The Director General

Maisons-Alfort, 8 March 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

On 19 March 2015, ANSES received a formal request from the Directorate General for Competition, Consumer Affairs and Fraud Control (DGCCRF) to undertake the following expert appraisal: "request for an opinion on the migration of mineral oil compounds into food from recycled paper and cardboard packaging".

1. BACKGROUND AND PURPOSE OF THE REQUEST

Materials intended to come into contact with food are regulated under European Commission Regulation (EC) No 1935/2004. Article 3 of this Regulation states that all materials and articles intended to come into direct or indirect contact with food shall be manufactured in compliance with good manufacturing practice so that, under normal or foreseeable conditions of use, they do not transfer their constituents to food in quantities which could endanger human health, or bring about an unacceptable change in the composition of the food, or bring about a deterioration in their organoleptic characteristics.

The majority of food contact materials (FCMs), including paper and cardboard, are not currently subject to any specific European regulations. In view of the absence of any European legislation, the French Directorate General for Competition, Consumer Affairs and Fraud Control (DGCCRF) has made data sheets available to industrial companies for each type of material, including the sheet on "organic materials based on plant fibres"[1], as well as the information memo no. 2006-156[2] specific to coated

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[1] http://www.economie.gouv.fr/dgccrf/Papiers-et-cartons

paper and cardboard, stating the criteria to be met in order to certify the suitability of these materials for contact with food. The DGCCRF regularly conducts investigations to verify the inertia of paper and cardboard intended to be placed in contact with food.

The issue of mineral oils in contact with food emerged following work by the Zurich Cantonal Laboratory (Switzerland), which revealed the presence of certain categories of mineral oils in dry food packaged in paper and cardboard packaging (Biedermann *et al.*, 2009). Mineral oils (mineral oil hydrocarbons - MOHs) are complex mixtures derived from crude oil. They are composed of mineral oil saturated hydrocarbons (MOSHs) and mineral oil aromatic hydrocarbons (MOAHs).

In its Opinion of 2012 on mineral oils found in food, EFSA identified the different sources of MOHs in food with, in particular, a significant contribution from recycled papers for dry food (EFSA 2012). EFSA considered exposure to MOSHs to be of concern and exposure to MOAHs as of particular concern. In 2012, EFSA also stressed the need to establish new toxicity reference values for these specific categories of mineral oil hydrocarbons.

In its opinion, EFSA recommended:

- Among the MOSHs, distinguishing between the compounds according to their structure and carbon number.
- Carrying out interlaboratory validation of the analytical methods.
- Determining food categories representative of exposure to MOHs.
- Identifying sources of contamination at various stages of food production.
- Using barrier materials to limit the migration of compounds from the packaging into the food.
- Selecting sources of recycled paper and cardboard.
- Avoiding the use of mineral oil hydrocarbon compounds in the composition of recycled paper and cardboard.
- Continuing investigations into microgranuloma formation in rats exposed to MOSHs.
- Studying the effect of oral exposure to MOSHs on the immune system.
- Conducting toxicological studies on MOSHs that consider their structure and mass rather than their physico-chemical properties (viscosity).
- Developing regulations, primarily at European Union level.

In view of the data available on this issue, ANSES was asked to:

1. Propose a definition for the MOHs (MOSHs and MOAHs) migrating from food contact materials, as well as a way to characterise them (in particular the MOAHs);
2. Rule on the analytical methods mentioned in the documents accompanying the formal request;
3. Rule on the risks associated with the presence of MOHs in food and, if possible, propose toxicity reference values (TRVs) for the MOHs;
4. If it should prove impossible to establish TRVs, ANSES was asked to provide an update on the priority work areas and the existing shortcomings from the point of view of characterising the chemical composition and toxicity of the MOHs.

2. ORGANISATION OF THE WORK

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[2] http://www.economie.gouv.fr/dgccrf/Papiers-et-cartons-enduits

The expert appraisal was carried out in accordance with French Standard NF X 50-110 "Quality in Expert Appraisals – General Requirements of Competence for Expert Appraisals (May 2003)".

The collective expert appraisal was conducted by the autonomous WG on "Assessment of substances and procedures subject to authorisation in human food" (ESPA WG) and adopted by the Expert Committee on "Assessment of physico-chemical risks in food" (CES ERCA). The methodological and scientific aspects of the work were presented to the ESPA WG at the meetings of 22 September 2016, 17 November 2016 and 15 December 2016, and then validated on 19 January 2017 and 15 February 2017 after review and validation of the document by the CES ERCA at the meeting of 11 January 2017. In the framework of this work, a hearing took place with the association Foodwatch on 30 November 2016. A hearing also took place with the association Club MCAS (materials for food contact and health), whose members include industrial companies in the paper and cardboard packaging sector, on 5 January 2017.

ANSES analyses the links of interest declared by the experts prior to their appointment and throughout the work, in order to avoid potential conflicts of interest with regard to the matters dealt with as part of the expert appraisal. The experts' declarations of interests are made public via the ANSES website (www.anses.fr).

3. ANALYSIS AND CONCLUSIONS

3.1. Origin of mineral oil hydrocarbons (MOHs) in food

3.1.1. Origin of the MOHs used in food production

Because of their broad spectrum of applications, mineral oils can be found in foods at different stages of food production, whether intentionally (food additives and pesticides) or not (lubricants, cleaning products, *etc.*). In its report of 2012, EFSA identified the different sources of MOHs in food. Among the sources that can arise from the food production chain, it mentioned:

- Food additives and processing aids regulated by Directive No 95/2/EC[3] and the Ministerial Order of 19 October 2006[4]). Certain mineral oils are authorised for use in the food sector as:
    - mould release agents for cooking industrial bakery products,
    - coating agents for confectionery (excluding chocolate) and certain fruits (papaya, mango, melon and avocado),
    - anti-dust agents, to avoid the formation of dust during the storage of cereal grain,
    - components of cheese rinds.
- Lubricants, cleaning products[5] and engine oils used in factories or for agricultural machinery,
- Pesticides: certain paraffinic mineral oils are authorised within the European Union as miticides and insecticides for treating certain crops (potatoes, fruit trees). Their use is governed by Regulation (EC) No 1107/2009[6]. Certain mineral oils are also authorised as adjuvants in the formulation of plant protection preparations.

3.1.2. Migration of MOHs from food contact materials

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[3] European Parliament and Council Directive No 95/2/EC of 20 February 1995 on food additives other than colours and sweeteners.

[4] Ministerial Order of 19 October 2006 on the use of processing aids in the manufacture of certain foods.

[5] French Order of 8 September 1999 on fraud and falsification with regard to processes and products used for cleaning materials and items intended to come into contact with foodstuffs, products and beverages for human and animal consumption.

The involvement of food packaging as a significant source of migration of MOHs into foods has now been documented (EFSA, 2012).

- Paper and cardboard packaging

The issue of migration of MOHs into food packaged in paper and cardboard packaging was originally revealed through the work of the Zurich Cantonal Laboratory (Kantonales Labor Zürich (KLZH)), made accessible by numerous publications (e.g. Biedermann *et al.*, 2009). In a German study conducted on 119 dry foods (pasta, semolina, rice, biscuits, cereals, *etc.*) packaged in paper and cardboard materials and stored for 2 to 3 months at room temperature, the authors observed that the mineral oils that had migrated into the foods were composed of 10 to 20% aromatic hydrocarbons (MOAHs), with mean MOSH concentrations ranging from 0.5 to 24 mg/kg of food depending on the food (Vollmer *et al.* 2011). These same samples analysed after 4 and then 16 months of storage (corresponding to the best before date, or BBD) at room temperature had higher concentrations, demonstrating that migration continues throughout the storage (Biedermann *et al.* 2013a). Lastly, measurements of residual MOSH and MOAH levels in dry and fatty foods, as well as in the paper and cardboard packaging containing them, were published in 2015 by the association Foodwatch in France, Germany and the Netherlands; this study confirmed the results previously published on this topic[7].

The problem of MOHs does not only concern packaging in direct contact with food, since some studies have shown that MOHs could also migrate from corrugated cardboard used for food transport and storage (Biedermann *et al.* 2011a, Barp *et al.* 2015).

