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Re-evaluation of fatty acids (E 570) as a food additive

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Abstract

The EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS) provides a scientific opinion re-evaluating the safety of fatty acids (E 570) when used as a food additive. The food additive includes caprylic- (C8), capric- (C10), lauric- (C12), myristic- (C14), palmitic- (C16), stearic- (C18) and oleic acid (C18:1), present alone or in combination. In 1991, the Scientific Committee on Food (SCF) established a group acceptable daily intake (ADI) 'not specified' for the fatty acids (myristic, stearic, palmitic and oleic acid). The fatty acids (E 570) are absorbed in the same way as the free fatty acids from the regular diet. They show low acute toxicity. The available studies on subchronic toxicity were limited but there was no evidence for toxic effects at doses up to 10% in the diet (equivalent to 9,000 mg lauric acid/kg body weight (bw) per day). The Panel considered that the fatty acids (E 570) did not raise a concern for genotoxicity. Data on chronic toxicity, reproductive toxicity and developmental toxicity were too limited to reach a conclusion on these endpoints. The Panel noted that the contribution of fatty acids (E 570) represented on average only 1% of the overall exposure to saturated fatty acids from all dietary sources (food additive and regular diet). Based on the approach described in the conceptual framework for the risk assessment of certain food additives re-evaluated under Commission Regulation (EU) No 257/2010 and taking into account the considerations mentioned above, the Panel concluded that the food additive fatty acids (E 570) was of no safety concern at the reported uses and use levels.

Keywords: food additive, caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, oleic acid

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Summary

Following a request from the European Commission, the EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS) was asked to re-evaluate the safety of fatty acids (E 570) when used as a food additive. The Panel was not provided with a newly submitted dossier and based its evaluation on previous evaluations and reviews, additional literature that became available since then and the data available following a public call for data. The Panel noted that not all original studies on which previous evaluations were based were available for this re-evaluation.

Fatty acids (E 570) is authorised as a food additive in the European Union (EU) in accordance with Annex II and Annex III to Regulation (EC) No 1333/2008 on food additives and specific purity criteria have been defined in the Commission Regulation (EU) No 231/2012. The EU Scientific Committee on Food in 1991 established a group acceptable daily intake (ADI) 'not specified' for four fatty acids (myristic-, stearic-, palmitic- and oleic acid). The Panel noted that that caprylic acid, capric acid and lauric acid, which can also be present in the food additive E 570, were not included within the fatty acids considered in the Scientific Committee on Food (SCF) evaluation in 1991.

Caprylic acid (FL-No. 08.010), capric acid (FL-No. 08.011), lauric acid (FL-No. 08.012), myristic acid (FL-No. 08.016), palmitic acid (FL-No. 08.014), stearic acid (FL-No. 08.015) and oleic acid (FL-No. 08.013) are included in the Union list of flavourings (Commission Implementing Regulation (EU) No 872/2012). In 1999, the Joint FAO/WHO Expert Committee on Food Additives (JECFA) reviewed caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid and stearic acid as flavouring substances (JECFA, 1999) as well as oleic acid (JECFA, 2000) and considered them as 'no safety concern at current levels of intake when used as a flavouring agent'. The EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids (CEF) considered oleic acid (FL-No. 08.013) as a supporting substance for the assessment of other flavouring substances.

Caprylic acid (PM Ref. 14320 and 41960), capric acid (PM Ref. 15095 and 45940), lauric acid (PM Ref. 19470 and 63280), myristic acid (PM Ref. 22350 and 67891), palmitic acid (PM Ref. 22780 and 70400), stearic acid (PM Ref. 24550 and 89040) and oleic acid (PM Ref. 22763 and 69040) are included in the Union list of authorised substances that may be intentionally used in the manufacture of plastic layers in plastic materials and articles (Annex I to Commission Regulation (EU) No 10/2011). Furthermore, they are permitted in cosmetic products (European Commission database-CosIng). Caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid and oleic acid are included in the European Union Register of feed additives (Regulation (EC) No 1831/2003).

The EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA) proposed dietary reference values (DRVs) for fats (EFSA NDA Panel, 2010). As regards the specific fatty acids (E 570) that were the subject of the present opinion the proposal was as follows: intake of saturated fatty acid (caprylic-, capric-, lauric-, myristic-, palmitic- and stearic acid) as low as possible; no DRV was set for *cis*-monounsaturated fatty acids (oleic acid).

The European Food Safety Authority (EFSA) also peer reviewed the initial risk assessments carried out by the competent authority of the rapporteur Member State Ireland, for the pesticide active substance fatty acids C7–C18 (approved under Regulation (EC) No 1107/2009 as Fatty acids C7–C20) (EFSA, 2013). It was concluded that exposure to fatty acids derived from the use as plant protection products would be considered of low toxicological concern and no reference values would be needed if the different groups of fatty acids could be considered of food grade quality.

The food additive (E 570) is considered in Commission Regulation (EU) No 231/2012 as a group of substances composed of six linear saturated fatty acids (caprylic-, capric-, lauric-, myristic-, palmitic- and stearic acid) and one *cis*-monounsaturated fatty acid (oleic acid), but that no indication is given as regards the level at which an individual acid may be present in the group, except that the total percentage of the fatty acids should not be less than 98%.

Caprylic-, capric-, lauric-, palmitic-, myristic-, stearic- and oleic acid, like other fatty acids are readily and extensively absorbed from the gastrointestinal tract and are further metabolised to carbon dioxide, which is finally excreted via exhalation.

Caprylic-, capric-, lauric-, palmitic-, myristic-, stearic- and oleic acid have a low acute toxicity.

From the available feeding studies on subchronic toxicity, although limited, the Panel considered that there was no evidence for toxic effects of fatty acids at dose levels up to 10% in the diet (equivalent to 9,000 mg lauric acid/kg body weight (bw) per day). However, these studies were not conducted according to the current guidelines.

Despite the absence of specific studies for the evaluation of structural and numerical chromosomal aberrations, the Panel considered the available genotoxicity data did not raise a concern for

genotoxicity for caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid used as a food additive.

Only limited data on chronic toxicity were available and because of the limitations of these studies, no valuable conclusion could be drawn. Insufficient data were available on reproductive toxicity of fatty acids and the limited data on developmental toxicity were available only for caprylic acid.

From the available human data available, the Panel noted that ingestion of fatty acids may have some adverse health effects. However, in these studies, the fat intake was high compared to the intake from the use of fatty acids (E 570) as a food additive. Therefore, these studies were not relevant for the safety assessment of the food additive E 570.

Although some of the fatty acids which are included in the food additive E 570, have been reported to be recognised by specific cellular receptors, the Panel considered it outside the scope of this evaluation to review the extensive literature available about the effects of free fatty acids the consequence of binding their specific receptors. The Panel considered that although it is relevant to consider the possible effects resulting from this binding, this should be done in the perspective of a comparison between the exposure resulting from the use of free fatty acids used as a food additive and their intake from the regular diet. Given that the amount of fatty acids in food from all sources present in the regular diet markedly exceeds the intake of free fatty acids resulting from their use as a food additive, the Panel considered that receptor binding with fatty acids derived from their use as food additive was unlikely to give rise to adverse effect(s)

Only a limited number of usage levels (8 out of 67 food categories in which E 570 is authorised) was available for the exposure assessment, which might indicate a limited use of E 570 as a food additive in Europe. The Panel noted that the information from the Mintel Global New Products Database (GNPD) supported the observation that fatty acids (caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid) are apparently not used in all food categories in which the food additive E 570 is authorised.

The Panel noted that the dietary exposure to E 570 was low (on average 1%) compared to the total daily exposure to saturated fatty acids via the regular diet, either free or incorporated into glycerides and phospholipids. The intake of fatty acids (E 570) as well as that of saturated fatty acids via the regular diet, were far below 9,000 mg/kg bw per day (for lauric acid) at which no adverse effects were observed in subchronic toxicity studies. According to the EU specifications, E 570 may contain only one particular fatty acid or a mixture of the seven fatty acids which are authorised within the food additive. Accordingly, the exposure assessment referred to E 570 and not to a specific fatty acid.

According to the conceptual framework for the risk assessment of certain food additives re-evaluated under Commission Regulation (EU) No 257/2010 (EFSA ANS Panel, 2014) and given that:

- the safety assessment carried out by the Panel was limited to the use and use levels received from industry in eight food categories out of 67 food categories in which fatty acids (E 570) is authorised;
- fatty acids used as a food additive (E 570) were absorbed in the same way as the free fatty acids from the regular diet;
- fatty acids used as a food additive (E 570) were metabolised in the same way as fatty acids when derived from lipid molecules present in the regular diet;
- the toxicity database was limited, however, no adverse effects were observed in subchronic toxicity studies up to 10% in the diet (equivalent to 9,000 mg lauric acid/kg bw per day);
- there was no genotoxicity concern for these fatty acids;
- the contribution of fatty acids (E 570) represented on average only 1% of the overall exposure to saturated fatty acids from all dietary sources (food additive and regular diet);

the Panel concluded that the food additive fatty acids (E 570) was of no safety concern at the reported uses and use levels.

The Panel recommended that:

- the European Commission considers lowering the current limits for toxic elements (arsenic, lead and mercury) in the EU specifications for fatty acids (E 570) in order to ensure that fatty acids (E 570) as a food additive will not be a significant source of exposure to those toxic elements in food;
- since only data for eight out of the 67 food categories in which fatty acids (E 570) is authorised were available, more information on uses and use levels should be made available to the Panel in order to perform a more accurate exposure assessment.

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1. Introduction

The present opinion deals with the re-evaluation of the safety of 'fatty acids (E 570)' when used as a food additive.

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

1.1.1. Background

Regulation (EC) No 1333/2008[1] of the European Parliament and of the Council on food additives requires that food additives are subject to a safety evaluation by the European Food Safety Authority (EFSA) before they are permitted for use in the European Union (EU). In addition, it is foreseen that food additives must be kept under continuous observation and must be re-evaluated by EFSA.

For this purpose, a programme for the re-evaluation of food additives that were already permitted in the EU before 20 January 2009 has been set up under Regulation (EU) No 257/2010[2]. This Regulation also foresees that food additives are re-evaluated whenever necessary in light of changing conditions of use and new scientific information. For efficiency and practical purposes, the re-evaluation should, as far as possible, be conducted by group of food additives according to the main functional class to which they belong.

The order of priorities for the re-evaluation of the currently approved food additives should be set on the basis of the following criteria: the time since the last evaluation of a food additive by the Scientific Committee on Food (SCF) or by EFSA, the availability of new scientific evidence, the extent of use of a food additive in food and the human exposure to the food additive taking also into account the outcome of the Report from the Commission on Dietary Food Additive Intake in the EU[3] of 2001. The report 'Food additives in Europe 2000[4]' submitted by the Nordic Council of Ministers to the Commission, provides additional information for the prioritisation of additives for re-evaluation. As colours were among the first additives to be evaluated, these food additives should be re-evaluated with a highest priority.

In 2003, the Commission already requested EFSA to start a systematic re-evaluation of authorised food additives. However, as a result of adoption of Regulation (EU) No 257/2010 the 2003 Terms of References are replaced by those below.

1.1.2. Terms of Reference

The Commission asks EFSA to re-evaluate the safety of food additives already permitted in the Union before 2009 and to issue scientific opinions on these additives, taking especially into account the priorities, procedures and deadlines that are enshrined in the Regulation (EU) No 257/2010 of 25 March 2010 setting up a programme for the re-evaluation of approved food additives in accordance with the Regulation (EC) No 1333/2008 of the European Parliament and of the Council on food additives.

1.2. Information on existing authorisations and evaluations

Fatty acids (E 570) is authorised as a food additive in the EU in accordance with Annex II and Annex III to Regulation (EC) No 1333/2008 on food additives and specific purity criteria have been defined in the Commission Regulation (EU) No 231/2012[5].

The EU SCF established a group acceptable daily intake (ADI) 'not specified' for the fatty acids (myristic-, stearic-, palmitic- and oleic acid) (SCF, 1991). The Panel noted that that caprylic acid, capric acid and lauric acid, which can also be present in the food additive E 570, were not included within the fatty acids considered in the SCF evaluation in 1991.

[1] Regulation (EC) No 1333/2008 of the European Parliament and of the Council of 16 December 2008 on food additives. OJ L 354, 31.12.2008

[2] Commission Regulation (EU) No 257/2010 of 25 March 2010 setting up a programme for the re-evaluation of approved food additives in accordance with Regulation (EC) No 1333/2008 of the European Parliament and of the Council on food additives. OJ L 80, 26.3.2010, p. 19–27.

[3] COM(2001) 542 final.

[4] Food Additives in Europe 2000, Status of safety assessments of food additives presently permitted in the EU, Nordic Council of Ministers, TemaNord 2002:560.

[5] Commission Regulation (EU) No 231/2012 of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council. OJ L 83, 22.3.2012, p. 1–295.

Caprylic acid (FL-No. 08.010), capric acid (FL-No. 08.011), lauric acid (FL-No. 08.012), myristic acid (FL-No. 08.016), palmitic acid (FL-No. 08.014), stearic acid (FL-No. 08.015) and oleic acid (FL-No. 08.013) are included in the Union list of flavourings (Commission Implementing Regulation (EU) No 872/2012[6]). The Joint FAO/WHO Expert Committee on Food Additives (JECFA) reviewed caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid and stearic acid as flavouring substances (JECFA, 1999) as well as oleic acid (JECFA, 2000) and considered them as 'no safety concern at current levels of intake when used as a flavouring agent'. EFSA considered oleic acid (FL-No. 08.013) as a supporting substance for the assessment of other flavouring substances (EFSA CEF Panel, 2013).

Caprylic acid (PM Ref. 14320 and 41960), capric acid (PM Ref. 15095 and 45940), lauric acid (PM Ref. 19470 and 63280), myristic acid (PM Ref. 22350 and 67891), palmitic acid (PM Ref. 22780 and 70400), stearic acid (PM Ref. 24550 and 89040) and oleic acid (PM Ref. 22763 and 69040) are included in the Union list of authorised substances that may be intentionally used in the manufacture of plastic layers in plastic materials and articles (Annex I to Commission Regulation (EU) No 10/2011[7]). Furthermore, they are permitted in cosmetic products (European Commission database-CosIng[8]). Caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid and oleic acid are included in the European Union Register[9] of feed additives (Regulation (EC) No 1831/2003[10]).

The EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA) proposed dietary reference values (DRVs) for fats (EFSA NDA Panel, 2010). As regards the specific fatty acids (E 570) that are the subject of the present opinion the proposal was as follows: intake of saturated fatty acid (caprylic-, capric-, lauric-, myristic-, palmitic- and stearic acid) as low as possible; no DRV was set for cis-monounsaturated fatty acids (oleic acid).

EFSA also peer reviewed the initial risk assessments carried out by the competent authority of the rapporteur Member State Ireland, for the pesticide active substance fatty acids C7–C18 (approved under Regulation (EC) No 1107/2009[11] as fatty acids C7–C20) (EFSA, 2013). It was concluded that exposure to fatty acids derived from the use as plant protection products would be considered of low toxicological concern and no reference values would be needed if the different groups of fatty acids could be considered of food grade quality.

The JECFA's specifications for octanoic acid (a synonym of caprylic acid) were revised to include infrared test conditions and the reference spectrum (JECFA, 2016).

2. Data and methodologies

2.1. Data

The Panel was not provided with a newly submitted dossier. EFSA launched public calls for data,[12,13] to collect relevant information from interested parties.

