National Institute for Public Health
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Synopsis

Mineral oils in food; a review of toxicological data and an assessment of the dietary exposure in the Netherlands

Mineral oils can be intentionally added to foods after they have been refined or they can end up in food as contaminants. In recent years, such substances have sparked controversy as they can be harmful to health. In 2012, the European Food Safety Authority (EFSA) concluded that the intake of mineral oils via food is of potential concern.

Within the context of this discussion, it's important to distinguish between saturated hydrocarbons (MOSH) and aromatic hydrocarbons (MOAH) in mineral oils, as they have different harmful effects. RIVM research indicates that the limited number of studies on MOSH published since 2012 seem to somewhat reduce the concerns expressed by EFSA. RIVM calculations also indicate that no health effects are to be expected if people are exposed to MOSH via food. Therefore, the focus should be more on MOAH, as some of these substances are carcinogenic. It is however not possible to specify whether the daily intake of these substances is too high, as there is no existing health-based guidance value for MOAH.

Carcinogenic MOAH are mainly found in crude or insufficiently refined mineral oils and in oils that have been heated. Not all sources from which MOAH can end up in food contain carcinogenic MOAH. The total MOAH concentration therefore does not provide information on whether the intake of MOAH is actually harmful. According to RIVM, it's important to determine the specific sources from where MOAH ends up in food. Measures can then be taken to avoid harmful sources as much as possible. An example of a source of contamination is provided by jute bags that are treated with oil and used for packaging cocoa beans.

The contribution from another source of contamination, namely paperboard packaging made from recycled materials, seems to be less of a problem in the Netherlands. These materials are often used for packaging dry foods such as rice, pasta, breakfast cereals, and chocolate sprinkles. The intake calculations carried out make it clear that intake via these foods makes only a small contribution to the total exposure to mineral oils via food. Measures aimed at reducing the exposure resulting from the use of paperboard packaging would therefore have only a limited effect.

Keywords: mineral oil hydrocarbons (MOH), MOSH, MOAH, risk assessment, toxicity, dietary exposure, margin of exposure (MOE), food contact materials, paperboard packaging.

Publiekssamenvatting

Minerale oliën in voedsel; een overzicht van de toxicologische gegevens en een beoordeling van de inname via voedsel in Nederland

Minerale oliën kunnen in gezuiverde vorm bewust aan voedsel worden toegevoegd, of er als verontreiniging in terechtkomen. De laatste jaren is er ophef over ontstaan omdat ze schadelijk voor de gezondheid kunnen zijn. In 2012 oordeelde de European Food Safety Authority (EFSA) dat de inname van minerale oliën via voeding mogelijk zorgwekkend is.

In de discussie is het belangrijk een onderscheid te maken tussen verzadigde koolwaterstoffen (MOSH) en aromatische koolwaterstoffen (MOAH) in minerale oliën, omdat de schadelijke effecten daarvan verschillen. In het beperkte aantal studies dat sinds 2012 over MOSH is verschenen, wordt de zorg van EFSA iets afgezwakt, zo blijkt uit RIVM-onderzoek. Ook zijn volgens berekeningen van het RIVM geen gezondheidseffecten te verwachten als mensen via voedsel aan MOSH worden blootgesteld. Het RIVM wil zich meer op MOAH richten, omdat sommige kankerverwekkend zijn. Het is alleen niet mogelijk om aan te geven of mensen er te veel van binnenkrijgen omdat voor MOAH geen gezondheidskundige norm bestaat.

De kankerverwekkende MOAH zitten vooral in ruwe of onvoldoende gezuiverde minerale oliën en in oliën die verhit zijn geweest. Ze zitten niet in alle bronnen vanwaaruit MOAH in voedsel terecht kunnen komen. Het totale MOAH-gehalte geeft daarom geen informatie over de vraag of de inname van die MOAH schadelijk is. Volgens het RIVM is het zinvol te achterhalen wat de bronnen zijn vanwaaruit MOAH in voedingsmiddelen terechtkomen. Dan kunnen maatregelen genomen worden om schadelijke bronnen zoveel mogelijk te vermijden. Een voorbeeld van een verontreinigende bron zijn jute zakken die met olie zijn behandeld en waarin cacaobonen worden verpakt.

De bijdrage van een andere verontreinigende bron, kartonnen verpakkingen van gerecycled materiaal, lijkt in Nederland mee te vallen. Droge levensmiddelen zoals rijst, pasta, ontbijtgranen en hagelslag worden hier vaak in verpakt. De inname via deze levensmiddelen blijkt in de innameberekeningen slechts een kleine bijdrage te leveren aan de totale blootstelling aan minerale oliën door voedsel. Maatregelen om de blootstelling vanuit kartonnen verpakkingen te beperken, zullen dus een beperkt effect hebben.

Kernwoorden: minerale olie koolwaterstoffen, MOSH, MOAH, risicobeoordeling, toxiciteit, innameberekening, ‘margin of exposure’ (MOE), voedselcontactmaterialen, karton verpakkingen.

Contents

Summary

In 2012, the European Food Safety Authority (EFSA) published a scientific opinion on mineral oil hydrocarbons (MOH) in food, and concluded, based on the data available, that the exposure to MOH via food in Europe was of potential concern. Among the many sources of MOH to enter food, EFSA identified migration of MOH from recycled paperboard packaging into food as a potentially significant contributor to the total exposure of consumers to MOH. In 2015, the Non-Governmental Organisation (NGO) Foodwatch published an investigation into the occurrence of MOH in paperboard packaged foods on the Dutch market such as rice, pasta, breakfast cereals and chocolate sprinkles, and concluded that many of these products contain MOH.

In order to assess the need for possible regulatory measures regarding MOH in food, RIVM screened the new toxicity data on MOH published since the latest opinion of EFSA in 2012 and performed an exposure assessment to mineral oil saturated hydrocarbons (MOSH) and mineral oil aromatic hydrocarbons (MOAH) via food in the Netherlands, which are the two fractions of MOH. For this, Dutch food consumption data of persons aged 2 to 69 years were combined with the data provided by Foodwatch and occurrence data from the EFSA opinion, for foods not analysed by Foodwatch.

The relatively low number of new studies contributed only moderately to the risk assessment of MOH. No new *in vivo* toxicity studies on MOAH were found. New *in vivo* studies on MOSH showed that its bioaccumulation highly depends on the structure and size of MOSH, and varies per tissue. MOSH found in liver of both rat and human consists mainly of highly isomerised naphthenes of $>C_{25}$. The fraction of MOSH that is most potent to induce hepatic microgranulomas in Fischer 344 rats were the n-alkanes of $>C_{25}$. After a diet rich in this fraction, these n-alkanes were also found to accumulate in rat liver. The newly available data was reviewed in an attempt to identify a potential mechanism of action leading to the formation of microgranulomas and associated inflammation in Fischer rats and their possible relevance for humans, but could not conclude on that. Two sub-chronic oral immunotoxicity studies in rats showed no impact of MOSH mixtures on the immune response, indicating that the immunotoxic effects seen after parenteral injections are not relevant for long term dietary intake of MOSH.

Although some new toxicity studies are available, addressing some of the uncertainties as indicated by EFSA, a potential change in the Reference Point can only be decided on in coherence with the studies that have already been evaluated. Therefore, the current Reference Point as selected by EFSA (2012b) was used in the risk assessment of the calculated exposure estimates of MOSH. This Reference Point for MOSH is a no-observed adverse effect level (NOAEL) of 19 mg/kg bw/d, based on the induction of microgranulomas in the liver of rats in a short-term study. The median exposure levels for MOSH, calculated for the Dutch population, resulted in margins of exposure (MOEs) of 190 in children aged 2 to 6 years, and 450 in persons aged 7 to 69 years;

corresponding MOEs for the high (P95) exposure were 90 and 160, respectively. The MOE of 90 for the high exposure of young children is somewhat lower than the minimal MOE of 100 that is generally considered to indicate that exposure is of no health concern. However, since the toxicological Reference Point is based on effects observed after repeated exposure and the MOE is only temporarily slightly below 100, and given the questionable toxicological relevance for humans of the liver microgranulomas observed in rats, it was concluded that the estimated exposure levels to MOSH were of no health concern for the Dutch population. Note that EFSA identified two specific uses of white oils (as release agents for bread and rolls and for spraying of grains) as of potential concern. A preliminary assessment for the Dutch population included in this report supports this observation.

The exposure to MOAH was about 15% of the exposure to MOSH. MOAH is potentially mutagenic and carcinogenic and therefore the exposure to MOAH via food is a reason for concern. However, EFSA did not derive a Reference Point to be used for a risk characterisation of MOAH. As not all MOAH are mutagenic, reduction of potential risks of exposure to MOAH would likely be most effective by identifying and then avoiding MOAH contamination from sources that contain these mutagenic MOAH, like crude or combusted mineral oils.

In the EFSA opinion of 2012, the use of recycled paperboard for food packaging was identified as a source that could contribute significantly to the total exposure of MOH. The estimated exposure to MOSH and MOAH via paperboard packaged foods as calculated in this report was however shown to be limited compared to the exposure via the total diet: only 2% at the median exposure level up to around 15% for 2 to 6-year olds and 18% for persons aged 7 to 69 at the upper intake percentiles (P95, largely due to high consumers of pasta). The real percentages may even be lower, as the exposure via paperboard packaged foods is very likely overestimated. It was assumed that all relevant foods were always packaged in paperboard, while other types of packaging (mainly plastic) are also quite common. Additionally, MOH concentrations in dry rice and pasta were directly linked to the consumed amounts of these foods, which are however based on consumption of prepared foods, which contain a large amount of water. Reducing the exposure to MOSH and MOAH via paperboard packaging will therefore only have a limited effect on the total exposure to these compounds.

1 Introduction

Mineral oils (mineral oil hydrocarbons - MOH) are complex mixtures of hydrocarbons, derived from crude oil. They consist of two fractions: mineral oil saturated hydrocarbons (MOSH) and mineral oil aromatic hydrocarbons (MOAH). MOSH consist of linear and branched alkanes (paraffins) and largely alkylated cyclo-alkanes (naphthenes). MOAH include largely alkylated polyaromatic hydrocarbons. For food, hydrocarbons in the range of C_{10}-C_{50} are relevant. Because of the large variation in both carbon number and structure, MOH in food can appear in thousands of different chemical structures.

Consumers are exposed to mineral oils via food. MOH can occur in food as a result of both contamination and intentional addition. Contamination can occur via many sources, among which food contact materials, lubrication oils from machinery used in harvesting or food-processing, unburned fuel oil in exhaust, debris of tires, etcetera. MOH can be added to food as authorised food additive or can be used as processing aids, e.g. as release agents for bakery ware, and anti-dusting agents for grain in silo's.

In 2012, the European Food Safety Authority (EFSA) published a scientific opinion on mineral oil hydrocarbons (MOH) in food, and concluded that due to lack of data on specific structural groups of MOH, it was not possible to propose a 'tolerable daily intake' (TDI) for MOH in food (EFSA, 2012b). However, with the data available, exposure to MOH via food in Europe was considered of potential concern. Among the many sources for MOH in food, EFSA identified migration of MOH from recycled paperboard packaging into food as a potentially significant contributor to the total exposure of consumers to MOH.

Mineral oils are present in recycled paperboard due to incomplete removal of printing inks used in e.g. newspapers during the recycle process. MOH are transferred to the packaged food mainly by evaporation. Even food packaged in paperboard from fresh (non-recycled) paper fibers can become contaminated, due to use of large cardboard boxes to pack the smaller paperboard packages during storage and transport. These larger boxes are usually made of recycled cardboard and mineral oils evaporated from these boxes can pass straight through the paperboard food packaging into the food. MOH from printing inks used to print the cardboard or paperboard boxes, as well as from exhaust gases (e.g. during transportation) can contribute to the MOH in paperboard packaged food as well.

In 2015, the Non-Governmental Organisation (NGO) Foodwatch published a study into the occurrence of MOH in paperboard packaged foods on the Dutch market such as rice, pasta, breakfast cereals and chocolate sprinkles, and concluded that many of these products contain mineral oils (Foodwatch, 2015). The paperboard was either made of fresh or recycled fibres.

In order to assess the need for possible regulatory measures as regards MOH in food, the Dutch Ministry of Public Health, Welfare and Sports (VWS) requested RIVM to screen new toxicity data published since the latest opinion of EFSA in 2012. These data are briefly summarised in this report.

Furthermore, an intake assessment was performed for the Dutch population, using the occurrence data generated by Foodwatch (2015; 2016a,b) together with those in other foods from the EFSA opinion (2012b). With the available data on the occurrence of MOH in food, an estimation was made of the contribution of specific foods often packed in paperboard packages to the total dietary intake of MOSH and MOAH in the Netherlands.

2 Literature search

A literature screening was performed using the search engine of SCOPUS. The following words were used: ‘mineral oil’ OR ‘MOSH’ OR ‘MOAH’, present in the title of the publication. The search was restricted to articles published between 2012 and 2017, i.e. after the EFSA evaluation in 2012 (EFSA, 2012b).

Studies investigating the kinetics or toxicity of MOH were selected and considered for further evaluation. Selection was based on the title of the paper and information derived from the abstract. Only studies examining clear-cut toxicological endpoints (or reviews), which may be relevant for a risk assessment, were included and are described in section 3.

In addition, two scientific reports were used as extra sources of information:

1. a report on the bioaccumulation and toxicity of MOSH, drafted by the Norwegian Institute of Public Health (NIPH), the French National Institute for Agricultural Research (INRA), and the Swiss Kantonales Labor Zürich (KLZH) in 2017,
2. a recently published evaluation report by the French Agency for Food, Environmental and Occupational Health & Safety (ANSES). However, this report was not considered in detail. It was screened for any additional publications on toxicity, possibly not identified through the above described SCOPUS search.

3 Toxicological data

3.1 Highlights from the latest EFSA opinion (EFSA 2012)

MOSH and MOAH have low acute oral toxicity and this was considered not relevant given the level of exposure from food. In 90-day studies in rat, MOSH have been shown to bioaccumulate and lead to the formation of microganulomas in mesenteric lymph nodes (MLN) and liver. The microgranulomas in the liver were associated with inflammatory reactions and were considered possibly relevant to humans. Accumulation occurs due to slow biotransformation, specifically of the branched and cyclic alkanes with a carbon chain between C_{16} and C_{35}, whereas *n*-alkanes are more efficiently eliminated via metabolism.