The main sources of mineral oils in paper and cardboard are offset printing inks[8] directly applied to the paper and cardboard food packaging, as well as those found in packaging following the recycling of paper and cardboard (Biedermann and Grob 2010, Biedermann *et al.* 2011b). Newspapers and other printed media entering the recycling chain have been identified as the main sources of mineral oils in recycled paper and cardboard food packaging (Biedermann and Grob 2010, Biedermann *et al.* 2011b). In addition, it appears that paper and cardboard packaging produced from recycled fibres contains higher levels of mineral oils than packaging produced from virgin fibres (Lorenzini *et al.* 2010).

Nevertheless, printing inks are not the only source of MOHs in paper and cardboard food packaging. The hot-melt glues and adhesives used to stick the paper and cardboard boxes used as food containers consist partly of hydrocarbons that can migrate into food (Lommatzsch *et al.* 2016, Pivnenko *et al.* 2016).

- Plastic packaging

Certain mineral oils are authorised for use as additives (use as a lubricant) in plastic food contact materials (see Section 3.2.1). Plastic materials can therefore also constitute a source of migration of MOHs into food (Biedermann-Brem *et al.* 2012).

- Other packaging

Jute and sisal sacks used to carry certain foods such as rice and cocoa have also been identified as a source of contamination of foods by mineral oils (Grob *et al.* 1991, EFSA 2005). In order to soften the fibres used to manufacture these bags, these fibres are treated by immersion in a "batching oil" before being spun. Lastly, the presence of mineral oils has been identified in canned tuna, sardines and anchovies in olive oil as a result of cross-contamination during the manufacture of food cans from the oils used to lubricate equipment (Grob *et al.* 1997).

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[6] Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market.

[7] Results searchable from: http://www.foodwatch.org/fr/s-informer/topics/emballages-toxiques/dernieres-actus/huiles-minerales-dans-nos-aliments-tous-les-resultats/

[8] Offset inks are oil-based inks with high viscosity.

3.2. Regulations

3.2.1. Food contact materials

- Plastic materials

As detailed above, certain mineral oils are authorised for use as additives in plastic food contact materials. Regulation (EU) No 10/2011 on plastic materials and articles intended to come into contact with food authorises the use of the following compounds:

- Refined paraffinic waxes derived from petroleum-based or synthetic hydrocarbon feedstocks[9], of low viscosity (CAS No. 95858):
  - Average molecular mass at least equal to 350 Da.
  - Viscosity at 100°C at least equal to 2.5 cSt (2.5×10^{-6} m²/s).
  - Mineral hydrocarbon content with a carbon number less than 25, lower than 40% (m/m).
  - Specific migration limit of 0.05 mg of substance per kg of food.
  - The regulation prohibits their use for items in contact with fatty foods for which ethanol 50% is the regulatory simulant[10].
- Refined waxes derived from petroleum-based or synthetic hydrocarbon feedstocks, of high viscosity (CAS No. 95859), with the following specifications:
  - Average molecular mass at least equal to 500 Da.
  - Viscosity at 100°C at least equal to 11 cSt (11×10^{-6} m^2/s).
  - Mineral hydrocarbon content with a carbon number less than 25, lower than 5% (m/m).
  - Generic specific migration limit of 60 mg of substance per kg of food.
- White mineral oils, paraffinic, derived from petroleum-based hydrocarbon feedstocks (CAS No. 95883) with the following specifications:
  - Average molecular mass at least equal to 480 Da.
  - Viscosity at 100°C at least equal to 8.5 cSt (8.5×10^{-6} m^2/s).
  - Mineral hydrocarbon content with a carbon number less than 25, lower than 5% (m/m).
  - Generic specific migration limit of 60 mg of substance per kg of food.

- Rubber materials

The French Order of 9 November 1994 on rubber materials and items in contact with foodstuffs, food products and beverages authorises the use of the following compounds:

- Food-grade paraffinic mineral oils (CAS No. 8042-47-5) with the following specifications:
  - Specific migration limit of 0.3 mg/kg for paraffinic oils and 3 mg/kg for hydrogenated oils.
- Food-grade paraffinic waxes, including microcrystalline (CAS No. 08002-74-2), with the following specifications:
  - Specific migration limit of 0.3 mg/kg for paraffinic waxes and 3 mg/kg for hydrogenated waxes.

- Paper and cardboard materials

For paper and cardboard materials intended to come into contact with food, there is no European regulatory limit relative to the authorised levels of mineral oil hydrocarbons. To compensate for the absence of regulations, the DGCCRF has developed information memos[11] intended for industrial companies whose aim is to clarify the rules for verifying and monitoring the suitability of paper and

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[9] Basic products.

[10] The food simulants and the general and specific assignment of simulants to food are defined in Annex III of Regulation (EU) No 10/2011. Ethanol 50% has been assigned to fatty foods.

[11] Information memo No. 2004-64 of 6 May 2004 on food contact materials and information memo No. 2006-156 of 20 July 2006 concerning coated paper and cardboard.

cardboard for contact with food. These memos make reference to the requirements for purity, the limits of acceptability and the analytical methods for different substances or groups of substances.

In addition, the Council of Europe is currently working on the drafting of a technical guide on paper and cardboard, which will propose migration restrictions and limits for various known impurities, mineral oils in particular.

Lastly, the German Federal Institute of Risk Assessment (BfR) has prepared three drafts for the 22nd Order on mineral oils since 2011[12]. The third and most recent draft presented in July 2014 gives the requirements, which have been listed below (Table 1), for materials based on paper and cardboard intended for contact with food and manufactured with recycled paper and cardboard. The materials must not be placed on the market if the maximum levels shown in the table below are not complied with:

Table 1: Maximum levels of mineral oil hydrocarbons proposed in the third draft of the 22nd German Order

Substances	Application criteria	Maximum levels in the materials	Specific migration limit in foods
MOSH	- C16-C25 for contact with dry and non-fatty foods stored at room temperature or lower - C16-C35 for all other types of contact	24 mg/kg paper and cardboard	2 mg/kg for the MOSHs C20-C35
MOAH	- C16-C25 for contact with dry and non-fatty foods stored at room temperature or lower - C16-C35 for all other types of contact	6 mg/kg paper and cardboard	0.5 mg/kg For the MOAHs C16-C35

These limits are not currently in force and the BfR is now working on a fourth version of its 22nd Order.

3.2.2. Food additives and processing aids

As detailed above, certain mineral oils are also authorised as food additives. This is particularly the case with microcrystalline wax (E905), which may be used *quantum satis*[13] for the surface treatment of confectionery (except for chocolate), chewing gum and certain fruits (melon, papaya, mango, avocado)[14]. In 2013, EFSA reassessed this additive and concluded that its use was unlikely to lead to a risk to consumer health (EFSA 2013). Regulation (EU) No 231/2012[15] establishes the technical specifications of the additive E905. This additive is in particular characterised by its viscosity and its benzo[a]pyrene content (see details in Annex 1).

Lastly, in France, certain mineral oils are authorised as mould release agents and anti-dust agents and are included in Annex IA of the Ministerial Order of 19 October 2006[16] (see details in Annex 2).

3.2.3. Pesticides

Certain mineral oils are authorised as insecticides and miticides for treating certain crops (potatoes and fruit trees: citrus fruit, pome fruits and stone fruits). Their use is governed by Regulation (EC) No

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[12] http://www.bmel.de/SharedDocs/Downloads/Service/Rechtsgrundlagen/Entwuerfe/Entwurf22VerordnungBedarfsgegnstaende.pdf?__blob=publicationFile Consulted on 25 November 2016

[13] Quantity strictly necessary to achieve the desired effect.

[14] European Parliament and Council Directive No 95/2/EC of 20 February 1995 on food additives other than colours and sweeteners.

[15] Commission Regulation (EU) No 231/2012 of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council.

[16] Ministerial Order of 19 October 2006 on the use of processing aids in the manufacture of certain foods.

1107/2009. This is the case with four paraffin oils (CAS No. 64742-46-7, CAS No. 72623-86-0, CAS No. 97862-82-3 and CAS No.8042-47-5)[17], which were approved on 1 January 2010 for a duration of 10 years.

3.3. Physico-chemical characterisation of mineral oils

3.3.1. Composition of mineral oils and associated products

The generic term mineral oil hydrocarbons (MOHs) encompasses a broad range of chemical compounds in terms of types, physico-chemical properties and potential toxicity profiles. These toxicological profiles are usually established on the scale of a group of chemical compounds or, when it is possible to isolate them, on the scale of a compound.