The Panel based its assessment on information submitted to EFSA following the public calls for data, information from previous evaluations and additional available literature up to February 2017. Attempts were made at retrieving relevant original study reports on which previous evaluations or reviews were based, however not always these were available to the Panel.

The EFSA Comprehensive European Food Consumption Database (Comprehensive Database[14]) was used to estimate the dietary exposure.

[6] Commission implementing Regulation (EU) No 872/2012 of 1 October 2012 adopting a 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.

[7] Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food. OJ L 12, 15.1.2011, p. 1–89.

[8] Available online: http://ec.europa.eu/consumers/cosmetics/cosing/index.cfm?fuseaction=search.simple

[9] Available online: http://ec.europa.eu/food/food/animalnutrition/feedadditives/comm_register_feed_additives_1831-03.pdf

[10] Regulation (EC) No 1831/2003 of the European Parliament and the Council of 22 September 2003 on additives for use in animal nutrition. OJ L 268, 18.10.2003, 29–43.

[11] Regulation (EC) No 1107/2009 of the European Parliament and the Council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC. OJ L 309, 24.11.2009, 1–50.

[12] Call for scientific data on miscellaneous food additives permitted in the EU and belonging to several functional classes. Published: 14 February 2012. Available from: http://www.efsa.europa.eu/en/dataclosed/call/120215

[13] Call for food additives usage level and/or concentration data in food and beverages intended for human consumption. Published: 12 October 2015. Available from: http://www.efsa.europa.eu/en/data/call/151012

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

The Mintel's Global New Products Database (GNPD) is an online resource listing food products and compulsory ingredient information that should be included in labelling. This database was used to verify the use of fatty acids (E 570) in food products.

2.2. Methodologies

The assessment was conducted in line with the principles described in the EFSA Guidance on transparency in the scientific aspects of risk assessment (EFSA Scientific Committee, 2009) and following the relevant existing Guidances from the EFSA Scientific Committee.

The EFSA Scientific Panel on Food Additives and Nutrient Sources added to Food (ANS) assessed the safety of fatty acids (E 570) as a food additive in line with the principles laid down in Regulation (EU) 257/2010 and in the relevant guidance documents: Guidance on submission for food additive evaluations by the SCF (2001).

When the test substance was administered in the feed or in the drinking water, but doses were not explicitly reported by the authors as mg/kg body weight (bw) per day based on actual feed or water consumption, the daily intake was calculated by the Panel using the relevant default values as indicated in the EFSA Scientific Committee guidance document (EFSA Scientific Committee, 2012) for studies in rodents or, in the case of other animal species, by JECFA (2000). In these cases, the daily intake is defined in the text as 'equivalent to'.

Dietary exposure to fatty acids (E 570) from their use as a food additive was estimated combining the food consumption data available within the EFSA Comprehensive European Food Consumption Database with the reported use levels submitted to EFSA following a call for data. Different scenarios were used to calculate exposure (see Section 3.3.1). Uncertainties on the exposure assessment were identified and discussed.

In the context of this re-evaluation, the Panel followed the conceptual framework for the risk assessment of certain food additives re-evaluated under Commission Regulation (EC) No 257/2010 (EFSA ANS Panel, 2014).

3. Assessment

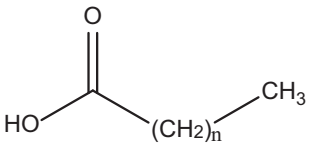
3.1. Technical data

3.1.1. Identity of the substance

According to Commission Regulation (EU) No 231/2012, the food additive fatty acids (E 570) includes linear fatty acids: caprylic acid (C8), capric acid (C10), lauric acid (C12), myristic acid (C14), palmitic acid (C16), stearic acid (C18) and oleic acid (C18:1), which according to the European Commission can be present alone or in combination in the food additive E 570. The chemical names, molecular formulas, CAS Registry Numbers, molecular weights and synonyms for the seven specified fatty acids covered by E 570 are listed in Table 1.

The CAS Registry Number 90990-08-2 and the EC (EINECS) number 292-769-6 have been assigned to the 'Fatty acids C-8-18' (EC inventory, online).

The general structural formula of the saturated fatty acids included in E 570 is depicted in Figure 1.



Caprylic acid n = 6
Capric acid n = 8
Lauric acid n = 10
Myristic acid n = 12
Palmitic acid n = 14
Stearic acid n = 16

Figure 1: General structural formula of the saturated fatty acids included in E 570

The structural formula of oleic acid (9-*cis*-octadecenoic acid) is given in Figure 2.

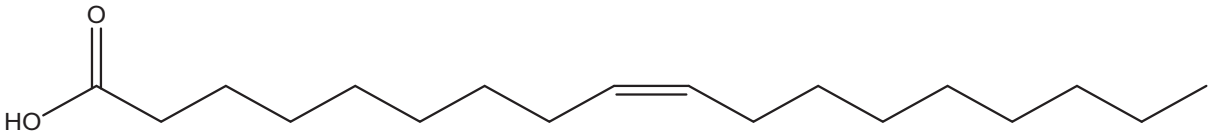


Figure 2: Structural formula oleic acid

Table 1: Identity of the fatty acids included in E 570

Common name	Chemical name	Molecular formula	Molecular weight (g/mol)	CAS registry number/EC number	Synonyms
Caprylic acid	Octanoic acid	$C_8H_{16}O_2$	144.21	124-07-2/204-677-5	1-Heptanecarboxylic acid; Caprylic acid; Octylic acid; *n*-Caprylic acid; *n*-Octanoic acid; *n*-Octoic acid; *n*-Octylic acid
Capric acid	Decanoic acid	$C_{10}H_{20}O_2$	172.26	334-48-5/206-376-4	1-Decanoic acid; 1-Nonanecarboxylic acid; Capric acid; Caprinic acid; Caprynic acid; Decoic acid; Decylic acid; *n*-Capric acid; *n*-Decanoic acid; *n*-Decoic acid; *n*-Decylic acid
Lauric acid	Dodecanoic acid	$C_{12}H_{24}O_2$	200.32	143-07-7/205-582-1	Lauric acid (8CI); 1-Dodecanoic acid; 1-Undecanecarboxylic acid; Dodecylic acid; Laurostearic acid; Vulvic acid; *n*-Dodecanoic acid
Myristic acid	Tetradecanoic acid	$C_{14}H_{28}O_2$	228.37	544-63-8/208-875-2	Myristic acid (8CI); 1-Tetradecanoic acid; 1-Tridecanecarboxylic acid; *n*-Tetradecan-1-oic acid; *n*-Tetradecanoic acid; *n*-Tetradecoic acid
Palmitic acid	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.42	57-10-3/200-312-9	Palmitic acid (7CI,8CI); 1-Pentadecanecarboxylic acid; Cetylic acid; Palmitinic acid; Pentadecanecarboxylic acid; *n*-Hexadecanoic acid; *n*-Hexadecoic acid
Stearic acid	Octadecanoic acid	$C_{18}H_{36}O_2$	284.48	57-11-4/200-313-4	Stearic acid (8CI); 1-Heptadecanecarboxylic acid; 1-Octadecanoic acid; Rubber Grade Stearic Acid; *n*-Octadecanoic acid
Oleic acid	*cis*-9-Octadecenoic acid	$C_{18}H_{34}O_2$	282.46	112-80-1/204-007-1	9-Octadecenoic acid (*Z*)-; Oleic acid (8CI); 9-Octadecenoic acid, (*Z*)-; 9-*cis*-Octadecenoic acid; 9*Z*-Octadecenoic acid; *Z*-9-Octadecenoic acid; *cis*-9-Octadecenoic acid; *cis*-Oleic acid; *cis*-Δ9-Octadecenoic acid; Δ9-*cis*-Octadecenoic acid; Δ9-*cis*-Oleic acid

CAS: Chemical Abstracts Service.

3.1.2. Specifications

The specifications for 'fatty acids' (E 570) as a food additive according to Commission Regulation (EU) No 231/2012 are given in Table 2. The Panel noted that JECFA has not prepared specifications for 'Fatty acids' (INS No 570).

Table 2: Specifications for fatty acids (E 570) as a food additive according to Commission Regulation (EU) No 231/2012

	Commission Regulation (EU) No 231/2012
Definition	Linear fatty acids, caprylic acid (C8), capric acid (C10), lauric acid (C12), myristic acid (C14), palmitic acid (C16), stearic acid (C18), oleic acid (C18:1)
Assay	Not less than 98% by chromatography
Description	A colourless liquid or white solid obtained from oils and fats
Identification	Individual fatty acids can be identified by acid value, iodine value, gas chromatography
Purity	
Residue on ignition	Not more than 0.1%
Unsaponifiable matter	Not more than 1.5%
Water content	Not more than 0.2% (Karl Fischer method)
Arsenic	Not more than 3 mg/kg
Lead	Not more than 1 mg/kg
Mercury	Not more than 1 mg/kg

The Panel noted that, according to the EU specifications, impurities of the toxic elements arsenic, lead and mercury are accepted up concentrations of 3, 1, and 1 mg/kg, respectively. Contamination at those levels could have a significant impact on the exposure to these metals, for which the exposure already are close to the health based guidance values or benchmark doses (lower confidence limits) established by EFSA (EFSA CONTAM Panel, 2009, 2010, 2012a,b,c, 2014).

3.1.3. Manufacturing process

No detailed information on the manufacturing of the food grade substances was identified.

According to CIR (1987) and Anneken et al. (2012), the fatty acids – caprylic-, capric-, lauric-, myristic-, palmitic-, stearic and oleic acid – are usually produced by hydrolysis of common animal and vegetable fats and oils and later fractionation of the individual fatty acids. Caprylic- and capric acid may be produced from coconut oil by saponification and fractional distillation of the coconut oil triglycerides (Lewis, 2007a,b). Sources of myristic acid are coconut oil and other vegetable oils; the substance also occurs in sperm whale oil (Lewis, 2007c). Palmitic acid is produced from palm oil and other natural fats (Lewis, 2007d). Sources of stearic acid are animal fats and oils, as well as some vegetable oils (Lewis, 2007e; Merck Index, 2013a). Oleic acid may be prepared from olive oil (Merck Index, 2013b).

3.1.4. Methods of analysis in food

A number of methods for the analysis of individual fatty acids in food have been described in literature. An overview of the available methods was given not only by Mukherjee and Weber (1993), but also by Sun et al. (2011). These methods include:

- thin-layer chromatography (TLC) for lauric- and oleic acid for detection purposes only (Purghazi et al., 2007);
- high-performance liquid chromatography (for the estimation of total free and bound in glycerides fatty acids) equipped with a fluorescence detector after (1) formation of their *p*-bromophenacyl esters (Reed et al., 1984 as referred to by Mukherjee and Weber, 1993; Elliot et al., 1989; as referred to by Mukherjee and Weber, 1993), (2) a post-column reaction with a pH indicator (Bigley and Grob, 1986 as referred to by Mukherjee and Weber, 1993), (3) derivatisation with 3-bromomethyl-6,7-dimethoxy-1-methyl-2(1*H*)-quinoxalinone and limit of detection (LOD) of 10–15 fmol/10 μL (Yamaguchi et al., 1985 as referred to by Sun et al., 2011) or 6,7-dimethoxy-1-methyl-2(1*H*)-quinoxalinone-3-propionylcarboxylic acid hydrazide and LOD of 3–6 fmol/10 μL (Yamaguchi et al., 1990 as referred to by Sun et al., 2011), (4) derivatisation with 4-aminomethyl-6,7-dimethoxycoumarin and LOD of 20–50 fmol/10 μL (Sasamoto et al., 1996 as referred to by Sun et al., 2011), (5) derivatisation with 1-[2-(*p*-toluenesulfonate)-ethyl]-2-phenylimidazole-[4,5-*f*]-9,10-phenanthrene and LOD of 26–77 fmol/10 μL (You et al., 2007; as referred to by Sun et al., 2011), (6) derivatisation with 4-[2-(*N,N*-dimethylamino)ethylaminosulfonyl]-7-(2-aminoethylamino)-2,1,3-benzoxadiazole and LOD of 110–660 fmol/10 μL (Tsukamoto et al., 2005 as referred to by Sun et al., 2011), (7) derivatisation with 6-oxy-(acetyl piperazine)

fluorescein and LOD of 1–64 fmol/10 μL (Du et al., 2007 as referred to by Sun et al., 2011), (8) derivatisation with 6-oxy-(ethylpiperazine)-9-(2-methoxycarbonyl) fluorescein (Du et al., 2008), with a LOD between 1 and 2 nmol of fatty acid/L edible oil equal to 0.14 μg/L for caprylic acid and 0.57 μg/L for stearic acid; and (9) derivatisation with 2-(2-(anthracen-10-yl)-1*H*-phenanthro[9,10-d]imidazol-1-yl)ethyl 4-methylbenzenesulfonate and identification with online post-column atmospheric pressure chemical ionisation/mass spectrometry (APCI/MS) with a LOD of 23 fmol/10 μL equal to 0.65 μg stearic acid/L extract from raisins and hawthorn (Sun et al., 2011);

- gas chromatography (GC) with flame ionisation detector (FID) following an initial isolation step, i.e. via ion exchange resins, alumina (Mukherjee and Weber, 1993) or via magnetic solid-phase extraction with LODs 7.22–26.26 ng/mL oil (Wei et al., 2013). Sharma and Bindal, 1987; (as referred to by Mukherjee and Weber, 1993) and Blanco-Gomis et al. (2001, as referred to by Sun et al., 2011) selectively esterified free fatty acids with BF_3 and BCl_3, respectively, and analysed the resulting methylesters with a LOD of 7–25 ng/10 μL for the BCl_3 method. Williams and Macgee (1983, as referred to by Mukherjee and Weber, 1993) analysed their trimethylammonium salts ending up to a pyrolytic transformation into their methyl esters. Determination of the methyl esters of the free fatty acids via GC was also possible after an alkali reaction and GC/FID detection (Berchmans and Hirata, 2008; as referred to by Sun et al., 2011) or acidic reaction and GC/MS detection (Hejazi et al., 2009 as referred to by Sun et al., 2011). Yang et al. (2009) as referred to by Sun et al., 2011) have volatilised free fatty acids by using N,O-bis(trimethylsilyl)trifluoroacetamide in order to be subsequently measured via GC/MS analysis with a LOD of 1–12 ng/10 μL;
- supercritical fluid chromatography with Fourier transform infrared detection (Hellgeth et al., 1986 as referred to by Mukherjee and Weber, 1993).

The Panel noted that the analytical methods available cannot distinguish between added and naturally occurring free fatty acids.

3.1.5. Reaction and fate in food

The Panel considered that saturated fatty acids are rather stable and that decomposition may occur only at high temperatures. According to data in literature, thermal decomposition of saturated fatty acids occurs in a temperature range of around 480–550°C (Gouveia de Souza et al., 2004; Bagoria et al., 2012). Therefore, it was considered unlikely that such decomposition occurs in food during food processing.

Fatty acids are unstable in the presence of atmospheric oxygen. As with fats, autoxidation of fatty acids leads to the formation of hydroperoxides, which decompose to oxygen-containing products such as aldehydes, ketones, and hydroxy compounds. The effect of atmospheric oxygen on fatty acids depends primarily on the temperature, the number of double bonds, and the molecular structure. Saturated fatty acids show little tendency to undergo autoxidation, whereas unsaturated fatty acids, and especially polyunsaturated acids, are very susceptible to autoxidation (Anneken et al., 2012).

3.2. Authorised uses and use levels

Maximum levels of fatty acids (E 570) have been defined in Annex II to Regulation (EC) No 1333/2008[15] on food additives, as amended. In this document, these levels are named maximum permitted levels (MPLs).