Previously, acceptable daily intakes (ADIs) had been derived by Scientific Committee on Food (SCF) (1995), the Joint Expert Committee of Food Additives (JECFA) (2002) and EFSA (2009). JECFA proposed the allocation of medium/low viscosity MOSH into three classes: Class I, II and III, based on the viscosity (primarily a function of the molecular weight), and the ADIs were determined for each class (Table 1).

Table 1 Classification of mineral oil hydrocarbons according to JECFA (2002).

Substance name	ADI (mg/ kg bw)	Viscosity at 100°C (mm^2/s)	Average relative molecular weight
High viscosity[1]	0-20	>11	>500
Medium/low viscosity (Class I)[2]	0-10	8.5-11	480-500
Medium/low viscosity (Class II)[3]	0-0.01	7.0-8.5	400-480
Medium/low viscosity (Class III)[4]	0-0.01	3.0-7.0	300-400

[1] Paraffinic oil
[2] Paraffinic oil or paraffinic oil hydrotreated (catalytic hydrogenation)
[3] Crude naphthenic oil, hydrotreated (catalytic hydrogenation)
[4] Crude naphthelic or paraffinic oil, hydrotreated (catalytic hydrogenation)

EFSA highlighted that these ADIs need to be revised, based on new available toxicokinetic studies, lack of sufficient chemical characterisation of the individual mixture components used in the toxicological studies, and the lack of toxicological relevance for humans of the effects in MLN observed in Fischer 344 rats (EFSA, 2012b). In particular, EFSA proposed an assessment of MOSH mixtures by considering molecular mass range and subclass composition (e.g. n-, branched- or cyclo-alkanes), instead of viscosity.

In the absence of more precise information, a margin of exposure (MOE) approach was put forward for MOSH, using as Reference Point, the most critical NOAEL of 19 mg/kg bw/d (test material: low/medium melting point wax, consisting primarily of *n*-alkanes) based on microgranuloma formation in the liver of Fischer 344 rats. Only for white mineral oils, which are used as release agents for bread and rolls and for spraying of

grains, a higher NOAEL of 45 mg/kg bw/d was defined as Reference Point in the MOE calculation.

In contrast to MOSH, MOAH are sufficiently metabolised and are not known to accumulate in tissues (EFSA, 2012b). However, their presence in the food chain as contaminants is considered to be of concern, as MOAH are potentially mutagenic and carcinogenic. Specifically, due to MOAH, all MOH mixtures are mutagenic, if they are not refined (EFSA, 2012b). No critical level could be established for the MOAH fraction because of their classification as genotoxic carcinogens and the lack of carcinogenicity studies performed on MOAH mixtures (EFSA, 2012b).

3.2 Newly published studies since the EFSA (2012) opinion

3.2.1 Toxicokinetics and bioaccumulation potential

Boogard and his co-workers (2012) compared the kinetics of a low viscosity white oil (P15H) in female Fischer 344 rats (n=60) and female Sprague-Dawley rats (n=45), by administrating a single oral dose of 0, 20, 200 or 1.500 mg/kg bw, and in human female volunteers (dosed 1 mg/kg bw; n=9). Comparing the study design and results, this study was already summarised in the EFSA opinion of 2012, only there it was an unpublished study report provided by Concawe, referred to as: 'Bakker, 2011'. The AUC[1] and the C_{max}[2] measured in blood, as well as the concentrations in the liver were significantly (3 to 4 times) higher in the Fischer 344 rats when compared to the Sprague-Dawley rats with the same dosis, suggesting a higher bioavailability in this strain. The authors propose that the toxicity discrepancies between the two animal strains, i.e. hepatic granulomas observed in the Fischer 344 rats at 200 and 2000 mg/kg bw, are due to this difference in bioavailability.

Barp *et al.* (2014 - Part I) examined the concentration of MOH in human tissues, in terms of concentration and molecular weight distribution. The levels were determined in samples from various organs from 37 post-mortem patients during autopsy. The results revealed high accumulation of MOSH in the organs of all patients that seemed to increase proportionally with age. Mean values recorded were 223 mg/kg in MLN, 131 mg/kg in liver, 130 mg/kg in adipose tissue, 93 mg/kg in spleen and 12 mg/kg in lung. Calculated from the weight of tissue and multiplied by the MOSH concentrations found, it was estimated that a quarter of the subjects had a total body burden of more than 5 g MOSH. The MOSH in the liver and the spleen were different from those in the MLN and fat tissue. Fat tissue and MLN contained an almost identical composition of MOSH, consisting of an 'unresolved hydrocarbon hump' (iso-alkanes, branched and cyclic MOSH), with primarily C_{23}-C_{24}, but ranging from C_{16}-C_{35}. *n*-Alkanes (mainly of plant origin -odd numbered) were also seen in fat and MLN, but at much lower levels.
In liver and spleen, MOSH consisted again of an unresolved hydrocarbon hump, but with other carbon chain length, mainly C_{25}-C_{27}, and with a huge range from C_{18}-C_{46}. *n*-Alkanes did not accumulate at all in liver and spleen. It was suggested that n-alkanes are selectively taken up and eliminated from the human body.

[1] AUC: Area Under the Curve
[2] Cmax: the peak serum concentration

Differences between subjects were more pronounced for the liver/spleen and less for the fat/MLN. No MOAH was detected (except for a very low amount in one subject), suggesting that they are not accumulating. Not reported in the publication of this study, but mentioned in Barp *et al.* (2017b), is that no hepatic microgranulomas were found in any of the livers examined in this study.

This work was continued (Biedermann *et al*., 2015 - Part II), with the aim to qualitatively compare the composition of accumulated hydrocarbons found in human tissues with several mineral oil products, to which humans are expected to be exposed to. The characterisation was performed by two-dimensional gas chromatography (GCxGC). The MOSH composition in the liver (comparable to the spleen) consisted primarily of 'unresolved material'. Some branched paraffins could be characterised, as well as multibranched hydrocarbons. Most of the material was however expected to consist primarily of naphthenes. For the MLN and fat tissue, only a low proportion of *n*-alkanes were identified. Multibranched hydrocarbons were seen, and some cyclic species (*n*-alkyl cyclopentanes and cyclohexanes), as well as naphthenes with more rings. Comparison with plots obtained from mineral oil products indeed confirmed that the accumulated MOSH in the tissues are originating from mineral oils. The low or almost absent levels of *n*-alkanes in the tissues (although present in mineral oils), demonstrate their metabolic elimination. Metabolic elimination seems to occur also within other MOSH classes (slightly branched paraffins, multibranched paraffins and naphthenes), though more selectively. The unresolved accumulated residue consists of highly isomerised, hydrocarbons (primarily naphthenes).

In its opinion on mineral oils in food, EFSA (2012) highlighted the need for further toxicological and kinetic information, as regards to such substances. For these purposes two different repeated-dose experiments were performed, as requested by EFSA, which have been discussed in the scientific report by INRA, KLZH and NIPH (Cravedi *et al.* 2017) on bioaccumulation and toxicity of MOSH in rats. A short summary of the studies' results, as well as the group's conclusions are presented here. In both tests, female Fischer 344 rats were fed MOSH, but as different mixtures.

In the first experiment (Barp *et al*., 2017a, Cravedi *et al.* 2017), female Fischer 344 rats were exposed for 30, 60, 90 and 120 consecutive days, at dose levels of 40, 400 and 4000 mg/kg feed (corresponding to approximately 2, 22 and 222 mg/kg body weight). As testing material a broad MOSH mixture was used, which ranged from C_{14} to C_{50}, but with significant amounts of hydrocarbons only up to C_{21}. The mixture consisted of 31% *n*-alkanes and little-branched paraffins, 9.9% multibranched paraffins and 59% naphthenes.

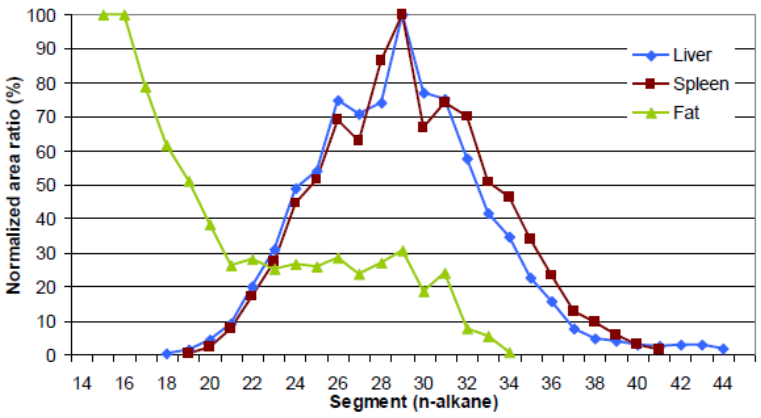
The MOSH levels were analysed in several organs: liver, spleen, adipose tissue and the remaining carcass (after removal of the GI tract) on different time points of exposure (30, 60, 90 and 120 days) and after a 30 day post-treatment period (only for the 90-days exposure group). The results showed that the mean MOSH concentrations were highest for the liver (~50% of the total recovered dose), followed by the adipose

tissue, the spleen and, and the carcass. High concentrations in the liver of animals are in line with previous observations in older studies. When exposure stopped, the MOSH decreased significantly in the liver, spleen, and carcass, but remained unchanged in the adipose tissue.

The accumulation diminished significantly with increasing dose, probably due to a decreased uptake; hence, the authors conclude that a linear extrapolation from high doses to low doses for tissue concentrations would actually result in an underestimation of tissue concentrations. In addition, again due to a probable decreased uptake, the total recovery of MOSH, reduced from 10.9% of the administered dose after 30 days exposure, to 6.2% after 120 days and 3.9% after a 90-day exposure period followed by a 30-day depuration period, at the dose of 40 mg/kg feed.

Given the estimated human daily dietary exposure to MOSH of 0.03 to 0.3 mg/kg bw, as estimated by EFSA in 2012, the authors calculated the expected concentrations in the human organs, if extrapolated directly from the levels measured in the rat study. Even when the lowest dose in the rat study (resulting in the highest accumulation) is used, the calculated values highly underestimated the accumulation, based on the comparison of the estimated versus the actual measured levels in humans from the study of Barp *et al*., (2014). This fact questions the direct extrapolation from animal data to humans.

The MOSH detected in the liver and spleen were comparable as regards to their carbon content, ranging from C_{19} to C_{40}, and with a maximum retention at C_{29} (Figure 1).



Fat: abdominal adipose tissue.

Figure 1 Recovery of MOSH with regard to molecular mass: areas for single carbon segments in tissues divided by those in the feeds and normalised on the maximum (40 mg/kg exposure during 90+30 days) (Figure as taken from EFSA, 2017).

Surprisingly, although the significant fraction of MOSH in the feed was up to approximately C_{21}, such MOSH species were almost completely absent from the two organ tissues, suggesting their fast elimination. This was not the case for the MOSH in the adipose tissue, with a

maximum retention at C_{15}. MOSH with C_{22} to C_{34} were only recovered at considerably lower quantities in the adipose tissue.
In the second experiment (Barp *et al.* 2017b, Cravedi *et al.* 2017), the aim was to elucidate whether the maximum relative accumulation in liver and spleen is indeed with MOSH containing around C_{29} atoms, and the potential impact of *n*-alkanes on hepatic granuloma formation. For these purposes, three different MOSH mixtures were used, as following:

- S-C25 (branched and cyclic MOSH, only ~27% contained more carbon atoms> C_{25}),
- L-C25 (C_{25}-C_{45} branched and cyclic MOSH, deparaffinated oil free of *n*-alkanes), and
- L-C25W (1:1 mix of L-C25 with a wax with similar mass range).

The female rats were fed one of the three different mixtures for 120 days, at doses of 0, 400, 1000 and 4000 mg/kg feed (0, 22, 55 and 222 mg/kg bw/d).

Again here the liver was the major organ of MOSH accumulation for all mixtures, with 73 to 85% of the total recovered material, followed by the spleen and adipose tissue. Only very low levels were recorded again in the carcass. In all tissues, the highest MOSH contents were measured for the S-C25 fraction, the lowest ones for the L-C25 fraction, except for the spleen, in which the residues of L-C25W were even lower (Figure 2).

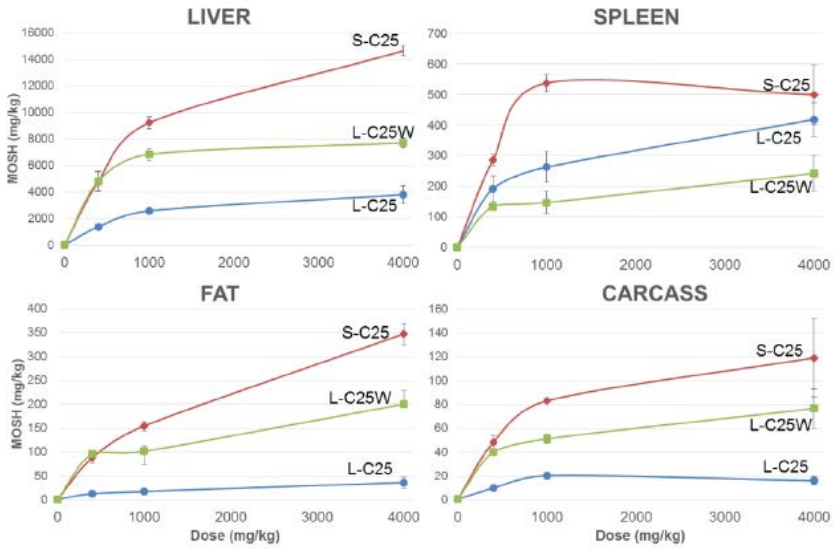


Figure 2: MOSH concentrations (mg/kg) measured in liver, spleen, adipose tissue and carcass of Fischer rats exposed for 120 days to S-C25, L-C25 or L-C25W (Figure as taken from EFSA, 2017).

As seen previously with the broad MOSH mixture, in the adipose tissue (and carcass), lower molecular weight MOSH were detected, as compared to the liver and spleen. In liver and spleen accumulation was higher for the MOSH C_{26}-$_{30}$ than C_{20}-$_{25}$.