The assessment of the health risk for consumers exposed to these compounds as a result of migration from recycled paper and cardboard is linked to the ability to isolate these compounds or classes of compounds and establish a clear dichotomy between some of these derivatives. In particular, the n-alkanes naturally present in certain foods need to be distinguished from those migrating from packaging.

Mineral oils are derived from crude oil, coal, natural gas, or biomass after the application of processes such as distillation, extraction of aromatics, fractionation, hydrogenation and other means of purification. Thus, mineral oils of high, low or medium viscosity, white mineral oils, liquid paraffins, food-grade mineral oils and microcrystalline waxes are all derived from crude oil but have very different compositions depending on the source of the crude oil and the refining processes applied.

Mineral oils are complex mixtures comprising:

- saturated hydrocarbons (MOSHs) composed of paraffins (*i.e.* linear or branched alkanes) and naphthenes (*i.e.* monocyclic or polycyclic alkylated or non-alkylated cycloalkanes),

- aromatic hydrocarbons (MOAHs), monocyclic or polycyclic and predominantly alkylated. A very minor fraction of non-alkylated polycyclic aromatic hydrocarbons (PAHs) may be present.

The MOHs may also include sulphur and nitrate derivatives, present in minor quantities; crude oil is generally treated with desulphurisation/denitrification processes to reduce the presence of these compounds (EFSA 2012).

The term "mineral oil hydrocarbons (MOHs)" excludes the hydrocarbons naturally present in food. The latter are predominantly n-alkanes with an odd number of carbon atoms (with carbon chains comprising between 21 and 35 carbon atoms), which enables them to be distinguished analytically from MOHs (Biedermann and Grob 2012a,b) consisting of n-alkanes with a homogeneous proportion of odd and even carbon numbers. MOHs are also characterised by an odd or even number of carbon atoms and by the presence of a large number of isomers whose chain lengths are mostly between C10 and C50. Thus, EFSA indicates in its opinion that the possible number of mineral oil hydrocarbons can exceed 100,000 for those with less than 20 carbon atoms, and increases exponentially with the carbon number. The limitation at C50 is also a consequence of the analytical techniques used for characterisation. Indeed, the most efficient technique is based on gas chromatography, whose main limitation is the volatilisation ability of the injected compounds (EFSA 2012).

3.3.2. Physico-chemical characteristics of MOHs and associated products

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[17] Commission Implementing Regulation (EU) No 540/2011 of 25 May 2011 implementing Regulation (EC) No 1107/2009 of the European Parliament and of the Council as regards the list of approved active substances.

The composition of the mineral oils depends on the origin of the initial product (crude oil, coal, natural gas, *etc.*) and the refining processes used. Mineral oils derived from these processes are generally characterised by their physical properties (boiling temperature, density, dielectric constant, viscosity, *etc.*).

As an example, several parameters distinguish refined paraffins of medium and low viscosity, authorised by Regulation (EU) No 10/2011[18], from high-viscosity paraffins: viscosity at 100°C, average molecular mass, and a limitation concerning the fraction of low-mass hydrocarbons *i.e.* containing less than 25 carbon atoms. The white mineral oils also listed in this Regulation are qualified on similar properties (see details in Section 3.2.1).

The JECFA monograph (JECFA 2013) supplements these criteria for mineral oils with viscosity between 8.5 and 11 cSt used as additives, with determination of the extractable polycyclic aromatic hydrocarbons (PAHs) using UV spectrometry in reference to a standard solution of naphthalene. For microcrystalline waxes (E905), characterisation criteria that supplement those presented above are available. In particular, a definition of the extractable PAH content is given; it is measured according to a global approach that relies on the UV absorbance of the extract. This approach was subsequently amended by Regulation (EU) No 231/2012, which specifies a limit for benzo[a]pyrene (50 µg/kg of product) excluding any other PAH from the analysis. However, in its opinion of 2013 concerning medium-viscosity oils, EFSA stressed the need to supplement this measurement of benzo[a]pyrene by characterising three other PAHs[19] (EFSA 2008, EFSA 2013).

Nevertheless, all the criteria/specifications used to describe the classes of "mineral oils" as initial products are global approaches providing no information on the chemical structure of the compounds present. Indeed, EFSA (2012) highlighted the weakness of viscosity as a parameter to define the chemical composition of a mineral oil. It has been demonstrated that a mixture of n-alkanes may have identical kinematic viscosity properties to a mixture of polycyclic aromatic compounds[20].

3.3.3. Chemical characteristics of MOHs migrating from paper and cardboard packaging

The mineral oils found in foods can come from multiple sources (natural origin, contamination in the production chain, food contact materials). However, for any given food, it is complicated to distinguish the MOHs derived from paper and cardboard packaging that are the subject of this formal request.

Nevertheless, recent work has helped provide some indications as to the chemical characteristics of the MOHs that come from paper and cardboard packaging. The hydrocarbons found in recycled paper and cardboard food packaging are characterised by a carbon chain between C12 and C50 long. Among these hydrocarbons, it has been shown that those contained in printing inks (one of the main sources of MOHs in paper and cardboard) have a chain length between C13 and C30 (centred on C18-C19) (Lorenzini *et al.* 2010, Barp 2014, Pivnenko *et al.* 2016).

In addition, the combined presence in the foods of MOHs and diisopropylnaphthalene (DIPN), which is a solvent mainly used for carbonless paper, is sometimes observed. Given the fact that this substance has no significant application in the area of food packaging, its presence in food is a sign of MOH contamination from paper and cardboard packaging made from recycled fibres (Zhang *et al.* 2008, Lorenzini *et al.* 2013, Barp *et al.* 2015).

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[18] Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food. Official Journal of the European Union, L12/1 to L 12/89.

[19] In its Opinion of 2008, EFSA considered that the risk assessment should be based on the mass sum of the PAHs for which there are oral carcinogenicity data available, and proposed a selection of four PAHs as markers of exposure and effects of PAHs in food: benzo[a]anthracene (BaA), benzo[a]pyrene (BaP), benzo[b]fluoranthene (BbF) and chrysene (CHR).

[20] American Petroleum Institute, 1961. Selected values of properties of hydrocarbons and related compounds. Research Project 44, Department of Chemistry, Agricultural and Mechanical College of Texas, TX, USA.

Unlike food-grade mineral oils, which are treated in such a way as to be free of MOAHs, processing aids (inks, glues, *etc.*) and recycled paper and cardboard contain MOAHs. Indeed, MOAHs account for between 15 and 35% of the MOHs migrating from recycled paper and cardboard (EFSA 2012, Barp 2014). Using two-dimensional gas chromatography to analyse samples of newspaper and recycled paper and cardboard packaging, Biedermann and Grob demonstrated that the MOAH fraction consisted mainly of compounds with 1 to 3 aromatic rings (Biedermann and Grob 2010).

Insofar as recycled paper and cardboard packaging is not used in contact with liquid food, the problem of mineral oils mainly concerns dry food (EFSA 2012). In this case, migration takes place through a phenomenon of gas-phase transfer. Therefore, only compounds that are sufficiently volatile can contaminate food. The main studies on the subject show that migration is significant for hydrocarbons with a chain length up to C24 (Lorenzini *et al.* 2010). The migration of compounds from packaging into foods has however been observed for longer-chain hydrocarbons (up to C28) (Vollmer *et al.* 2011).

Lastly, because this formal request is only concerned with the migration into food of mineral oil compounds (MOSHs and MOAHs) from recycled paper and cardboard packaging, the ESPA WG limited the scope of its investigation to compounds that can migrate by gas phase, *i.e.* mineral oil hydrocarbons with a chain length less than or equal to 28 carbon atoms. Specifically with regard to MOAHs, the available data suggest that those found in paper and cardboard packaging primarily consist of compounds with 1 to 3 aromatic rings. Thus, the toxicology appraisal presented in Section 3.5 focuses on MOHs with a chain length less than or equal to 28 carbon atoms and MOAHs whose structure contains 1 to 3 aromatic rings.

3.4. Existing analytical methods for characterising mixtures

3.4.1. General principles for measuring hydrocarbons of mineral origin

Mineral oil saturated hydrocarbons (MOSHs) are the main constituents of mineral oils and the easiest fraction to analyse. For this reason, MOSHs are often used to search for the presence of mineral oils in food. However, their analysis is not sufficient to assess the level of contamination. Analysis of MOHs takes place in two steps. An initial high-performance liquid chromatography (HPLC) step is used to isolate a fraction of saturated hydrocarbons and a fraction of aromatic hydrocarbons. Most of the time (Biedermann and Grob 2012a), these two fractions are analysed by gas chromatography coupled with flame ionisation detection (GC/FID).