Currently, fatty acids (E 570) is included in the Group I of food additives authorised at *quantum satis* (QS).

Table 3 summarises foods that are permitted to contain fatty acids (E 570) as set by Annex II to Regulation (EC) No 1333/2008.

[15] Commission Regulation (EC) No 1333/2008 of the European Parliament and of the Council of 16 December 2008 on food additives. OJ L 354, 31.12.2008, p. 16.

Table 3: MPLs of fatty acids (E 570) in foods according to the Annex II to Regulation (EC) No 1333/2008

Food category number	Food category name	E-number/ Group	Restrictions/ exception	MPL (mg/L or mg/kg as appropriate)
01.3	Unflavoured fermented milk products, heat-treated after fermentation	Group I		*quantum satis*
01.4	Flavoured fermented milk products including heat-treated products	Group I		*quantum satis*
01.6.3	Other creams	Group I		*quantum satis*
01.7.1	Unripened cheese excluding products falling in category 16	Group I	Except mozzarella	*quantum satis*
01.7.5	Processed cheese	Group I		*quantum satis*
01.7.6	Cheese products (excluding products falling in category 16)	Group I		*quantum satis*
01.8	Dairy analogues, including beverage whiteners	Group I		*quantum satis*
02.2.2	Other fat and oil emulsions including spreads as defined by Council Regulation (EC) No 1234/2007 and liquid emulsions	Group I		*quantum satis*
02.3	Vegetable oil pan spray	Group I		*quantum satis*
03	Edible ices	Group I		*quantum satis*
04.2.1	Dried fruit and vegetables	Group I		*quantum satis*
04.2.2	Fruit and vegetables in vinegar, oil, or brine	Group I		*quantum satis*
04.2.4.1	Fruit and vegetable preparations excluding compote	Group I		*quantum satis*
04.2.5.4	Nut butters and nut spreads	Group I		*quantum satis*
04.2.6	Processed potato products	Group I		*quantum satis*
05.1	Cocoa and Chocolate products as covered by Directive 2000/36/EC	Group I	Only energy-reduced or with no added sugar	*quantum satis*
05.2	Other confectionery including breath freshening microsweets	Group I		*quantum satis*
05.3	Chewing gum	Group I		*quantum satis*
05.4	Decorations, coatings and fillings, except fruit-based fillings covered by category 4.2.4	Group I		*quantum satis*
06.2.2	Starches	Group I		*quantum satis*
06.3	Breakfast cereals	Group I		*quantum satis*
06.4.2	Dry pasta	Group I	Only gluten free and/or pasta intended for hypoproteic diets in accordance with Directive 2009/39/EC	*quantum satis*
06.4.4	Potato Gnocchi	Group I	Except fresh refrigerated potato gnocchi	*quantum satis*
06.4.5	Fillings of stuffed pasta (ravioli and similar)	Group I		*quantum satis*
06.5	Noodles	Group I		*quantum satis*
06.6	Batters	Group I		*quantum satis*
06.7	Precooked or processed cereals	Group I		*quantum satis*
07.1	Bread and rolls	Group I	Except products in 7.1.1 and 7.1.2	*quantum satis*

Food category number	Food category name	E-number/ Group	Restrictions/ exception	MPL (mg/L or mg/kg as appropriate)
07.2	Fine bakery wares	Group I		*quantum satis*
08.3.1	Non-heat-treated processed meat	Group I		*quantum satis*
08.3.2	Heat-treated processed meat	Group I	Except *foie gras, foie gras entier, blocs de foie gras, Libamáj, libamáj egészben, libamáj tömbben*	*quantum satis*
08.3.3	Casings and coatings and decorations for meat	Group I		*quantum satis*
09.2	Processed fish and fishery products including molluscs and crustaceans	Group I		*quantum satis*
09.3	Fish roe	Group I	Only processed fish roe	*quantum satis*
10.2	Processed eggs and egg products	Group I		*quantum satis*
11.2	Other sugars and syrups	Group I		*quantum satis*
12.1.2	Salt substitutes	Group I		*quantum satis*
12.2.2	Seasonings and condiments	Group I		*quantum satis*
12.3	Vinegars	Group I		*quantum satis*
12.4	Mustard	Group I		*quantum satis*
12.5	Soups and broths	Group I		*quantum satis*
12.6	Sauces	Group I		*quantum satis*
12.7	Salads and savoury-based sandwich spreads	Group I		*quantum satis*
12.8	Yeast and yeast products	Group I		*quantum satis*
12.9	Protein products, excluding products covered in category 1.8	Group I		*quantum satis*
13.2	Dietary foods for special medical purposes defined in Directive 1999/21/EC (excluding products from food category 13.1.5)	Group I		*quantum satis*
13.3	Dietary foods for weight control diets intended to replace total daily food intake or an individual meal (the whole or part of the total daily diet)	Group I		*quantum satis*
13.4	Foods suitable for people intolerant to gluten as defined by Regulation (EC) No 41/2009	Group I	Including dry pasta	*quantum satis*
14.1.2	Fruit juices as defined by Directive 2001/112/EC and vegetable juices	Group I	Only vegetable juices	*quantum satis*
14.1.3	Fruit nectars as defined by Directive 2001/112/EC and vegetable nectars and similar products	Group I	Only vegetable nectars	*quantum satis*
14.1.4	Flavoured drinks	Group I		*quantum satis*
14.1.5.2	Other	Group I	Excluding unflavoured leaf tea; including flavoured instant coffee	*quantum satis*
14.2.3	Cider and perry	Group I		*quantum satis*
14.2.4	Fruit wine and made wine	Group I		*quantum satis*
14.2.5	Mead	Group I		*quantum satis*
14.2.6	Spirit drinks as defined in Regulation (EC) No 110/2008	Group I	Except whisky or whiskey	*quantum satis*

Food category number	Food category name	E-number/ Group	Restrictions/ exception	MPL (mg/L or mg/kg as appropriate)
14.2.7.1	Aromatised wines	Group I		*quantum satis*
14.2.7.2	Aromatised wine-based drinks	Group I		*quantum satis*
14.2.7.3	Aromatised wine-product cocktails	Group I		*quantum satis*
14.2.8	Other alcoholic drinks including mixtures of alcoholic drinks with non-alcoholic drinks and spirits with less than 15% of alcohol	Group I		*quantum satis*
15.1	Potato-, cereal-, flour- or starch-based snacks	Group I		*quantum satis*
15.2	Processed nuts	Group I		*quantum satis*
16	Desserts excluding products covered in categories 1, 3 and 4	Group I		*quantum satis*
17.1[(a)]	Food supplements supplied in a solid form including capsules and tablets and similar forms, excluding chewable forms	Group I		*quantum satis*
17.2[(a)]	Food supplements supplied in a liquid form	Group I		*quantum satis*
17.3[(a)]	Food supplements supplied in a syrup-type or chewable form	Group I		*quantum satis*
18	Processed foods not covered by categories 1–17, excluding foods for infants and young children	Group I		*quantum satis*

MPL: Maximum permitted level.
(a): FCS 17 refers to food supplements as defined in Directive 2002/46/EC of the European Parliament and of the Council excluding food supplements for infants and young children.

According to Annex III, Part 1, Part 2, Part 3, Part 4 and Part 5 (section A) of Regulation (EC) No 1333/2008, fatty acids (E 570) is also authorised as a carrier in food additives in glazing agents for fruit with a maximum level at QS, as a food additive in food additives with a maximum level in all food additives preparations at QS, as a food additive in food enzymes with a maximum level in the products (final food and beverages) at QS, as a food additive in food flavourings with a maximum level in all flavourings at QS, and as a food additive in nutrients (all nutrients except nutrients containing unsaturated fatty acids) with a maximum level in the products at QS.

3.3. Exposure data

3.3.1. Reported use levels or data on analytical levels of fatty acids (E 570)

Most food additives in the EU are authorised at a specific MPL. However, a food additive may be used at a lower level than the MPL. Therefore, information on actual use levels is required for performing a more realistic exposure assessment, especially for those food additives for which no MPL is set and which are authorised according to *QS* as is the case for fatty acids (E 570).

In the framework of Regulation (EC) No 1333/2008 on food additives and of Commission Regulation (EU) No 257/2010 regarding the re-evaluation of approved food additives, EFSA issued a public call[16] for occurrence data (usage level and/or concentration data) on fatty acids (E 570). In response to this public call, updated information on the actual use levels of fatty acids (E 570) in foods was made available to EFSA by industry. These data do not include information on the composition of the fatty acids actually used as food additive E 570. No analytical data on the concentration of fatty acids (E 570) in foods were made available by the Member States.

[16] Call for food additives usage level and/or concentration data in food and beverages intended for human consumption. Published 27 March 2013. http://www.efsa.europa.eu/en/dataclosed/call/130327.htm

Summarised data on reported use levels in foods provided by industry

Industry provided EFSA with data on use levels (n = 30) of fatty acids (E 570) in foods for eight out of the 67 food categories in which fatty acids (E 570) is authorised.

Updated information on the actual use levels of fatty acids (E 570) in foods was made available to EFSA by FoodDrinkEurope (FDE), the International Chewing Gum Association (ICGA), the Association of the European Self-Medication Industry (AESPG), Dr Loges Naturheilkunde neu endtdecken and KRÜGER GmbH & Co.

The Panel noted that 11 usage levels on food supplements referred to a niche product. Since other usage levels were available for this food category, the Panel decided to exclude them from the specific *food supplements consumers only scenario* (Section 3.4).

Appendix A provides data on the use levels of fatty acids (E 570) in foods as reported by industry.

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

The Mintel GNPD is an online database, which monitors product introductions in consumer packaged goods markets worldwide. It contains information of over 2 million food and beverage products of which more than 800,000 are or have been available on the European food market. The Mintel GNPD started covering EU's food markets in 1996, currently having 20 out of its 28 member countries and Norway presented in the Mintel GNPD.[17]

For the purpose of this Scientific Opinion, the Mintel GNPD[18] was used for checking the labelling of food products containing fatty acids (caprylic, capric, lauric, myristic, palmitic, stearic and oleic acid) within the EU's food products as the Mintel GNPD shows the compulsory ingredient information presented in the labelling of products. Within the Mintel GNPD, it was not always possible to distinguish between fatty acids added to a food product as a food additive or as an ingredient for fortification purposes.

According to the Mintel GNPD, fatty acids (caprylic, capric, lauric, myristic, palmitic, stearic and oleic acid) are labelled on food products (n = 414) of bakery, dairy, sauces & seasonings, snacks, meals & meal centers, desserts & ice cream, sugar & gum confectionery, vitamins & dietary supplements, baby food (fatty acids added for fortification purpose), and sweeteners & sugar.

Appendix B presents the percentage of the food products labelled with fatty acids (caprylic, capric, lauric, myristic, palmitic, stearic and oleic acid) out of the total number of food products per food sub-category according to the Mintel GNPD food classification between 2011 and 2016. The percentages were less than 5% in all food subcategories. The overall percentage of food products labelled with fatty acids (caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid), considering the food subcategories with at least one food to which these fatty acids were added according to the label, was 0.09%.

3.3.3. Food consumption data used for exposure assessment

EFSA Comprehensive European Food Consumption Database

Since 2010, the EFSA Comprehensive European Food Consumption Database (Comprehensive Database) has been populated with national data on food consumption at a detailed level. Competent authorities in the European countries provide EFSA with data on the level of food consumption by the individual consumer from the most recent national dietary survey in their country (cf. Guidance of EFSA on the Use of the EFSA Comprehensive European Food Consumption Database in Exposure Assessment' (EFSA, 2011a). New consumption surveys recently[19] added to the Comprehensive database were also taken into account in this assessment.[20]

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

[17] Missing Bulgaria, Cyprus, Estonia, Latvia, Lithuania, Luxembourg, Malta and Slovenia.
[18] http://www.gnpd.com/sinatra/home/ accessed on 14/10/2016.
[19] Available online: http://www.efsa.europa.eu/en/press/news/150428.htm
[20] Available online: http://www.efsa.europa.eu/en/datexfoodcdb/datexfooddb.htm

the EFSA Comprehensive Database represents the best available source of food consumption data across Europe at present.

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

Table 4: Population groups considered for the exposure estimates of fatty acids (E 570)

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

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

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

Food categories considered for the exposure assessment of fatty acids (E 570)

The food categories in which the use of fatty acids (E 570) is authorised were selected from the nomenclature of the EFSA Comprehensive Database (FoodEx classification system), at the most detailed level possible (up to FoodEx Level 4) (EFSA, 2011b).

Some food categories or their restrictions/exceptions are not referenced in the EFSA Comprehensive Database and could therefore not be taken into account in the present estimate. This may have resulted in an underestimation of the exposure. This was the case for 13 food categories (Appendix C).The food categories which were not taken into account are described below (in ascending order of the FCS codes):

- 01.7.6. Cheese products (excluding products falling in category 16);
- 02.3 Vegetable oil pan spray;
- 06.4.2. Dry pasta: only gluten free and/or pasta intended for hypoproteic diets in accordance with Directive 2009/39/EC;
- 06.4.4. Potato gnocchi: except fresh refrigerated potato gnocchi;
- 06.6. Batters;
- 06.7. Precooked or processed cereals;
- 08.3.3. Casings and coatings and decorations for meat;
- 12.1.2. Salt substitutes;
- 14.1.3. Fruit nectars, only vegetable nectars;
- 14.2.4. Fruit wine and made wine;
- 14.2.5. Mead;
- 14.2.7.2. Aromatised wine-based drinks;
- 14.2.7.3. Aromatised wine-product cocktails.

For the following food category, the restriction which applies to the use of fatty acids (E 570) could not be taken into account, and therefore the whole food category was considered in the exposure assessment. This may have resulted in an overestimation of the exposure:

- 05.1. Cocoa and cocoa products, only energy-reduced or with no added sugar.

In addition, for the following food categories: FC 17.1, 17.2 and FC 17.3 Food supplements, in solid, liquid and syrup-type or chewable form, which were used only in the specific exposure scenario including food supplements, the restrictions which apply to the use of fatty acids (E 570) could not be taken into account, and therefore the whole food category (FC 17) was considered in the exposure assessment.

Considering that the food category 18 (Processed foods not covered by categories 1–17, excluding foods for infants and young children) is extremely unspecific (e.g. composite foods), processed foods, prepared or composite dishes belonging to the food category 18 were reclassified under food categories in accordance to their main component. Therefore, food category 18 is not taken into account as contributor to the total exposure estimates.

Food categories 13.2, 13.3 and 13.4 were not considered in exposure assessment (as explained in section 3.4.1).

For all scenarios, 42 food categories were not taken into account because no concentration data were provided for these food categories to EFSA (Appendix C). For the remaining food categories, the refinements considering the restrictions/exceptions as set in Annex II to Regulation No 1333/2008 were applied.

Overall, for the maximum level and the refined exposure scenarios, data for eight authorised food categories were available for the present exposure assessment to fatty acids (E 570) (Appendix C).

3.4. Exposure estimate

3.4.1. Exposure to fatty acids (E 570) from its use as a food additive

The Panel noted that according to the EU specifications the food additive (E 570) may contain only one particular fatty acid or a mixture of the fatty acids authorised within the E 570. Accordingly, the exposure assessment refers to E 570 and not to a specific fatty acid.

