of the Procedure

6.1. Application of the Procedure to 19 Simple Aliphatic Sulphides and Thiols Evaluated by the JECFA (JECFA, 2000; JECFA, 2004b)

According to JECFA 15 substances belong to structural class I and four to structural class II using the decision tree approach presented (Cramer et al., 1978).

None of the substances could be anticipated to be metabolised to innocuous products and all were evaluated via the B-side of the Procedure. The estimated daily per capita intakes of the 19 flavouring substances are below the threshold of concern for structural class I and II, and a No Observed Adverse

Effect Level (NOAEL) exists to provide an adequate margin of safety to the estimated intake as flavouring substances (step B4).

Step B4

For erythro- and threo-3-mercapto-2-methylbutanol [FL-no: 12.291(3-mercapto-2-methyl-1-butanol)], the NOEL of 0.7 mg/kg body weight per day for the structurally related substance 2-mercapto-3-butanol [FL-no: 12.024] from a 92-day study in rats fed by gavage (Cox et al., 1974) provides an adequate margin of safety (> 10.000) in relation to known levels of intake of this agent.

This NOEL is also appropriate for the structurally related agents (±)-2-mercapto-2-methylpentan-1-ol [FL-no: 12.241], 3-mercapto-2-methylpentan-1-ol (racemic) [FL-no: 12.238], 3-mercapto-2-methylpentanal [FL-no: 12.239], and (±)-ethyl 3-mercaptobutyrate [FL-no: 12.255], because they are all acyclic thiols with oxidized side-chains that are anticipated to undergo oxidation or hydrolysis and subsequent metabolism via similar metabolic pathways.

For 4-mercapto-4-methyl-2-pentanone [FL-no: 12.169], the NOEL of 1.9 mg/kg bw per day for the structurally related substance 3-mercapto-2-pentanone [FL-no: 12.031] administered to rats by gavage in a 92-day study (Morgareidge, 1971) provides an adequate margin of safety (> 10,000) in relation to known levels of intake of this agent.

For ethyl 4-(acetylthio)butyrate [FL-no: 12.257], the NOEL of 6.5 mg/kg bw per day reported in a 13-week study in rats (Shellenberger, 1970) fed with the structurally related substance ethylthioacetate [FL-no: 12.018] provides an adequate margin of safety (>10,000) in relation to known levels of intake of this agent.

For 2-(methylthio)ethanol [FL-no: 12.179], the NOEL of 1.4 mg/kg bw per day reported in a 13-week study in rats (Cox et al., 1979) fed by gavage with the structurally related substance 2-(methylthiomethyl)-3-phenylpropenal [FL-no: 12.087] provides an adequate margin of safety (>10,000) in relation to known levels of intake of this agent. This NOEL is also appropriate for the structurally related agent ethyl-5-(methylthio)valerate [FL-no: 12.212], which is also an acyclic sulphide with an oxidized side-chain that is anticipated to undergo oxidation and subsequent metabolism via similar pathways.

For 2,3,5-trithiahexane [FL-no: 12.198], the NOEL of 0.3 mg/kg bw per day reported in a 13-week study (Mondino, 1981) in rats fed with the structurally related substance 3-methyl-1,2,4-trithiane [FL-no: 15.036] provides an adequate margin of safety (> 10,000) in relation to known levels of intake of this agent.

For diisopropyl trisulphide [FL-no: 12.280], the NOEL of 4.8 mg/kg bw per day reported in a 13-week study (Morgareidge and Oser, 1970a) in rats fed by gavage with the structurally related substance dipropyl trisulphide [FL-no: 12.023] provides an adequate margin of safety (>100,000) in relation to known levels of intake of this agent.

For diallyl trisulphide [FL-no: 12.009] and dipropyl trisulphide [FL-no: 12.023], the NOELs of 4.6 mg/kg bw per day and 4.8 mg/kg bw per day, respectively were reported in a 90 days study (Morgareidge and Oser, 1970a; Morgareidge and Oser, 1970b) at a single dose, which gave adequate margins of safety for [FL-no: 12.013, 12.020, 12.045, 12.074 and 12.155]. The dose that had no effect is more than 10.000 times greater than the estimated per capita intake in Europe and more than 100,000 times higher than the estimated per capita intake in the United States.

No adequate NOEL was available for diallyl sulphide [FL-no: 12.088] or a related substance, therefore no adequate margin of safety can be provided. Accordingly, the evaluation of the substance proceeds to step B5.

Step B5

For diallyl sulphide [FL-no: 12.088] the intake is estimated to be 0.4 µg/*capita*/day in the USA, which is lower than 1.5 µg/day, therefore the JECFA has concluded that there is no safety concern based on the intake data.

In conclusion the JECFA evaluated all 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 19 simple aliphatic sulphides and thiols with the outcome of the JECFA-evaluations are summarised in Table 10 of this FGE.

6.2. Application of the Procedure to Aliphatic and Alicyclic Mono-, Di-, Tri-, and Polysulphides with or without Additional Oxygenated Functional Groups Evaluated by EFSA in FGE.08Rev5 (EFSA CEF Panel, 2012b)[12]

The application of the Procedure is based on intakes estimated on the basis of the MSDI approach.

For the candidate substance methyl methanethiosulphonate [FL-no: 12.159] (the only substance in subgroup X), there is an indication of a genotoxic potential *in vitro*. Furthermore, for three candidate substances (in subgroup III), 2-methylbutane-2-thiol [FL-no: 12.172], 2-methylpropane-2-thiol [FL-no: 12.174] and ethyl-2-mercapto-2-methyl propanoate [FL-no: 12.304] and one candidate substance (in subgroup VII), 2,4,4-trimethyl-1,3-oxathiane [FL-no: 16.057], a concern for genotoxicity was also identified based on experimental evidence for [FL-no: 12.174] and the structural similarity among these four substances. Therefore, in the absence of further genotoxicity data, the Panel concluded that the Procedure could not be applied to these five substances.

For four candidate substances, 3-mercaptooctanal [FL-no: 12.268] (subgroup III), 3-mercaptodecanal [FL-no: 12.269] (subgroup III), methanedithiol diacetate [FL-no: 12.271] (subgroup VIII) and 3,5-dimethyl-1,2-dithiolane-4-one [FL-no: 12.295] (subgroup V) no data on use as flavouring substances in Europe are available. Therefore, no intakes in Europe can be estimated and accordingly the Panel concluded that the Procedure could not be applied to these four substances.

Thus, for in total nine candidate substances the Procedure could not be applied: [FL-no: 12.159, 12.172, 12.174, 12.268, 12.269, 12.271, 12.295, 12.304 and 16.057].

For the safety evaluation of the remaining 71 candidate substances from chemical groups 20 and 30 the Procedure as outlined in Annex I was applied, based on the MSDI approach. The stepwise evaluations of the 71 substances evaluated through the Procedure are summarised in Table11.

Step 1

The candidate substances were classified following the procedure established by Cramer et al. (Cramer et al., 1978). For the 71 candidate substances evaluated through the Procedure, 42 substances were classified into structural class I, 19 substances were classified into structural class II and 10 substances were classified into structural class III.

Step 2

Step 2 requires consideration of whether metabolic pathways exist to metabolise the candidate substances to innocuous products at the expected levels of intake. The candidate substances may be biotransformed to reactive metabolites, such as thiols, sulphoxides and sulphones and, in consequence,

[12] The text is taken verbatim from the indicated reference source, but text related to subgroups not included in the present FGE has been removed

they are not predicted to be metabolised to innocuous products. Therefore, the evaluation of all 71 candidate substances proceeds via the B-side of the Procedure scheme.