The MOSH found in the tissues were characterised by HPLC-GC-FID and two-dimensional GC (GCxGC). In liver and spleen, MOSH in the range of C_{26}-C_{30} more strongly accumulated than those in the range of C_{20}-C_{25}. *n*-Alkanes and *n*-alkyl monocyclic naphthenes were generally enriched in

adipose tissue. In liver and spleen, *n*-alkanes up to C_{25} were eliminated, but strongly accumulated at around C_{30}. Based on this profile, it was hypothesised that crystallization protects these wax components against metabolism and elimination. Compared to the animal data, accumulation of *n*-alkanes from vegetable sources, such as apples (wax consists of 19-36% *n*-alkanes, mainly C_{27} and C_{29}), into human tissues seems low, perhaps because of low absorption due to their presence in crystalline form, hindering absorption of these compounds.

Besides the information on bioaccumulation in the publications of Barp *et al.* (2017a+2017b), both studies included toxicological examinations; these results are summarised in paragraph 3.2.2 and 3.2.3.

3.2.2 Repeated dose toxicity

Adenuga *et al*. (2014), exposed Sprague-Dawley rats to 0, 500, 2500 and 5000 mg/kg bw/d of a C_{10}-C_{13} solvent (42% paraffins, 58% naphthenes)[3], not containing MOAH, by gavage for 90 consecutive days. Some elevations were recorded in serum levels of liver enzymes (alanine aminotransferase, gamma glutamyltransferase, total bilirubin) in the medium and high dose groups, nevertheless, not accompanied by histopathological findings. Hepatic enlargement and centrilobular hypertrophy observed were considered as adaptive response to treatment with hydrocarbons. Kidney effects seen in the male rats are well known as specific for light-hydrocarbon induced nephrotoxicity in male rats. The estimated BMDL was 1857 mg/kg bw/d, based on increased levels of alanine aminotransferase in the serum. Such a value is in line with other studies performed with comparable hydrocarbons in this specific strain, demonstrating lack of systemic toxicity at doses up to 1000 mg/kg bw/d.

In §3.2.1, the study of Barp *et al.* (2017a) is described, in which female Fischer 344 rats were dosed a broad MOSH mixture (C_{14}-C_{50}) for up to 120 consecutive days, at dose levels of 40, 400 and 4000 mg/kg feed (2, 22 and 222 mg/kg bw).
As regards to potential toxicity, the results revealed an increased absolute and relative liver weight at the highest dose of 4000 mg/kg feed. Such changes were reversible, 30 days post-treatment. The histopathological examination showed increased granulomas in the liver of rats only at the highest dose level of 4000 mg/kg feed. The effect was visible only after 90 or 120 days of exposure, and could still be seen after the 30-day recovery period.

The other study of Barp *et al.* (2017b) described in §3.2.1, in which female Fischer 344 rats were dosed 3 different fractions of MOSH mixtures for 120 consecutive days, at dose levels of 400, 1000 and 4000 mg/kg feed (22, 55 and 222 mg/kg bw), also included toxicity data.
Results revealed that liver and spleen weights were significantly increased in both L-C25 and L-C25W groups, at all dose levels, but not in the S-C25 group. In the histopathological examination, hepatic granulomas and lymphoid cell clusters in the liver were observed in the

[3] Conventional name: hydrocarbons, C10-C13, *n*-alkanes, iso-alkanes, cyclics, <2% aromatics

high dose group fed with S-C25, and in all three dose groups fed L-C25W. The L-C25 MOSH, which is the only *n*-alkane free mixture, did not induce the formation of liver granulomas in none of the groups, indicating a correlation between the granulomas and exposure to *n*-alkanes.
To analyze the link between accumulation and different toxicological endpoints including the formation of hepatic microgranulomas and the change in immune function, immunological testing was done. Results are described in §3.2.3 (Cravedi *et al.* 2017)

3.2.3 Immunotoxicity

In the 2 experiments described by Barp *et al.* (2017a, 2017b, Cravedi *et al.* 2017) with rats exposed to various MOSH mixtures, as described in §3.2.1 and §3.2.2 of this report, additional testing on the toxicity for the immune system was performed (described in Cravedi *et al.* 2017), as follows: 5 days before the end of the experiments, all rats exposed for 120 days were injected (i.v. in the first experiment; s.c. in the second experiment) with an antigen (KLH). After euthanasia, blood samples were taken and KLH-specific antigen IgM were determined in the serum. No significant differences between the groups were observed for KLH-specific IgM antibodies concentrations in serum due to MOSH exposure, neither after exposure to up to 4000 mg/kg feed (222 mg/kg bw) of the broad mixture of MOSH, nor to the three “narrow” MOSH mixtures.

Kimber and Carillos (2016) performed a literature review in order to examine whether oral exposure to mineral oils can induce adverse effects on the immune system, in particular autoimmune responses. The results of their search show that such effects are not observed in experimental animals when mineral oils are administered orally. Although some epidemiological data indicate a possible association, actual mineral oil levels were not measured. Data indicate that parenteral administration of aliphatic hydrocarbons at high doses may result in autoimmune responses, albeit with a lack of dose-response relationships. Even so, this is not relevant for dietary exposure.

In a more recently performed study, Andreassen *et al*. (2017) used the arthritis susceptible Dark Agouti (DA) rats, so as to elucidate further any potential correlation between autoimmune responses and MOSH dietary exposure. This experiment was also part of the first experiment described by Cravedi *et al.* (2017), see §3.2.1. The animals were given the test material in the feed for 90 days. Two different test materials were used: pristane (4000 mg/kg bw) or a MOSH mixture (C_{14}-C_{50}; 40, 400, 4000 mg/kg bw). Apart from the feeding study, some animals received a single intradermal injection of pristane (200 µL) as positive control. Markers previously reported to be associated with arthritis development were analysed; levels of cytokines and rheumatoid factor IgG and IgM. Whereas rats intradermally injected with pristane developed arthritis, none of the animals given pristane or MOSH in the feed developed arthritis symptoms (clinical or biological markers) at any dose level applied. The fact that intradermal injections resulted in arthritis symptoms in all treated rats, suggests that the route of administration plays a fundamental role in the development of such effects.

3.2.4 *Endocrine disruption*

Tarnow *et al*. (2016) assessed the estrogenic potential of 15 different MOH samples of variable composition, containing MOSH, but also high contents of MOAH, in three in vitro tests: E-screen, an estrogen responsive luciferase assay and in a transcriptional assay of selected estrogen responsive genes. Out of the 15, 10 samples gave a positive response, which was proportional to the quantity of MOAH (16% or higher), indicating that the observed responses may be linked to the aromatic hydrocarbons *per se*. This was confirmed in a subsequent experiment with isolated fractions of MOSH and MOAH. The study demonstrates that MOAH could potentially be endocrine disruptors, and hence, further research shall elucidate this further.

3.2.5 *Mode of action*

A recent evaluation of the existing data on MOSH toxicity was performed, in an attempt to first identify a potential mechanism of action (MoA) leading to the formation of microgranulomas, and second examine the relevance of this effect for humans (IPCS human relevance framework-HRF) (Adenuga *et al.*, 2017).

The hypothesized MoA is shown schematically below and it was evaluated with a systematic weight of evidence analysis, in accordance to the Bradford Hill considerations:

Key event 1: Intestinal absorption→ Key event 2: liver deposition and retention→key event 3: inflammatory cell tissue infiltration due to retained material in the liver

The authors conclude that the liver microgranulomas seen in the Fischer 344 rats are in fact formed through an inflammatory reaction to the retained MOH fraction.

In order to determine whether this MoA is relevant for humans, qualitative and quantitative differences between the two species were examined, with human data collected over the last 30 years. In respect to key events 1 and 2, no qualitative differences were identified. The hydrocarbons in the range of C_{22}-C_{28}, are absorbed and retained in several tissues also in humans. However, there is hitherto no evidence that granulomas are also formed in humans, indicating a qualitative difference as regards to key event 3. According to the authors, this is further supported by quantitative differences between the two species, which also point towards the direction that this mechanism is most likely not relevant for humans. Overall, the Fischer 344 rat MoA was not considered to be relevant to humans, consistent with data showing no evidence for the formation of epithelioid granulomas with humans even in cases of massive ingestion of MOH.

3.2.6 *Epidemiological data*

One publication (Miligi *et al*., 2013) was identified in the public domain, which associated an increased risk for children leukaemia with parental exposure to aromatic and aliphatic hydrocarbons in occupational settings, amongst which mineral oils. Nonetheless, routes of exposure do not include the oral one and also no details are given in the composition of mineral oils linked to such effects. Therefore, the value of this study within the context of this report is considered ambiguous.

4 Conclusions and discussion on MOH toxicity

4.1 Summary of new studies

An assessment of any recent toxicological and kinetic studies on MOH (MOSH and MOAH), conducted from 2012 onwards, resulted in the identification of only a few studies that have examined the accumulation potential and toxicity of MOSH. No studies for MOAH were detected, except for one study *in vitro*. The main effort regarding mineral oils and their presence in the food chain is currently focused on other aspects than their toxicity. Such aspects include the development of appropriate analytical techniques to characterise the complex mixtures, identification of contamination sources and definition of measures to mitigate the migration of mineral oils in food products.

The main conclusions of the findings are as follows:

- Only few new toxicity/kinetic studies were performed from 2012 up to now, and predominately for MOSH.
- A study with human post-mortem samples revealed a high accumulation of MOSH in human tissues, such as liver, fat and mesenteric lymph nodes (MLN), through life-long exposure (Barp *et al.* 2014).
- Characterisation of the accumulated fractions in human tissues showed low levels of *n*-alkanes, suggesting that *n*-alkanes are not absorbed very well and/or efficiently metabolised and eliminated. MOSH in human liver are analysed as a cloud of unresolved highly isomerised hydrocarbons; mainly naphthenes were observed. (Biedermann *et al.* 2015)
- The MOSH composition in human fat tissue and lymph nodes is similar, with maximum concentrations for the C_{23}-C_{24} hydrocarbons, but different from that in liver and spleen, with maximum concentrations for the C_{25}-C_{28} hydrocarbons (Barp *et al.* 2014, Biedermann *et al.* 2015)
- In animal studies, accumulation of MOSH in tissues is also observed, but not proportionally to the administered doses. Therefore, extrapolation from high doses to low doses for determinations of tissue MOSH levels may lead to an underestimation of the actual tissue concentrations (Boogard *et al.* 2012, Barp *et al.* 2017a).
- MOSH accumulation in female Fischer 344 rats showed higher blood and equivalent higher liver values of MOSH than in Sprague Dawley rats at the same dose level. This indicates that internal blood levels are a more appropriate surrogate for the risk assessment of MOSH than the external dose, and that there is a difference in bioavailability between the two strains (Boogard *et al.*, 2012).
- In liver and spleen, accumulation of MOSH of C_{26}-C_{30} is higher than that of C_{20}-C_{25}. With regard to *n*-alkanes, those up to C_{25} were eliminated from liver and spleen, but those around C_{30} accumulated strongly. It was hypothisised that crystallisation slows down the biotransformation and elimination of *n*-alkanes (Barp *et al.* 2017b).

- Hepatic microgranulomas were not seen in female Fischer 344 rats exposed to MOSH of longer-chains (>C_{25}), branched and cyclic, and virtually free of *n*-alkanes. However, when these longer chains were mixed 1:1 with wax (>C_{25}, 80% *n*-alkanes), hepatic microgranulomas were seen in low, mid and high dosed animals. Furthermore, although treatment with shorter-chains (<C_{25}; mainly branched and cyclic MOSH) resulted in the highest accumulation of total MOSH in the liver, hepatic microgranulomas were seen only at the highest dose in these rats. This indicates that hepatic microgranuloma formation in the female Fischer 344 rat does not depend on accumulation of total amount of MOSH but rather on the accumulation of *n*-alkanes in the liver (Barp *et al.* 2017b). It was hypothisised that maybe the crystallisation of *n*-alkanes plays a role in triggering the inflammatory response seen in the hepatic microgranulomas in Fischer 344 rats (Barp *et al.* 2017b).
- MOSH exposure, irrespective of mixture tested, had no impact on the immune response following antigen challenge (Cravedi *et al.* 2017), nor did it induce arthritis by the oral route (Andreassen *et al.* 2017).
- Using mode of action/human relevance framework (MoA/HRF) analysis for MOH-induced epithelioid granulomas, the Fischer 344 rat MoA was not considered to be relevant to humans (Adenuga *et al.* 2017).
- No new toxicity data were detected for MOAH, apart from one *in vitro* study, which indicated that MOAH could potentially be endocrine disruptors, and hence, further research shall elucidate this further.

4.2 Risk characterisation of MOSH

For MOSH, the NOAEL of 19 mg/kg bw has been used as Reference Point for the risk assessment by the EFSA CONTAM Panel (EFSA, 2012b). This NOAEL is based on microgranuloma formation in the liver of Fischer 344 rats exposed to a low/medium melting point wax, consisting primarily of *n*-alkanes. In the EFSA opinion however, no ADI was derived from this NOAEL, as (and this is quoted from EFSA, 2012b:)

"*several uncertainties regarding the extrapolation from data on experimental animals to humans exist, in particular the relevance of these lesions for humans and the sensitivity of humans in comparison with the most sensitive species tested, Fischer 344 rats.*
It is assumed that the accumulation of MOSH plays an important role in microgranuloma formation both in rats and humans. The low and intermediate melting point waxes, which consist mainly of n-alkanes that do accumulate to a much lesser extent than branched- and cyclic-alkanes, are the most potent mixtures tested. This fact would indicate that unknown mechanisms other than accumulation of MOSH per se are involved in the pathogenesis of the microgranuloma formation."

The new study in Fischer 344 rat of Barp *et al.* (2017b) showed that, although the branched-alkanes and naphthenes (C_{25}-C_{30}) were indeed more prone to accumulate in the liver, it was the *n*-alkane fraction (chain length >C_{25}) that seemed to induce the hepatic microgranulomas,

where the branched-alkanes and naphthenes (C_{25}-C_{30}) did not. It can be concluded that microgranuloma formation in the Fischer 344 rat is related to accumulation of n-alkanes with chain length >C25 rather than to accumulation of total MOSH in the liver.
n-Alkanes are hardly found in human liver tissues, nor in other human tissues (Biedermann *et al.* 2015). This suggests that these *n*-alkanes are not absorbed very well and/or are efficiently metabolised and eliminated in humans, at the dietary intake levels. Evaluation of an analysis on the mode of action/human relevance framework (MoA/HRF) for MOH-induced epithelioid granulomas in the Fischer rat, using modified Bradford Hill considerations, led to the conclusion that the mode of action was not relevant to humans (Adenuga 2017). It is however difficult to judge the value of this analysis and this is beyond the scope of this report.