The Panel estimated chronic exposure to fatty acids (E 570) for the following population groups: infants; toddlers, children, adolescents, adults and the elderly. Dietary exposure to fatty acids (E 570) was calculated by multiplying fatty acids (E 570) concentrations for each food category (Appendix C) with their respective consumption amount per kilogram of body weight for each individual in the Comprehensive Database. The exposure per food category was subsequently added to derive an individual total exposure per day. These exposure estimates were averaged over the number of survey days, resulting in an individual average exposure per day for the survey period. Dietary surveys with only 1 day per subject were excluded as they are considered as not adequate to assess repeated exposure.

This was carried out for all individuals per survey and per population group, resulting in distributions of individual exposure per survey and population group (Table 4). Based on these distributions, the mean and 95th percentile of exposure were calculated per survey and per population group. The 95th percentile of exposure was only calculated for those population groups where the sample size was sufficiently large to allow this calculation (EFSA, 2011a). Therefore, in the present assessment, the 95th percentile of exposure for infants from Italy and for toddlers from Belgium, Italy and Spain were not included.

Exposure assessment to fatty acids (E 570) was carried out by the ANS Panel based on (1) maximum reported use levels provided to EFSA (defined as the *maximum level exposure assessment scenario*) and (2) reported use levels (defined as the *refined exposure assessment scenario*). These two scenarios are discussed in detail below. Exposure scenarios can consider only food categories for which data were available to the Panel.

These scenarios do not consider the consumption of food supplements (FC 17.1, 17.2 and FC 17.3), which is covered in an additional scenario detailed below (*food supplements consumers only scenario*), nor foods for special medical purposes (FSMP). FSMP consumed may be very diverse; they cannot be considered because of very limited information on consumption. Eating occasions belonging to the food categories 13.2, 13.3 and 13.4 were therefore reclassified under food categories in accordance to their main component.

Therefore, only five out of the eight authorised food categories for which reported use levels were available, were used for the exposure assessment to fatty acids (E 570).

A possible additional exposure from the use of fatty acids (E 570) as a carrier in food additives, food additive in food additives, in food enzymes, in flavourings and in nutrients in accordance with Annex III to Regulation (EC) No 1333/2008 (Part 1, Part 2, Part 3, Part 4 and Part 5 (section A)) was not considered in any of the exposure assessment scenarios, due to the absence of information on use levels.

Maximum level exposure assessment scenario

The *regulatory maximum level exposure assessment scenario* is based on the MPLs as set in Annex II to Regulation (EC) No 1333/2008. As fatty acids (E 570) is authorised according to QS in all food categories, a *maximum level exposure assessment scenario* was estimated based on the maximum reported use levels provided by industry, as described in the EFSA Conceptual framework (EFSA ANS Panel, 2014). The maximum reported use levels used are listed in Appendix C.

The Panel considers the exposure estimates derived following this scenario as the most conservative as it is assumed that that the population group will be exposed to fatty acids (E 570) present in food at the maximum reported use levels over a longer period of time, and assuming that fatty acids (E 570) are only used in the food categories for which data were submitted by industry.

Refined exposure assessment scenario

The refined exposure assessment scenario of fatty acids (E 570) was based on typical and maximum use levels of food additive.

Appendix C summarises the concentration levels of fatty acids (E 570) used in the refined exposure assessment scenario. Based on the available data set, the Panel calculated two refined exposure estimates based on different model populations:

- The brand-loyal consumer scenario: It was assumed that a consumer is exposed long-term to fatty acids (E 570) present at the maximum reported use level for one food category. This exposure estimate is calculated as follows:
  - Combining food consumption with the maximum of the reported use levels for the main contributing food category at the individual level.
  - Using the mean of the typical reported use levels for the remaining food categories.
- The non-brand-loyal consumer scenario: It was assumed that a consumer is exposed long-term to fatty acids (E 570) present at the mean reported use levels in food. This exposure estimate is calculated using the mean of the typical reported use levels for all food categories.

Specific exposure assessment scenario: *Food supplements consumers only scenario*

Fatty acids (E 570) is authorised in the food category 17 Food supplements as defined in Directive 2002/46/EC, excluding food supplements for infants and young children. As exposure via food supplements may deviate largely from the one via food, and that the number of food supplement consumers may be low depending on populations and surveys, this additional scenario was calculated in order to reflect additional exposure to fatty acids (E 570) from food supplements compared to the exposure to the food additive excluding these sources.

This scenario was estimated assuming that consumers only of food supplements were exposed to fatty acids (E 570) present at the maximum reported use level on a daily basis via consumption of food supplements. For the remaining food categories, the mean of the typical reported use levels was used.

As food category 17 does not include food supplements for infants and toddlers (Regulation (EC) No 1333/2008), exposure to fatty acids (E 570) from food supplements was not estimated for these two population groups.

Appendix C summarises the concentration levels of fatty acids (E 570) used in the specific exposure assessment scenario.

Dietary exposure to fatty acids (E 570)

Table 5 summarises the estimated exposure to fatty acids (E 570) from its use as a food additive in six population groups (Table 4) according to the different exposure scenarios. Detailed results per population group and survey are presented in Appendix D.

Table 5: Summary of dietary exposure to fatty acids (E 570) from its use as a food additive in the *maximum level exposure assessment scenario* and in the refined exposure scenarios, in six population groups (minimum–maximum across the dietary surveys in mg/kg bw per day)

	Infants (12 weeks–11 months)	Toddlers (12–35 months)	Children (3–9 years)	Adolescents (10–17 years)	Adults (18–64 years)	The elderly (≥ 65 years)
Maximum level exposure assessment scenario						
Mean	0.5–10.4	1.5–54.4	5.3–61.6	6.1–45.8	1.8–19.3	0.5–5.4
95th percentile	0.4[(a)]–64.7	9.0–196.0	23.7–153.8	24.3–107.9	10.3–68.4	2.9–23.5
Refined estimated exposure assessment scenario						
Brand-loyal scenario						
Mean	0.5–10.4	1.5–54.4	5.2–61.6	6.1–45.8	1.8–19.3	0.5–5.4
95th percentile	0.4[(a)]–64.7	9.0–196.0	22.4–153.8	24.3–107.9	10.3–68.4	2.9–23.5
Non-brand-loyal scenario						
Mean	0.4–5.3	1.1–27.7	3.1–31.7	3.3–23.5	1.0–9.9	0.3–2.9
95th percentile	0.4–33.8	8.9–99.1	13.7–79.0	12.5–54.8	5.2–35.4	1.7–13.2

(a): In one dietary survey, the flavoured drinks largely contributed to the mean exposure, but since their consumption was reported by less than 5% of subjects, the P95th percentile resulted to be lower than the mean exposure level.

In the *maximum level exposure assessment scenario*, mean exposure to fatty acids (E 570) from its use as a food additive ranged from 0.5 mg/kg bw per day in infants and the elderly to 61.6 mg/kg bw per day in children. The 95th percentile of exposure to fatty acids (E 570) ranged from 0.4 mg/kg bw per day in infants to 196.0 mg/kg bw per day in toddlers.

In the refined *brand-loyal scenario*, the exposure estimates were similar as those reported for the maximum level exposure assessment scenario. This was due to the fact that only two food categories, flavoured drinks and chewing gum, had different mean and maximum use levels (Appendix C). Since the flavoured drinks were frequently the main contributor to the individual exposure to fatty acids (E 570), the maximum level was mostly assigned to this food category which is normally consumed in a large amount. In consequence, the difference in exposure between maximum level and *brand-loyal scenario*, which was predominantly due to the consumption of chewing gum, was negligible. In the refined *non-brand-loyal scenario*, mean exposure to fatty acids (E 570) from its use as a food additive ranged from 0.3 mg/kg bw per day in the elderly to 31.7 mg/kg bw per day in children. The 95th percentile of exposure to fatty acids (E 570) ranged from 0.4 mg/kg bw per day in infants to 99.1 mg/kg bw per day in toddlers.

In all three exposure scenarios and population groups, the main contributing food category to the total mean exposure estimates was flavoured drinks. For infants and toddlers, an additional important contributing food category was cocoa and chocolate products.

The main food categories contributing to the exposure to fatty acids (E 570) are presented in Appendix E.

In the *food supplements consumers only scenario*, mean exposure to fatty acids (E 570) from its use as a food additive ranged for children from 7.8 to 50.2 mg/kg bw per day and from 4.8 to 8.8 mg/kg bw per day for adults. The 95th percentile of exposure to fatty acids (E 570) ranged for children from 30.8 to 76.6 mg/kg bw per day and for adults from 17.4 to 29.3 mg/kg bw per day.

Uncertainty analysis

Uncertainties in the exposure assessment of fatty acids (E 570) have been discussed above. In accordance with the guidance provided in the EFSA opinion related to uncertainties in dietary exposure assessment (EFSA, 2007), the following sources of uncertainties have been considered and are summarised in Table 6.

Table 6: Qualitative evaluation of influence of uncertainties on the dietary exposure estimate, excluding the *food supplements consumers only scenario*

Sources of uncertainties	Direction[(a)]
Consumption data: different methodologies/representativeness/underreporting/misreporting/no portion size standard	+/–
Use of data from food consumption survey of a few days to estimate long-term (chronic) exposure for high percentiles (95th percentile)	+

Sources of uncertainties	Direction[a]
Correspondence of reported use levels and analytical data to the food items in the EFSA Comprehensive Food Consumption Database: uncertainties to which types of food the levels refer to	+/–
Uncertainty in possible national differences in use levels of food categories	+/–
Concentration data: • levels considered applicable for all items within the entire food category • unclear representativeness of foods on the EU market	 + +/–
Consumption data considered in the refined exposure assessment: 2–46% of the amount of food consumed (grams per kg body weight) corresponding to five food categories (out of 67 authorised food categories) taken into account	–
Food categories selected for the exposure assessment: exclusion of food categories due to missing FoodEx linkage (n = 13/67 food categories)	–
Food categories selected for the exposure assessment: inclusion of food categories without considering the restriction/exception (n = 1/67 food categories)	+
Food categories included in the exposure assessment: no additional data available for authorised food categories (n = 42/67 food categories)	–
Maximum level exposure assessment scenario: • exposure calculations based on the maximum reported use levels (reported use from industry) assuming fatty acids (E 570) is not used in the food categories for which no use levels were submitted • foods which may contain fatty acids (E 570) according to Annex III to Regulation (EC) No 1333/2008 not taken into account	 + –
Refined exposure assessment scenarios: • exposure calculations based on the maximum or mean levels (reported use from industry) • foods which may contain fatty acids (E 570) according to Annex III to Regulation (EC) No 1333/2008 not taken into account	 +/– –

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

Overall, assuming that the food additive is not used in the food categories for which no use levels were reported, the Panel considered that the uncertainties identified would, in general, result in an overestimation of the exposure to fatty acids (E 570) as a food additive in European countries in all exposure scenarios. This assumption of non-use was supported by the observation that fatty acids (E 570) is authorised as a Group I food additive in 67 food categories (Table 3). Since, all these food categories correspond to the general Group I food additives authorisation, fatty acids (E 570) may not necessarily be used in some of these food categories, and that may explain why reported use levels of fatty acids (E 570) were only available for eight food categories. The Panel noted that the information from the Mintel GNPD supported the observation that fatty acids included in fatty acids (E 570) (caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid) are apparently not used in all food categories in which they are authorised (Section 3.3.2).

In addition, regarding the *food supplements consumers only scenario*, the Panel considered that the uncertainties would result in an overestimation of the exposure to fatty acids (E 570) as a food additive, given that the calculations were based on consumers only of food supplements and assuming a long-term brand loyalty consumption of these supplements on a daily basis.

In none of the exposure scenarios, the use of fatty acids (E 570) according to Annex III to Regulation No 1333/2008 was considered. Neglecting this source of exposure may have resulted in an underestimation of exposure to fatty acids (E 570) in all scenarios.

3.4.2. Exposure via regular diet

In 2010, the EFSA NDA Panel issued an opinion on the DRVs for fats, including saturated fatty acids, polyunsaturated fatty acids, monounsaturated fatty acids, trans fatty acids, and cholesterol (EFSA NDA Panel, 2010). This opinion estimated fatty acid intakes in energy % (E%) among European populations for several age classes and by gender. The intake estimates were based on concentration

data derived from national reports and published overviews, and consumption data from individual based on food consumption surveys. Intake via food supplements was not taken into account.

For saturated fatty acids, which were considered the most representative for the food additive E 570, the NDA Panel estimated an average E% intake from all fats (including free fatty acids) of between 11 E % and 13 E% in infants, 10% and > 15% in children, and < 9% and 26% in adults and the elderly.

Based on these estimates, the ANS Panel calculated a minimum and maximum mean intake of saturated fatty acids derived from all fats, expressed in mg/kg bw per day as follows:

- the minimum and maximum mean E% of saturated fatty acids intakes derived from all fats were identified among different European countries and age groups (according to Table 4);
- in order to express these intake values in mg/kg bw per day, the values of the total energy intakes (all components from food) (kcal/day) corresponding to the respective age groups and countries (EFSA NDA Panel, 2013), the standard energy yield of fat of 9 kcal/g and default body weight values were used.

As a result, the minimum–maximum mean intakes of saturated fatty acids derived from all fats (mg/kg bw per day) were approximately 2,500–3,000 mg/kg bw per day in infants, 1,100–1,300 mg/kg bw per day in toddlers, 900–1,300 mg/kg bw per day in children, 400–700 mg/kg bw per day in adolescents, 400–1,000 mg/kg bw per day in adults and 300–1,300 mg/kg bw per day in the elderly.

The Panel noted that monounsaturated fatty acids were not included in the calculation of the exposure to fatty acids, derived from all fats, from the regular diet resulting in an underestimation of the exposure to fatty acids from this source.

The Panel noted that the actual level of free fatty acids derived from all fats in foods depends on the nature of the foodstuffs (Appendix F). For example, in refined oils it is well below 0.5% (w/w) while in the oil fraction of fresh fish it can be up to 3.7% (w/w) (expressed as oleic acid). The percentage of free fatty acids tends to increase by food processing (e.g. in heated oils up to 1.6% or up to 4.3% (as oleic acid) in the oil fraction of fresh minced fish), packaging or storage (e.g. up to 5.2% in cereals stored for 9 months in polypropylene bags).

Based on the estimated intakes of saturated fatty acids via the regular diet and those of fatty acids (E 570) as a food additive (Section 3.4.1, *non-brand-loyal* scenario), the Panel estimated that at the mean level the intake of fatty acids (E 570) as a food additive may contribute on average 1% to the overall dietary exposure to saturated fatty acids derived from all fats.

3.5. Biological and toxicological data

It is recognised that free fatty acids may specifically bind G-protein coupled receptors (GPCR) (FFAR1-4, GPR84), which are widely expressed (Alvarez-Curto and Milligan, 2016). Although expressed on a variety of cells, the presence of these free fatty acid receptors on key cell types, which regulate both metabolic and immunological processes, provides a link between the regulation of energy homoeostasis and the control of immunological and inflammatory responses. As a result of binding, various types of effects, sometime opposite (den Besten et al., 2013; Frommer et al., 2015; Miao et al., 2015; Rocha et al., 2017), have been reported for different free fatty acids depending on, e.g. their chain length, being saturated or not, and their degree of unsaturation (FAO, 2008).

Therefore, free fatty acids may be critical signalling molecules due to their role as agonists for different members of the family of free fatty acids receptors, therefore an imbalance in the amount of fatty acids ingested may have consequence for the organism, including peptide hormone secretion, blood coagulation, inflammation, hepatic toxicity and increased susceptibility to allergy. (Hoak, 1994; Temme et al., 1999; Cai et al., 2014; van Elten et al., 2015; Miyamoto et al., 2016).