Even with the best resolution currently available, gas chromatography (one- and two-dimensional) cannot individually separate mineral oil hydrocarbons; they are therefore identified by envelopes ("Humps" or "Unresolved Complex Mixtures" - UCMs).

The main difficulties related to the analysis of mineral oil hydrocarbons are as follows:

- the pre-separation of mineral oil saturated hydrocarbons (MOSHs) and mineral oil aromatic hydrocarbons (MOAHs),
- the elimination of potentially interfering compounds such as waxes (esters of fatty alcohols and fatty acids), terpenes, olefins and the fats found naturally in foods,
- the re-concentration of the extracts to reach a limit of quantification between 0.1 and 1 mg/kg,
- the definition of the start and end of the integration of the chromatographic signal (humps), which can be observed over several minutes.

The lengths of the hydrocarbon chains present in the MOSHs can be characterised by comparison with the retention time of reference n-alkane mixtures. However, for the same carbon number, branched hydrocarbons will be eluted in GC in the conditions used before their linear equivalents (equal to two fewer carbon atoms). For MOAHs, it is more complicated to determine the chain lengths by comparing their retention times in GC with those of the n-alkanes, because of their different chemical structure.

3.4.2. HPLC-GC/FID methods developed by the Zurich Cantonal Laboratory for analysing MOSHs and MOAHs

This method corresponds to the one described in the publication by Biedermann and Grob (2012a). It is cited in the EFSA report (2012) as a reference method for the analysis of MOHs.

- Principle

After extraction (of the packaging or the foodstuff) using a non-polar organic solvent (hexane, for example) or a mixture of polar/non-polar solvents (ethanol/hexane, for example), the organic extracts are purified by HPLC in order to isolate a fraction of saturated hydrocarbons and a fraction of aromatic hydrocarbons that will be analysed directly by GC/FID with online coupling (see chromatograms in Figure 1).

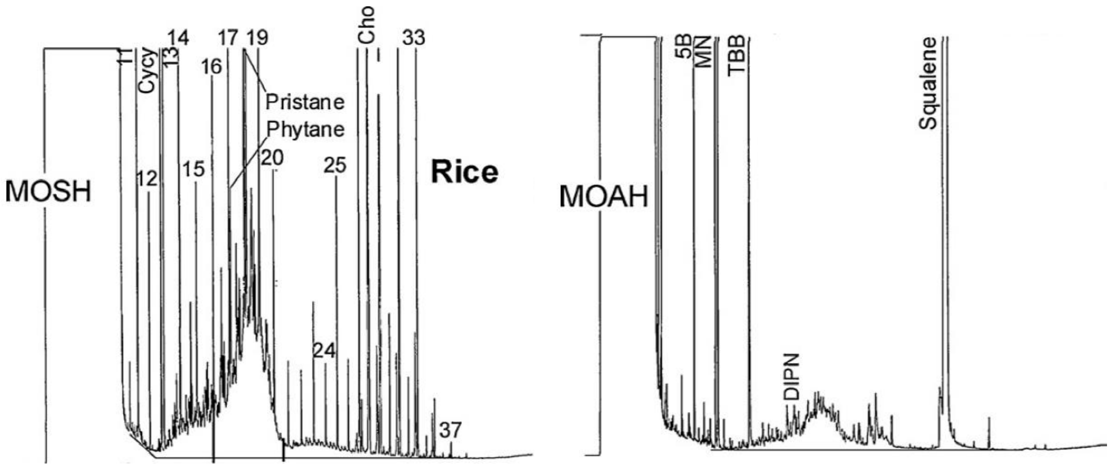

Figure 1: Example of the chromatograms obtained by GC/FID from analysis of a sample of rice packaged in printed paper and cardboard packaging (Biedermann and Grob 2012a).

- Method performance

The limit of quantification (LOQ) of MOSHs or MOAHs in recycled paper and cardboard is of the order of 5 mg/kg. In foods it depends on the fat content of the food, since the amount of fat injected into the HPLC column should not exceed 20 mg.

For foods with a low fat content (rice, semolina, pasta), the LOQ is of the order of 0.1 mg/kg after concentration of the extract before analysis.

For foods containing around 20% fat (muesli, biscuit), the LOQ is 0.5 mg/kg, whereas for fats and oils, the LOQ is around 2.5 mg/kg. These limits of quantification can be lowered by applying a pre-purification step on a column consisting of activated silica and activated alumina (Fiselier and Grob 2009).

The uncertainty is estimated to be less than 20% but it depends on the shape of the GC envelope of the MOHs and the analyst’s experience in integrating this kind of signal.

3.4.3. Method developed by the BfR in collaboration with the Zurich Cantonal Laboratory (BfR 2012)

- Principle

After extraction by a non-polar organic solvent (or a mixture of polar/non-polar solvents), the organic extracts are purified by liquid chromatography, on a silica column treated with silver nitrate, to isolate a fraction of saturated hydrocarbons and a fraction of aromatic hydrocarbons. These fractions are then analysed by a large-volume injection GC/FID system, possibly after re-concentration of the solvent volume.

- Comparison of the results obtained with the HPLC-GC/FID technique

A comparison of this method with the one described in the study by Biedermann and Grob (2012a) was carried out (Fiselier *et al.* 2013). This comparison was conducted on the basis of the analysis of a batch of rice containing around 12 mg/kg of MOSHs and 3 mg/kg of MOAHs, some recycled cardboard, and rapeseed oil containing 50 mg/kg of Gravex 913 mineral oil. The results obtained show that when the methods are well controlled, they lead to equivalent results.

3.4.4. Method used by the laboratory of the Marseille Joint Laboratory Service (SCL)

In December 2013 the laboratory of the Marseille SCL was tasked with conducting work on the subject of mineral oil saturated and aromatic hydrocarbons derived from printing inks in packaging materials.

The method used for this study was that of the BfR (BfR 2012) in which certain steps had been modified: optimisation of the elution volume of the MOSHs, adaptation of the injection volume to the device used. However, a few criticisms were identified by the laboratory of the Marseille SCL. These related to preparation of the silica impregnated with silver nitrate, elution of the MOSH and MOAH fractions, and the phase of evaporation and resolution of the standard peaks. The sensitivity of the analysis was estimated in comparison with the sample supplied in the analysis kit provided by the BfR (around 1000 mg/L of MOSHs and MOAHs).

3.4.5.Techniques used to characterise the mixtures of hydrocarbons of mineral origin

The methods described previously can be used to quantify the two hydrocarbon fractions and estimate the approximate chain lengths present. They cannot, however, identify the species present.

It is nonetheless possible to achieve partial characterisation of chemical structures using more sophisticated analyses involving two-dimensional couplings such as GCxGC, generally consisting of a first non-polar column that separates the hydrocarbons according to their boiling temperature, followed by a second polar column separating the compounds according to their polarity; the reverse is also used (Biedermann and Grob 2015). For the MOSH and MOAH fractions not separated in GCxGC, a pre-fractionation step by HPLC is needed to separately analyse the two saturated and unsaturated fractions. This fractionation may be automated to lead to the HPLC-GCxGC (Adam *et al.* 2007). When a time-of-flight mass spectrometer (Q-TOF) is connected to these couplings, a better characterisation of the compounds may be possible (Ávila *et al.* 2011) through the measurement of their exact mass.

The GCxGC analysis of the MOSH fraction provides a partial separation of the cycloalkanes, the multi-branched paraffins, and the sparsely-branched n-alkanes or alkanes, which makes it possible to differentiate naphthenic oil and paraffinic oil. Analysis of the MOAH fraction leads to the separation of the aromatic compounds according to the number of their rings, for example, the alkyl-benzenes, the alkyl-naphthalenes and the benzothiophenes, as shown in Figure 2 (Biedermann and Grob 2012b).