Another uncertainty in the risk assessment identified in the EFSA opinion was that it was not known whether long term oral exposure could induce autoimmune responses, as seen after intraperitonal and intradermal injections, but not seen in short term oral exposure. Two new sub-chronic oral immunotoxicity studies showed that MOSH mixtures had no impact on the immune response (Cravedi *et al.* 2017, Andreassen *et al.* 2017), indicating that the immunotoxic effect seems to be route specific and also that immunotoxicity is not a relevant effect for long term dietary intake of MOSH.

Overall, the new studies on MOSH address some of the uncertainties identified in the EFSA opinion of the CONTAM Panel (EFSA, 2012b). Results of the new studies should however be considered in coherence with the studies that have already been evaluated by EFSA. Therefore, no proposal for revision of the Reference Point will be made in the current report. For the risk assessment of MOSH for the Dutch population, the NOAEL of 19 mg/kg bw/d, used as Reference Point by the EFSA CONTAM Panel, was used for calculating the margins of exposure (MOE's).

The uncertainty about the relevance of liver microgranulomas for humans, and the sensitivity of humans in comparison with the most sensitive species tested (i.e. Fischer 344 rats), was characterised by EFSA (2012b) as an "uncertainty with potential to cause over-estimation of the risk".

4.3 Risk characterisation of MOAH

MOAH are potentially mutagenic and carcinogenic, but neither for MOAH mixtures, nor for mineral oils, dose-response data on the carcinogenicity is available. Therefore, it was not possible to establish a Reference Point (EFSA 2012b). The new toxicity studies do not include any *in vivo* dose-response toxicity data on MOAH either, so this is not changed.

The limited data on toxicokinetics of MOAH available in the EFSA opinion of 2012 indicate that MOAH are well absorbed and are rapidly distributed to all organs. The data also indicate that MOAH are extensively metabolised and do not bioaccumulate (EFSA 2012b).

Studies on mixtures of MOAH in EFSA (2012b) were restricted to genotoxicity studies *in vitro,* and dermal repeated dose carcinogenicity studies in mice. Studies on the genotoxicity of mineral oils have mainly been performed in the Salmonella typhimurium mutagenicity assay (AMES-test). With the exception of highly purified oil varieties consisting of alkanes and naphthenes (so, virtually free of MOAH), all mineral oils are mutagenic in this assay. The same outcome applies to the dermal carcinogenicity tests in mice (skin-painting studies): highly refined oils do not induce tumours in the skin, but all other mineral oils do (EFSA 2012b). Furthermore two dietary carcinogenesis studies on 3 types of mineral oils that were all food-grade, i.e. virtually free of MOAH, did not cause an increase in the incidence of tumours (EFSA 2012b).

In its opinion of 2012, the EFSA CONTAM Panel stated that the mutagenicity of MOH is caused mainly by aromatic 3-7 fused ring MOAH, including alkylated and non-alkylated Polycyclic Aromatic Hydrocarbons (PAH). The non-alkylated PAH is a minor fraction of the MOAH and mainly formed by heating of the oil. These non-alkylated PAH are covered by monitoring programmes in food (EFSA, 2012b).

Alkylated PAH are not covered by the PAH-monitoring programmes. The effect of alkylation on the genotoxic activity of PAH is highly dependent on both size and location of the substituents; whilst methyl-substitution of aromatics with few rings might enhance biological activity, on the other hand particularly bulky ring-substitutions would tend to prevent bio-activation and intercalation with DNA (EFSA, 2012b). Some highly alkylated MOAH can however act as tumour promoters. The genotoxic activity is the combined effect of the simultaneous presence of all of the MOAH, which individually on a molecular level might express additivity, synergy or antagonism (e.g. benzo[a]pyrene may be inhibited by less active MOAH). Hence, it was concluded by EFSA that it is not possible to sum up the activity of a number of single fractions, nor meaningful to establish health-based guidance values based on studies on individual components (EFSA 2012).

One of the objectives of this report was to explore the possibility of using marker molecules for the MOAH risk assessment. In view of the lack of new data on the toxicity of MOAH, this is however not possible.

5 Preliminary dietary exposure assessment of MOSH and MOAH in the Netherlands

In this chapter, an exposure assessment to MOSH and MOAH via food (including beverages) in the Netherlands is described. In addition to the intake of MOSH and MOAH via the total diet, an exposure assessment for MOSH and MOAH via food items packaged in paperboard was performed. The approach taken to estimate the exposure is addressed in sections 5.1, 5.2 and 5.3. In section 5.4, the exposure levels to MOSH and MOAH in the Netherlands are reported and they are discussed in section 5.5 in relation to the uncertainties of the exposure assessment.

5.1 Input data

5.1.1 Food consumption data

The exposure to MOSH and MOAH via food was calculated with food consumption data of two food consumption surveys performed in the Netherlands covering the general population from age 2 up to 69. These surveys include the Dutch National Food Consumption Survey (DNFCS)-Young children, which covers the dietary habits of young children aged 2 to 6 and was conducted in 2005 and 2006 (Ocké *et al.*, 2008). The other food consumption survey was performed in 2007 to 2010 among persons aged 7 to 69; the DNFCS 2007-2010 (van Rossum *et al.*, 2011). The foods recorded in the food consumption databases are coded via different food coding systems. In the present study, the food codes of the Dutch Food Composition Database NEVO[4] were used. For a more detailed description of both surveys, see Appendix A.

5.1.2 Concentration data

Two sources of concentration data were used in the exposure assessment of MOSH and MOAH: concentrations published by Foodwatch in 2015 and 2016 (Foodwatch, 2015; 2016a,b) and by the EFSA Panel on Contaminants in the Food Chain (CONTAM) in 2012 (EFSA, 2012b). These data are described in more detail below.

5.1.2.1 **Foodwatch**

In 2015, Foodwatch published concentrations of MOSH and MOAH in food packaged in paperboard purchased on the Dutch market (Foodwatch, 2015). These foods covered both foods packaged with or without a (plastic) interior lining. Different brands of rice, pasta, breakfast cereals (cornflakes) and chocolate sprinkles were sampled, as well as one brand of oatmeal, cacao powder and flavoured sprinkles. Furthermore, one brand per following cereal product was sampled: breadcrumbs, corn starch, couscous, semolina and a whole wheat grain product. The concentrations of MOSH ranged from <0.2 mg/kg (limit of detection; LOD) to 133 mg/kg in one brand of white pasta. Corresponding figures for MOAH were <0.2 mg/kg (LOD) to 5 mg/kg in another brand of white pasta. The limit of quantification (LOQ) was not reported.

[4] http://nevo-online.rivm.nl/

In 2016, Foodwatch also published concentrations of MOSH and MOAH in chocolate bunnies (Foodwatch, 2016a) and chocolate Santa Clauses (Foodwatch, 2016b), both packaged in aluminium foil. These products were purchased on the German market. Concentrations of MOSH and MOAH ranged from 0.6 to 21.2 mg/kg and <0.5 (LOD) to 2.9 mg/kg, respectively. Also here, no LOQs were reported.

The MOSH and MOAH analyses in foods packaged in paperboard, as well as in the chocolate bunnies were performed by an accredited laboratory (DIN EN ISO 17025). The Foodwatch report on the concentrations of MOSH and MOAH in chocolate Santa Clauses does not report the laboratory that performed the analyses, but is very likely the same.

For an overview of the individual MOSH and MOAH concentrations reported by Foodwatch, see Appendix B.

5.1.2.2 EFSA CONTAM Panel

The concentration data published by the EFSA CONTAM Panel in 2012 were obtained via a call for data (EFSA, 2012b). In total, 1 455 single analytical data were submitted, of which 338 originated from Germany, France and Italy (all regarding vegetable oils) and the remaining 1 117 were submitted by KLZH, Kantonales Labor Zürich in Switzerland. The data from KLZH were related to foods belonging to different food groups, including bread, meat, eggs, seafood, sugar and confectionery. All data related to the presence of MOSH. No analytical data on MOAH were made available to EFSA.

The EFSA CONTAM Panel reported that some of the MOSH concentrations submitted for the food groups ‘bread and rolls’ and ‘grains for human consumption’ (mainly rice) were very high (EFSA, 2012b). The Panel explained these high concentrations due to the use of specific production practices based on food grade white oils. Therefore, it identified two mean MOSH concentrations for these two food groups: a background and a ’high level’ mean concentration[5]. The background concentrations were used in the current study.

For an overview of the MOSH concentrations published by the EFSA CONTAM Panel (EFSA, 2012b), which were used in the current exposure assessment, see Appendix C.

5.1.2.3 Concentration data used in the exposure assessment

In the current exposure assessment, priority was given to the concentration data of Foodwatch (2015; 2016a,b). These were supplemented with the concentrations reported by the EFSA CONTAM Panel (EFSA, 2012b) to obtain more complete exposure estimates of MOSH and MOAH (total diet estimates of exposure). Additionally, the exposure was also calculated using only the Foodwatch data for paperboard packaged foods (pasta, rice, breakfast cereals, cereal products, and sprinkles). This was done to determine the intake of MOSH and MOAH due to this type of packaging. There was one very

[5] These concentrations were derived by modelling the distribution of MOSH concentrations per food group using a maximum likelihood log-normal fitting (EFSA, 2012b).

high level of MOSH reported by Foodwatch in one type of pasta: 133 mg/kg (Appendix B). This level was very likely an example of contamination with white oil, due to either the use of MOH as a release agent or for spraying of grains, given the very low corresponding MOAH level of only 0.6 mg/kg (0.5% of total MOH) and the fact that white oils are virtually free of MOAH (EFSA, 2012b). Therefore, this MOSH level of 133 mg/kg was included in the exposure assessment of the total dietary intake, but not in the exposure assessment of intake via paperboard packaged foods (i.e. not included in the calculation of the mean concentration for the food group 'pasta'). The corresponding level of MOAH was however included; as white oil is virtually free of MOAH, the presence of MOAH in this sample was assumed to be derived from migration via the paperboard packaging rather than from use of white mineral oils as release agent or for spraying of grains.

The exposure to MOSH and MOAH was calculated according to the medium bound (MB) scenario; samples with reported concentrations below the limit of detection (LOD) or quantification (LOQ) were assumed to contain MOSH and MOAH at half the LOD or LOQ (whichever was applicable). However, the EFSA CONTAM Panel only reported mean MOSH concentrations for the lower (LB) and upper bound (UB) scenarios. These concentrations were calculated by assigning either zero (LB) or the relevant LOD or LOQ (UB) to the samples with a level below LOD or LOQ. For the current exposure assessment, these LB and UB mean values were averaged to obtain an approximation of the 'medium bound' concentration. These 'MB' concentrations were subsequently used in the exposure assessment. The EFSA CONTAM Panel did not receive any concentration data of MOAH in food (section 5.1.2.2). However, the Panel estimated the MOAH concentration originating from recycled paper and board at approximately 15% of the MOSH concentration. This percentage was therefore used in this report to derive mean MB MOAH concentrations from the corresponding mean MOSH concentrations reported by the EFSA CONTAM Panel. Based on the concentration data published by Foodwatch, the concentration of MOAH was on average 13% of the MOSH concentration, which is comparable to the value of 15% estimated by EFSA.

The Foodwatch concentration data of MOSH and MOAH included samples with a concentration below LOD (0.2 or 0.5 mg/kg). According to the MB scenario, these non-detect samples were assumed to contain the compounds at a concentration of half this limit value (= 0.1 or 0.25 mg/kg). After imputing the non-detect samples with this concentration, MOSH and MOAH concentrations were included in the exposure assessment by fitting a lognormal distribution to the samples with a positive (≥ LOD) concentration per food group and by modelling the non-detect samples as a proportion of samples below LOD. This approach is recommended in the refined long-term exposure assessment (EFSA, 2012a). To model the concentrations in this way, the 'NonDetectSpike LogNormal' option within MCRA was used (van der Voet *et al.*, 2015; de Boer *et al.*, 2016). A mean concentration was subsequently calculated from both the positive and medium bound imputed values per food group and used in the long-term exposure

assessment[6]. Figure 3 shows an example of a NonDetectSpike-LogNormal distribution fitted to the Foodwatch MOSH concentration data of the food groups 'chocolate' and 'breakfast cereals'.

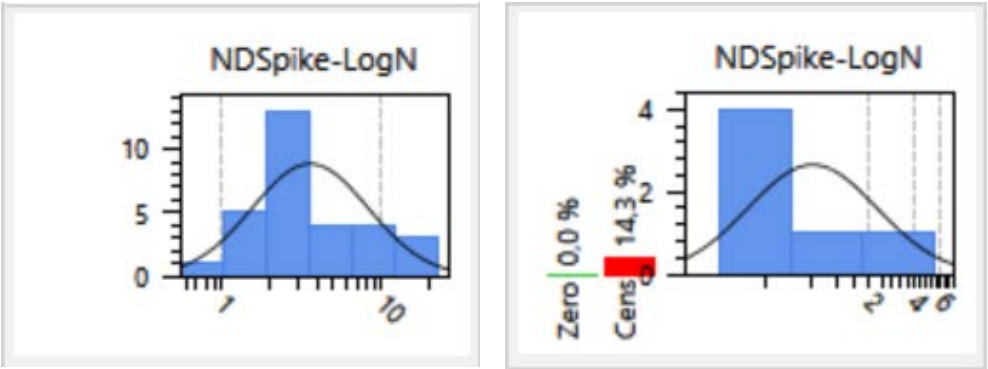


Figure 3. Example of a 'NonDetectSpike LogNormal' distribution fitted to the MOSH concentrations of the food group 'chocolate' (A) (0% non-detect samples) and 'breakfast cereals' (B) (14% non-detect samples)

For fitting a lognormal distribution to the positive concentrations, at least two of such samples should be available for a certain food group. In our assessment, the number of positive MOSH samples ranged from 5 for the food group 'cereal products' to 30 for the food group 'chocolate'. The corresponding numbers for MOAH were 2 for the food group 'chocolate sprinkles' and 10 for the food group 'chocolate' (Appendix B).

The concentration data obtained from the EFSA CONTAM Panel (EFSA, 2012b) and those for oatmeal, cacao powder and fruit-flavoured sprinkles obtained from Foodwatch (Appendix B) consisted of only one value. These concentrations were included as such (so-called empirical modelling) in the exposure assessment. Appendix D lists the simulated mean MOSH and MOAH concentrations for the relevant food groups as used in the exposure assessment.