Step B3

The 42 substances in structural class I have estimated European daily *per capita* intakes ranging from 0.0012 to 6.1 µg, which is below the threshold of concern of 1800 µg/person/day. The 19 substances evaluated through the Procedure in structural class II have estimated European daily *per capita* intakes ranging from 0.0024 to 2.4 µg, which is below the threshold of concern for class II of 540 µg/person/day. The 10 substances in structural class III have estimated European daily *per capita* intakes from 0.012 to 6.1µg, which is below the threshold of concern for class III of 90 µg/person/day. Accordingly, all 71 candidate substances proceed to step B4 of the Procedure.

Step B4

No adequate studies on candidate substances are available. Repeated-dose toxicity studies are available on some supporting substances, which, with very few exceptions, have been carried out testing only one dose, giving rise to no observed adverse effects. The results of the adequate studies on supporting substances show a relatively high degree of variability in the reported No Observed Adverse Effect Levels (NOAELs), ranging from 0.06 to 250 mg/kg bw/day.

The 20 candidate substances in subgroup I can be represented by the supporting substance dimethyl sulphide [FL-no: 12.006], for which an adequate 90-day subchronic study is available, indicating that no adverse effects were produced by the highest oral dose tested (250 mg/kg bw/day), which can be considered a NOAEL. The combined estimated daily *per capita* intake of 10 µg for the 18 candidate substances in subgroup I corresponds to 0.17 µg/kg bw/day at a body weight of 60 kg. Thus, a margin of safety of 1.5×10^6 can be calculated. The 20 candidate substances in subgroup I are accordingly not expected to be of safety concern at the estimated levels of intake.

Within subgroup III, adequate 90-day subchronic studies are available for four supporting secondary thiols, 2-mercapto-3-butanol [FL-no: 12.024], cyclopentanethiol [FL-no: 12.029], 2,3- and 10-mercaptopinane [FL-no:12.035] and 2,6-(dimethyl)thiophenol [FL-no: 12.082], which can be considered representative of the 11 candidate substances evaluated through the Procedure in this subgroup. In the four studies, no adverse effects were produced by the highest oral dose tested ranging from 0.06 up to 0.7 mg/kg bw/day. By adopting a conservative approach the lowest value (0.06 mg/kg bw/day) can be considered a NOAEL. The combined estimated daily *per capita* intake of 1.13 µg for the 11 candidate substances evaluated through the Procedure in subgroup III corresponds to 0.019 µg/kg bw/day at a body weight of 60 kg. Thus, a margin of safety of 3×10^3 can be calculated. The 11 candidate substances in subgroup III are accordingly not expected to be of safety concern at the estimated levels of intake.

Within subgroup V, adequate 90-day subchronic studies are available for two supporting substances dicyclohexyl disulfide [FL-no: 12.028] and benzyl methyl disulfide [FL-no: 12.068], which can be considered representative of the four candidate substances in this subgroup evaluated through the Procedure. In the two studies, no adverse effects were produced by the highest oral dose tested: 0.23 and 1.15 mg/kg bw/day. By adopting a conservative approach, the lowest value (0.23 mg/kg bw/day) can be considered a NOAEL. The combined estimated daily *per capita* intake of 0.6 µg for the four candidate substances evaluated through the Procedure in subgroup V corresponds to 0.01 µg/kg bw/day at a body weight of 60 kg. Thus, a margin of safety of 2.3×10^4 can be calculated. The four candidate substances in subgroup V are accordingly not expected to be of safety concern at the estimated levels of intake.

Within subgroup VI, no adequate toxicity study from which a NOAEL could be established was available, neither on the candidate substances nor on supporting substances. Therefore, the Panel

concluded that additional data are required for the eight tri-, tetra- and polysulphides in subgroup VI [FL-no: 12.093, 12.094, 12.097, 12.100, 12.112, 12.116, 12.164 and 12.167].

Within subgroup VIII, an adequate 90-day subchronic study is available for one supporting substance, ethyl thioacetate [FL-no: 12.018], which can be considered representative of the eight candidate substances evaluated through the Procedure in this subgroup. In the study, no adverse effects were produced by the highest oral dose tested: 6.63 mg/kg bw/day. Therefore, the NOAEL is concluded to be 6.63 mg/kg bw per day for ethyl thioacetate. The combined estimated daily *per capita* intake of 2.4 µg for the eight candidate substances in subgroup VIII corresponds to 0.04 µg/kg bw/day at a body weight of 60 kg. Thus, a margin of safety of 1.7×10^5 can be calculated. The eight candidate substances in subgroup VIII are accordingly not expected to be of safety concern at the estimated levels of intake.

The conclusion from step B4 is that for the 43 candidate substances belonging to subgroups I, III, V and VIII, and evaluated through the Procedure, adequate NOAELs exist for the candidate substance or for structurally related substances providing adequate margins of safety at the estimated levels of intake. Therefore, these candidate substances are not expected to be of safety concern at the levels of exposure estimated by the MSDI approach.

For the eight candidate substances belonging to subgroup VI [FL-no: FL-no: 12.093, 12.094, 12.097, 12.100, 12.112, 12.116, 12.164 and 12.167] additional toxicity data are required.

The evaluations of the aliphatic and alicyclic mono-, di-, tri-, and polysulphides are summarised in Table 11.

6.3. EFSA Considerations

The 19 JECFA evaluated simple aliphatic sulphides and thiols are distributed into five subgroups of structurally related substances. The sub-grouping is in compliance with the one used in FGE.08Rev5 (see Section 2.1.2 and Table 1).

Althoug the available data are limited the Panel considered that for the remaining 19 substances in FGE.74Rev3 the genotoxicity data do not preclude evaluating these substances through the Procedure.

The Panel agrees with the application of the Procedure as performed by the JECFA for five simple aliphatic sulphides and thiols, namely [FL-no: 12.238, 12.239, 12.255, 12.257 and 12.291].

For 14 substances [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.088, 12.155, 12.169, 12.179, 12.198, 12.212, 12.241 and 12.280] the Panel did not agree with the application of the Procedure by the JECFA for the following reasons:

The JECFA derives a NOAEL of 1.4 mg/kg bw per day reported in a 13-week study in rats (Cox et al., 1979) fed by gavage with 2-(methylthiomethyl)-3-phenylpropenal [FL-no: 12.087]. The Panel did not agree with the JECFA that 2-(methylthiomethyl)-3-phenylpropenal [FL-no: 12.087] is structurally related to 2-(methylthio)ethan-1-ol [FL-no: 12.179] or ethyl-5-(methylthio)-valerate [FL-no: 12.212]. The JECFA derived a NOAEL of 0.3 mg/kg bw per day reported in a 13-week study (Mondino, 1981) in rats fed with 3-methyl-1,2,4-trithiane [FL-no: 15.036]. The Panel does not agree with the JECFA that 2,3,5-trithiahexane [FL-no: 12.198] is structurally related to 3-methyl-1,2,4-trithiane [FL-no: 15.036].

However, in the first revision of FGE.74, FGE.74Rev1, all substances have been distributed to subgroups with respect to sulphur-containing functional groups, according to FGE.08 and following revisions. The JECFA evaluated substances 2-(methylthio)ethan-1-ol and ethyl-5-(methylthio)valerate [FL-no: 12.179 and 12.212] have been allocated to subgroup I, *Acyclic sulphides*, and 2,3,5-trithiahexane [FL-no: 12.198] has been allocated to subgroup V, *Acyclic and cyclic disulphides*. Appropriate NOAELs exist for these two subgroups, as is argued in FGE.08Rev5. Since based on

these NOAELs adequate margins of safety can be calculated for [FL no: 12.179, 12.198 and 12.212], in line with the JECFA, the Panel also concludes that these substances are not expected to be of safety concern at the estimated levels of intake.