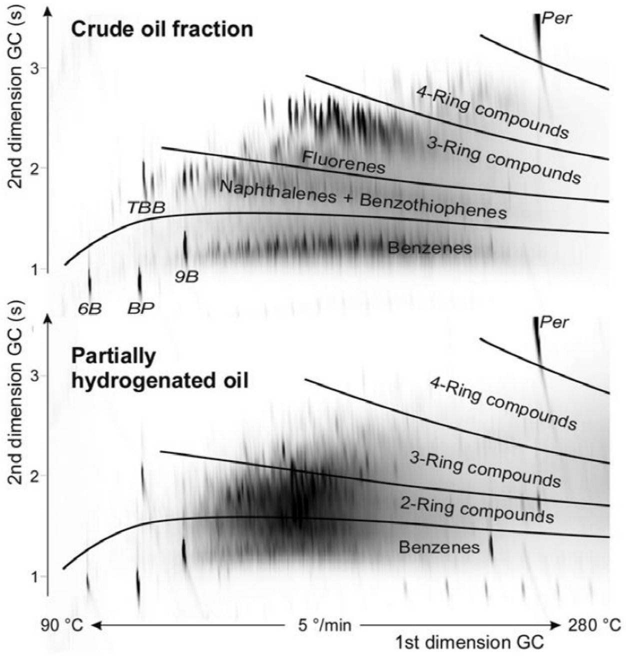

Figure 2: Chromatograms obtained by GC×GC on the MOAH fraction of a crude oil (top figure) and a partially hydrogenated oil (bottom figure) (Fiselier *et al.* 2013) (Biedermann and Grob 2012a).

Thus, the analysis of the aromatic fraction of different crude oils shows a mass predominance of mono-aromatic (45%) and di-aromatic (33%) compounds compared to polycyclic compounds (Weng *et al.* 2015). The fraction of mono-aromatic compounds (UCM-1) can be divided into three classes and 43 sub-classes according to their molecular weight with chemical structures such as alkyl-benzene, mono-aromatic substituted by one cycloalkane, or mono-aromatic substituted by more than one cycloalkane (see Figure 3).

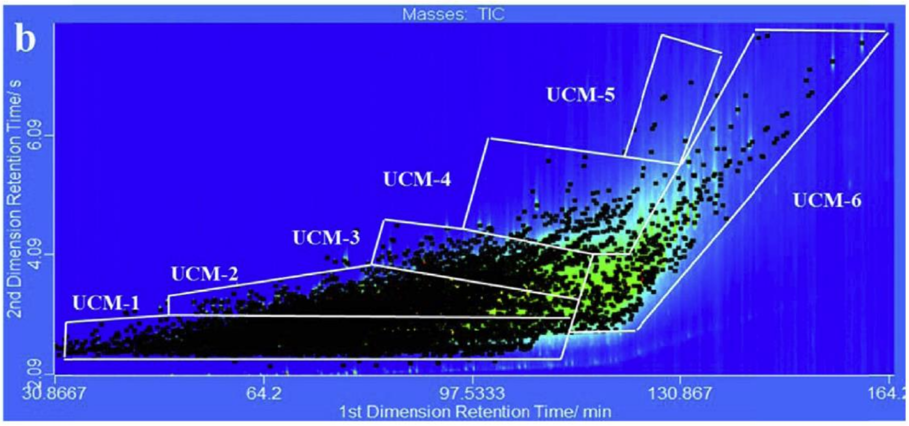

Figure 3: Chromatogram of an aromatic fraction of "Well CI 20-138" oil obtained by GCxGC/TOF-MS (Weng *et al.* 2015)

Lastly, it should be noted that recent publications cite the use of a dual FID and MS/MS detection step after the HPLC/GC fractionation. Here, the mass spectrometry makes it possible to verify the mineral origin of the MOSH and MOAH fraction by examining the chromatographic profile and searching for hopanes, which are petrogenic markers (Zoccali *et al.* 2016).

In conclusion, these techniques help group together into classes the hydrocarbons present in the sample. It is however unrealistic, given the number of compounds present in these complex mixtures, to expect to accurately identify each one.

3.5. Toxicological data concerning MOHs

3.5.1. Toxicological data relating to MOSHs

- General toxicity

Alkanes are metabolised by the cytochrome P450 system, transformed into alcohols and then into the corresponding fatty acids, some of which follow the normal β-oxidation pathway. This reaction is slower when the n-alkanes are branched or cyclic. Because of a slow biotransformation in the liver, in particular for the branched or cyclic alkanes, MOSHs with a carbon number between 16 and 35 can accumulate in different tissues such as fat, the lymph nodes, the spleen and the liver.

A recent study (Barp *et al.* 2017) conducted in female Fischer F344 rats fed for 30, 60, 90 (with or without a 30-day recovery period) or 120 days with a diet containing a mixture of MOSHs (C14 to C50, consisting of low and high viscosity, highly liquid paraffins) seemed to indicate that the accumulation potential of the MOSHs depends more on the chemical structure of these compounds than their number of carbon atoms. Indeed, it appears that naphthenes are the predominant compounds in the liver and the spleen, while the n- and iso-alkanes predominate in adipose tissue. The authors of this study also had reservations as to the transposition of the results from rats to humans because of the observed differences in the concentration and distribution of MOSHs in these samples, and those reported in the same tissues in a previously published study (Barp *et al.* 2014b and Biedermann *et al.* 2015b), which focused on human samples. While the authors also suggest that the critical effect selected for determining the ADI (hepatic microgranuloma) may not be the most relevant, no evidence was provided to support this assumption. The nature of these results does not call into question the relevance of the previous assessments (EFSA 2012).

In rats, the estimated half-life in blood for white oils (P15) is between 23 and 59 hours, but this corresponds to only one case of a readily degradable mineral oil (Cnubben 2010 and Van Stee 2011). The toxicokinetics data show that the n-alkanes and cyclo-alkanes are readily absorbed when the dose administered is low. In rats, the absorption varies from 90% for compounds with low molecular weight (C14-C18) down to about 25% for compounds consisting of 26 to 29 carbon atoms. For an equal number of carbon atoms, absorption is lower for the branched alkanes (EFSA 2012).

Numerous sub-chronic (90 days) and chronic (two years) toxicity studies by the oral route (often by gavage) have been carried out in mice and rats. The mineral oil mixtures used were characterised by varying proportions of MOAHs and linear alkanes (n-undecanes), branched alkanes and cyclic alkanes (decahydronaphthalene). The studies on the mixtures were carried out with products whose compositions varied greatly (as is often the case with these oils) but none can be considered representative of the reality of their use in food for human consumption (EFSA 2012).

In general, these studies never reported toxicity or notable histopathological changes. Besides a few limited haematological changes, and only at high doses of between 500 and 1000 mg/kg bw/day, the effects most often observed for substance mixtures with a carbon number between 10 and 13 were an increase in relative liver weight and nephrotoxicity related to alpha-2-microglobulin. These effects were considered to be adaptive (on the liver) or specific to the animal used for the study (nephrotoxic effect related to the presence of alpha-2-microglobulin in male rats only) and probably not transposable to humans[21]. Histiocytosis in the mesenteric lymph nodes has also been reported, but this was considered to be non-specific. Indeed, it is usually observed in the case of poorly-absorbed compounds with high molecular weight, and never progresses to effects causing the development of a pathological condition in the case of long-term exposure. Moreover, no inflammation or immunotoxic effects have been found in

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[21] The accumulation in the kidney of alpha-2-microglobulin is specific to male rats.

association with the formation of this histiocytosis; this effect was not therefore selected as toxic and/or relevant for assessing these substances in humans (EFSA 2012).

In rats, images of fibrosis and the formation of liver microgranulomas consisting of aggregations of macrophages surrounded by inflammatory cells and occasionally necrotic cells have been reported. These reactions could suggest evolution towards a more serious pathology and were therefore selected to define a no observed adverse effect level (NOAEL) in rats. For this effect, the lowest NOAEL selected was 19 mg of MOSH/kg body weight/day in the case of a study on low and intermediate viscosity oils (EFSA, 2012).

Very few repeated dose oral toxicity studies have been published since the work by EFSA (2012). More recently, in a study by Adenuga *et al.* (2014), Sprague-Dawley rats were exposed for 90 days by gavage to a C10-C13 solvent not containing MOAHs (composed of 42% chlorinated paraffins and 58% naphthenes (cycloalkanes)) at doses of 0, 500, 2500 and 5000 mg/kg body weight/day. Hepatomegaly and centrilobular hypertrophy were observed in all the treated groups, but these effects were considered to be the adaptive consequences of enzyme induction induced by these hydrocarbons. The clinical data showed increases in the rates of alanine aminotransferase (ALT), gamma glutamyltransferase (γGT) and total bilirubin in the medium and high dose groups, suggesting potential hepatobiliary effects with exposure to high doses. Increases in absolute kidney weight were only observed in male rats and were also associated with renal lesions indicative of α-2-µglobulin-mediated nephropathy. Modelling of the reference dose made it possible to estimate a BMDL[22] of 1857 mg/kg body weight/day based on the increase in serum ALT[23] for this C10-C13 solvent not containing MOAHs. This value is consistent with studies on similar hydrocarbon substances, which show a lack of systemic effects at doses of up to 1000 mg/kg body weight/day for the Sprague-Dawley strain of rat.