5.2 Food mapping

Mapping is the process of matching the foods for which concentration data are available to those recorded in the food consumption databases. The majority of the MOSH and MOAH concentrations available for this study were those reported by the EFSA CONTAM Panel (EFSA, 2012b) (see section 5.1.2.2). These concentrations were classified according to level 1 of the FoodEx1 classification system (EFSA, 2011). This system consists of four hierarchical levels, with level 4 representing the most refined (e.g. bread) and level 1 the most aggregated (e.g. grains and grain based products) classification level. The MOSH concentrations were classified in 26 FoodEx1 food groups. In the current exposure assessment, concentrations of 20 FoodEx1 food groups were used (Appendix B); for the other six food groups, data from Foodwatch were used or no consumption was recorded in the food consumption databases (breast milk and potato flakes). The relevant foods recorded

[6] For example, if a food group consists for 10% of non-detect samples, the mean medium bound concentration was calculated as 0.1 x medium bound imputed value + 0.9 x mean concentration of lognormal positive distribution.

in the food consumption databases were assigned to one of these FoodEx1 food groups. Some specific decisions made were:

- The FoodEx1 food group 'vegetable products' was mapped to vegetables coded as glass/can and deep-frozen. Vegetables consumed raw or after cooking were not considered relevant for the exposure to MOSH and MOAH. No concentration of MOSH and MOAH were available for fruit products. Therefore, the concentrations of this food group were also mapped to fruit products coded as glass/can and deep-frozen. Also in this case, fruits consumed raw were not considered.
- The FoodEx1 food group 'chocolate (cocoa) products' was mapped to chocolate bars and chocolate coated confectionary.

The concentrations of Foodwatch (2015; 2016a,b) were also mapped to the most appropriate foods recorded in the two food consumption databases, taking into account similarities in packaging and consistency:

- The concentrations in breadcrumbs, corn starch, couscous, semolina and a whole wheat grain product were grouped as 'cereal products', and mapped to similar foods recorded in the food consumption databases[7].
- The concentrations in chocolate bunnies and chocolate Santa Clauses were mapped to the consumption of chocolate as such.
- The concentrations in oatmeal were also mapped to muesli products, based on comparable consistency.
- The concentrations in breakfast cereals were mapped to different kinds of flaky breakfast cereals.

5.3 Long-term dietary exposure assessment

Long-term exposure to MOSH and MOAH was estimated using the Monte Carlo Risk Assessment Software (MCRA) release 8.2 (de Boer, Goedhart *et al.* 2016) with the observed individual means (OIM) method. In this model, daily consumption patterns of individuals are multiplied with the mean concentration per consumed food, and summed over foods per day per individual. All daily estimated exposures are adjusted for individual body weight, resulting in a distribution of daily exposures per individual. For more details, see Appendix E.

Calculated exposures to MOSH and MOAH using this model were expressed in "mg/kg body weight (bw) per day", and weighted for small deviances in socio-demographic factors and season. The exposure distribution of persons aged 7 to 69 was also corrected for day of the week. No correction weights for day of the week were available within the DNFCS-Young children database. Weights were those used by Ocké *et al.* (2008) and van Rossum *et al.* (2011).

The exposure was calculated for two age groups: children aged 2 to 6 and persons aged 7 to 69. The reported percentiles of the long-term exposure distribution were the 50th (median, P50) and 95th (P95).

[7] These products have the same consistency; they all consist of scatterable particles that are relatively dry, and low in fat but rich in starch. The level of minderal oils did not show any correlation with fat content nor with particle size and therefore, the variation in the level of contamination between these products was considered being more likely due to differences in processing of the grain than to different food types. MOSH and MOAH concentrations in these products were therefore treated as repeated measurements within the same food group

By using the bootstrap approach, the uncertainty around the exposure percentiles caused by the sample size of the food consumption data was quantified (Efron 1979, Efron and Tibshirani 1993). The uncertainty of the concentration database can only be quantified when per food group more concentrations are available. This was true for part of the concentrations from Foodwatch (Appendix B). For the majority of the concentration data, only one mean concentration value per food group was available. The uncertainty due to the sample size of the concentration data could therefore only be quantified to a limited extent via bootstrapping. A description of the bootstrap is given in Appendix F.

5.4 Dietary exposure results

Exposure percentiles
Table 2 lists the median (P50) and high (P95) long-term exposure to MOSH and MOAH in children aged 2 to 6 and in persons aged 7 to 69 via the total diet and via paperboard packaged foods.

The median and high levels of exposure to MOSH and MOAH in 2- to 6-year olds were approximately a factor 2 higher than in persons aged 7 to 69 via both exposure sources (Table 2). Considering the uncertainty around the exposure estimates due to the size of the food consumption and concentration databases (section 5.3), the high levels (P95) of exposure to MOSH and MOAH could be as high as respectively 0.40 and 0.028 mg/kg bw per day in 2- to 6-year olds via the total diet.

Comparing the intake of MOSH and MOAH via the consumption of paperboard packaged foods with that via the total diet showed that the dietary exposure to MOSH and MOAH was only about 2% of the total exposure at the median level (P50), both in 2- to 6-year olds and in persons aged 7 to 69 years. At the high level of exposure (P95, largely due to the high consumers of pasta), this percentage increased to around 15% for 2 to 6-year olds and 18% for persons aged 7 to 69.

Contribution food groups to the exposure
Figure 4 shows the contribution of the food groups to the total exposure distribution of MOSH via the total diet. Food groups that contributed at least 10% to the exposure to MOSH in children aged 2 to 6 were ‘confectionery (non-chocolate)’, ‘pasta’, ‘ice and desserts’ and ‘vegetable products’. For persons aged 7 to 69, two food groups contributed at least 10% to the exposure to MOSH: ‘pasta’ and ‘herbs, spices and condiments’.

If only the exposure to MOSH via paperboard packaged foods was considered, the food group ‘pasta’ was the main contributor to the exposure to MOSH. Percentages of contribution increased to 75% and 84% in children aged 2 to 6 and persons aged 7 to 69, respectively.

The concentrations of MOAH used in the exposure assessment were (partly) predefined as a fixed fraction of the MOSH concentrations. The same food groups are therefore expected to contribute to the exposure to MOAH in the same order of magnitude.

Table 2. Exposure to MOSH and MOAH via food in the Dutch population

Concentration data[1]	Population and exposure (mg/kg bw per day)			
	2-6 years		7-69 years	
	P50	P95	P50	P95
MOSH				
Total diet	0.098 [0.087-0.10]	0.21 [0.17-0.40]	0.042 [0.035-0.045]	0.12 [0.093-0.25]
Paperboard packages	0.0015 [0.0011-0.0019]	0.033 [0.016-0.053]	0.00075 [0.00057-0.00084]	0.023 [0.0083-0.035]
MOAH				
Total diet	0.014 [0.013-0.014]	0.026 [0.025-0.028]	0.0055 [0.0052-0.0060]	0.014 [0.013-0.016]
Paperboard packages	0.00023 [0.00017-0.00028]	0.0037 [0.0017-0.0068]	0.00013 [0.00010-0.00014]	0.0024 [0.00089-0.0049]

Note: 2.5% lower – 97.5% upper confidence limits of the percentiles of exposure are reported between brackets.
[1] Total diet: MOSH and MOAH concentrations of the EFSA CONTAM Panel (EFSA, 2012b) and Foodwatch (2015; 2016a,b); Paperboard package: MOAH and MOAH concentration of Foodwatch analysed in foods packaged in paperboard boxes (pasta, rice, breakfast cereals, cereal products and sprinkles). For more details, see section 5.1.2.

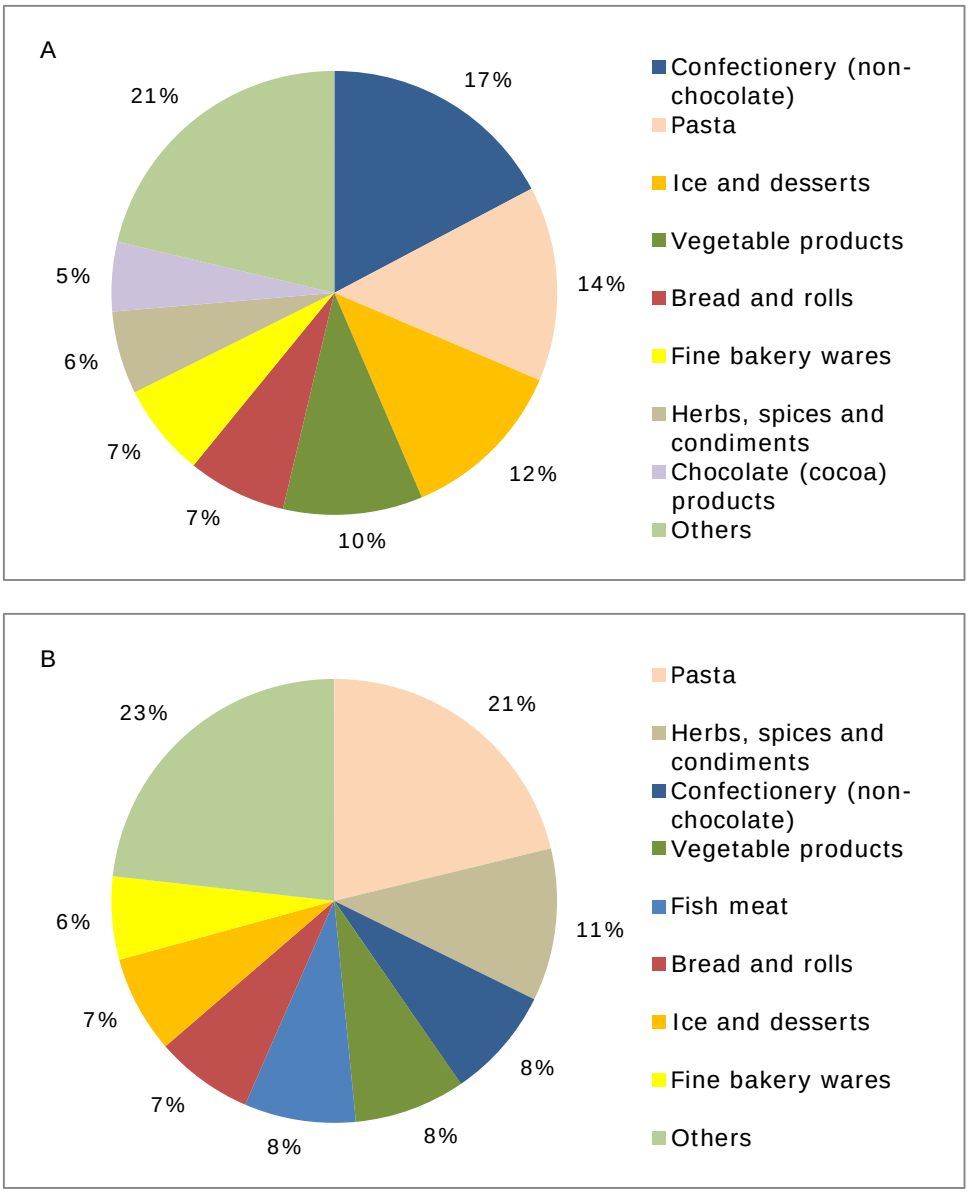


Figure 4. Contribution (%) of food groups, with a contribution of at least 5%, to the total exposure distribution to MOSH in children aged 2 to 6 (A) and persons aged 7 to 69 (B) via the total diet.

5.5 Discussion of the exposure assessment

5.5.1 Comparison with intake reported by the EFSA CONTAM Panel

In 2012, the EFSA CONTAM Panel reported on the exposure to MOSH via food in different population groups (EFSA, 2012b). These exposure results were based on the concentration data as described in section 5.1.2.2 of this report combined with food consumption data of different national dietary surveys of European countries, including the Netherlands. Exposure estimates for the Netherlands were based on food consumption data from the DNFCS-Young children, also included in the current study (section 5.1.1), and the DNFCS 2003 (Ocké *et al.* 2005) covering adults aged 19 to 30. Exposure results reported by the EFSA CONTAM Panel (EFSA, 2012b) are reported in Table 3 for the age

groups relevant for this study, as well as the exposure estimates of the current study for the total diet. In general, the exposure estimates of the current study were in the same order of magnitude as those reported by the EFSA CONTAM Panel (EFSA, 2012b).

Table 3. Mean, median (P50) and high (P95) exposure to MOSH via food in children and adults as estimated by EFSA CONTAM Panel (EFSA, 2012b) and in the current study

Population	Exposure (mg/kg bw per day)	
EFSA CONTAM Panel (EFSA, 2012b)		
	Mean	P95
Toddlers[1,2]	0.038-0.19[3]	0.18-0.26
Other children	0.083-0.17	0.14-0.32
Adolescents	0.066-0.096	0.063-0.20
Adults	0.031-0.068	0.059-0.12
Elderly	0.031-0.059	0.058-0.096
Current study (via total diet)		
	P50	P95
2-6 years	0.098[4]	0.21
7-69 years	0.042	0.12

[1] Toddlers: 12-35 months of age; other children: 3-9 years; adolescents: 10-17 years; adults: 18-64 years; elderly: 65-74 years
[2] Only the exposure results reported by the EFSA CONTAM Panel (EFSA, 2012b) for toddlers, other children and adults are partly based on Dutch food consumption data. The EFSA CONTAM Panel (EFSA, 2012b) did not report specific exposure results per national dietary survey.
[3] Ranges refer to the lowest (lower bound) and highest (upper bound) exposure estimated across national dietary surveys (section 5.1.2.3).
[4] Medium bound estimates of exposure (section 5.1.2.3)

Exposure estimates at the national level are usually lower than those reported by Panels of EFSA (Sprong & Boon, 2015; Boon *et al.*, 2017). The reasons for this could be a more precise mapping of foods consumed to those analysed, lower (national) concentrations of the compound in foods and/or the use of a more advanced model to assess the long-term exposure. The reason that the exposure levels of the present study were within the range of those assessed by the EFSA CONTAM Panel was very likely due to the use of predominantly the same concentration data as the EFSA CONTAM Panel (EFSA, 2012b). Because of this, also the possibility to refine the assessment via a more precise mapping of foods was very limited. Furthermore, the model used to estimate the exposure was also the same as used by EFSA CONTAM Panel.

The exposure estimates were higher in young children compared to the older population groups both in the current study and as estimated by the EFSA CONTAM Panel (Table 3). The reason for this was that children consume more food per kg bodyweight and not because they consume specific foods that contain higher levels of MOSH (and MOAH).