For the diallyl sulphide [FL-no: 12.088] the JECFA evaluated this substance at step B5 of the Procedure to be of no safety concern as the estimated intake in the USA of 0.4 µg/*capita*/day is below 1.5 µg/day. 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 µg per person per day. However, this substance is allocated subgroup I, for which an appropriate NOAEL (reported for dimethylsulphide [FL-no: 12.006]) exist, as is demonstrated in FGE.08Rev5. The NOAEL of 250 mg/kg bw/day provides a margin of safety of 4.3 x 10^6 based on a European MSDI of 3.5 µg/*capita*/day and accordingly the Panel concludes that this substance is not expected to be of safety concern at the estimated level of intake.

Industry indicated that for the two tertiary thiols, 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169] and 2-mercapto-2-methylpentan-1-ol [FL-no: 12.241], both from subgroup III, the JECFA derives a NOAEL from 90-day studies performed with secondary thiols (3-mercapto-3-butanol [FL-no: 12.031] (Morgareidge, 1971) and 2-mercapto-3-butanol [FL-no: 12.024] (Cox et al., 1974), respectively. The Panel did not agree with the JECFA that the tertiary and secondary thiols are sufficiently structurally related for a reading across with respect to deriving a NOAEL. Accordingly, the Panel concluded at step B4 that further data are required for the evaluation of [FL-no: 12.169 and 12.241].

For the eight substances in subgroup VI (acyclic tri- and polysulphides) [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155 and 12.280], 90-day studies were available on [FL-no: 12.009 and 12.023], but the studies were not considered adequate for deriving a NOAEL (Morgareidge and Oser, 1970a; Morgareidge and Oser, 1970b) (see FGE.08Rev3 Section 8.2 (There are no data on stability of test substances and no results reported from histopathological examinations)). It has also been concluded that tri- and poly-sulphides cannot be covered by NOAELs for disulphides, due to the formation of more reactive metabolites than is the case for the disulphides. Accordingly, the Panel concluded at step B4 (contrary to JECFA) that further data are required for the tri- and polysulphides [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155 and 12.280].

In summary: No safety concern was identified for the following substances: [FL-no: 12.088, 12.179, 12.198, 12.212, 12.238, 12.239, 12.255, 12.257 and 12.291]. For the remaining substances [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155, 12.169, 12.241 and 12.280], additional toxicity data is requested.

An overview of the EFSA considerations is given in Table 5 below.

Table 5: Overview of Supporting Substances Providing Adequate NOAEL for the Procedure Step B4

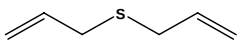
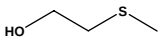
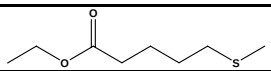
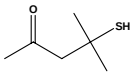
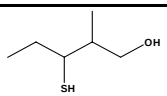
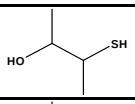
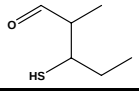
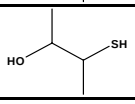
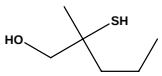
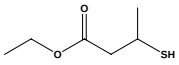
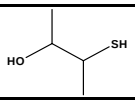
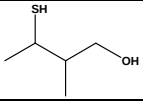
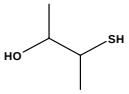
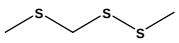
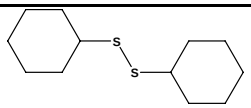
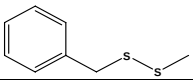
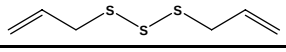
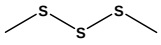
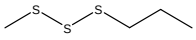
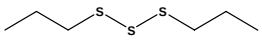
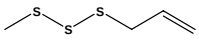
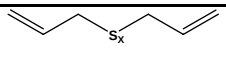
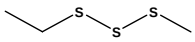
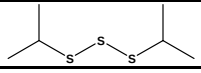
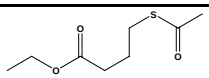
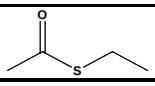
FL-no:	Register name	Structural formula	NOAEL provider
I	***Acyclic sulphides***		
12.088	Diallyl sulphide		
12.179	2-(Methylthio)ethan-1-ol		
12.212	Ethyl-5-(methylthio)valerate		
III	***Monothiols***		

Table 5: Overview of Supporting Substances Providing Adequate NOAEL for the Procedure Step B4

12.169	2-Methyl-4-oxopentane-2-thiol		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.238	3-Mercapto-2-methylpentan-1-ol		
12.239	3-Mercapto-2-methylpentanal		
12.241	2-Mercapto-2-methylpentan-1-ol		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.255	Ethyl 3-mercaptobutyrate		
12.291	3-Mercapto-2-methyl-1-butanol		
V	***Acyclic and cyclic disulphides***		
12.198	2,3,5-Trithiahexane		 and 
VI	***Acyclic tri- and polysulphides***		
12.009	Diallyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.013	Dimethyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.020	Methyl propyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.023	Dipropyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.045	Methyl allyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.074	Diallyl polysulfides	 X=2,3,4 or 5	No adequate NOAEL available for step B4 in the Procedure – additional data required
12.155	Methyl ethyl trisulfide		No adequate NOAEL available for step B4 in the Procedure – additional data required
12.280	Diisopropyl trisulphide		No adequate NOAEL available for step B4 in the Procedure – additional data required
VIII	***Thioesters***		
12.257	Ethyl 4-(acetylthio) butyrate		

The results of the evaluation of the 19 candidate substances in this FGE have been included in Table 10.

CONCLUSION

In Flavouring Group Evaluation 74, Revision 3 (FGE.74Rev3) the EFSA considered 12 simple aliphatic sulphides and thiols evaluated by the JECFA at its 61st meeting and seven trisulphides and one monosulphide in a group of simple aliphatic and aromatic sulphides and thiols evaluated at its 53rd meeting. Accordingly the consideration dealt with 19 JECFA evaluated substances.

The Panel concluded that the 19 substances in the JECFA flavouring group of simple aliphatic sulphides and thiols are structurally related to a group of aliphatic and alicyclic mono-, di-, and polysulphides with or without additional oxygenated functional groups evaluated by EFSA in the Flavouring Group Evaluation 08, Revision 5 (FGE.08Rev5).

This revision is made due to additional genotoxicity data have been submitted for 4-mercapto-4-methyl-2-pentanone [FL-no: 12.169]. In addition, for seven substances [FL-no: 12.009, 12.020, 12.045, 12.169, 12.238, 12.239 and 12.291] new data on specifications have been included. Industry also submitted additional information intended to support the derivation of a NOAEL for substances in subgroup VI (tri- and poly-sulphides).

In previous versions of this FGE a concern with respect to genotoxicity was identified for two candidate substances [FL-no: 12.169 and 12.241]. Additional genotoxicity data have now become available for 2-methyl-4-oxopentane-2-thiol [FL-no: 12.169], which is considered to be supporting for [FL-no: 12.241]. Althoug the available data are limited the Panel considered that for the remaining 19 substances in FGE.74Rev3 the genotoxicity data do not preclude evaluating these substances through the Procedure.

The Panel agrees with the application of the Procedure as performed by the JECFA for five of the 19 aliphatic sulphides and thiols [FL-no: 12.238, 12.239, 12.255, 12.257 and 12.291]. For these five substances the Panel concluded, similar to the JECFA that these would not pose any safety concern at the current levels of exposure based on the MSDI approach. For three substances [FL-no: 12.179, 12.198 and 12.212] the Panel reached the same conclusion, but used a NOAEL from a different study as the one used by the JECFA.