- Immunotoxicity

The authors of one review sought to determine whether oral exposure to MOSHs could induce immunotoxic effects and in particular signs of autoimmunity (Kimber and Carillos, 2016). These effects could come from a possible adjuvant effect of the MOSHs. An exhaustive review of the available literature led the authors to conclude that:

  - the autoimmune effects reported by the parenteral route appear only after exposure to very high doses (of the order of 0.5 ml administered by intraperitoneal route in animals or after subcutaneous injections in human cosmetic surgery);
  - in these studies, no dose-effect relationship was observed, with the effects appearing only at the highest dose tested;
  - there is no indication that this type of effect appears after oral administration. In most cases it related to inhalation or dermal exposure.

3.5.2. Toxicological data relating to MOAHs

- Toxicokinetics

Although there is little information on the toxicokinetics of MOAHs, the available data indicate that these compounds are readily absorbed and rapidly distributed to all organs. MOAHs are largely metabolised and do not bioaccumulate (EFSA 2012). Many MOAHs with at least three aromatic rings and little or no alkylation, as well as their heterocyclic analogues, can be activated by the cytochrome P450s into chemically-reactive genotoxic carcinogens such as epoxides (EFSA 2012).

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[22] Benchmark dose limit.

[23] The benchmark response (BMR) considered is a two-fold increase in serum ALT compared to the average of the control group.

- Genotoxicity/mutagenesis

Mineral oil products rich in 3 to 7 ring polycyclic aromatic hydrocarbons (PAHs), such as unrefined and acid- or hydro-treated oils, are mutagenic (EFSA 2012). Given the complexity of the mineral oils and the processes used to produce them, the International Agency for Research on Cancer (IARC) (1984) deemed it useful to adopt a classification based on the processes used to obtain them (see classification in Annex 4). In the framework of this work, it was demonstrated that the samples of vacuum distillates (class 1), solvent-refined oils (class 3), hydro-treated oils (class 4), and used hardening and crankcase oils (class 7.2) are mutagenic with regard to *Salmonella* Typhimurium in the presence and absence (Class 7.2 only) of an exogenous source of metabolic activation. In contrast, during Ames testing, samples of a white oil (class 5), a refined hardening oil (class 7.1) and unused crankcase oils (class 7.1) did not prove to be mutagenic with regard to the strain TA98 of *Salmonella* Typhimurium in the presence or absence of an exogenous system of metabolic activation (IARC 1984, 1987).

In addition, naphthenic distillates (crude or treated with sulphuric acid) containing around 12% (m/m) of PAHs exhibited mutagenic activity in the presence of metabolic activation with regard to *Salmonella* Typhimurium (Granella & Clonfero, 1991). Similarly, samples of mineral oil assessed in a test with *Salmonella* Typhimurium in the presence of an exogenous system of metabolic activation were mutagenic, with the results showing a correlation between the amplitude of the mutagenesis and their PAH levels (Brooks *et al.*, 1995). Moreover, a highly refined white oil with a very low aromatic compound content did not produce DNA adducts, whereas this mechanism has been observed with both an oil refined without solvent and an oil extracted by solvent (but not hydrogenated) (Ingram *et al.* 2000). The rate of formation of DNA adducts obtained with an extracted oil is much lower than that obtained with a non-extracted oil. Indeed, during fractionation and examination of the DNA adducts formed by the 2-3 ring and 4-6 ring aromatic fractions, it seems that the main DNA adducts produced by a non-extracted oil may be attributable to 2 or 3 ring aromatic components. The activity of the non-extracted mineral oils seems to be largely due to substituted 3 and 4 ring polycyclic aromatic compounds (Ingram and Philips, 1993). This means that the MOAHs considered in the framework of this formal request (*i.e.* MOAHs containing mainly 1 to 3 rings), potentially have mutagenic activity.

The mutagenic activity of 15 samples of high-viscosity naphthenic mineral oils and 12 samples of used lubricants, as well as their recycled products, was assessed with the Ames test using both the standard technique and Blackburn's modification (Granella, 1995). The polycyclic aromatic hydrocarbon content was also evaluated as a polynuclear aromatic fraction (PAF) and total PAH fraction. Nevertheless, the presence of mutagens was not correlated to the PAF, nor to the total PAH fraction, which emphasises the difficulty of predicting the mutagenicity of mineral oils. It should however be noted that most naphthenic oils and certain recycled oils contain components that inhibit the exogenous metabolism system used in the Ames test, with the risk of potentially generating "false negative" results (Granella, 1995).

In general, the mutagenicity of MOHs mainly seems to be induced by the presence of MOAHs with between 2 and 7 rings and including alkylated and non-alkylated polycyclic aromatic hydrocarbons (PAHs). The MOAHs with a maximum of 3 rings that are concerned in the specific context of this opinion potentially have mutagenic activity. Nevertheless, in some studies, the induction of mutagenesis was not correlated to the polynuclear aromatic fraction, nor to the total PAH fraction, which emphasises the difficulty of predicting the mutagenic nature of mineral oils.

- Subchronic and chronic toxicity (EFSA, 2012)

Numerous subchronic oral studies have been conducted in mice and rats with MOAHs in a mixture (petroleum distillates, extract of jet fuel[24]) or pure (naphthalene, fluorene, pyrene, 1- and 2-

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[24] Mixture used in the field of aviation.

methylnaphthalene, diisopropylnaphthalene, tetrahydronaphthalene, acenaphthene and benzo[b]thiophene).

Concerning the mixtures, haematological effects and an increase in relative liver weight associated with histological changes (atrophy of the thymus, prostate and seminal vesicles, fibrosis and low cellularity in the bone marrow, hepatic fibrosis and necrosis) have been reported, mainly in rats. Mice appear less sensitive. The lowest NOAEL was 20 mg/kg body weight/day in a 90-day study in rats.

For the pure products, toxicity was observed at lower doses. Essentially, comparable effects were reported (increase in liver weight, haematological effects), with the most pronounced effect being observed in a 90-day study in rats exposed orally to benzo[b]thiophene. In particular, the histological examination showed in female rats an epithelial hyperplasia in the kidney, and in males an increase in the number of binucleate hepatocytes at the dose of 0.34 mg/kg body weight/day. A NOAEL of 0.04 mg/kg body weight/day was determined from this study.

- Carcinogenicity

Concerning the carcinogenicity of MOHs in general, most of the studies have been carried out by the dermal route. The only carcinogenicity study conducted by the oral route was carried out with a liquid paraffin, which caused no increase in the incidence of tumours (Shoda *et al.*, 1997). In view of the small number of studies conducted by the oral route, carcinogenicity studies by other routes were analysed to provide an indication of the biological activity. Indeed, while the dermal route of exposure does not immediately seem really relevant in the framework of this opinion, it is of particular interest in the discrimination of genotoxic and non-genotoxic effects and may make it possible to "compare" the carcinogenic activity of mineral oils that have been refined to different degrees.

It appears that an unused engine oil applied to the skin of mice resulted in no tumourigenic response (Saffiotti and Shubik, 1963, Kane *et al*., 1984). In contrast, a used engine oil had a stronger tumourigenic activity than an unused oil; the solvent extraction of the polycyclic aromatic hydrocarbons had virtually eliminated the tumourigenic activity (Grimmer *et al.*, 1982). Similarly, a used cutting oil was shown to be more active than a comparable unused oil (Gilman and Vesselinovitch, 1955; Jepsen *et al.*, 1977). In addition, hydro-treated naphthenic oils, associated with a decrease in the PAH content, did not induce carcinogenesis (McKee *et al*., 1989). Concerning more specifically the aromatic fraction (MOAHs), by-products of solvent refining called aromatic oils also induced a significant increase in the incidence of skin tumours (Gradiski *et al.*, 1983).