Although some of the fatty acids, which are included in the food additive E 570, have been reported to be recognised by these receptors, the Panel considered it outside the scope of this evaluation to review the extensive literature available about these receptors.

3.5.1. Absorption, distribution, metabolism and excretion

Caprylic acid

Human studies

Schwabe et al. (1964) studied the absorption and oxidation of orally ingested or intravenous (i.v.) injected ^{14}C-carboxyl-labelled caprylic acid, in 12 normal volunteers (medical students, no further details). The absorption and oxidation was determined by monitoring the expired $^{14}CO_2$ for 50 min

after administration. In both trials, 2–3 μCi of the test item was given to each volunteer with a time interval of 3 days between oral or i.v. treatment. There was no significant difference in the mean cumulative recovery of $^{14}CO_2$ after oral or intravenous administration of labelled caprylic acid: 15.4% vs 15.7% of the applied radioactivity, respectively. A slight increase in the delay time (the time period between application and first measurement of radioactivity) was reported after oral administration (3–6 min) vs i.v. injection (1–2 min). In both trials, the maximal radioactivity in expired air was measured 15–30 min after administration. In further experiments, it was shown that co-application of glucose, orally or by co-infusion of 1 g unlabelled caprylic acid did not inhibit caprylic acid oxidation based on expired $^{14}CO_2$ measurements. The authors concluded that caprylic acid is readily absorbed from the gastrointestinal tract and rapidly oxidised.

In the studies by Chen et al. (2003) and by Sanaka et al. (2006), the absorption of ^{13}C-labelled caprylic acid was evaluated using the gastric emptying breath test. According to the authors, this test allowed indirect assessment of gastric emptying by monitoring exhaled $^{13}CO_2$. ^{13}C-Caprylic acid was absorbed only after it was emptied from the stomach, and then was oxidised to $^{13}CO_2$. The methodological relevance of the breath test was based on the assumption that intestinal absorption and hepatic oxidation progress rapidly. After an overnight fasting, breath samples were taken from six male healthy volunteers (30–39 years) who received, via an oesophago-gastro-duodenoscope, 100 mg ^{13}C-caprylate dissolved in 20 mL normal saline, in the descending portion of the duodenum. Maximal exhalation rate of $^{13}CO_2$ was found 15 min after application. Beyond 60 min post-dose, $^{13}CO_2$ excretion diminished in an exponential manner, characterised by a half-excretion time of 71.3 ± 12.7 min.

Capric acid

Animal studies

Blomstrand (1955) studied the intestinal absorption of [1-^{14}C]-capric acid in four male rats (strain not given) with cannulated thoracic lymph ducts after gavage of olive oil containing 1.1% of the free [1-^{14}C]-capric acid (no further information given). The lymph (in four experimental animals) and expired air (in three experimental animals) were collected for 24 h, after which the animals were sacrificed and the radioactivity of the whole gastrointestinal tract plus faeces was measured. The applied radioactivity was almost completely absorbed within 24 h from the gastrointestinal tract (96–100%) and 3–16% of the label was recovered in the lymph lipids from the thoracic duct. About 2% of the activity found in the lymph lipids was in the form of phospholipids but 98% in the form of triglycerides; only traces of free capric acid were detected. Large amounts of applied radioactivity (19% within 8 h and 46% within 21 h) occurred in the expired air in form of $^{14}CO_2$. Overall, free capric acid was nearly completely absorbed from the gastrointestinal tract and mainly metabolised and excreted in the form of exhaled CO_2; minor amounts were transported via the thoracic-duct lymph and incorporated into triglycerides.

Borgström (1955) studied the transport form of ^{14}C-lipid in blood samples taken from the portal vein and from the inferior vena cava blood. Samples were collected 1.5 h after administration of 1 mL olive oil containing 28% free ^{14}C-capric acid administered via gavage to two adult rats (no further details provided). Higher radioactivity of blood lipids was detected in the portal vein (2.3- or 4-fold increase compared to vena cava), mostly in the form of free fatty acids.

Vallot et al. (1985) studied, by perfusion of isolated duodenojejunal loops in male Wistar rats (n = 4), the modification of both the portal vein and intestinal lymph absorption of capric acid in the presence of a monoglyceride (monopalmitin) and long-chain fatty acids (oleic- or palmitic acid). In the experiments, 10–15 μCI (52 mCi/mmol) of 1-^{14}C-capric acid was added to each lipid emulsion. Blood samples were collected from the cannulated portal vein at 5 min intervals for 1 h after infusion of 90 μmol of labelled capric acid. During this time period, 80% of the applied radioactivity disappeared from the intestinal loop and 11% was recovered in the mucosa at termination (after 1 h). About 35% of the capric acid was transported directly into the bloodstream mainly in form of free fatty acids. Capric acid oxidation in the mucosa was evident since $^{14}CO_2$ was detected in portal venous blood. The authors concluded that labelled capric acid was rapidly and mainly absorbed via the portal route. When, in further experiments, labelled capric acid was infused into the loop of intestinal lymph fistulated rats (n = 3), a very low percentage of radioactivity (< 1%) was recovered in the lymph during a sampling period of 6 h. The intestinal absorption and mucosal oxidation of capric acid was modulated by the co-application of palmitic- or oleic acid.

Lauric acid

Animal studies

Göransson (1965) administered i.v. 0.5 mL rat serum containing 0.1 μeq [1-^{14}C]-labelled lauric acid together with 0.1 μeq [9,10-^{3}H]-labelled palmitic acid to fasted and refed rats (no further details given). The disappearance of the two different labels (^{14}C vs ^{3}H) was measured 1–5 min after injection. Lauric acid disappeared more rapidly from blood than palmitic acid. Data on tissue distribution of the two labels indicated that lauric acid was more rapidly oxidised than palmitic acid. Similar results were reported in the re-fasted rats.

Myristic acid

Animal studies

Göransson (1965) administered i.v. 0.5 mL rat serum containing 0.1 μeq [1-^{14}C]-labelled myristic acid together with 0.1 μeq [9,10-^{3}H]-labelled palmitic acid to fasted and refed rats (no further details given). The disappearance of the two different labels (^{14}C vs ^{3}H) was measured 1–5 min after injection. Myristic acid disappeared more rapidly from blood than palmitic acid. Data on tissue distribution of the two labels indicated that myristic acid was more rapidly oxidised than palmitic acid. Similar results were reported in the non-fasted rats.

Rioux et al. (2000) compared the metabolism of myristic and palmitic acids in cultured hepatocytes obtained from male Sprague–Dawley rats. [1-^{14}C]-Labelled fatty acids were incubated for 12 h with 24-h cultured hepatocytes. Myristic acid was taken up more rapidly than palmitic acid (about 87% and about 69%, respectively; $p < 0.05$). After 4 h of incubation, 13% of the initial myristic acid label was still in the medium while after 12 h of incubation there was almost no myristic acid left in the medium (0.9%). Incorporation into cellular lipids, however, was similar after the same time (33.4 ± 2.8% and 34.9 ± 9.3%, respectively, of initial radioactivity). In the early phase of incubation (30 min), myristic acid was more rapidly incorporated into cellular triglycerides than was palmitic acid (7.4 ± 0.9% and 3.6 ± 1.9% of initial radioactivity respectively). However, after 12 h incubation, the radioactivity of cellular triglycerides, cellular phospholipids, and secreted triglycerides was significantly higher with palmitic acid as precursor. Myristic acid oxidation was significantly higher than that of palmitic acid, with 14.9 ± 2.2% and 2.3 ± 0.6% of the initial radioactivity respectively, being incorporated into β-oxidation products after 4 h. Myristic acid was also elongated to palmitic acid to a greater degree (12.2 ± 0.8% of initial radioactivity after 12 h) than palmitic acid was to stearic acid (5.1 ± 1.3% of initial radioactivity after 12 h). The combination of elongation and β-oxidation resulted in the rapid disappearance of C14:0 in hepatocytes whereas C16:0 was esterified to form glycerolipids.

Palmitic acid

Animal studies

Coniglio and Cate (1959) administered 100 mg [1-^{14}C]-palmitic acid in 1 mL of olive oil to fed male Sprague–Dawley rats (tissue from two to four animals were pooled per time point) via a stomach tube (no further details given). The animals were sacrificed 3, 6 or 24 h after administration. The intestine was washed out with water and lipids of the intestinal contents, intestinal tissue and the liver were extracted and analysed for distribution of the ^{14}C in various types of lipids (phospholipids, glycerides and fatty acids). The total activity in the intestinal contents was predominantly as free fatty acids at both 3 and 6 h after administration. However, up to 18% of the radioactivity was found in the triglyceride fraction at these times. In the intestinal tissue, radioactivity was associated largely with triglycerides, but at 6 and 24 h, this fraction contained decreasing amounts of radiolabel and there was a progressive increase in radioactivity associated with phospholipids. In the liver, however, the activity was greater in phospholipids than in triglycerides measured 3 h after application; minor amounts of label were also found in mono- and diglycerides as well as cholesterol esters.

Laurell (1959) studied the distribution of radiolabelled free palmitic acid in non-fasted or fasted male albino rats (n = 3 each; strain not given) after i.v. injection of 8–10 μCi of ^{14}C- labelled palmitic acid in 1 mL 10% albumin or in 1 mL rat serum. Twenty minutes after injection, rats were sacrificed and liver, muscle and adipose tissues were removed and lipids extracted and analysed. Radioactivity in other organs and carcass was measured only in one rat. In non-starved rats, the total recovery ranged between 83% and 94%. Most activity was found in the liver: 40–43% of applied radioactivity (20–28% in triglycerides, 15–19% in phospholipids and only traces of free palmitic acid), 6–8% in adipose tissues but less than 1% in 2 g muscle (mainly as triglycerides in both tissues). Only 8–11% of applied radioactivity

was detected in all other internal organs (mainly digestive tract) but 5–8% in the skin and 21–28% in carcass. Similar results were obtained in starved rats; however, the total recovery was reduced to 50–59% which might be related to rapid metabolism and exhalation of radiolabel (not measured). Overall, most of the radioactivity was found in the liver after i.v. injection of labelled free palmitic acid.

Olivecrona (1962) administered 5 μCi of ^{14}C-labelled palmitic acid (bound to albumin) via i.v. injection to male Sprague–Dawley rats (number of animals not stated). Data showed that 30% of the label was taken up by the liver within 5 min where almost 97% of the label was present in the esterified form. More label appeared in the hepatic neutral fat fatty acids than in the phospholipid fraction and it disappeared more rapidly from neutral fat, suggesting rapid turnover. Similar relations were found for plasma neutral fat fatty acids versus phospholipids. One h after injection 3% of the applied radioactivity was detected in adipose tissue. Eighty minutes after application the levels of ^{14}C in plasma and liver lipids were similar suggesting equilibration of the two pools. However, the carcass lipid pool was still lower than plasma and liver pool even after 24 h.

Clandinin et al. (1988), administered [1-^{13}C]-palmitic acid and [16-^{13}C]-palmitic acid (in capsule form) with a breakfast meal to four male healthy volunteers (22–27 years old) at a dose 0.5–0.75 mg ^{13}C/kg bw. The $^{13}CO_2$ was measured in exhaled air. Chain shortening of palmitic acid was examined by comparing oxidation rates of [1-^{13}C]-palmitic acid vs [16-^{13}C]-palmitic acid. The whole-body rate of oxidation of [1-^{13}C]-palmitic acid was significantly greater than that observed for [16-^{13}C]-palmitic acid. These results suggested that up to 34% of dietary palmitic acid consumed may be subjected to chain shortening.

Stearic acid

Animal studies

Carroll and Richards (1958) studied the digestibility of stearic acid in rats (Sprague–Dawley and Wistar rats; only limited information was provided on methods and animals). The animals were fed a diet containing 10% unlabelled stearic acid (equivalent to 11,800 mg/kg bw per day) for 16 days. The digestibility of stearic acid was measured by analysis of ingested fat and the fat excreted via faeces. The authors calculated a digestibility of 24%. For comparison, a digestibility of 40% was obtained in similar trials with palmitic acid and 73% with oleic acid.

De Leo and Foti (1961) administered 200 μCi 1-^{14}C labelled stearic acid in 1 mL olive oil via gavage to four male Wistar rats. The results showed the formation of labelled cholesterol, which was widely distributed in the organism within 24 h after administration; the highest activity of cholesterol was noted in liver and serum.

Oleic acid

Animal studies

Bergström et al. (1954) studied the intestinal absorption of oleic acid in five male rats (strain not given) with cannulated thoracic lymph ducts, after gavage of 0.5 mL/rat of [1-^{14}C]-oleic acid (no further data). The lymph and expired air were collected for 24 h, after which the animals were sacrificed and radioactivity in the whole gastrointestinal tract plus faeces were measured. A mean of 78% of the fed activity (range: 69–92%) was absorbed in 24 h and 50% of the applied radioactivity was recovered in the lymph lipids from the thoracic duct. About 2% of the activity found in the lymph lipids was in the form of phospholipids and 98% in the form of glycerides. Only 4% of applied activity was exhaled via labelled CO_2 in cannulated rats but 17% in a rat without a thoracic duct fistula. The authors concluded that oleic acid was transported via the thoracic lymph duct and incorporated mainly into triglycerides.

Carroll and Richards (1958) measured the digestibility of oleic acid by analysis of ingested fat and of fat excreted via faeces in rats (Sprague–Dawley and Wistar rats; only limited information was provided on methods and animals) fed a diet containing 10% (equivalent to 11,800 mg/kg bw per day) unlabelled oleic acid for 16 days. The authors calculated a digestibility of 73%, but lower values were obtained in similar trials with saturated fatty acids: 40% digestibility for palmitic acid and only 24% for stearic acid.

In a similar experiment by Gallagher et al. (1965) with male Sprague–Dawley rats, the thoracic lymph duct was cannulated prior to application via a gastrostomy tube of 0.1 mL [U-^{14}C]-oleic acid (purity 99.5%) diluted with unlabelled oleic acid (no further details given). The lymph was collected for 12 h after feeding the labelled oleic acid. Thereafter, the animals were sacrificed and the radioactivity in the whole gastrointestinal tract plus faeces was determined. After application into the stomach, the mean absorption of applied radioactivity from the gastrointestinal tract was 82% ($n = 12$) and radioactivity detected in the lymph lipid within 12 h after application was 69% (mainly in lymph

obtained the first 4 h). Only a small amount of label in the lymph lipid was identified as free oleic acid (4%) but essentially in triglycerides. After application into the duodenum (n = 22) the absorption of applied radioactivity decreased to 68%.

Kern and Borgström (1965) studied the absorption of [9,10-^{3}H]-labelled oleic acid in albino rats (16 animals; weight, 150–200 g). A suspension (3 mL) of the test item containing skimmed milk, glucose, unlabelled oleic acid or labelled oleic acid (3 μCi) was administered via gavage, and the animals were sacrificed 2, 4, 6 or 9 h after application. The gastrointestinal tract along with its contents were removed and analysed for radioactivity. In the gastrointestinal tract, after the given time intervals, 72%, 62%, 49% or 29% respectively, of the applied radioactivity was detected. Significantly lower values were measured in similar trials after application of a suspension containing the bile salt sodium taurodesoxycholate (120 μM).