For diallyl sulphide [FL-no: 12.088] the JECFA evaluated this substance at step B5 to be of no safety concern as the estimated intake in the USA is 0.4 µg/*capita*/day, which is below 1.5 µg/day. 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 µg per person per day. However, the Panel decided that this substance could be allocated to subgroup I, for which a supporting substance [FL-no: 12.006] provides a NOAEL. Based on the intake estimate (MSDI) for diallyl sulphide [FL-no: 12.088] and this NOAEL an adequate margin of safety of 4.3×10^6 could be calculated.

For the two tertiary thiols [FL-no: 12.169 and 12.241] the Panel did not agree with the JECFA that appropriate studies were available for deriving NOAELs, and accordingly the Panel concluded that additional data are required for these two substances.

For the eight tri- and polysulphides [FL-no: FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155 and 12.280], the Panel concluded that the additional information submitted by the Industry was insufficient to evaluate the safety of the tri- and polysulphides. Therefore there is still a need for a 90-day study for these substances.

For one substance use levels have been provided by the Industry. The mTAMDI figure calculated for the substance [FL-no: 12.291] is below the threshold of concern for the structural class I. For the remaining 18 substances use levels must be provided. These are needed to calculate the mTAMDIs in

order to identify those flavouring substances that need more refined exposure assessment and to finalise the evaluation.

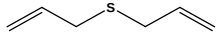
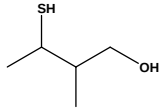
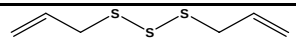
In order to determine whether the conclusion for the JECFA evaluated substances can be applied to the materials of commerce, it is necessary to consider the available specifications. Specifications including purity criteria and identity are available for all the JECFA evaluated substances.

Thus, for 10 candidate substances in FGE.74Rev3 [FL-no: 12.009, 12.013, 12.020, 12.023, 12.045, 12.074, 12.155, 12.169, 12.241 and 12.280] evaluated through the Procedure, the Panel concluded that additional toxicity data are required.

For the remaining nine JECFA evaluated simple aliphatic sulphides and thiols [FL-no: 12.088, 12.179, 12.198, 12.212, 12.238, 12.239, 12.255, 12.257 and 12.291] the Panel agrees with the JECFA conclusion “No safety concern at estimated levels of intake as flavouring substances” based on the MSDI approach.

SUMMARY OF GENOTOXICITY DATA

Table 6: Genotoxicity Data (*in vitro / in vivo*) evaluated by JECFA (JECFA, 2000; JECFA, 2004b)

FL-no JECFA-no	EU Register name JECFA name	Structural formula	End-point	Test system	Concentration	Results	Reference	Comments
In vitro								
12.088 458	Diallyl sulfide		Reverse mutation	*S. typhimurium* TA100	0.004 - 0.44 µg/ml	Negative[(a)]	(Eder et al., 1982)	The Panel noted that the publication by Eder et al., 1982a is not the correct paper to quote from. It has not been possible for EFSA to identify the correct paper.
12.291 1289	3-Mercapto-2-methyl-1-butanol		Reverse mutation	*S. typhimurium* TA1535, TA97, TA98, TA100, TA102	50 – 5000 µg/ plate	Negative[(a)]	(Gocke, 1997)	The racemate (Erythro- and threo-3-Mercapto-2-methyl-1-butanol) was used in the toxicological evaluation.
In vivo								
12.009 587	Diallyl trisulfide		*In vivo* mouse micronucleus test	Mouse	0.33 – 0.67 mM/kg (59 - 120 mg/kg)[(b)]	Negative	(Marks et al., 1992)	Insufficient quality. Mixture of three substances was tested.

(a): With and without metabolic activation from S9.
(b): Study used a mixture of allyl sulphide, allyl disulphide and allyl trisulphide in the respective ratio, 68:20:12.

Table 7: Genotoxicity Data (*in vitro*) EFSA / FGE.08Rev5 (EFSA CEF Panel, 2012b)

Chemical Name [FL-no]	Test system	Test Object	Concentration	Result	Reference	Comments
Subgroup I – Acyclic Sulphides						
(Diallyl sulphide [12.088])	Ames test	*S. typhimurium* TA100	0.004 – 0.44 µg/ml	Negative (±S9)	(Eder et al., 1982)	Review. No details on method and results reported. Only TA100 used.
	Sister chromatid exchange	Chinese hamster ovary cells	200 - 600 µg/ml	Positive[(a)]	(Musk et al., 1997)	Limited quality of study. Insufficiently reported.
	Chromosomal aberrations	Chinese hamster ovary cells	200 - 600 µg/ml	Positive[(a)]	(Musk et al., 1997)	Limited quality of study. Insufficiently reported.
Di-(1-propenyl)-sulfid (mixture) [12.298]	Ames test	*S. typhimurium* TA98, TA100, TA102, TA1535, TA1537	1 – 100 µg/plate	Negative[(a)]	(Stien, 2005)	Un-published GLP study. Study considered valid.
Subgroup II – Cyclic Sulphides						
Tetrahydrothiophene [15.102]	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537	50 – 5000 µg/plate	Negative (±S9)	(Pennwalt Corporation, 1987a)	Validity of this study cannot be fully evaluated (only abstract provided).
	Cytogenetic assay	Human lymphocytes	12.5 – 125 µg/ml	Negative (±S9)	(Pennwalt Corporation, 1987a)	Validity of this study cannot be fully evaluated (only abstract provided).
	HPRT assay	Chinese hamster ovary cells	100 – 200 µg/ml	Negative (±S9)	(Pennwalt Corporation, 1987a)	Validity of this study cannot be fully evaluated (only abstract provided).
	Sister chromatid exchange	Chinese hamster ovary cells	15.63 – 125 µg/ml	Negative (±S9)	(Pennwalt Corporation, 1987b)	Validity of this study cannot be fully evaluated (only abstract provided).
	Unscheduled DNA synthesis	Human epithelial cells	2.5 – 5120 µg/ml	Negative (±S9)	(Pennwalt Corporation, 1987a)	Validity of this study cannot be fully evaluated (only abstract provided).
(1,4-Dithiane [15.066])	Ames test	*S. typhimurium* TA98, TA100	0.8 – 100 µ mol/plate (96.2 - 12024 µg/plate)	Positive (-S9) Negative (+S9)	(Lee et al., 1994)	Only two strains were tested, otherwise acceptable study.
	Sister chromatid	Chinese hamster ovary cells	2000 µM (240 µg/ml)	Negative	(Lee et al., 1994)	Insufficient quality.

Table 7: Genotoxicity Data (*in vitro*) EFSA / FGE.08Rev5 (EFSA CEF Panel, 2012b)