5.5.2 Concentration data and food mapping

The exposure assessment of MOSH and MOAH described in this report is subject to different sources of uncertainty. The main sources were related to the concentration data used and the way in which these data

were mapped to the foods recorded in the food consumption databases. In this section, the most important aspects of these two uncertainties are discussed.

Concentration data
The majority of the concentration data used in the exposure assessment was derived from the EFSA CONTAM Panel, which were related to concentrations analysed in foods available on the European market (EFSA, 2012b). These data were mainly derived from one enforcement laboratory and were for a large part obtained via targeted sampling, which may have caused a bias to higher concentrations. Comparing the concentrations of Foodwatch with those of the EFSA CONTAM Panel showed indeed that the concentrations reported by the Panel tended to be higher. The MOSH concentrations of the FoodEx1 food groups 'grain milling products' and 'grains for human consumption' (referring predominantly to rice) were significantly higher than the concentrations in comparable foods obtained from Foodwatch. The average of the LB and UB MOSH concentrations in these food groups were 9.3 and 132 mg/kg respectively (EFSA, 2012b), whereas the average modelled concentrations based on concentrations reported by Foodwatch were 5.4 and 1.4 mg/kg, respectively (Appendix D). The average modelled concentration in chocolate analysed by Foodwatch was also lower than the one for the comparable FoodEx1 food group 'chocolate (cocoa) products': 5.1 versus 11 mg/kg (Appendix C and D). An exception was the MOSH concentration in pasta. The average modelled concentration used in the current assessment, based on data from Foodwatch, was 30.4 mg/kg (Appendix D), whereas the average of the mean LB and UB concentrations reported by the EFSA CONTAM Panel was 11 mg/kg (EFSA, 2012b). The reason for this was a high MOSH concentration in one pasta brand: 133 mg/kg (Appendix B). This high average level was also the reason why pasta contributed highly to the exposure via the total diet in both population groups (Figure 4). Removing this concentration resulted in a mean modelled MOSH concentration of 11.2 mg/kg (Appendix D; footnote 2): comparable to the level reported by the EFSA CONTAM Panel (EFSA, 2012b). These differences in concentrations between Foodwatch and those reported by the EFSA CONTAM Panel may also have been partly due to the way in which the concentrations of Foodwatch were included in the present study (section 5.1.2). However, as the arithmetic mean MOSH concentrations were very close to the modelled ones, this is not likely (Appendix B and D).

Another potential source of uncertainty related to the concentration data of the EFSA CONTAM Panel (EFSA, 2012b) was the use of medium bound (MB) concentrations based on reported lower (LB) and upper bound (UB) concentrations of MOSH (section 5.1.2). This may potentially have resulted in an erroneous estimation of the exposure if the LB and UB concentrations differ greatly. This was however not the case. The LB and UB concentrations of MOSH were identical for 10 out of the 20 FoodEx1 food groups considered in this assessment, and the LB concentration was more than 90% of the UB concentration for eight food groups. For the FoodEx1 food groups 'sausages' and 'legumes, beans, dried', the LB concentration was 80% and 67% of the UB concentration, respectively. As the difference between the LB and UB concentrations

was overall very small, a possible erroneous estimation of the exposure due to use of MB concentrations based on reported LB and UB concentrations was considered negligible.

When assessing the long-term exposure, mean concentrations are used assuming that the concentrations to which people are exposed over a longer period of time will level out. In the present study, mean concentrations per food group were used. This may have introduced an uncertainty in the exposure estimates as the concentrations could vary considerably between brands of foods within a food group as made evident by the analysed concentrations of Foodwatch (Appendix B). If concentrations in a certain brand tend to be always higher or lower than the mean concentration of the food group and a consumer is loyal to that brand, the exposure may potentially be either much higher or lower than calculated based on a mean concentration. However, it is unclear whether high or low levels within a certain brand will remain low or high in time or will fluctuate. Given the information available, use of the mean concentration per food group was therefore considered the best possible approach.

Regarding the data of Foodwatch (Appendix B), the number of foods included in this assessment packaged using paperboard boxes was very limited, making it uncertain how well these foods cover all foods packaged in this manner (Appendix B). For example, the analyses in cacao powder, fruit-flavoured sprinkles and oat meal/muesli were based on just one sample. The maximum number of samples was 30 for the food group ‘chocolate’. Additionally, the mean concentrations obtained from the EFSA CONTAM Panel (EFSA, 2012b) were often also based on a limited number of foods. For example, the mean MOSH and MOAH concentrations of the FoodEx1 food groups ‘snack food’ and ‘sugar’ were based on just four samples.

In the exposure assessment, the exposure to MOSH and MOAH via consumption of fresh fruits and vegetables was not considered, as no realistic concentrations of MOSH and MOAH were available from the EFSA CONTAM Panel opinion (EFSA, 2012b) or Foodwatch (2015; 2016a,b). This may have resulted in an underestimation of the exposure, as MOH can be present due to natural occurrence, spraying of crops or via protective coating on the surface of fruits and vegetables. MOSH and MOAH in processed fruits and vegetables (glass, canned or deep-frozen) have however been considered.

Food mapping
Food mapping is another important source of uncertainty in this exposure assessment. This uncertainty was due to the choices made when mapping the foods analysed to those recorded in the food consumption databases given the time constraints and the limitations in detail of both the analysed and recorded foods.

In the assessment, only foods consumed as such were mapped to analysed food groups. For example, the concentrations in cacao powder were only included in the exposure assessment when consumption of cacao powder was recorded as such in the food consumption databases. Foods containing this product as an ingredient, like chocolate custard,

were not included. Note that chocolate bars, chocolate coated confectionery and chocolate paste were included in the assessment via the EFSA FoodEx1 food group ‘chocolate (cocoa) products’.

It was furthermore assumed that foods analysed by Foodwatch (such as pasta, rice, sprinkles and cereal products) were all consumed as packaged in paperboard, due to the absence of information of this type of packaging in the food consumption databases. This has likely resulted in an overestimation of the exposure, as people consume these foods also in other types of packaging, such as plastic. Additionally, the MOSH and MOAH concentrations of Foodwatch in dry white rice and pasta were directly mapped to the consumed amounts of these foods in the food consumption databases. These amounts are however based on the consumption of prepared foods, which contain a large amount of water, and not on the dry product. The exposure to MOSH and MOAH via these dietary sources has been overestimated.

Concentrations of MOSH and MOAH in vegetable products were only mapped to the consumption of vegetables and fruits recorded as glass/can or deep-frozen in the food consumption databases (section 5.2). This is a potential source of underestimation. During the collection of the food consumption data, this information was not always recorded because the respondent was not aware of the type of packaging. In those cases, the consumption was recorded as consumed as such (possibly after cooking).

Based on the uncertainties listed above regarding the concentration data and food mapping, the reported exposure estimates of MOSH and MOAH are highly uncertain, and should at the most be regarded as an indication of the exposure to MOSH and MOAH via food in Dutch consumers.

5.5.3 Modelling of exposure

In this study, the Observed Individual Means (OIM) model was used to assess the long-term exposure to MOSH and MOAH. This is the same model as used by the EFSA CONTAM Panel (EFSA, 2012b). In this model, the distribution of individual mean intakes over the person-days present in the food consumption is taken as a proxy for the long-term intake distribution. Given the limited number of person-days present in a food consumption database per person, in our case two (Appendix A), and the variation in daily food consumption patterns within an individual, the distribution of mean exposures over individuals obtained with OIM will often be too wide in comparison to distributions of ‘true’ long term exposure (Goedhart *et al.*, 2012). This results in exposures that are about right in the middle of the exposure distribution, but are too high in the upper tail and too low in the lower tail of the exposure distribution.

If a more realistic exposure estimate is warranted, for example if the exposure is close to the health-based guidance value or results in an insufficiently large margin of exposure, more advanced models as available in MCRA, may be used to assess the ‘true’ long-term exposure (Boon & van der Voet, 2015). Given the indicative character of this exposure assessment, modelling the exposure via a more advance long-

term exposure model was not considered. The contribution of the food groups to exposure will remain the same when using more advance models.

Furthermore, since the uncertainty of the sample size of the concentration database could only be included in the exposure assessment for the Foodwatch concentration data (section 5.3), the width of the confidence interval due to this uncertainty was underestimated.

5.5.4 *'High level' versus background concentration MOSH*

In the current assessment, the background concentration of MOSH as modelled by the EFSA CONTAM Panel for the FoodEx1 food group 'bread and rolls' was used (EFSA, 2012b). Apart from this concentration, the EFSA CONTAM Panel also modelled 'high level' concentrations in this food group, as well as in the FoodEx1 food group 'grains for human consumption' (section 5.1.2). The MOSH concentration in this latter food group were predominantly based on those in rice. The EFSA CONTAM Panel used the mean concentrations for both food groups with high levels of MOSH in two separate 'high exposure' scenarios to also "consider that specific consumers might be exposed during long periods to bread or cereal grains with high levels of MOSH, due to restricted choice in the food supply or to brand loyalty". Since these high concentrations were explained to arise from the use of food grade white oils, which are virtually free of MOAH, these 'high exposures' were only calculated for MOSH (EFSA, 2012b).

If these 'high level' concentrations are used in the current exposure assessment, the exposure is completely driven by these two food groups due to the very high concentrations of MOSH: 532 and 977 mg/kg for 'bread and rolls' and 'grains', compared to the background concentrations of 1.8 mg/kg (EFSA 2012b) and 1.4 mg/kg (rice, appendix D), respectively. Consumption of highly contaminated bread will result in exposure levels of MOSH of about 2-3 mg/kg bw per day. As the consumption of rice is less predominant in the Netherlands, the exposure to highly contaminated rice will result in an exposure to MOSH of about 0.2 mg/kg bw per day.

5.5.5 *Paperboard packaged food versus total diet*

The EFSA CONTAM Panel noted that the use of recycled paperboard as packaging material may be a significant source of dietary exposure to MOSH and MOAH (EFSA, 2012b). This was the reason why Foodwatch monitored pasta, rice, breakfast cereals, cereal products and cacao powder packaged in paperboard boxes, as well as a typical Dutch food items as chocolate and fruit-flavoured sprinkles.

The exposure to MOSH and MOAH via paperboard packaged foods was also calculated and shown to be very limited compared to the exposure via the total diet: only 2% at the median exposure level. This contribution was higher at upper intake percentiles (P95, largely due to high consumers of pasta); around 15% for 2 to 6-year olds and 18% for persons aged 7 to 69.

This relative low contribution of paperboard packaged foods to the total exposure to MOSH and MOAH seems to conflict with data presented in Figure 4, which indicates that pasta already contributed for 14% (young children) and 21% (persons aged 7-69) to the total dietary intake of MOSH. However, not all MOSH in pasta is derived from paperboard; it can also be derived from other sources, such as the use of white oils. This was the case in the pasta-sample with a high MOSH concentration (133 mg/kg) combined with a low MOAH concentration (0.6 mg/kg). This MOSH concentrations was excluded from the intake calculation via paperboard packaged foods.

Foodwatch made a distinction between foods packaged in paperboard made of recycled and fresh fibers. Taking into account only the concentration data of food that was packaged in recycled paperboard resulted in comparable exposure estimates as those considering all foods packaged in paperboard derived from either recycled or fresh fibers. The average MOSH- and MOAH-levels were higher in foods packaged in recycled paperboard, but this paperboard is apparently not used for all types of dry foods. The Foodwatch data showed that it is used for pasta, breakfast cereals and other cereal products, but not for e.g. rice and sprinkles.

The current intake assessment for the Dutch population does not support the conclusion of the EFSA CONTAM Panel (EFSA, 2012b) that the exposure to MOSH from migration into dry foods packaged in recycled paperboard may contribute significantly to the total dietary exposure.

6 Risk assessment and conclusions

6.1 Risk assessment of MOSH

Conform the EFSA opinion, by lack of an ADI, the MOE approach was used for the risk assessment of MOSH. The NOAEL for the most potent MOSH grades for microgranulomas of the liver, which was 19 mg/kg bw per day for low and intermediate melting point waxes was selected as the Reference Point (EFSA 2012b). MOEs were calculated by dividing this NOAEL by the calculated intake levels for MOSH for the Dutch population. Results are listed in Table 4.

Table 4. Margins of exposure (MOEs) calculated for the exposure to MOSH (median and high (P95) consumption)

Population	Exposure[1] (mg/kg bw/day)		MOE[2] (NOAEL[3]: 19 mg/kg bw/day)	
	P50	**P95**	**P50**	**P95**
2-6 years	0.098	0.21	190	90
7-69 years	0.042	0.12	450	160

[1] Medium bound estimates of exposure (see section 5.1.2.3)
[2] MOE: Margin of Exposure
[3] NOAEL (non-observed adverse effect level) which was used as Reference Point

For a negligible health risk, the MOE must have a minimum value, depending on the nature of the critical endpoint on which the NOAEL is based that is used as Reference Point. The EFSA CONTAM Panel did not mention the minimum value of MOE needed to consider an exposure to MOSH as of 'no concern' (EFSA, 2012b). For not-genotoxic substances, a MOE of at least 100 is usually considered to indicate an exposure of no concern.

Median exposure levels to MOSH via the total diet resulted in MOEs of 190 in children aged 2 to 6, and 450 in persons aged 7 to 69;the corresponding MOEs for the high (P95) exposure were 90 and 160, respectively. The MOE of 90 for the high exposure of young children is somewhat lower than the minimal value of 100. However, since the toxicological Reference Point is based on effects observed after repeated exposure and the MOE is only temporarily slightly below 100, and given the questionable toxicological relevance for humans of the liver microgranulomas observed in rats, it was concluded that the estimated exposure levels to MOSH were of no health concern for the Dutch population.

For white mineral oils used as release agents for bread and rolls and for spraying of grains (including rice), a higher NOAEL of 45 mg/kg bw per day was selected as Refference Point for the MOE calculation by the EFSA CONTAM Panel (2012b). A separate 'high level' intake assessment for these uses was performed by the EFSA CONTAM Panel (2012b). The MOEs were mostly below 100 and sometimes even lower than 10, and the panel identified these uses of white oils as of potential concern. The preliminary assessment for the Dutch population supports this

conclusion for the food item ‘bread and rolls’. EFSA recommended to include these food items in monitoring activities.

In Barp *et al.* (2017b), it was suggested that mineral release agents are no longer used for bakery ware. New monitoring data are needed to assess if this exposure is still of potential concern.