Hamilton (1971) studied the absorption of oleic acid *in vivo* by the rat jejunum, measuring the uptake, incorporation into triglyceride and transport out of the intestine by tied loops of jejunum and, *in vitro,* by measuring the incorporation into triglyceride by jejuna slices. For that purpose, 0.25 mL of lipid solution in 15 mM sodium taurocholate containing 2.4 mg/mL of [^{14}C]-oleic acid (purity 96%) at pH 5.8 or 7.2 (emulsion at pH 5.8 or micellar solution at pH 7.2) were placed in two different rat jejunal loops of male Sprague–Dawley rats. The loops were washed-out at 'zero' time and at 10 or 60 s, 5 or 30 min. For each trial, 4–10 rats were used. There was a rapid initial uptake of labelled oleic acid into the mucosa, with a slower incorporation into triglyceride and disappearance out of the loop. The absorption of radioactivity at pH 5.8 and pH 7.2 were similar at all time points. Absorption was related only to the total oleic acid concentration, regardless of physical form (i.e. emulsion or micellar solution) in which the oleic acid was present.

Cunningham and Lawrence (1977) administered by gavage 15 μCi [^{3}H]-labelled oleic acid in 1 mL olive oil to four male Wistar rats (100 g bw). After 24 h, 93% of the applied radioactivity was absorbed from the gastrointestinal tract, only 7% of the radioactivity was detected in the faeces (minor amounts were present in bile from separately treated animals collected over 3 days), 4% of the radioactivity was excreted via the urine.

Hollander et al. (1984) studied the intestinal absorption of oleic acid [1-^{14}C]-labelled, purity > 99%, activity 56 Ci/mol) at low intraluminal concentrations and the influence of luminal factors on its absorption, in male Sprague–Dawley rats. The labelled test solution was applied into the small bowel via an inflow tube at 1 cm distal to the common bile duct; a glass tube, 50 cm distal to the infusion site, was used to cannulate the intestine for sampling the outflow for a period of 1 h in 10 min aliquots followed by analysis of radioactivity. The labelled oleic acid was diluted with unlabelled oleic acid (purity 99%). Concentrations of 30–2,500 μM oleic acid in a 10 mM sodium taurocholate micellar solution (pH 6.5) were tested (n ≥ 3 per trial). Absorption of oleic acid increased as the pH was decreased and taurocholate concentrations were increased or as the thickness and resistance of the unstirred water layer were diminished or following addition of lysolecithin. The additions of Tween-80 (polysorbate 80), lecithin, linoleic-, linolenic- or arachidonic acid to the perfusate decreased the rate of absorption of oleic acid. The authors concluded that oleic acid absorption displayed apparent saturation kinetics which was due to unstirred layer effects, limited aqueous solubility of oleic acid and possible saturation of cytosol fatty acid binding proteins. Factors which increased oleic acid's protonated concentration or diminished the unstirred layer resistance enhanced its absorption rate. On the contrary, factors which enhanced its micellar solubility or interfere with its transfer out of the cell membrane decreased its overall rate of absorption.

Human studies

Pinto Correia et al. (1963) studied the absorption of oleic acid in 15 healthy human volunteers (no further details given). The test subjects received a test meal containing 30 μCi ^{139}I-C_1-labelled oleic acid suspended in an emulsion of 90 mL milk and 10 mL olive oil, followed by 150 mL milk. Blood samples were collected 1, 2, 4, 6, 12 and 24 h after ingestion and stools for up to 120 h. Radioactivity in blood samples reached a maximal mean value of 2.8% of applied activity per litre of blood 4 h after treatment. Data on radioactivity excreted via faeces indicated that 3.1% (range: 0.03–8.6%) of applied radioactivity was detected in the faeces.

Overall, the Panel considered that caprylic-, capric-, oleic-, lauric-, palmitic-, myristic- or stearic acid like other fatty acids were readily and extensively absorbed from the gastrointestinal tract. After absorption, fatty acids were either metabolised or incorporated into chylomicrons, which enter the systemic circulation. Ultimately, fatty acids, either incorporated into glycerides and phospholipids, were

catabolised via the β-oxidation pathway and the tricarboxylic acid cycle to carbon dioxide which is finally excreted via exhalation.

3.5.2. Acute oral toxicity

Smyth et al. (1962) studied the acute oral toxicity of undiluted caprylic- or capric acid (mixed isomers tested) in groups of five non-fasted male Carworth–Wistar rats. Based upon mortalities during the 14-day observation period, the authors reported median lethal dose (LD_{50}) values of 1,300 mg/kg bw for caprylic acid and 3,300 mg/kg bw for capric acid (no further details available).

Jenner et al. (1964) administered, by gavage, increasing doses of caprylic acid to groups of five male and five female fasted Osborn–Mendel rats (dose range 8,190–12,370 mg/kg bw). The treated rats showed depression and diarrhoea during the 2-week observation period. The rats died between 4 h and 9 days after application. The authors calculated a LD_{50} of 10,080 mg/kg bw for male and female rats (no further details given).

International-BioResearch (1974, as referred to by CIR, 1987) determined the acute oral toxicity in groups of five male albino rats. Animals were administered by gavage lauric-, myristic-, palmitic- or stearic acid with increasing doses of up to 10,000 mg/kg bw and oleic acid up to 20,000 mg/kg bw. It was observed that for all these fatty acids the LD_{50} value was above the maximum level tested.

No mortality was observed in five albino rats gavaged with 5 g/kg bw oleic acid (commercially supplied); clinical signs were not reported during the 7-day post-exposure period (CTFA, 1978, as referred to in CIR, 1987).

The Panel noted that caprylic-, capric-, oleic-, lauric-, palmitic-, myristic- or stearic acid have a low acute toxicity.

3.5.3. Short-term and subchronic toxicity

Rats

Male Osborne–Mendel albino rats (5 rats/group) weighing between 40 and 50 g were fed 0 or 10% (equivalent to 0 and 9,000 mg/kg bw per day) lauric acid in their diet for 18 weeks (Fitzhugh et al., 1960). Animals were weighed weekly and mortalities and physical condition, appearance and behaviour were observed during the study period. According to the authors, haematological examinations (no details provided) were performed at the end of the study. Organ weights were recorded and gross pathology performed. No adverse effects were observed on any of the measured parameters.

Albino rats (10 animals of both sexes and mixed strain per group) were given a rice diet with 10% (equivalent to 9,000 mg/kg bw per day) capric-, lauric- or palmitic acid for a maximum of 150 days (Mori, 1953). Interim sacrifices were performed throughout the experiment and stomachs were examined for gross lesions. According to the author, no remarkable changes were detected in the forestomach or glandular stomach.

Rats (Holtzman strain) were exposed to a diet containing 50.4% palmitic- or stearic acid for 24 weeks (Herting and Crain, 1958; Herting et al., 1959). Control group was given 50% lard. Rats were sacrificed at 8-week intervals and examined grossly. A microscopic 'foreign body-type reaction' in adipose tissue has been observed from the first sacrifice (8 weeks). The Panel noted that the doses were very high.

No data were available for caprylic, myristic and oleic acid.

Overall, the Panel considered that there was no evidence for toxic effects of fatty acids in subchronic toxicity feeding studies at dose levels up to 10% in the diet (equivalent to 9,000 mg/kg bw per day). However, the Panel noted that the available studies were not conducted according to the current guidelines.

3.5.4. Genotoxicity

In vitro

Caprylic acid

In the study by Litton Bionetics (1976), caprylic acid (purity 98%) was evaluated for its mutagenicity in the bacterial reverse mutation assay using *Salmonella* Typhimurium strains TA1535, TA1537 and TA1538, and for induction of mitotic recombination in *Saccharomyces cerevisiae* strain D4 using both the plate incorporation and pre-incubation methods in the absence and presence of liver S9 metabolic activation from rat mouse and monkey. Concentrations used, selected from survival tests, were 2.5, 1.25 or 0.625 μg/plate for the bacterial reverse mutation assay and 13, 6.5 or 3.25 μg/plate for induction of mitotic recombination. Negative results were obtained for both mutagenicity and

mitotic recombination. The Panel noted that the set of *S.* Typhimurium tester strains was limited and that the mitotic recombination assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

In the study by Zeiger et al. (1988), caprylic acid (purity 99%) was assessed for its mutagenicity in the reverse mutation assay using *S.* Typhimurium strains TA1535, TA1537, TA97, TA98 and TA100 up to a maximum concentration of 3,333 μg/plate in dimethylsulfoxide (DMSO), both in the absence and presence of rat and Chinese hamster S9 metabolic activation at 10% and 30%, and no mutagenicity was observed. The Panel noted that the study complies with current OECD Guideline No 471 (OECD, 1997) with the exception that tester strains *S.* Typhimurium TA102 or *Escherichia coli* WP2uvrA bearing AT mutation were not used.

In the study by Heck et al. (1989), caprylic acid (unknown purity) was tested for induction of gene mutation in the bacterial reverse mutation assay *S.* Typhimurium strains TA98, TA100, TA1535, TA1537 and TA1538 with and without S9 metabolic activation at single concentration of 5,000 μg/plate and for the induction of unscheduled DNA synthesis (UDS) in hepatocytes freshly obtained from male Fisher rats at final concentration of 300 μg/mL. Negative results were obtained in both assays. However, the Panel noted that the reporting of data was marginal and that tester strains *S. Typhimurium* TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Capric acid

Szybalski (1958) reported that capric acid (unknown purity) in a bacterial mutation test (no details reported) did not induce streptomycin-independent revertants of *E. coli* using the paper disc method. The Panel noted that the method employed does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

Oda et al. (1978) assessed the DNA-modifying effects of capric acid at concentration of 18 μg/disk by the Rec-assay (DNA repair test) using *Bacillus subtilis* mutant strain M45 rec-, unable to repair DNA damage and the wild-type strain H17 rec+ as control. Negative results were obtained. However, the Panel noted that the results obtained are of limited relevance since this assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

In the study by Zeiger et al. (1988), capric acid (purity 99%) was assessed for its mutagenicity in the reverse mutation assay using *S.* Typhimurium strains TA1535, TA1537, TA97, TA98 and TA100 up to a maximum concentration of 666 μg/plate in DMSO, both in the absence and presence of rat and Chinese hamster S9 metabolic activation at 10% or 30%, and no mutagenicity was observed. The Panel noted that the study complies with current OECD Guideline No 471 (OECD, 1997) with the exception that tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Lauric acid

Parry et al. (1981) reported that lauric acid (no details on purity provided), in *Saccharomyces cerevisiae* D6, induced aneuploidy at concentrations from 10 to 200 μg/mL. However, analysis of crossing over of chromosomes in the same cell cultures revealed no effects. No data were available on cytotoxicity but it was stated that increased yeast cell lethality might influence the results on aneuploidy. The Panel noted that the test does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

In the study by Zeiger et al. (1988), lauric acid (purity 99%) was assessed for its mutagenicity in the reverse mutation assay using *S.* Typhimurium strains TA1535, TA1537, TA97, TA98 and TA100 up to a maximum concentration of 666 μg/plate in DMSO, both in the absence and presence of rat and Chinese hamster S9 metabolic activation at 10% or 30%, and no mutagenicity was observed. The Panel noted that the study complies with current OECD Guideline 471 No 471 (OECD, 1997) with the exception that tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Myristic acid

In the study by Zeiger et al. (1988), myristic acid (purity 98%) was assessed for its mutagenicity in the reverse mutation assay using *S.* Typhimurium strains TA1535, TA1537, TA97, TA98 and TA100 up to a maximum concentration of 3,333 μg/plate in DMSO, both in the absence and presence of rat and Chinese hamster S9 metabolic activation at 10% or 30%, and no mutagenicity was observed. The Panel noted that the study complies with current OECD Guideline No 471 (OECD, 1997) with the exception that tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Heck et al. (1989) tested myristic acid (unknown purity) in the bacterial reverse mutation assay in the presence and absence of metabolic activation using *S.* Typhimurium strains TA98, TA100, TA1535, TA1537 and TA1538 at concentrations up to 10,000 μg/plate. Negative results were obtained. Furthermore, myristic acid did not induce gene mutations in the mouse lymphoma assay (L5178Y TK+/− cells) at concentrations of up to 62 μg/mL (without metabolic activation) or 125 μg/mL (with metabolic activation) and UDS in hepatocytes freshly obtained from male Fisher rats at final concentration of 300 μg/mL. However, the Panel noted that the results obtained are of limited validity since only one concentration was used in both assays, reporting of data was marginal and that tester strains *S. Typhimurium* TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Stearic acid

Parry et al. (1981) reported that stearic acid (no details provided) did not induce aneuploidy in *S. cerevisiae* D6 at concentrations up to 500 μg/mL. Additionally, analysis of crossing over of chromosomes in the same treated cell cultures revealed no effects. The Panel noted this assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

Blevins and Taylor (1982) reported that stearic acid (unknown purity) did not induce mutagenic effects in a bacterial reverse mutation assay with *S.* Typhimurium strains TA98, TA100, TA1535, TA1537 and TA1538 with and without rat liver metabolic activation, at a concentration of 50 μg/plate. However, the Panel noted that the results obtained are of limited validity since only one concentration was used, reporting of data was marginal and that tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

Crebelli et al. (1985) tested stearic acid (unknown purity) in the plate incorporation assay with *S.* Typhimurium strains TA98, TA100, TA1535 and TA1537 in the presence of metabolic activation (S9 from Aroclor induced male Sprague-Dawley rats) at concentrations of 40-1,000 μg/plate. DMSO was used as solvent. The test included positive controls. No mutagenic activity was found. The Panel noted that only treatments in the presence of S9 metabolic activation were used and that tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not used.

In the study by Shimuzu et al. (1985), stearic acid (60% purity) was assessed for its mutagenicity in a bacterial reverse mutation assay using *S.* Typhimurium strains TA98, TA100, TA1535, TA1537, TA1538 and *E. coli* WP2uvrA. The pre-incubation test was performed in the presence and absence of metabolic activation (liver S9 from polychlorinated biphenyl induced male rats) at concentrations of 1–1,000 μg/plate (seven concentrations) including negative, vehicle and positive control. The test substance did not induce an increase in revertants colonies compared to the concurrent negative control. The Panel noted that the study complies with current OECD Guideline 471 No 471 (OECD, 1997).

Oleic acid

Cotruvo et al. (1977) assessed the mutagenic potential of oleic acid (no details reported) in a bacterial reverse mutation assay in *S.* Typhimurium strains TA98, TA100, TA1535, TA1536, TA1537 and TA1538, and in the *Saccharomyces cerevisiae* D3 mitotic recombination assay with and without metabolic activation. No genotoxic activity was reported in both assays at maximum solubility of 99 μg/mL. The Panel noted that the tester strains *S. Typhimurium* TA102 or *E. coli* WP2uvrA bearing AT mutation were not used and that the mitotic recombination assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

Mortelmans et al. (1986) tested oleic acid (technical grade) in the bacterial reverse mutation assay, in *S.* Typhimurium strains TA98, TA100, TA1535 and TA1537 in the presence and absence of metabolic activation (S9-mix from hamster or rat liver). The test substance was dissolved in DMSO and tested up to the toxicity threshold (3.3–10 μg/plate without metabolic activation and 333 μg/plate with metabolic activation). No mutagenic activity was observed. The Panel noted that the tester strains *S.* Typhimurium TA102 or *E. coli* WP2uvrA bearing AT mutation were not employed.