Chemical Name [FL-no]	Test system	Test Object	Concentration	Result	Reference	Comments
	exchange			(±S9)		
Subgroup III – Monothiols						
2-Methylpropane-2-thiol [12.174]	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	10000 µg/plate	Negative (±S9)	(Phillips Petroleum Company, 1990a)	Validity of this study cannot be fully evaluated (only abstract provided).
	Forward mutational MLTK assay	L5178Y/tk+/- mouse lymphoma cells	1000 µg/ml	Positive (-S9) Negative (+S9)	(Phillips Petroleum Company, 1990a)	Validity of this study cannot be fully evaluated (only abstract provided).
	Sister chromatid exchange	Chinese hamster ovary cells	1350 µg/ml	Negative (+S9)[(b)]	(Phillips Petroleum Company, 1990a)	Validity of this study cannot be fully evaluated (only abstract provided).
(Allyl mercaptan [12.004])	Modified Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	0.005 – 1.5 µl/ml (4.6 – 1400 µg/ml)	Negative (±S9)	(Eder et al., 1980)	Acceptable quality.
(Benzyl mercaptan [12.005])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	3.6 mg/plate (3600 µg/plate)	Negative (±S9)	(Wild et al., 1983)	Review. Methods and results insufficiently documented.
(2-Mercaptopropionic acid [12.039])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	3.6 mg/plate (3600 µg/plate)	Negative (±S9)	(Wild et al., 1983)	Review. Methods and results insufficiently documented.
(Benzenethiol [12.080])	Ames test	*S. typhimurium* TA98, TA100	25 – 500 µg/plate	Negative (±S9)	(LaVoie et al., 1979)	Insufficient quality (only two strains were used, and all doses -except the lowest dose - were toxic).
Subgroup IV – Dithiols						
(1,2-Ethanedithiol [12.066])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	5 doses up to 5000 µg/plate	Negative (±S9)	(Phillips Petroleum Company, 1990b)	Validity cannot be fully evaluated (only abstract provided).
	Sister chromatid exchange	Chinese hamster ovary cells	0.5 - 50 µg/ml	Positive (±S9)	(Pence et al., 1982)	Acceptable quality.
	Forward mutational assay	L5178Y/tk+/- mouse lymphoma cells	150 µg/ml	Positive (-S9)	(Pence et al., 1982)	Positive only at cytotoxic concentrations.
	Forward mutational	L5178Y/tk+/- mouse lymphoma	1 µg/ml	Negative	(Pence et al., 1982)	Insufficiently

Table 7: Genotoxicity Data (*in vitro*) EFSA / FGE.08Rev5 (EFSA CEF Panel, 2012b)

Chemical Name [FL-no]	Test system	Test Object	Concentration	Result	Reference	Comments
	assay	cells		(+S9)		documented.
Subgroup V – Acyclic Di-, Tri-, and Poly-sulphides						
(Diallyl disulphide [12.008])	Modified Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	0.0015 – 0.15 µg/ml	Negative (±S9)	(Eder et al., 1980)	Acceptable quality.
	Sister chromatid exchange	Chinese hamster ovary cells	2 - 25 µg/ml	Weakly positive (±S9)	(Musk et al., 1997)	Limited quality. Insufficiently reported.
	Chromosomal aberrations	Chinese hamster ovary cells	2 - 25 µg/ml	Positive (±S9)	(Musk et al., 1997)	Limited quality. Insufficiently reported.
(Dimethyl disulphide [12.026])	Ames test	*S. typhimurium* TA98, TA100, TA102	0.000011 – 1.1 mmol/plate (1.04 - 104000 µg/plate)	Negative (±S9)	(Aeschbacher et al., 1989)	Limited quality (only 3 strains used).
(Phenyl disulphide [12.043])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	3.6 mg/plate (3600 µg/plate)	Negative (±S9)	(Wild et al., 1983)	Review. Methods and results insufficiently documented.
(Benzyl disulphide [12.081])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538	3.6 mg/plate (3600 µg/plate)	Negative (±S9)	(Wild et al., 1983)	Review. Methods and results insufficiently documented.
Dibutyl disulphide [12.111]	Forward mutational assay	Mouse lymphoma cells	NR	Negative (-S9)	(Dooley et al., 1987)	Validity cannot be fully evaluated (only abstract provided).
Subgroup VIII – Thioesters						
(Methylthio 2-(acetyloxy)propionate [12.203])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, *E. Coli* WP2uvrA	0.156-5.0 mg/plate (156-5000 µg/plate	Negative (±S9)	(Watanabe and Morimoto, 1989a)	Acceptable quality.
(Methylthio 2-(propionyloxy) propionate [12.227])	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, *E. Coli* WP2uvrA	0.156 – 5.0 mg/plate (156 - 5000 µg/plate)	Negative (±S9)	(Watanabe and Morimoto, 1989b)	Acceptable quality.
Subgroup X – Sulfoxides/Sulphones and Sulphonates						
Methyl methane-thiosulfonate [12.159]	Ames test	*S. typhimurium* TA98, TA100, TA1535, TA1537, TA1538, TA2637	0.6 – 60 µg/plate	Negative (-S9)	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Ames test	*S. typhimurium* TA98, TA100,	2 – 600 µg/plate	Negative	(Dorange et al., 1983)	Test is not appropriate for

Table 7: Genotoxicity Data (*in vitro*) EFSA / FGE.08Rev5 (EFSA CEF Panel, 2012b)

Chemical Name [FL-no]	Test system	Test Object	Concentration	Result	Reference	Comments
		TA1535, TA1537, TA1538, TA2637		(+S9)		antimicrobial agents[(f)].
	Ames test	*S. typhimurium* TA98, TA100, TA2637	0.6 – 60 µg/plate	Negative (-S9)	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Ames test	*S. typhimurium* TA98, TA100, TA2637	0.6 – 200 µg/plate	Negative (+S9)	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Ames test	*S. typhimurium* TA98, TA100, TA2637	NR	Negative[(c)]	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Ames test	*S. typhimurium* TA98, TA100, TA2637	0.6 – 200 µg/plate	Negative[(d)]	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Yeast assay	*S. cerevisiae* Strain D7	1– 300 µg/ml	Negative (±S9)	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
	Yeast assay	*S. cerevisiae* Haploid strain N123	1– 100 µg/ml	Negative (±S9)	(Dorange et al., 1983)	Test is not appropriate for antimicrobial agents[(f)].
(Methylsulfinyl methane [12.175]) (synonym: dimethylsulfoxid, DMSO)	Ames test	*S. typhimurium* TA97, TA98, TA100	100000 – 300000 µg/plate	Negative (±S9)	(Brams et al., 1987)	Insufficient method (3 strains and 3 concentrations only).
	Ames test	*S. typhimurium* TA97, TA98, TA100, TA1535, TA1537	100 – 10000 µg/plate	Negative (±S9)	(Zeiger et al., 1992)	Acceptable quality.
	Ames test	*S. typhimurium* TA97, TA98, TA100, TA102, TA104, TA1535, TA1538, *E. Coli* WP2	0.1 – 0.4 ml/plate (100000 - 400000 µg/plate)	Negative (-S9)	(Hakura et al., 1993)	Good quality study.
	Ames test	*S. typhimurium* TA1537, TA2637, *E. Coli* WP2uvrA	0.1 – 0.4 ml/plate (100000 - 400000 µg/plate)	Positive (-S9)[(e)]	(Hakura et al., 1993)	Good quality study. Positive at high doses with reduced bacterial survival. Doses routinely used in Ames test were negative.

NR: Not reported.
(a): With and without metabolic activation at clearly cytotoxic concentrations.
(b): A statistically significant increase in the number of SCEs per chromosome was seen at 1350 µg/ml and the 450 µg/ml dose level in the presence of metabolic activation; but no significant increase was seen in the remaining dose levels, and no dose level showed a two fold increase in SCEs; therefore, t-butyl mercaptan is not considered to be mutagenic.
(c): With 100 µl/plate fecalase.
(d): With 100 µl/plate S9 metabolic activation and 100 µl/plate fecalase. Negative results reported after 2 days of incubation. Results for TA98 test strain were positive after 5 days of incubation
(e): Positive results obtained at doses where lethal toxicity was observed. Negative results obtained at doses routinely used in Ames test.

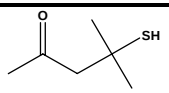
(f): Thiosulphonates in general, and methyl methane thiosulphonate in particular, are non-specific antimicrobial agents that are active at low concentrations on prokaryotic bacteria, as well as on yeast and other eukaryotic fungi. This was even pointed out by Dorange et al. (1983). Therefore bacterial test systems and yeast assays are not appropriate to evaluate genotoxicity of thiosulphonates.