Similarly, fractions with a high boiling point from oils having undergone a catalytic cracking step, which are also classified as aromatic oils, induced a greater number of skin tumours in mice, while supplementary fractionation helped establish that the carcinogenic activity is concentrated in the aromatic fraction of the oils. Despite the possible correlation, the aromatic compound content by itself is not currently considered predictive of carcinogenic activity. In addition, it should be noted that the composition varies depending on the batches.

Moreover, promoting actions have also been detected for certain petroleum fractions. Indeed, highly alkylated MOAHs can also act as tumour promoters, without being directly carcinogenic (*i.e.* initiators) (Shubik and Saffiotti, 1955; Saffiotti and Shubik, 1963). For example, some MOAHs, such as naphthalene, are very probably carcinogenic through a non-genotoxic mode of action, involving cytotoxicity and proliferative regeneration.

In addition, the overview proposed by Achten and Andersson (2015) helps visualise the PAHs and their 2- or 3-ring derivatives taken individually, for which toxicological data (carcinogenicity and acute toxicity) are available (see Annex 5). It confirms that some PAHs and alkylated derivatives, keto-, nitro- and amino-PAHs present sufficient evidence of carcinogenic activity. Lastly, unrefined or relatively unrefined oils whose benzo[a]pyrene content is a few mg/kg are classified as Group 1 carcinogens by the IARC, while highly refined oils are part of Group 3, *i.e.* not classifiable as to their carcinogenicity to humans (INRS, 2012)**[25]**.

In conclusion, the only carcinogenicity study using the oral route was carried out on a liquid paraffin, which caused no increase in the incidence of tumours. By the dermal route, various MOAHs and derivatives (vacuum distillate fractions or naphthenic or paraffinic oils, acid-treated naphthenic or paraffinic oils) have induced tumours. On the other hand, highly refined white oils have generally not led to any significant increase in tumour incidence in the species tested by the dermal route. Nevertheless, the PAH content by itself cannot be considered predictive of carcinogenic activity.

- Reprotoxicity

There are very few studies concerning the toxicity of MOAHs for reproduction. Maternal toxicity (significant increase in the number of females without viable offspring at the end of the gestation period and lower litter size), as well as foetal toxicity (decreases in foetal body weight and incomplete ossification of the skull bones) and an increase in costal cartilage development anomalies and the occurrence of cleft palate in pups were observed with a heavy paraffinic distillate aromatic extract (EFSA 2012).

In the framework of a 28-day study conducted in female Sprague-Dawley rats that received tetralin**[26]**, after mating, by daily oral administration from D6 to D19 post-coitum inclusive, no treatment-related maternal mortality or clinical signs were observed, while the average feed consumption and body weight were considerably reduced in the group treated with the maximum dose of 135 mg/kg body weight/day (OECD, 2004). In addition, a 13-week study in rats exposed to tetralin by inhalation revealed no adverse effect on vaginal cytology, sperm or reproductive organs (OECD, 2004). Lastly, in a 13-week study by inhalation of tetralin performed in mice, no effect on vaginal cytology or sperm was noted, but atrophy of the uterus and ovary were found in the absence of systemic toxicity.

Thus, in view of the available data, an effect on reproduction cannot be totally excluded.

- Endocrine-disrupting potential

An endocrine-disrupting potential was investigated in MOH samples of varying composition (Tarnow *et al.*, 2016). The *in vitro* models (E-screen, ER luciferase, transcriptional assay) mainly used cells of human origin. In some cases, these effects may appear at concentrations ranging between 5.4 and 54 mg/L, which are comparable to the concentrations that have been found in foods (1 to 60 mg MOAH/kg of food).

Of the 15 MOHs tested, 10 resulted in oestrogenic type effects. These concerned products containing more than 16% MOAHs. A separation study showed that the endocrine-disrupting activity in a given sample of MOHs was due to the MOAHs and not to the MOSHs. Similarly, only the MOAHs were able to induce transcription of genes CYP 1A1 and 1B1 and to influence phase 1 metabolism through signalling involving the aryl hydrocarbon receptor (AhR). Furthermore, in this study, the number of carbon atoms constituting the MOAHs or MOSHs did not seem to be a discriminating criterion.

Nevertheless, although on the basis of these *in vitro* studies the endocrine-disrupting potential of the MOHs (particularly those rich in MOAHs) cannot be ruled out, additional studies are required to confirm these findings.

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[25] Estimate of the carcinogenic potential of mineral oils. ND 2356-227-12. INRS, 2012.

[26] Tetralin is an aromatic hydrocarbon with formula $C_{10}H_{12}$, whose structure is similar to that of naphthalene, except that one of the two rings is saturated.

3.6. Conclusions of the ESPA WG

3.6.1. Definition of mineral oils

Insofar as this formal request concerns the migration of mineral oil compounds (MOSHs and MOAHs) into food from recycled paper and cardboard packaging, and in view of the information provided by EFSA (2012) relating firstly to the types of food placed in contact with the recycled paper and cardboard, and secondly to the migration studies reported in the literature, the ESPA WG limited the scope of its investigation to compounds that can migrate by gas phase, *i.e.* mineral oil hydrocarbons with a chain length not exceeding 28 carbon atoms. With regard to aromatic compounds specifically, the available data suggest that the MOAHs found in paper and cardboard packaging primarily consist of compounds with 1 to 3 aromatic rings. This toxicological expert appraisal therefore focused on MOAHs consisting of 1 to 3 aromatic rings.

The ESPA WG wishes to point out that the definition proposed above is suited to dry foods, which are the main foods placed in direct contact with paper and cardboard packaging. Nevertheless, other types of foodstuffs than dry foods, in particular fatty foods, may be packaged in direct contact with paper and cardboard packaging (e.g. breaded frozen fish[27], pizza, *etc.*). For fatty foods, migration of MOHs by direct contact seems possible. As migration is not limited to gas-phase migration, this implies that these foods could potentially be contaminated by a wider spectrum of compounds, in particular those with a carbon chain exceeding 28 carbon atoms.

3.6.2. Overview of the existing analytical methods

The proposed methods have been established according to the same general principle: extraction, fractionation by liquid chromatography (HPLC), analysis by GC/FID. These methods make it possible to separate the saturated hydrocarbons from the aromatic hydrocarbons but cannot differentiate MOSHs from POSHs[28] (polyethylene or polypropylene oligomers) or PAOs[29] (poly-alpha-olefins). However, the shape of the envelope from the chromatogram, the peaks above the envelope and the chain lengths of the compounds resulting from the chromatographic analysis may enable a tentative identification of the hydrocarbon types detected in simple cases.

When several sources of contamination are superimposed, the hydrocarbon profile may become difficult to elucidate in the absence of any knowledge of the raw materials used. It is especially difficult to integrate signals on the chromatograms, for which it is necessary to remove the peaks relating to any n-alkanes naturally present in plant-based foods.

The method of the Zurich Cantonal Laboratory enables the last two stages to be automated by using online coupling of HPLC-GC/FID. Separation of the MOSH and MOAH fractions using HPLC (method of the Zurich Laboratory) is more efficient than that obtained by liquid chromatography (method of the BfR). Indeed, the Marseille SCL had to change the elution volumes recommended by the BfR's method for the MOSH fraction in order to maintain an effective separation of the two fractions.

However, these two methods were compared in a publication through the analysis of three samples conducted by two different laboratories (Fiselier *et al.* 2013). Overall, the data showed that the levels estimated by the two methods are comparable when implemented by expert laboratories, although these conclusions are based on a limited number of samples. Nevertheless, in view of the data available, the

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[27] In the sheet on "organic materials based on plant fibres" published by the DGCCRF, it is mentioned that "*frozen foods are considered to be dry foods if the foods are not frozen or defrosted in their packaging; otherwise, they are considered to be moist and fatty foods*": http://www.economie.gouv.fr/dgccrf/materiaux-organiques-a-base-fibres-vegetales

[28] POSHs are oligomers known to migrate from polyethylene and polypropylene packaging.

[29] Compounds used as lubricants, resins and adhesives.

method using the online HPLC-GC/FID coupling developed by the Zurich Cantonal Laboratory seems more robust than the one developed by the BfR.

Lastly, in the case of the method optimised by the laboratory of the Marseille SCL, it would be useful to produce a characterisation and validation dossier for the method, taking into account the changes made to the BfR method concerning the following points: study of the non-specific signal (in blank samples), sensitivity, recovery rates of MOSHs and MOAHs, and precision. The test reports provided by the SCL present relatively high MOAH levels compared to the associated MOSH levels, which is not in agreement with the maximum percentage of MOAHs present in a mineral oil, commonly estimated at around 15 to 35% (EFSA 2012).