Parry et al. (1981) reported that oleic acid (no details provided) induced aneuploidy in *S. cerevisiae* D6 at concentrations from 100 to 500 μg/mL. However, analysis of crossing over of chromosomes in the same cell cultures revealed no effects. No data were available on cytotoxicity but increased yeast cell lethality might influence the results on aneuploidy. The Panel noted that the test does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

Osawa and Namiki (1982) assessed the DNA-modifying effects of oleic acid (unknown purity) at concentration of 100 or 1,000 μg/disk by the Rec-assay (DNA repair test) using *Bacillus subtilis* mutant strain M45 rec-, unable to repair DNA damage and the wild-type strain H17 rec+ as control. Negative results were obtained. However, the Panel noted that the results obtained were of limited relevance

since this assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

Kinsella (1982) studied the induction *in vitro* of forward mutations after exposure to oleic acid by the mammalian cell gene mutation assay in V79 6-thioguanine resistant Chinese hamster lung fibroblasts without metabolic activation. At a concentration of 1 μg/mL, no increase in the mutation frequency compared to the negative concurrent control was observed. In further experiments using also V79 cells, sister chromatid exchange (SCE) analysis and quantitation of induced chromosomal aberrations in growing cells exposed simultaneously to 5-bromo-2'-deoxyuridine and the test agents were performed. No DNA damage and no clastogenic effects were detected at concentrations of 2.5, 5 or 10 μg oleic acid/mL. However, the Panel noted that the study design had significant limitations which included single concentration in the mammalian cell gene mutation assay, very low concentrations (1–10 μg/mL), the absence of treatment in the presence of S9 metabolic activation and no indication of sampling time. On this basis the results obtained were not considered reliable for risk assessment.

Shimuzu et al. (1985), in a bacterial reverse mutation assay using *S. Typhimurium* strains TA98, TA100, TA1535, TA1537 and TA1538, and *E. coli* WP2uvrA assessed the mutagenicity of oleic acid (99.9% purity) dissolved in acetone. The pre-incubation test was performed in the presence of metabolic activation (liver S9 from polychlorinated biphenyl induced male rats) at concentrations of 1-5,000 μg/plate (eight concentrations) including negative, vehicle and positive control. In TA1537 and TA1538, the test substance was tested up to the cytotoxic doses (100 μg/plate). The test substance did not induce any increase in revertant colonies compared to the concurrent negative control. The Panel noted that the study complied with current OECD Guideline No 471 (OECD, 1997).

In the study by Higgins et al. (1999), oleic acid (99% purity) was assessed for the induction of the SCE in the Indian muntjac fibroblasts, complexed with 2% bovine serum albumin without metabolic activation at a single concentration of 50 μM. Analytical control revealed significant uptake of oleic acid by the cells but no genotoxic effects were reported. The Panel noted severe limitations of the study protocol which included the use of a single concentration, treatments performed only in the absence of S9 metabolism, inadequate selection of the highest concentration which was low and did not induce appropriate reduction of mitotic index. In addition, the SCE assay does not belong to the genotoxicity tests recommended for regulatory purposes (EFSA Scientific Committee, 2011).

In vivo

Fang et al. (2007) administered oleic acid (purity 99%), coconut oil or linoleic acid at a dose of 500 mg/kg bw per day by gavage for 30 days to young adult BD VI rats (4–6 rats/sex per group). The animals were sacrificed and the blood, liver, colon, prostate (males) and breast tissue (females) was collected. Etheno-DNA adducts (1,*N*(6)-etheno-2'-deoxyadenosine (εdA) and 3,*N*(4)-etheno-2'-deoxycytidine (εdC)) were measured in white blood cells and colon, liver, mammary and prostate tissue by immunoaffinity/^{32}P-post-labelling. These adducts are highly miscoding DNA lesions in mammalian cells, involved in the initiation of the carcinogenic processes through specific point mutations. Results obtained indicated that etheno-DNA adducts were significantly elevated in colon cells in both sexes and in white blood cells in female animals only, following treatment with linoleic acid compared to the levels induced by treatments with oleic acid and coconut oil. In contrast, oleic acid, unexpectedly, induced significant increases of both etheno-DNA adducts levels (εdA and εdC) in prostate and εdA adduct levels only, in mammary gland which were 3–9- and two fold, respectively, the levels observed for linoleic acid. However, all these increases were relatively small in absolute terms compared to those observed in the female colon following linoleic acid treatments (the Panel noted that there was no negative control group). The explanation provided by the authors indicate that the high adduct levels observed in colon and white blood cells seemed to be a direct effect after cellular uptake of free linoleic acid which, as an ω-6 polyunsaturated fatty acid, is a good substrate for lipid peroxidation leading to the induction of etheno-DNA adducts. Conversely, changes in the adduct level found in the 'distant organs (mammary gland and prostate)' following treatment with oleic acid could be due to altered membrane phospholipid composition. This was because prostate epithelial cells have the capacity to incorporate oleic acid into the membrane phospholipids and, thus oleic- and linoleic acid can compete for incorporation into membranes. Consequently, high intake of oleic acid may cause the replacement of membrane linoleic acid thus increasing the level of free linoleic acid and the consequent production of etheno-DNA adducts. The Panel agreed with this interpretation and considered that oleic acid was not directly involved in the production of etheno-DNA adducts. The Panel noted that linoleic acid does not belong to the fatty acids which are authorised in the food additive E 570.

Overall, the Panel noted that the fatty acids – caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid – did not show a mutagenic potential in the bacterial reverse mutation assay. Caprylic and myristic acid were negative in the UDS assay in rat hepatocytes *in vitro* and myristic acid was negative in the mouse lymphoma L5178Y $TK^{+}/^{-}$ cell mutation assay, although these results were considered of limited validity by the Panel due to significant experimental limitations. Concerning the etheno-DNA adducts observed in one *in vivo* study, the Panel considered that oleic acid was not directly involved in the production of etheno-DNA adducts in different organs of male and female rats. The Panel noted that specific studies for the evaluation of structural and numerical chromosomal aberrations were not available; however, the Panel considered that the available data did not raise a concern for genotoxicity for caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- or oleic acid used as a food additive.

3.5.5. Chronic toxicity and carcinogenicity

Mice

Szepsenwol (1978), Szepsenwol and Boschetti (1975) performed two long-term carcinogenicity studies in T.M. mice. Animals were fed a diet containing 1.5% of a mixture of oleic- and linoleic acid (composition unknown) during more than 400 days. The authors described increase of tumour incidences in the lung, stomach and brain nerve. However, the Panel noted that the report of this study was poorly documented, only one dose was tested, and no information on the composition of the mixture was given. The Panel also noted that linoleic acid does not belong to the fatty acids authorised as the food additive E 570. Due to uncertainties in the design of the study, the Panel considered that this study cannot be taken into account for hazard characterisation.

In the study of El-Khatib and Cora (1981), 55 mice were administered a diet containing oleic acid (10 g of a mixture of 1.5 g oleic acid/100 g corn oil dispersed in 100 g feed (equivalent to 2,250 mg/kg bw per day), and 36 mice were fed with normal diet. Throughout the study, randomly selected mice were killed and examined after 6, 12, 18, 21 and 24 months. According to the authors, colon adenocarcinomas, which metastasised to the lung and muscle, were found in 8% (3/36) of the treated mice. Metastatic colon adenocarcinoma was found in the lung of one of these mice and in the muscle of the second one. In the third mouse, in addition to colon adenocarcinoma, polycystic kidney was also found. No more data were reported on histopathological examinations. Because only one dose was tested, and limited information on tumours in treated mice was given, the Panel considered that this study cannot be taken in account in the hazard characterisation.

Overall, the Panel noted that only limited data on chronic toxicity were available and that, because of the limitations of these studies, no reliable conclusion could be drawn.

Initiation-promotion studies

The Panel noted a series of studies which had investigated potential modulating effects of various fatty acids (lauric acid, oleic acid, myristic acid, capric acid, caprylic acid, stearic acid and palmitic acid) administration on the development of cancer in animal models treated with an initiating agent (Dayton et al., 1977; Jain et al., 1980; Traul et al., 1981; Hogan and Shamsuddin, 1984; Sakaguchi et al., 1984; Cohen et al., 1986; Habib et al., 1987; Lasekan et al., 1990; Hubbard and Erickson, 1991; Numata et al., 1994; Zusman et al., 1997; Kimura, 2002). With the exception of linoleic at high doses, most of these studies have reported protective effects of free fatty acids. The Panel considered these studies not relevant for the risk assessment of fatty acids (E 570) as a food additive.

3.5.6. Reproductive and developmental toxicity

Reproductive toxicity

Rats

Young rats (n = 4/sex per group) were fed 16 weeks premating 15% oleic acid in the diet (equivalent to 7,500 mg/kg bw per day). The animals were mated within the test group and four litters were born (Carroll and Noble, 1957). The study was very limited (number of animals and reporting). The Panel considered that this study cannot be used for the hazard characterisation.

Developmental toxicity

Rats

Narotsky et al. (1994) studied the developmental toxicity of caprylic acid in a screening test (a limited number of parameters were tested). Pregnant Sprague–Dawley rats were administered by gavage with caprylic acid (purity 99.5%) in corn oil on gestation days (GD) 6–15 at dose levels of 0 (20 animals), 1,125 mg/kg bw per day (16 animals) or 1,500 mg/kg bw per day (16 animals). The dams were allowed to deliver and their litters were examined through post-natal day 6. The treatment induced maternal toxicity: at the low dose level, rales were observed in 15/16 rats and dyspnoea in 4/16; similar incidences were found at the high dose. Five low-dosed rats and seven high-dosed rats died during the treatment period while no deaths were observed in the controls. Maternal body weight gain was significantly and dose dependently reduced. According to the authors the maternal effects might be influenced by application error; the authors stated that 'death may have been due to paragastric intubation'. The treatment did not affect the number of implantations/dam, perinatal loss, number of live pups at post-natal day 1 and pup weight at post-natal day 1 and 6. At the high dose level (1,500 mg/kg bw per day) but not at 1,125 mg/kg bw per day, the number of live pups at post-natal day 6 was statistically significantly reduced. This decreased viability occurred only in litters of dams with peripartum respiratory symptoms. External malformations were not observed. Overall, the Panel noted that in this study no developmental toxicity occurred at a maternal toxic dose of 1,125 mg/kg bw per day. Developmental toxicity at a dose of 1,500 mg/kg bw per day was related to maternal toxicity which may have been induced by application error rather than by the test item. However, the Panel noted that the oral limit dose according to OECD Guideline 414 is 1,000 mg/kg bw per day. The Panel also considered that the study design of this study was too limited to conclude on prenatal developmental toxicity.

In the study of Scott et al. (1994), a group of 12 pregnant Sprague–Dawley rats received via gavage a single dose of 2,700 mg caprylic acid (undiluted)/kg bw at GD 12. The control group (10 animals) was untreated. Caesarean section was performed at GD 20 and implantations, resorptions and dead fetuses were counted. The fetuses were removed, sexed, weighed and examined for external malformations. Two-third of the fetuses were examined for internal malformations and the others for skeletal malformations. The treatment resulted in severe maternal toxicity (no further information provided) but did not induce any developmental toxicity except a decrease in fetal weight (no data about statistical significance given) which was attributed to maternal toxicity. Based on these findings, the authors concluded that caprylic acid did not induce developmental toxicity. The Panel noted that the design of this study was very limited (low number of animals and inappropriate dosing regimen).

The Panel noted that insufficient data were available on reproductive toxicity of fatty acids. Limited data on developmental toxicity were available only for caprylic acid.

Overall, the Panel considered that data on reproductive and developmental effects were too limited to reach a conclusion on these endpoints.

3.5.7. Other studies

Observations in humans

Jain et al. (1980) conducted a case–control study of colorectal cancer in which 348 cases of colon cancer and 194 cases of rectal cancer were individually matched (by age, sex and neighbourhood of residence) to 542 population controls, and frequency matched to 535 hospital controls who had undergone an abdominal operation. According to the authors, the results of the study supported the hypothesis that high dietary fat intake is causally associated with cancers of the colon and rectum. The authors also stated that, although the high correlation among several nutrients made it difficult to single out a specific risk factor, higher risks were observed for saturated fat than for calories, total protein, total fat, oleic acid or cholesterol intake. The Panel considered that given the high fat intake and the low contribution of the fatty acids from the use as food additives, this study was not relevant for the assessment of fatty acids as a food additive E 570.

From the available data, it can be concluded that, in humans, a diet enriched in fat increased the level of blood lipids: clinical studies showed that a stearic acid-rich diet decreased plasma cholesterol concentrations (Dougherty et al., 1994); a lauric and myristic acid-rich diets produced a higher serum cholesterol concentration than does a palmitic acid-rich diet (Sundram et al., 1994); in comparison to oleic acid-rich diet, myristic acid and palmitic acid caused increased low-density lipoprotein cholesterol and apolipoprotein B levels and low ratios of high-density lipoprotein cholesterol to low-density

lipoprotein cholesterol ratios (Zock et al., 1994); a myristic acid-enriched diet raised in human subjects both the low-density lipoprotein and high-density lipoprotein cholesterol concentrations compared with oleic acid (Temme et al., 1997). In contrast, one study reported that a myristic acid containing fat was not more cholesterolaemic than was palmitic acid-rich diet in a controlled randomised dietary study (Tholstrup et al., 1994). The acute prothrombotic effect of fats high in myristic or stearic acid was not confirmed in a clinical study (Tholstrup et al., 1996). In a case report, eosinophilic pneumonia was caused by oral calcium stearate (Kurai et al., 2006); however, such effects were not confirmed by any other publication although calcium stearate is a common component of medical products.

Overall, from the available data, the Panel considered that fatty acids may have some adverse health effects. However, the Panel noted that the fat intake was high with regards to the intake from the use of fatty acids (E 570) as a food additive. Therefore, these studies were not relevant for the safety assessment of the food additive E 570.

3.6. Discussion

The Panel noted that the food additive (E 570) is considered in Commission Regulation (EU) No 231/2012 as a group of substances composed of six linear saturated fatty acids (caprylic-, capric-, lauric-, myristic-, palmitic- and stearic acid) and one *cis*-monounsaturated fatty acid (oleic acid), but that no indication is given as regards the level at which an individual acid may be present in the group, except that the total percentage of the fatty acids should not be less than 98%.

The Panel considered that caprylic-, capric-, oleic-, lauric-, palmitic-, myristic- or stearic acid like other fatty acids are readily and extensively absorbed from the gastrointestinal tract. After absorption, fatty acids are either metabolised or incorporated into chylomicrons, which enter the systemic circulation. Ultimately, fatty acids, either incorporated into glycerides and phospholipids, are catabolised via the β-oxidation pathway and the tricarboxylic acid cycle to carbon dioxide which is finally excreted via exhalation.

From the available feeding studies on subchronic toxicity, although limited, the Panel considered that there was no evidence for toxic effects of fatty acids at dose levels up to 10% in the diet (equivalent to 9,000 mg lauric acid/kg bw per day). However, the Panel noted that these studies were not conducted according to the current guidelines.

Specific studies for the evaluation of structural and numerical chromosomal aberrations were not available; however, the Panel considered the available genotoxicity data did not raise a concern for genotoxicity for caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- or oleic acid used as a food additive.

Only limited data on chronic toxicity were available and because of the limitations of these studies no valuable conclusion could be drawn.

Insufficient data were available to conclude on reproductive toxicity of fatty acids and limited data on developmental toxicity were available only for caprylic acid.

From the available human data, the Panel noted that ingestion of fatty acids may have some adverse health effects. However, the fat intake was high compared to the intake from the use of fatty acids (E 570) as a food additive. Therefore, these studies were not relevant for the safety assessment of the food additive E 570.