Table 8: Genotoxicity Data (*in vivo*) EFSA / FGE.08Rev5 (EFSA CEF Panel, 2012b)

Chemical Name [FL-no]	Test System	Test Object	Route	Dose	Result	Reference	Comments
Subgroup I – Acyclic Sulphides							
(Diallyl sulphide [12.088])	*In vivo* mouse micronucleus test	Mouse	Gavage	0.33 – 0.67 mM/kg (38 – 77 mg/kg)[1]	Negative	(Marks et al., 1992)	Insufficient quality. Mixture of three substances was tested.
Subgroup III – Monothiols							
(2-Mercaptopropionic acid [12.039])	*In vivo* Basc test	*Drosophila*	Dietary route	10 mM (1061 µg/ml)	Negative	(Wild et al., 1983)	Limited quality (insufficiently documented). The article compiles results obtained with 76 substances in 3 test systems.
Subgroup V – Acyclic and cyclic Disulphides							
(Allyl disulphide [12.008])	*In vivo* mouse micronucleus test	Mouse	Gavage	0.33 – 0.67 mM/kg (48 – 98 mg/kg) (a)	Negative	(Marks et al., 1992)	Insufficient quality. Mixture of three substances was tested.
Subgroup VI – Acyclic Tri- and Polysulphides							
(Diallyl trisulphide [12.009])	*In vivo* mouse micronucleus test	Mouse	Gavage	0.33 – 0.67 mM/kg (59 - 120 mg/kg) (a)	Negative	(Marks et al., 1992)	Insufficient quality. Mixture of three substances was tested.
Subgroup X – Sulphoxides/Sulphones and Sulphonates							
Methyl methane-thiosulfonate [12.159]	*In vivo* genetic mutation	*Nicotiana tabacum* seeds	-	2 - 4 mg/ml (2000 - 4000 µg/ml)	Negative	(Dorange et al., 1983)	Obscure test system(b). This assay cannot be regarded as standard test.
	In vivo genetic mutation	*Nicotiana tabacum* seeds	-	50 – 400 µg/ml	Negative	(Dorange et al., 1983)	Obscure test system(b). This assay cannot be regarded as standard test.

(a): Study used a mixture of allyl sulphide, allyl disulphide and ally trisulphide in the respective ratio, 68:20:12.
(b): Heterozygotic seeds were used. After exposure, the seeds were blotted on filter paper and planted in earthenware pots in medium normally used for planting tobacco. The leaves were analysed for alterations indicating genotoxicity.

Table 9: Summary of Additional Genotoxicity Data on 2-methyl-4-oxopentane-2-thiol

FL-no JECFA-no	EU Register name JECFA name	Structural formula	End-point	Test system	Concentration	Results	Reference	Comments
12.169	2-Methyl-4-oxopentane-2-thiol		Reverse mutation	*S. typhimurium* TA98, TA100, TA1535, TA1537 and TA102	5, 15. 81, 50, 158.1, 500, 1581 and 5000 µg/plate[(a)]	Negative	(Mc Garry, 2012)	Valid GLP study, in compliance with OECD 471 Guideline
					156.3, 312.5, 625.0, 1250, 2500 and 5000 µg /plate[(a,b)]	Negative		

(a): In the absence and presence of S9-mix metabolic bioactivation.
(b): Assay modified with pre-incubation in presence of S9-mix.

SUMMARY OF SAFETY EVALUATIONS

Table 10: Summary of Safety Evaluation by the JECFA (JECFA, 2000; JECFA, 2004b)

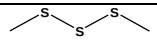
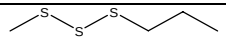
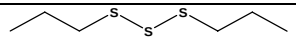
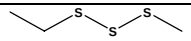
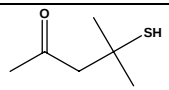
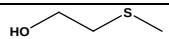
FL-no JECFA-no	EU Register name	Structural formula	EU MSDI [(a)] US MSDI (μg/*capita*/day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOAEL, genotoxicity)	EFSA conclusion on the material of commerce
12.013 582	Dimethyl trisulfide		1.1 0.02	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.020 584	Methyl propyl trisulfide		0.21 0.1	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.023 585	Dipropyl trisulfide		7.3 1	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.155 583	Methyl ethyl trisulfide		0.012 1	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.169 1293	2-Methyl-4-oxopentane-2-thiol		0.69 0.02	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.179 1297	2-(Methylthio)ethan-1-ol		0.97 0.9	Class I B3: Intake below threshold	d	No safety concern at the estimated level of intake based on the	No safety concern at the estimated level of intake based on the

Table 10: Summary of Safety Evaluation by the JECFA (JECFA, 2000; JECFA, 2004b)

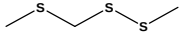
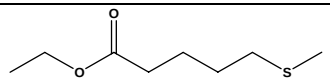
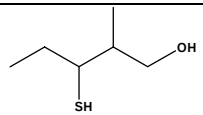
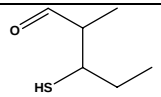
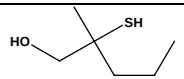
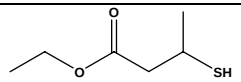
FL-no JECFA-no	EU Register name	Structural formula	EU MSDI [(a)] US MSDI (μg/*capita*/day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOAEL, genotoxicity)	EFSA conclusion on the material of commerce
				B4: Adequate NOAEL exists		MSDI approach.	MSDI approach.
12.198 1299	2,3,5-Trithiahexane		0.024 0.04	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.212 1298	Ethyl-5-(methylthio)valerate		1.8 2	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.238 1291	3-Mercapto-2-methylpentan-1-ol		0.85 0.7	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.239 1292	3-Mercapto-2-methylpentanal		2.6 4	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.241 1290	2-Mercapto-2-methylpentan-1-ol		0.012 4	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.255 1294	Ethyl 3-mercaptobutyrate		3.4 4	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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 10: Summary of Safety Evaluation by the JECFA (JECFA, 2000; JECFA, 2004b)

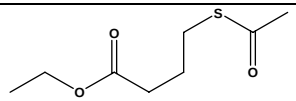
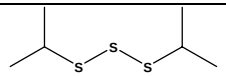
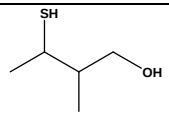
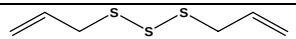
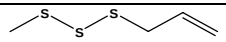
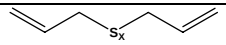
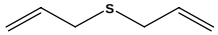
FL-no JECFA-no	EU Register name	Structural formula	EU MSDI [(a)] US MSDI (μg/*capita*/day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOAEL, genotoxicity)	EFSA conclusion on the material of commerce
12.257 1295	Ethyl 4-(acetylthio)-butyrate		3.4 4	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.280 1300	Diisopropyl trisulphide		0.24 0.007	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.291 1289	3-Mercapto-2-methyl-1-butanol		0.061 2	Class I B3: Intake below threshold B4: Adequate NOAEL exists	d	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.
12.009 587	Diallyl trisulfide		3.5 0.02	Class II B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.045 586	Methyl allyl trisulfide		0.012 0.9	Class II B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.074 588	Diallyl polysulfides	 X=2,3,4 or 5	1.2 0.02	Class II B3: Intake below threshold B4: Adequate NOAEL exists	d	Toxicity data required.	
12.088 458	Diallyl sulfide		3.5 0.4	Class II B3: Intake below	f	No safety concern at the estimated level of	No safety concern at the estimated level of

Table 10: Summary of Safety Evaluation by the JECFA (JECFA, 2000; JECFA, 2004b)

FL-no JECFA-no	EU Register name	Structural formula	EU MSDI [(a)] US MSDI (µg/*capita*/day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	EFSA conclusion on the named compound (Procedure steps, intake estimates, NOAEL, genotoxicity)	EFSA conclusion on the material of commerce
				threshold B4: No adequate NOAEL B5: Intake below 1.5 microg/person/day		intake based on the MSDI approach.	intake based on the MSDI approach.