6.2 Risk assessment of MOAH

MOAH are potentially mutagenic and carcinogenic and thus, their presence in foods is considered of concern. The common approach for the risk characterisation of genotoxic carcinogens is to calculate a margin of exposure (MOE), but neither for MOAH mixtures, nor for mineral oils, dose-response data on the carcinogenicity is available. Therefore it is not possible to establish a Reference Point that could be used for the calculation of the MOE (EFSA 2012). The new toxicity studies do not include any *in vivo* dose-response toxicity data on MOAH either, so this is not changed.

The mutagenicity of MOAH is caused mainly by aromatic 3-7 fused rings (Polycyclic Aromatic Hydrocarbons or PAH), with no or a low degree of alkylation. The non-alkylated PAH is a minor fraction of the MOAH. It is mainly formed by heating of the oil. These non-alkylated PAH are covered by monitoring programmes in food (EFSA, 2012b). In these programmes, specific non-alkylated PAH’s (benzo[a]pyrene, and/or the sum of 2, 4 or 8 PAH (PAH2, PAH4 or PAH8)) are analysed in food. The CONTAM Panel concluded that PAH4 and PAH8 are the most suitable indicators of PAH in food (EFSA 2008). Although PAH in food may result mainly from other sources (e.g. smoke flavourings, burning of food) than from mineral oils, it is worth mentioning that in the latest evaluation of PAH in food by EFSA, exposure to PAH due to dietary intake was considered of ‘low concern’ for the average consumer (conclusion drawn for exposure that have a Margin of Exposure (MOE) >10,000); while for high level consumers, MOE did vary from 9,500 to 10,800, indicating a possible need for risk management actions (EFSA 2008).

The markers selected for this analysis of PAH in food are based on examinations of PAH profiles in food and on evaluation of a carcinogenicity study of two coal tar mixtures in mice (EFSA 2008). Coal tar is produced through thermal destruction (pyrolysis) of coal. PAH profiles differ between pyrogenic and petrogenic PAH; pyrogenic being predominantly non-alkylated PAH, and petrogenic PAH being predominantly alkylated PAH (Stogiannidis *et al.* 2015). Therefore, the markers used in the PAH monitoring in food will cover the carcinogenicity caused by pyrogenic PAH (e.g. present in mineral oils that have been heated), but will only to a limited extent cover that of petrogenic PAH (that are originally present in (uncombusted) mineral oils).

Alkylated PAH are not covered by the PAH-monitoring programmes. Considering however that MOAH with long alkyl-chain substitutions are less likely to be mutagenic, the concern about MOAH coming from sources containing predominantly this type of MOAH will be considerably

lower than for MOAH from other sources, containing MOAH with short alkyl-chain substitutions. The refining technique used to obtain wax (wax being a fraction of oil predominantly consisting of long *n*-alkanes) is aimed to select compounds with long *n*-alkanes. Therefore, the MOAH in this fraction will most likely also be predominantly long-*n*-alkylated. Indeed, as measured with NMR-spectrometry, the relative proportion of aromatic protons in wax (that is; the proportion of the atomweight of atoms that are part of an aromatic ring) is much lower (i.e. <0,2% of total MOH, Lachenmeijer 2017) than the MOAH-content measured as relative weight fraction; normally in the range of 1-5% in waxes, as measured by (HP)LC-GC-FID, indicating that the MOAH in wax are highly alkylated, having no or low genotoxic potential.

Highly purfied oils have been shown to be non-mutagenic, both in vitro and in vivo. This would lead to the conclusion that the use of highly purified (i.e. food grade) oils in all processes of the food production chain would overcome the concern on carcinogenicity from MOAH from this source.

If the codes of practices[8] are complied with, as required by the European general food law (Reg (EC) No 178/2002), lubricants used in machinery for food processing should contain only mineral oils that are not classified as carcinogenic. For carcinogenicity classification and labelling of mineral oils, the European Chemical Agency (ECHA) accepted the use of IP346-assay (see Note L in Annex VI of Reg (EC) No 1272/2008). The IP346 assay is a gravimetric method, based on good solubility of the aromatic fraction in DMSO; after extraction of the mineral oil with DMSO and removal of solvent, the weight percent of the MOAH is determined. The IP346-assay was compared with the mice skin-painting test; refined mineral oils with an extractable fraction in the IP346 assay below 3% are shown tohave a good correlation with negative results in the mice skin-painting test (literature review of Concawe, 2016), and do not need to be classified as to their carcinogenicity. MOH-fractions not passing the <3% extractable weight in the IP346 test will be retreated to eliminate the aromatics. The IP346 is easy to perform and can therefore be used on all mineral oil products marketed.

It was noted that non-mutagenic refined mineral oils may become mutagenic after use (EFSA 2012b). High temperature processes, e.g. >800 °C can convert non-mutagenic mineral oil components into mutagenic PAH, and motor oils in addition to thermal decomposition may also pick up engine combustion products such that after use they may contain several orders of magnitude higher concentrations of PAH (EFSA 2012). It is not known to what extent this will contribute to the formation of mutagenic components and their contamination in food. Contribution of these pyrogenic sources is however included in the PAH monitoring programmes, as explained above, which were of low concern to the average consumer (EFSA 2008).

It can be concluded that the method currently used to determine MOAH in food (HPLC-GC/FID, or LC-GC/FID), is useful to measure the total

[8] Code of Practice and Technical Standard for food processors and suppliers to the public sector,

concentration of MOAH, but it does not make a distinction between the different types of MOAHs, and therefore it is of limited predictive value as to the carcinogenicity of the fraction.
Other information on MOAH is essential, either on the structure (subclasses) of MOAH, or on the source of the MOAH-contamination. Knowing the source of the MOAH in the food will contribute to the risk assessment and may be helpful to design effective risk mitigation measures. The monitoring action initiated by the European Commission, with a call for data on MOSH and MOAH content in various products and maybe at various stages of processing, will provide information on the sources of MOH in food and will therefore be expected to help formulate adequate risk mitigation measures.

6.3 Conclusions and considerations

- The new studies on the metabolism and toxicity of mineral oils published since 2012, do not lead to a change in the Reference Point for MOSH as selected by EFSA. They do however contribute to the discussion on the relevance for humans of liver granulomas, which is considered the critical effect for selection of the Reference Point in the 2012 EFSA report. In two new studies in rats on immunotoxicity, no indication was found that MOSH exposure via the oral route is associated with systemic autoimmune diseases or altered immune function as observed after some parenteral exposure.
- Dietary intake calculations for the Dutch population indicate that no health effects are to be expected if people are exposed to MOSH via food.
- For MOAH it is not possible to make a quantitative risk assessment, due to lack of data to derive a health-based guidance value. Furthermore it is not possible to point out marker molecules for the MOAH risk assessment. MOAH can be mutagenic and carcinogenic, but not all mineral oils from which MOAH can end up in food are carcinogenic. Risks of MOAH might be reduced by first identifying the sources of contamination, and then taking measures to avoid those sources that contain potentially carcinogenic MOAHs, such as crude and heated oils.
- The current intake assessment for the Dutch population does not support the conclusion of the EFSA CONTAM Panel (EFSA, 2012b) that the exposure to MOSH from migration into dry foods packaged in recycled paperboard may contribute significantly to the total dietary exposure. Paperboard packaged dry foods such as rice, pasta, breakfast cereals, and chocolate sprinkles make only a small contribution to the total exposure to mineral oils via food. Measures aimed at reducing the exposure resulting from the use of paperboard packaging would therefore have only a limited effect.

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8 References

Adenuga D, Carrillo JC, McKee RH, 2014. The sub-chronic oral toxicity of dearomatized hydrocarbon solvents in Sprague-Dawley rats. Regulatory Toxicology and Pharmacology, 2014; 70 (3):659-72.

Adenuga D, Goyak K, Lewis J, 2017. Evaluating the MoA/human relevance framework for F-344 rat liver epithelioid granulomas with mineral oil hydrocarbons. Critical Reviews in Toxicology 7:9, 750-766.

Ainsworth AJ, Fredrickson JR, Morbeck DE, 2017. Improved detection of mineral oil toxicity using an extended mouse embryo assay. Journal of Assisted Reproduction and Genetics, 2017; 34 (3):391-7.

Andreassen M, Hjertholm H, Cravedi JP et al., 2017. Effect of dietary pristane and other saturated mineral oils (mosh) on autoimmune arthritis in rats. Toxicology Reports, 2017; 4:104-12.

ANSES, 2017. Opinion of the French Agency for Food, Environmental and Occupational Health & Safety on the migration of mineral oil compounds into food from recycled paper and cardboard packaging. Request No 2015-SA-0070.

Barp L, Kornauth C, Wuerger T, 2014. Mineral oil in human tissues, Part I: Concentrations and molecular mass distributions Food and Chemical Toxicology, 2014; 72:312-21.

Barp L, Biedermann M, Grob K et al., 2017a. Accumulation of mineral oil saturated hydrocarbons (MOSH) in female Fischer 344 rats: Comparison with human data and consequences for risk assessment. Science of the Total Environment, 2017; 575:1263-78.

Barp L, Biedermann M, Grob K et al., 2017b. Mineral oil saturated hydrocarbons (MOSH) in female Fischer 344 rats; accumulation of wax components; implications for risk assessment. Science of the Total Environment 2017; 583: 319–33.

Biedermann M, Barp L, Kornauth C et al., 2015. Mineral oil in human tissues, part II: Characterization of the accumulated hydrocarbons by comprehensive two-dimensional gas chromatography. Science of the Total Environment, 2015; 506–507:644-55.

Boogaard PJ, Goyak KO, Biles RW et al., 2012. Comparative toxicokinetics of low-viscosity mineral oil in Fischer 344 rats, sprague-dawley rats, and humans - implications for an acceptable daily intake (adi). Regulatory Toxicology and Pharmacology, 2012; 63 (1):69-77.

Boon PE, Ruprich J, Petersen A, Moussavian S et al., 2009. Harmonisation of food consumption data format for dietary exposure assessments of chemicals analysed in raw agricultural commodities. Food and Chemical Toxicology 47: 2883-2889.

Boon PE, te Biesebeek JD, Donkersgoed G, 2017. Dietary exposure to lead in the Netherlands. RIVM Letter report 2016-0206. National Institute for Public Health and the Environment (RIVM), Bilthoven. Available online: www.rivm.nl.

Boon PE, van der Voet H, 2015. Probabilistic dietary exposure models. Reportnr: 2015-0191. National Institute for Public Health and the Environment (RIVM), Bilthoven. Available online: www.rivm.nl.

Concawe 2016. Critical review of the relationship between IP346 and dermal carcinogenic activity. Report 6/16, March 2016.

Cravedi JP, Grob K, Nygaard UC et al., (2017) Bioaccumulation and toxicity of mineral oil hydrocarbons in rats - specificity of different subclasses of a broad mixture relevant for human dietary exposures. INRI, KLZ, NIPH report doi:10.2903/sp.efsa.2017.EN-1090

de Boer WJ, Goedhart PW, Hart A et al., 2016. MCRA 8.2 a web-based program for Monte Carlo Risk Assessment. Reference Manual. December, 2016. Biometris, Wageningen UR, Food and Environmmental Research Agency (Fera) and National Institute for Public Health and the Environment (RIVM), Wageningen, Bilthoven, the Netherlands and York, UK.

EFSA, 2008. Polycyclic aromatic hydrocarbons in food. Scientific opinion of the panel on contaminants in the food chain. EFSA Journal 742:1-114. EFSA, 2012.

EFSA, 2011. Evaluation of the FoodEx, the food classification system applied to the development of the EFSA Comprehensive European Food Consumption Database. EFSA Journal 2011;9(3):1970, 27 pp. Available online: www.efsa.europa.eu

EFSA, 2012a. Guidance on the Use of Probabilistic Methodology for Modelling Dietary Exposure to Pesticide Residues. EFSA Journal 10(10):2839, 95 pp. Available online: www.efsa.europa.eu.

EFSA, 2012b. Scientific Opinion on Mineral Oil Hydrocarbons in Food. EFSA Panel on Contaminants in the Food Chain (CONTAM). EFSA Journal 2012;10(6):2704. [185 pp.] doi:10.2903/j.efsa.2012.2704. Available online: www.efsa.europa.eu

EFSA, 2015. Scientific Opinion on acrylamide in food. EFSA Panel on Contaminants in the Food Chain (CONTAM). EFSA Journal 13(6):4104, 321 pp. Available online: www.efsa.europa.eu.

EFSA, 2016. Scientific opinion on the risks for human health related to the presence of 3- and 2-monochloropropanediol (MCPD), and their fatty acid esters, and glycidyl fatty acid esters in food. EFSA Journal 2016;14(5): 4426. [159 pp.]. Available online: www.efsa.europa.eu.

Efron B, 1979. Bootstrap methods: another look at the jackknife. Annals of Statistics 7: 1-26.

Efron B, Tibshirani R, 1993. An introduction to the bootstrap. New York: Chapman & Hall.

Foodwatch, 2015. Besmetting van ons voedsel met minerale oliën. Resultaten van de foodwatch Test. Foodwatch Nederland, oktober 2015. Available online: https://www.foodwatch.org/fileadmin/foodwatch.nl/Onze_campagnes/Schadelijke_stoffen/Documents/Rapport_Besmetting_van_ons_voedsel_met_minerale_olie_-_testresultaten.pdf.

Foodwatch, 2016a. Mineröl in Schokoladen-Osterhasen. Foodwatch Deutschland, März 2016. Available online: https://www.foodwatch.org/fileadmin/foodwatch.nl/Onze_campagnes/Schadelijke_stoffen/Vitamin_research/Documents/Final_Schoko-Hasenreport.pdf

Foodwatch, 2016b. Mineröle in Schokoladen-Weihnachtsmännern. Foodwatch Deutschland, December 2016. (kan ik niet meer vinden op internet)

JECFA (Joint FAO/WHO Expert Committee on food Additives), 2002. Evaluation of certain food additives: fifty-ninth report of the Joint FAO/WHO Expert Committee on food Additives. WHO Technical Report Series 913, 2002.

Kimber I, Carrillo JC, 2016. Oral exposure to mineral oils: Is there an association with immune perturbation and autoimmunity? Toxicology, 2016; 344-346:19-25.