Although some of the fatty acids which are included in the food additive (E 570) have been reported to be recognised by specific cellular receptors, the Panel considered it outside the scope of this evaluation to review the extensive literature available about the effects of free fatty acids and the consequence of their binding to their specific receptors. The Panel considered that although it is relevant to consider the possible effects resulting from this binding, this should be done in the perspective of a comparison between the exposure resulting from the use of free fatty acids used as a food additive and their intake from the regular diet. Given that the amount of fatty acids in food from all sources present in the regular diet markedly exceeds the intake of free fatty acids resulting from their use as a food additive, the Panel considered that receptor binding with fatty acids derived from their use as food additive was unlikely to give rise to adverse effect(s).

For the exposure assessment of E 570, reported uses were available for only 8 out of 67 food categories in which E 570 is authorised. This might indicate a limited use of E 570 as a food additive in Europe. The Panel noted that the information from the Mintel GNPD supported the observation that fatty acids (caprylic-, capric-, lauric-, myristic-, palmitic-, stearic- and oleic acid) are apparently not used in all food categories in which the food additive E 570 is authorised.

The Panel noted that the dietary exposure to E 570 was low (on average 1%) when compared to the total daily exposure to saturated fatty acids via the regular diet (EFSA NDA Panel, 2010), either free or incorporated into glycerides and phospholipids. The intake of fatty acids (E 570), as well as that of saturated fatty acids via the regular diet, was far below 9,000 mg/kg bw per day (for lauric acid) at which in subchronic toxicity studies no adverse effects were observed. According to the EU specifications, E 570 may contain only one particular fatty acid or a mixture of the seven fatty acids, which are authorised within the food additive E 570. Accordingly, the exposure assessment referred to E 570 and not to a specific fatty acid.

4. Conclusions

According to the conceptual framework for the risk assessment of certain food additives re-evaluated under Commission Regulation (EU) No 257/2010 (EFSA ANS Panel, 2014) and given that:

- the safety assessment carried out by the Panel was limited to the use and use levels received from industry in eight food categories out of 67 food categories in which fatty acids (E 570) is authorised;
- fatty acids used as a food additive (E 570) were absorbed in the same way as the free fatty acids from the regular diet;
- fatty acids used as a food additive (E 570) were metabolised in the same way as fatty acids when derived from lipid molecules present in the regular diet;
- the toxicity database was limited, however, no adverse effects were observed in subchronic toxicity studies up to 10% in the diet (equivalent to 9,000 mg lauric acid/kg bw per day);
- there was no genotoxicity concern for these fatty acids;
- the contribution of fatty acids (E 570) represented on average only 1% of the overall exposure to saturated fatty acids from all dietary sources (food additive and regular diet);

the Panel concluded that the food additive fatty acids (E 570) was of no safety concern at the reported uses and use levels.

5. Recommendations

The Panel recommended that:

- the European Commission considers lowering the current limits for toxic elements (arsenic, lead and mercury) in the EU specifications for fatty acids (E 570) in order to ensure that fatty acids (E 570) as a food additive will not be a significant source of exposure to those toxic elements in food;
- since only data for eight out of the 67 food categories in which fatty acids (E 570) is authorised were available, more information on uses and use levels should be made available to the Panel in order to perform a more accurate exposure assessment.

Documentation provided to EFSA

1) AESGP (Association of the European Self-Medication Industry), 2016. Data on usage levels of Fatty acids (E 570) in foods in response to the EFSA call for food additives usage level and/or concentration data in food and beverages intended for human consumption (Batch 4). Submitted to EFSA on 27 May 2016.
2) Dr. Loges - Naturheilkunde neu entdecken, 2016. Data on usage levels of Fatty acids (E 570) in foods in response to the EFSA call for food additives usage level and/or concentration data in food and beverages intended for human consumption (Batch 4). Submitted to EFSA on 27 April 2016.
3) FDE (FoodDrinkEurope), 2016. Data on usage levels of Fatty acids (E 570) in foods in response to the EFSA call for food additives usage level and/or concentration data in food and beverages intended for human consumption (Batch 4). Submitted to EFSA on 31 May 2016
4) ICGA (International Chewing Gum Association), 2016. Data on usage levels of Fatty acids (E 570) in foods in response to the EFSA call for food additives usage level and/or

concentration data in food and beverages intended for human consumption (Batch 4). Submitted to EFSA on 31 May 2016.

5) KRÜGER GmbH & Co. KG, 2016. Data on usage levels of Fatty acids (E 570) in foods in response to the EFSA call for food additives usage level and/or concentration data in food and beverages intended for human consumption (Batch 4). Submitted to EFSA on 25 May 2016.
6) Pre-evaluation document prepared by DTU, March 2014.

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Abbreviations

ADI	acceptable daily intake
AESPG	Association of the European Self-Medication Industry
ANS	EFSA Scientific Panel on Food Additives and Nutrient Sources added to Food
APCI	atmospheric pressure chemical ionisation
bw	body weight

CAS	Chemical Abstracts Service
CEF	EFSA Panel on Food Contact Materials, Enzymes, Flavourings and Processing Aids
CIR	Cosmetic Ingredients Review
CONTAM	EFSA Panel on Contaminants in Food Chain
CTFA	Cosmetic, Toiletry and Fragrance Association
DMSO	dimethylsulfoxide
DRV	dietary reference value
εdA	1,*N*(6)-etheno-2′-deoxyadenosine
εdC	3,*N*(4)-etheno-2′-deoxycytidine
EINECS	European Inventory of Existing Chemical Substances
FAO	Food and Agriculture Organization of the United Nations
FCS	food categorisation system
FDE	FoodDrinkEurope
FFA	free fatty acid
FID	flame ionisation detector
FSMP	foods for special medical purposes
GC	gas chromatography
GD	gestation day
GLC	gas–liquid chromatography
GNPD	Global New Products Database
GPCR	G-protein coupled receptors
ICGA	International Chewing Gum Association
i.v.	intravenous
JECFA	Joint FAO/WHO Expert Committee on Food Additives
LD_{50}	median lethal dose
LOD	limit of detection
MPL	maximum permitted level
MS	mass spectrometry
NDA	EFSA Panel on Dietetic Products, Nutrition and Allergies
NMR	nuclear magnetic resonance
OECD	Organisation for Economic Co-operation and Development
QS	*quantum satis*
rh	relative humidity
SCE	sister chromatid changes
SCF	Scientific Committee on Food
TemaNord	is a publishing series for results of the often research-based work that working groups or projects under Nordic Council of Ministers have put in motion
TLC	thin-layer chromatography
UDS	unscheduled DNA synthesis
WHO	World Health Organization

Appendix A – Summary of reported use levels of fatty acids (E 570) provided by industry (mg/kg or mg/L as appropriate)

Appendix A can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2017.4785

Appendix B – Number and percentage of food products labelled with fatty acids (caprylic, capric, lauric, myristic, palmitic, stearic and oleic acid) out of the total number of food products present in the Mintel GNPD per food subcategory between 2011 and 2016

Appendix B can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2017.4785

Appendix C – Concentration levels of fatty acids (E 570) used in the maximum level exposure scenario, the refined exposure assessment scenarios and food supplements consumers only scenario (mg/kg or mL/kg as appropriate)

Appendix C can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2017.4785

Appendix D – Summary of total estimated exposure of fatty acids (E 570) from its use as a food additive for the maximum level exposure scenario and the refined exposure assessment scenarios per population group and survey: mean and 95th percentile (mg/kg bw per day)

Appendix D can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2017.4785

Appendix E – Main food categories contributing to exposure to fatty acids (E 570) using the maximum level exposure scenario and the refined exposure assessment scenarios (> 5% to the total mean exposure)

Appendix E can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2017.4785

Appendix F – Information on the percentage of free fatty acids in regular foods

Free fatty acids in foods

Triglycerides are the predominant component of most food fats and oils. Free fatty acids (FFA) and other components (i.e. mono- and diglycerides, phosphatides, sterols, fatty alcohols, fat-soluble vitamins, carotenoids) are present in relatively minor amounts.

The actual level of FFA in foods is dependent on the nature of the foodstuffs. FFA can be present either as natural constituents of the foods or derived from the acylglycerols present in the foodstuffs by enzymatically induced (activity of lipases) or thermally induced (heating, frying) reactions. The level depends on reaction-time, temperature and moisture content of the foods (Nawar, 1969; Mehasar et al., 2014).

FFA in oils

FFA in oils are formed by the hydrolysis of triacylglycerols during manufacturing. However, after extraction from the raw materials (i.e. vegetable or animal fats), the crude oil extracts are refined. During refining, the free fatty acids and other secondary products (e.g. phosphatides, pigments and odour compounds) are removed and the purity of triglycerides is increased from 95–96% to about 99% (Blanchard, 1992). However, as the refined oils are exposed to various condition such as storage or processing (heating or frying), FFA can be formed again (Nawar, 1969; Mehasar et al., 2014). Data in the literature indicate that in refined oil the level of FFA is well below 0.5% (in corn oil 0.05%; in olive oil 0.2%) (Blanchard, 1992; Mailer, 2006).

Dia et al. (2005) studied the physicochemical characteristics of virgin coconut oil produced by different methods. In their study the authors determined to FFA content in six commercial samples of virgin coconut oil. In these samples the FFA content varied from 0.06% to 0.32%.

El-Abassy et al. (2009) determined the FFA content in 18 brands of olive oil taken from the retail market. It was shown the percentage FFA, in terms of oleic acid, ranged from 0.14% to 0.40%.

Wei et al. (2013) determined the level of free fatty acids (i.e. C16:0, C18:0, C18:1, C18:2, C18:3, C20:0, C20:1 and C22:0) in rapeseed oil, soybean oil sunflower oil and coconut oil, taken from the retail market. The FFA level was determined in the fresh oil and after 10 days storage of the oil at 60°C. Data show that the total concentration of the FFAs in all fresh oils did not exceed 55 μg/mL (i.e. 0.06%). The highest concentration of total FFAs was found in sunflower oil (53.57 μg/mL; 0.058%) and the lowest concentration of total FFAs was found in corn oil (16.67 μg/mL; 0.18%).

The effect of storage of the oils for 10 days at 60°C on the level of FFA is shown in Table F.1.

Table F.1: Concentration of FFA (μg/mL oil) in vegetable oils, fresh and after 10 days of storage at 60°C

Type of oil	FFA in fresh oil (μg/mL)	% FFA in oil (average oil density: 0.917 g/mL)	FFA after storage (μg/mL)	% increase of FFA after storage
Rapeseed oil	53.15	0.058	63.55	19.6
Soybean oil	49.74	0.054	63.11	26.9
Sunflower oil	53.57	0.058	70.81	32.2
Corn oil	16.67	0.018	25.53	53.1

FFA: free fatty acid.

The authors attributed the increase in FFA to an enhanced degree of lipid peroxidation during the high-temperature storage.

In a study by Lucas-Torres et al. (2014), the thermal degradation of olive oil (from seven different cultivars) using conventional and microwave heating under the same experimental conditions (heating: 240 min/190°C) were compared. The results showed that during conventional heating the percentage level of FFA increased from a mean level of 0.09% to 1.56% under conventional heating and to 0.19% with microwave heating.

FFA in fish

Aro et al. (2000) studied the effects of season and processing on the free fatty acids level in the oil of Baltic Herring (*Clupea harengus membras*). A seasonal effect of the FFA content (expressed as oleic acid) in the oil was observed, varying from 3.2% (in Spring), 3.7% (in Summer), 1.4% (in Autumn) and 1.7% (in Winter). The authors also observed an effect on processing of the fish at factory level: when the fish was used to produce fried fish fillets (frying conditions: 2 min/250°C) the FFA level in the fish oil varied from 2.6% after catch, to 2.8% after filleting in the processing plant and to 1.4% after frying of the fillets. When the fish was used to produce fish burgers (frying conditions: 2 min/170°C), the evolution of the FFA level in the fish oil was as follows: 2.9% after gutting, 4.3% after mincing of the fillets and 1.4% in the fried burger.

FFA in milk and dairy products

Bovine milk typically contains ca 3.5–5 g fat/100 mL. The principal lipids of milk are triacylglycerides which may represent up to 98% of the total lipids. Other milk lipids are diacylglycerol (about 2% of the lipid fraction), cholesterol (less than 0.5%) and phospholipids (about 1%). The level of free fatty acids in milk fat amounts to about 0.1% (Jensen, 2002; Lindmark Månsson, 2008).

In milk, FFA are produced enzymatically by action of the naturally present lipoprotein lipase (LPL). (Fox and Stepaniak, 1993; Fox et al., 1993).

The major FFA found in milk fat are butyric acid (C4.0), caproic (C6.0), caprylic acid (C8:0), capric acid (C10:0), lauric acid (C12:0), myristic acid (C14:0), palmitic acid (C16:0), steric acid (C18:0), oleic acid (C18:1), linoleic (C18:2) and linolenic acid (C18:3) (Jensen et al., 1991). The FFA content of bovine milk is between 0.1 and 0.44 g/100 milk, equal to about, on average, 0.3% (w/w) (Keenan and Patton, 1995). Palmitic acid and stearic acid are the most abundant FFA. (Jensen et al., 1962).

De Jong et al. (1994) estimated the level of FFA in yoghurt with a total fat content of 3%. Following free fatty acids were identified: butyric acid (C4.0), caproic (C6.0), caprylic acid (C8:0), capric acid (C10:0), lauric acid (C12:0), myristic acid (C14:0), palmitic acid (C16:0), steric acid (C18:0) and oleic acid (C18:1). The total FFA content was determined to be 59.96 mg/L yoghurt, equal to 5.7% (w/w). Palmitic acid (C16:0) and oleic acid were present at the highest level (i.e. 17.13 mg/L and 15.65 mg/L respectively).

In cheese, the FFA are produced by lipases and esterases from lactic acid bacteria. The FFA levels in cheese vary very widely depending on the type of cheese. Based on data reported in the literature (Collins et al., 2003), the total amounts of FFA can vary between hard and soft cheeses: e.g. 0.356 g/kg cheese or 0.036% (w/w) in hard Edam cheese and up to 75.7 g/kg cheese or 7.6% (w/w) in some soft blue cheeses (e.g. Gamonedo blue).

Free fatty acids in cereals

Moltenberg et al. (1995) studied the effect of storage and heating on the content and composition of FFA (i.e. palmitic-, stearic-, oleic-, linoleic- and linolenic acid) of hulled and dehulled oat (*Avena sativa* L.) grains. Samples of three cultivars (Kapp, Mustang, and Svea) were stored at 30%, 55% and 80% relative humidity (rh) for 3.5 and 15.5 months before heating. After 3.5 months, the FFA content in the cultivar Kapp was 3.5 and 6.6 mg/g of dry matter when stored at 30% and 80% rh, respectively. After 15.5 months, FFA content increased to 5.4 and 11.3 mg/g of dry matter when stored at 30% and 80% rh, respectively.

To study the effect of heating, the oats were first soaked in water for 2 min and then heated in a baking oven (100°C, 10 min). During processing, the FFA content was reduced by an average of 50%. According to the authors, this reduction was due to complexing of fatty acids with starch and proteins. The total lipid content of oats did not change significantly during processing. The data demonstrated that the FFA content increased with increasing moisture content and time of storage, but decreased during heat processing.

Free fatty acids in a composite food model

Padmashree et al. (2012) studied the effect of the type of packaging material and storage time (3, 6 and 9 months at 37°C) on the evolution of the FFA content of a composite cereal bar that contained

roasted barley, wheat and puffed corn flour, soy protein concentrate, cacao butter and soy lecithin. Data showed an increase in the FFA levels from an initial value of 1.9% (expressed as oleic acid) up to 4.8% when packed in metallised polyester films, up to 4.9% when in paper-aluminium foil polyethylene laminate and up to 5.2% when in polypropylene.