(a): EU MSDI: Amount added to food as flavour in (kg / year) x 10^9 / (0.1 x population in Europe (= 375 x 10^6) x 0.6 x 365) = µg/*capita*/day.
(b): Thresholds of concern: Class I = 1800 µg/person/day, Class II = 540 µg/person/day, Class III = 90 µg/person/day.
(c): Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.
(d): No safety concern based on intake calculated by the MSDI approach of the named compound.
(e): Data must be available on the substance or closely related substances to perform a safety evaluation.
(f): Cleared by JECFA as intake below 1.5 µg/person/day.

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

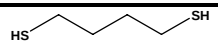
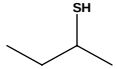
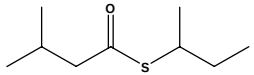
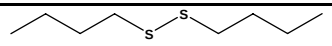
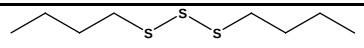
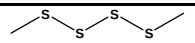
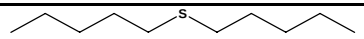
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	Outcome on the material of commerce [(f), (g) or (h)]	Evaluation remarks
12.103	Butane-1,4-dithiol		0.3	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.104	Butane-2-thiol		0.18	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.106	S-2-Butyl 3-methylbutanethioate		0.8	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.111	Dibutyl disulfide		0.37	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.112	Dibutyl trisulfide		0.12	Class I B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.116	Dimethyl tetrasulfide		0.016	Class I B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.117	Dipentyl sulfide		0.0037	Class I B3: Intake below threshold,	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

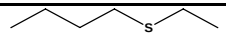
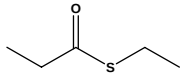
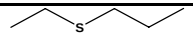
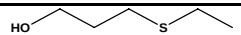
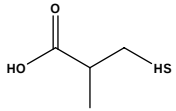
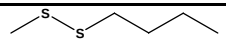
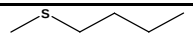
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	Outcome on the material of commerce [(f), (g) or (h)]	Evaluation remarks
				B4: Adequate NOAEL exists			
12.124	Ethyl butyl sulfide		0.037	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.125	Ethyl propanethioate		0.012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.127	Ethyl propyl sulfide		0.085	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.129	3-(Ethylthio)propan-1-ol		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.135	3-Mercapto-2-methylpropionic acid		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.151	Methyl butyl disulfide		0.0061	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.152	Methyl butyl sulfide		0.0024	Class I B3: Intake below threshold,	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

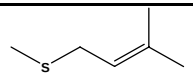
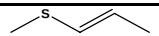
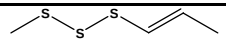
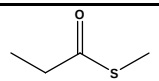
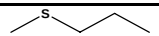
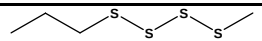
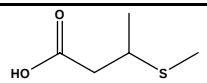
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	Outcome on the material of commerce [(f), (g) or (h)]	Evaluation remarks
				B4: Adequate NOAEL exists			
12.158	Methyl isoprenyl sulfide		0.0012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.163	Methyl prop-1-enyl sulfide		0.0097	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.164	Methyl prop-1-enyl trisulfide		0.0061	Class I B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.165	S-Methyl propanethioate		0.012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.166	Methyl propyl sulfide		0.0024	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.167	Methyl propyl tetrasulfide		0.0037	Class I B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.178	3-(Methylthio)butyric acid		0.12	Class I B3: Intake below threshold,	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

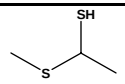
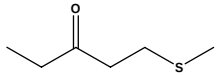
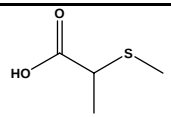
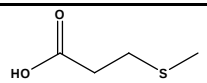
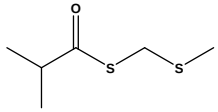
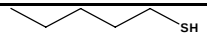
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	Outcome on the material of commerce [(f), (g) or (h)]	Evaluation remarks
				B4: Adequate NOAEL exists			
12.180	1-(Methylthio)ethane-1-thiol		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.181	1-(Methylthio)pentan-3-one		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.182	2-(Methylthio)propionic acid		0.011	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.183	3-(Methylthio)propionic acid		0.21	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.189	S-(Methylthiomethyl) 2-methylpropanethioate		0.061	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.191	Pentane-1-thiol		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

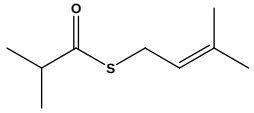
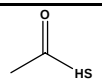
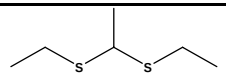
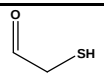
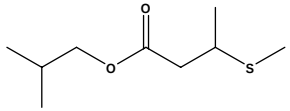
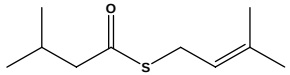
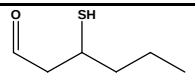
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
12.196	S-Prenyl thioisobutyrate		0.012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.199	Ethanethioic acid		0.0012	Class I B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.200	1,1-bis(Ethylthio)-ethane		0.0012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.205	Mercaptoacetaldehyde		0.011	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.214	Isobutyl-3-(methylthio)butyrate		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.221	S-Prenyl thioisopentanoate		0.012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.250	3-Mercaptohexanal		0.012	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

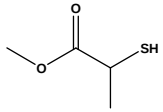
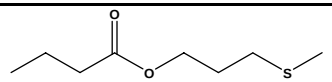
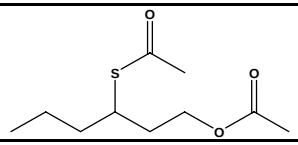
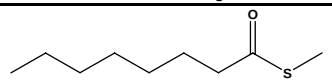
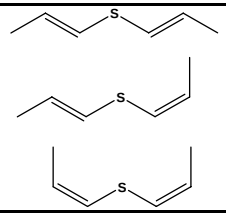
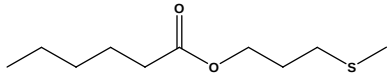
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
12.266	Methyl-2-mercaptopropionate		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	h	
12.277	3-(Methylthio)propyl butyrate		6.1	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.278	3-Acetyl-mercaptohexyl acetate		1.2	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.282	(S)-Methyl octanethioate		0.24	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	g	
12.298	Di-(1-propenyl)-sulfid (mixture)		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.299	3-(Methylthio)propyl hexanoate		0.061	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

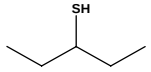
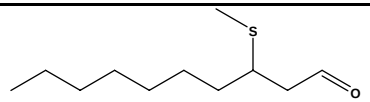
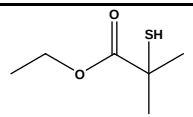
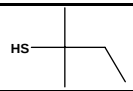
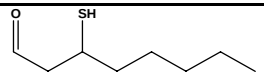
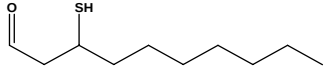
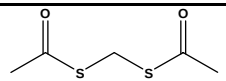
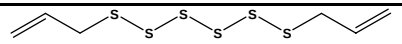
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
12.303	3-Pentanethiol		0.03	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.306	3-(Methylthio)-decanal		0.12	Class I B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.304	Ethyl-2-mercapto-2-methyl propanoate		0.012	Class I No evaluation			Pending update, as new genotoxicity data have become available.
12.172	2-Methylbutane-2-thiol		0.15	Class I No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
12.174	2-Methylpropane-2-thiol		0.0012	Class I No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
12.268	3-Mercaptooctanal			Class I No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
12.269	3-Mercaptodecanal			Class I No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
12.271	Methanedithiol diacetate			Class I No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
12.093	Diallyl hexasulfide		0.011	Class II B3: Intake below threshold, B4: No adequate	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