Miligi L, Benvenuti A, Mattioli S et al., 2013. Risk of childhood leukaemia and non-hodgkin's lymphoma after parental occupational exposure to solvents and other agents: The setil study. Occupational and Environmental Medicine, 2013; 70 (9):648-55.

Ocké MC, Hulshof KFAM, van Rossum CTM, 2005. The Dutch national food consumption survey 2003. Methodological issues. Archives of Public Health 63: 227-241.

Ocké MC, van Rossum CTM, Fransen HP et al., (2008). Dutch National Food Consumption Survey - Young children 2005/2006. Reportnr: 350070001/2008. National Institute for Public Health and the Environment (RIVM), Bilthoven. Available online: www.rivm.nl.

Sprong RC, Boon PE, 2015. Dietary exposure to cadmium in the Netherlands. Reportnr: 2015-0085. National Institute for Public Health and the Environment (RIVM), Bilthoven. Available online: www.rivm.nl.

Stogiannidis E, Laane R, 2015 Source characterization of Polycyclic Aromatic Hydrocarbons by using their molecular indices: An overview of possibilities. Chapter in: Withacre D. (Ed.) “Reviews of Environmental Contamination and Toxicology” Volume 234:50-133. ISBN 978 3 319 10638 0

Tarnow P, Hutzler C, Grabiger S et al., 2016. Estrogenic activity of mineral oil aromatic hydrocarbons used in printing inks. PLoS ONE, 2016; 11 (1).

van der Voet H, de Boer WJ, Kruisselbrink JW et al, 2015. The MCRA model for probabilistic single-compound and cumulative risk assessment of pesticides. Food and Chemical Toxicology 79: 5-12.

van Dooren MMH, Boeijen I, van Klaveren JD et al., 1995. Conversie van consumeerbare voedingsmiddelen naar primaire agrarische produkten (Conversion of consumed foods into raw agricultural commodities). Reportnr: 95.17. RIKILT-Instituut voor Voedselveiligheid, Wageningen UR, Wageningen. Available online: www.rikilt.wur.nl.

van Rossum CTM, Fransen HP, Verkaik-Kloosterman J et al., 2011. Dutch National Food Consumption Survey 2007-2010. Diet of children and adults aged 7 to 69 years. Reportnr: 350050006/2011. National Institute for Public Health and the Environment (RIVM), Bilthoven. Available online: www.rivm.nl.

Appendix A Description of consumption data used in the exposure assessment to MOSH and MOAH

DNFCS-Young Children 2005/2006 (Ocké *et al.*, 2008)
The target population of the DNFCS-Young Children 2005/2006 consisted of boys and girls aged 2 to 6 living in the Netherlands. Respondents were selected from representative consumer panels of Market Research Agency GfK. Panel characteristics, such as socio-demographic characteristics, are known to GfK. Persons in these panels participate in all types of surveys and were not specially selected on nutritional characteristics. Institutionalised persons were excluded, as well as children whose parents/carers did not have sufficient knowledge of the Dutch language. Per family, only one child was included to avoid correlations in dietary consumption patterns between children of the same family. In total, 1,634 children were invited to participate in the study, of which 1,279 consented (net response of 78%). During recruitment, the representativeness of the study population was monitored and, if necessary, the recruitment was adjusted for age and sex, education of the head of the household, level of urbanisation, place of residence and region. The study population was representative regarding socio-demographic characteristics (including region and education of the head of the household), but densely populated areas were slightly underrepresented.

The food consumption data were collected in the period October 2005 to November 2006 on two non-consecutive days (separated by about 8 to 13 days) via food diary. Parents/carers were visited at home by a trained employee of GfK. During the home visit, survey materials were presented and overall instructions were given.

Portion size of the foods and meals were estimated by using photographs, domestic measures (a small and a large spoon were supplied to standardise estimates), standard units, weight and/or volume. The usual volume of cups and glasses used was measured by the carer. All days of the week were equally represented, but the winter and autumn period were slightly overrepresented compared to the spring and summer period. National and/or religious holidays or holidays of the participants were not included in the survey.

DNFCS 2007-2010 (van Rossum *et al.*, 2011)
The target population of the DNFCS 2007-2010 consisted of people aged 7 to 69 living in the Netherlands. Pregnant and breast-feeding women, as well as institutionalised people were not included. Respondents were selected from representative consumer panels of GfK. A maximum of one person per household was included in the survey to avoid correlations in dietary consumption patterns between members of the same family. In addition, the panels only included people with sufficient knowledge of the Dutch language. In total, 5,502 individuals were invited to participate in the study, of which 3,819 consented (net response of 69%). Children were overrepresented in the study population and adults underrepresented.

The food consumption data were collected over a 3-year period from March 2007 to April 2010 via two non-consecutive 24-hour dietary recalls (separated by 2 to 6 weeks). Children aged 7 to 15 were interviewed face to face during home visits in the presence of at least one of the child's parents or carers. Participants aged 16 and over were interviewed by telephone, at dates and times unannounced to the participants.

Portion sizes of foods consumed were quantified in several ways: by means of quantities as shown on photos in a provided picture booklet, or in household measures, standard units, by weight and/or volume. The survey covered all days of the weeks and all four seasons. National and/or religious holidays or holidays of the participants were not included in the survey.

Appendix B MOSH and MOAH concentrations in food groups analysed by Foodwatch (2015; 2016a,b)

Food group	Product	Packaging material*	Concentration (mg/kg)[1,2]	
			MOSH	MOAH
Rice	Long grain rice	Fresh fiber	2.4	0.3
	Pandan rice	Fresh fiber	0.6	<0.2
	Quick-cooking rice	Fresh fiber	0.4	<0.2
	White rice	Fresh fiber	0.9	<0.2
	White rice biological	Fresh fiber	1.0	0.3
	White rice quick-cooking	Fresh fiber	0.9	<0.2
	Wholegrain rice	Fresh fiber	3.0	0.8
Pasta	Dora pasta	Recycled fiber	12.4	0.3
	Lasagne 1	Fresh fiber	3.6	0.6
	Lasagne 2	Recycled fiber	13.4	1.2
	Spaghetti	Recycled fiber	133	0.6
	Spaghetti biologic	Fresh fiber	<0.2	<0.2
	Tagliatella	Recycled fiber	3.6	<0.2
	Tagliolini	Recycled fiber	27.5	5.0
Breakfast cereals	Corn flakes 1	Recycled fiber	0.6	<0.2
	Corn flakes 2	Recycled fiber	0.4	<0.2
	Corn flakes 3	Recycled fiber	1.3	0.4
	Corn flakes 4	Recycled fiber	0.5	0.2
	Corn flakes 5	Recycled fiber	0.4	<0.2
	Corn flakes 6	Fresh fiber	<0.2	<0.2
	Corn flakes bio	Recycled fiber	5.1	1.2
Chocolate sprinkles	Dark 1	Fresh fiber	2.3	<0.5
	Dark 2	Fresh fiber	5.2	<0.5
	Dark 3	Fresh fiber	0.8	<0.5
	Dark 4	Fresh fiber	1.4	<0.5
	Dark bio	Fresh fiber	3.0	0.7
	Milk 1	Fresh fiber	0.8	<0.5
	Milk 2	Fresh fiber	4.5	0.8
Oat meal/muesli	Oat meal	Fresh fiber	0.8	<0.2
Cereal products[3]	Breadcrumbs	Fresh fiber	2.3	<0.2
	Corn starch	Recycled fiber	12.6	1.9
	Couscous	Recycled fiber	3.0	0.7
	Semolina	Fresh fiber	1.6	<0.2
	Whole wheat grain product	Recycled fiber	6.1	1.2
Fruit-flavoured sprinkles	Fruit-flavoured	Fresh fiber	<0.2	<0.2
Cacao powder	Cacao powder	Fresh fiber	9.4	0.7

Food group	Product	Packaging material*	Concentration (mg/kg)[1,2]	
			MOSH	MOAH
Chocolate	Bunny	alu-foil	1.3	<0.5
	Bunny	alu-foil	9.7	0.9
	Bunny	alu-foil	10.6	1.9
	Bunny	alu-foil	0.6	<0.5
	Bunny	alu-foil	8.4	0.5
	Bunny	alu-foil	3.1	<0.5
	Bunny	alu-foil	2.3	<0.5
	Bunny	alu-foil	1.6	<0.5
	Bunny	alu-foil	21.2	2.9
	Bunny	alu-foil	2.2	0.6
	Bunny	alu-foil	2.9	<0.5
	Bunny	alu-foil	1.8	<0.5
	Bunny	alu-foil	2.5	<0.5
	Bunny	alu-foil	2.9	0.6
	Bunny	alu-foil	19.7	2.6
	Bunny	alu-foil	4.7	<0.5
	Bunny	alu-foil	2.4	<0.5
	Bunny	alu-foil	8.2	<0.5
	Bunny	alu-foil	5.5	0.6
	Bunny	alu-foil	1.9	<0.5
	Santa Clause	alu-foil	2.9	<0.5
	Santa Clause	alu-foil	3.4	<0.5
	Santa Clause	alu-foil	12.1	1.0
	Santa Clause	alu-foil	1.7	<0.5
	Santa Clause	alu-foil	4.5	<0.5
	Santa Clause	alu-foil	2.9	<0.5
	Santa Clause	alu-foil	5.8	0.6
	Santa Clause	alu-foil	2.8	<0.5
	Santa Clause	alu-foil	2.7	<0.5
	Santa Clause	alu-foil	1.3	<0.5

* Assumption based on measured MOSH and MOAH content in the paperboard; fresh fibers were assumed if MOSH concentration was <175 mg/kg paper and no or low MOAH contamination

[1] Concentrations were derived from Foodwatch (2015; 2016a,b)

[2] Concentrations shown as "<0.2" or "<0.5" were reported as below the limit of detection

[3] Breadcrumbs, corn starch, couscous, semolina and whole wheat grain product are products with the same consistency; they all consist of scatterable particles that are relatively dry, and low in fat but rich in starch. The level of contamination with mineral oil did not show any correlation with fat content, nor with particle size, and therefore, the variation in the level of contamination between these products was considered coincidental, being more likely due to differences in processing or packaging of the grain, than that it is related to different types of food. The MOSH and MOAH concentrations measured in these products were therefore treated as repeated measurements within the same food group.

Appendix C MOSH and MOAH concentrations in food categories (FoodEx1 level 1) obtained from the EFSA CONTAM Panel (2012b)

Food Category (FoodEx1 level 1)	Concentration (mg/kg)[1]	
	MOSH	MOAH
Animal fat	23	3.5
Bread and rolls	1.8[2]	0.3
Chocolate (cocoa) products[3]	11	1.7
Confectionery (non-chocolate)	46	6.9
Dried fruits	1.2	0.2
Eggs, fresh	3.4	0.5
Fine bakery wares	4.6	0.7
Fish meat	21	3.2
Fish products (canned)	40	6.0
Herbs, spices and condiments	4.6	0.7
Ice and desserts	14	2.1
Legumes, beans dried	1.0	0.2
Livestock meat	1.9	0.3
Oilseeds	38	5.7
Sausages	8.1	1.2
Snack food	1.6	0.2
Sugars	3.6	0.5
Tree nuts	21	3.5
Vegetable oil	43	6.5
Vegetable products[4]	9.6	1.4

[1] Concentrations listed are the average of the lower and upper bound concentrations reported by the EFSA CONTAM Panel (2012b).
[2] The high, worst case, concentration of MOSH in bread and rolls was 532 mg/kg.
[3] Used for chocolate bars and chocolate coated confectionary. Chocolate consumed as such and cacao powder were mapped to foods analysed by Foodwatch (Appendix B).
[4] Concentrations were mapped to vegetables in glass/can or deep-frozen. Due to the absence of concentrations in fruit, this concentration was also mapped to fruits in glass/can.

Appendix D Mean concentrations of MOSH and MOAH of Foodwatch (2015; 2016a,b) used in the exposure assessment after parametric modelling per food group

Food group	Concentration (mg/kg)[1]	
	MOSH	MOAH
Rice	1.4	0.3
Pasta	30.4[2]	1.2
Breakfast cereals	1.2	0.4
Chocolate sprinkles	2.7	0.4
Cereal products[3]	5.4	0.8
Chocolate (bunnies and Santa Clauses)	5.1	0.6

[1] Concentration was only obtained via parametric modelling for those food groups with at least two positive measurements. This was not true for the food groups 'oat meal', 'fruit-flavoured sprinkles' and 'cacao powder'. For these food groups, the concentrations as listed in Appendix B were used in the exposure assessment.

[2] The mean concentration for the food group 'pasta' was 11.2 mg/kg in the exposure assessment using only the Foodwatch data for paperboard packaged foods.

[3] See footnote 3 of Appendix B.

Appendix E Modelling of long-term exposure using OIM

Long-term exposure was estimated using Monte Carlo Risk Assessment Software (MCRA) release 8.2 (de Boer *et al.*, 2016) with the Observed Individual Means (OIM) method. This method is frequently used to assess the exposure to food contaminants (e.g. EFSA, 2015, 2016). Briefly, daily consumption of foods of each individual in the food consumption database was multiplied with the relevant mean MOSH or MOAH concentration of that food. The intake was then summed per consumption day and per individual. To assess the long-term exposure, the exposures per individual were subsequently averaged over the two consumption days resulting in a two-day-average for each individual. Finally, the two-day-average estimated exposure for each individual was divided by its corresponding body weight. The calculation results in an exposure distribution of all two-days-averages from which exposure percentiles were obtained.

Appendix F Description of the bootstrap

There are different sources of uncertainty in dietary exposure assessments. One of these sources is the uncertainty due to the limited size of the datasets used in the exposure assessment. Typically, the smaller the dataset, the more uncertain the data are. This uncertainty can be quantified by using the bootstrap method (Efron 1979; Efron and Tibshirani 1993).

In this study, the bootstrap was used to assess of the food consumption database due to its sample size. For this, a bootstrap database is generated of the same size as the original database for the food consumption by sampling with replacement from the original dataset. This bootstrap database is considered as a database that could have been obtained from the original population if another sample had been randomly drawn. This bootstrap database is then used for the exposure calculations and derivation of the relevant percentiles. Repeating this process many times results in a bootstrap distribution for each percentile that allows for the derivation of confidence intervals around it. The bootstrap approach was used in this report by generating 100 food consumption bootstrap databases and calculating the long-term (with at least 10,000 iterations each) dietary exposure. Of the resulting bootstrap distributions, a 95% uncertainty interval was calculated per percentile by computing the 2.5% and 97.5% points of the empirical distribution.

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