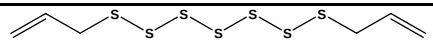
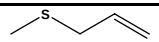
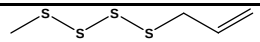
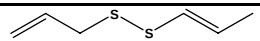
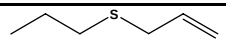
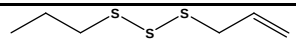
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [(d) or (e)]	Outcome on the material of commerce [(f), (g) or (h)]	Evaluation remarks
				NOAEL			
12.094	Diallyl heptasulfide		0.011	Class II B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.096	Allyl methyl sulfide		0.99	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.097	Allyl methyl tetrasulfide		0.012	Class II B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.098	Allyl prop-1-enyl disulfide		0.17	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.099	Allyl propyl sulfide		1.6	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.100	Allyl propyl trisulfide		0.12	Class II B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

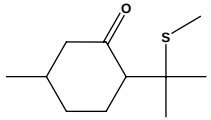
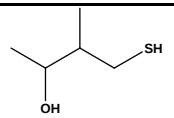
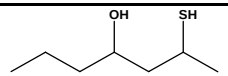
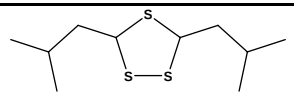
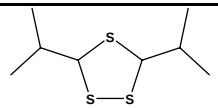
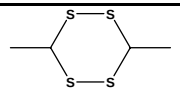
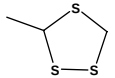
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
12.177	8-(Methylthio)-p-menthan-3-one		0.37	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.302	2-Butanol, 4-mercapto-3-methyl		0.061	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.305	2-Mercapto-4-heptanol		0.12	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.047	3,5-Di-isobutyl-1,2,4-trithiolane		0.024	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.048	3,5-Di-isopropyl-1,2,4-trithiolane		0.0061	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.056	3,6-Dimethyl-1,2,4,5-tetrathiane		0.0024	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.083	3-Methyl-1,2,4-trithiolane		0.0024	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

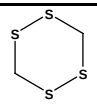
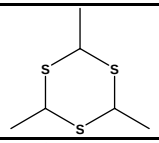
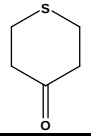
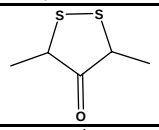
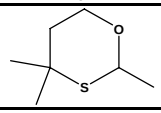
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
15.102	Tetrahydrothiophene		0.024	Class II B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		
15.103	1,2,4,5-Tetrathiane		0.073	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.110	2,4,6-Trimethyl-1,3,5-trithiane		0.0061	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.111	1,2,4-Trithiolane		2.4	Class II B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.125	4-Tetrahydrothiopyranone		0.12	Class II B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		Substance no longer supported by Industry (DG SANCO, 2012).
12.295	3,5-Dimethyl-1,2-dithiolane-4-one			Class II No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).
16.057	2,4,4-Trimethyl-1,3-oxathiane		0.0012	Class II No evaluation			Substance no longer supported by Industry (DG SANCO, 2012).

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

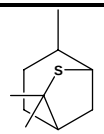
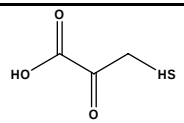
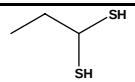
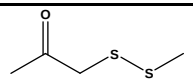
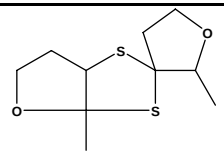
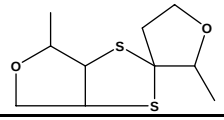
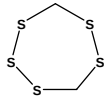
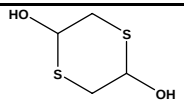
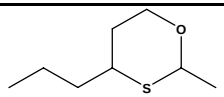
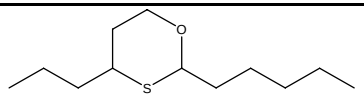
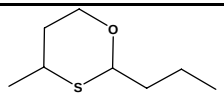
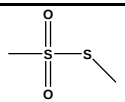
FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
12.120	2,8-Epithio-p-menthane		3.7	Class III B3: Intake below threshold, B4: No adequate NOAEL	Additional data required		No longer supported by Industry (DG SANCO, 2013).
12.136	3-Mercapto-2-oxopropionic acid		0.24	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.300	1,1-Propanedithiol		0.12	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.301	Methyl-2-oxo-propyl disulfide		0.061	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.007	Spiro(2,4-dithia-1-methyl-8-oxabicyclo[3.3.0]octane -3,3'-(1'-oxa-2'-methyl)-cyclopentane) and Spiro(2,4-dithia-6-methyl-7-oxabicyclo[3.3.0]octane -3,3'-(1'-oxa-2'-methyl)-cyclopentane)	 **I**  **II**	6.1	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	

Table 11: Summary of Safety Evaluation by the EFSA (FGE.08Rev5) (EFSA CEF Panel, 2012b)

FL-no	EU Register name	Structural formula	MSDI [(a)] (µg/capita/ day)	Class [(b)] Evaluation procedure path [(c)]	Outcome on the named compound [[(d)] or [(e)]]	Outcome on the material of commerce [[(f)], [(g)] or [(h)]]	Evaluation remarks
15.081	Lenthionine		0.012	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
15.134	2,5-Dihydroxy-1,4-dithiane		6.1	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
16.062	trans-2-Methyl-4-propyl-1,3-oxathiane		1.0	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
16.114	2-Pentyl-4-propyl-1,3-oxathiane		0.12	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
16.122	4-Methyl, 2-propyl, 1-3-oxathiane		0.24	Class III B3: Intake below threshold, B4: Adequate NOAEL exists	d	f	
12.159	Methyl methanethiosulfonate		0.061	Class III No evaluation			No longer supported by Industry (DG SANCO, 2013).

a): EU MSDI: Amount added to food as flavour in (kg / year) x 10^9 / (0.1 x population in Europe (= 375 x 10^6) x 0.6 x 365) = µg/*capita*/day.
(b): Thresholds of concern: Class I = 1800 µg/person/day, Class II = 540 µg/person/day, Class III = 90 µg/person/day.
(c): Procedure path A substances can be predicted to be metabolised to innocuous products. Procedure path B substances cannot.
(d): No safety concern based on intake calculated by the MSDI approach of the named compound.
(e): Data must be available on the substance or closely related substances to perform a safety evaluation.
(f): No safety concern at the estimated level of intake of the material of commerce meeting the specification requirement (based on intake calculated by the MSDI approach).

(g): 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.
(h): No conclusion can be drawn due to lack of information on the purity of the material of commerce.

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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
EFFA	European Flavour and Fragrance Association
EFSA	The 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 practice
HPRT	Hypoxanthine Phosphoribosyl transferase
ID	Identity
IP	Intraperitoneal
IR	Infrared spectroscopy
JECFA	The Joint FAO/WHO Expert Committee on Food Additives
MNBN	Micronucleated Binucleate cells
MS	Mass spectrometry
MSDI	Maximised Survey-derived Daily Intake
mTAMDI	Modified Theoretical Added Maximum Daily Intake
NCE	Normochromatic erythrocyte
NMR	Nuclear Magnetic Resonance
No	Number

NOAEL	No Observed Adverse Effect Level
NTP	National Toxicology Program
OECD	Organization for Economic Cooperation and Development
PCE	Polychromatic erythrocyte
RI	Replication Index
SCE	Sister chromatic exchange
SCF	Scientific Committee on Food
UDS	Unscheduled DNA Synthesis
WHO	World Health Organisation