3.6.3. Toxicity of MOSHs and MOAHs

- MOSHs

A "compound-by-compound" assessment is not a relevant approach in the case of excessively complex and variable mixtures containing a multitude of substances.

Because of a slow biotransformation, in particular for the branched or cyclic alkanes, MOSHs with a carbon number between 16 and 35 can accumulate in different tissues such as fat, the lymph nodes, the spleen and the liver.

MOSHs are characterised by very low acute toxicity (LD_{50}[30] most often greater than 5000 mg/kg body weight/day). In the many (sub)chronic toxicity studies available, including in long-term studies and even with high doses, the MOSHs (in a mixture or pure) did not show any particularly high toxic potential, nor was any immunotoxic effect shown after oral administration. However, considering the association of exposure to MOSHs with local inflammatory reactions, an effect on the formation of liver microgranulomas was retained, to identify a NOAEL of 19 mg/kg body weight/day. This value is highly protective in nature because, according to the data available on long-duration exposure, the occurrence of microgranulomas in the liver of rats does not evolve towards a cancerous process. In the absence of more precise data and indications of more severe toxicity in terms of its biological significance, and although the available studies were carried out using technical mixtures characterised by their physico-chemical properties (e.g. viscosity) and whose chemical composition was unknown, exposure to the MOSHs can be compared to this NOAEL (so-called margin of exposure approach). However, in the absence of toxicological studies conducted on mixtures representative of the MOSHs to which the consumer is exposed, it is not possible to propose a tolerable daily intake for this fraction.

In its 2012 report, EFSA conducted an assessment of the health risks associated with dietary exposure to MOSHs by applying the margin of exposure (MOE) approach. Selecting a NOAEL of 19 mg/kg body weight/day, MOE values between 59 and 680 were calculated. EFSA thus concluded that dietary exposure to MOSHs was potentially of concern.

- MOAHs

In general, the mutagenicity of the MOHs mainly seems to be induced by the presence of MOAHs containing between 3 and 7 rings and including alkylated and non-alkylated polycyclic aromatic hydrocarbons (PAHs). Nevertheless, in some studies, the induction of mutagenesis was not correlated to the polynuclear aromatic fraction, nor to the total PAH fraction, which emphasises the difficulty of predicting the mutagenicity of mineral oils.

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[30] Dose resulting in the death of 50% of the treated animals.

The only carcinogenesis study using the oral route was carried out with a liquid paraffin, which caused no increase in the incidence of tumours. On the other hand, via the dermal route, various MOAHs and derivatives have induced significant increases in tumours. However, the PAH content by itself cannot be considered predictive of carcinogenic activity.

At the present time, given the very small number of studies devoted to the effects of MOAHs on reproduction and development, and in view of the toxic effects observed in these studies, an effect on reproduction and development cannot be totally excluded.

MOAHs are complex mixtures formed of thousands of chemical compounds which can, to a certain extent, be resolved into very rudimentary fractions but not into single compounds. Nevertheless, EFSA's CONTAM Panel (2012) considered that the toxicological data collected on isolated MOSHs and MOAHs, found in relevant mixtures, were not appropriate for assessing the risks for mixtures of MOHs. In principle, therefore, there is no indicator compound that can characterise a particular mixture of MOAHs.

Due to their mutagenic and carcinogenic nature, no threshold can be proposed for mixtures of MOAHs. Theoretically, a margin of exposure (MOE) type approach should be used to characterise the risks of MOAHs, but there are no dose-response data on carcinogenesis or for the toxicological effects that precede tumour formation for these mixtures, which would enable a reference point to be established on which to base an MOE type calculation. In addition, application of the Threshold of Toxicological Concern (TTC) approach does not seem relevant for complex mixtures consisting of unknown substances (EFSA, 2012b).

In conclusion, given the scarcity of available data on the composition of the MOAH fraction, the potential toxicological effects and the insufficiently characterised human exposure to MOAHs, it is not currently possible to propose toxicity reference values and conduct a risk assessment for this class of MOHs.

3.7. Recommendations and outlook

3.7.1. Validate the existing analytical methods

In view of the sole inter-laboratory test conducted on a limited number of samples, it is important to validate and compare the analytical techniques currently used to analyse recycled paper and cardboard and foods, through the establishment of inter-laboratory tests. These should be carried out on a sufficient number of samples in order to obtain results allowing a robust statistical analysis.

3.7.2. Better characterise the chemical composition of MOHs

In order to better characterise the substances or groups of substances that can migrate into food from paper and cardboard packaging, more selective analytical strategies should be adopted, for example using two-dimensional gas chromatography (GCxGC) in order to characterise in greater detail the composition of the mixtures found in food and packaging. Detectors with higher specificity than the flame ionisation detection (FID) used in the reference methods presented in Section 3.4 should be considered. Multi-dimensional or high-resolution mass spectrometry may also be an appropriate solution.

3.7.3. Obtain additional data on food contamination by MOHs from recycled paper and cardboard packaging

In order to be able to assess the level of consumer exposure to MOHs, the ESPA WG recommends determining the levels of MOSHs and MOAHs present in commonly consumed foods, using the best suited analytical techniques. As most of the current data concern dry food, the ESPA WG stresses that the presence of MOHs in fatty foods that can be packaged in direct contact with paper and cardboard packaging (breaded frozen fish, pizza, *etc.*) should be investigated. In parallel, the WG recommends performing measurements of residual MOH levels in recycled paper and cardboard packaging in order to be able to characterise the potential quantities of MOHs able to migrate into foods.

3.7.4.Concerning the toxicity of MOSHs and MOAHs

The CES ERCA and the ESPA WG believe that better knowledge of the composition of the mixtures is a prerequisite for proposing toxicological recommendations.

In order to establish a TRV for the MOSHs, additional toxicology studies should be carried out on representative mixtures of MOSHs, migrating from paper and cardboard packaging, to which the consumer is exposed. This implies that these representative mixtures have previously been characterised in terms of their composition. Furthermore, in view of the recent study by Barp *et al.* (2017) relating to the bioaccumulation potential of MOSHs in rats, research on the influence of the chemical structure of the MOSHs on their bioaccumulation potential and toxicity should be pursued.

Given the genotoxic and mutagenic nature of some MOAHs, and their possible no-threshold effects, the ESPA WG and the CES ERCA believe that priority should be given to reducing the contamination of food by these compounds.

3.7.5. Limit consumer exposure to MOHs and more particularly to MOAHs

In order to limit consumer exposure to MOHs and in particular to MOAHs, the ESPA WG and the CES ERCA recommend acting initially on the main sources of mineral oils in paper and cardboard packaging, whether primary, secondary or tertiary packaging. In particular, the experts recommend the use of printing inks, glues, additives and processing aids free of MOAHs in the paper and cardboard packaging manufacturing process.

In addition, in view of the heavy contamination of paper and cardboard packaging made of recycled fibres, it is recommended that the levels of MOAHs in recycled fibres be limited. To this end, the WG proposes:

- Examining the feasibility of using printing inks, glues, additives and processing aids free of MOAHs[31] in the printing sector (magazines, newspapers and other graphic papers). Indeed, newspapers and other printed media entering the recycling chain have been identified as the main sources of mineral oils in recycled paper and cardboard food packaging.
- Conducting studies to identify the steps (sorting, paper pulp manufacture, *etc.*) in the recycling process that lead to the introduction of MOHs in recycled paper and cardboard packaging. This will help identify the technological levers that can be used to reduce the contamination of recycled fibres (more efficient sorting, reduction of cross-contamination, improvement of the de-inking process, *etc.*).

Lastly, in parallel, the ESPA WG and the CES ERCA recommend the use of barriers to limit the migration of MOHs from packaging into foods (see details in Annex 6) to concentrations below the limits of detection of the most sensitive validated analytical methods. Barriers should be used whose effectiveness has been recognised and previously tested according to a sufficiently robust methodology, such as the one proposed by Biedermann-Brem and Grob (2014).

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[31] Concentration below the threshold of detection.

4. AGENCY CONCLUSIONS AND RECOMMENDATIONS

The French Agency for Food, Environmental and Occupational Health & Safety endorses the CES ERCA's conclusions.

Dr Roger GENET

KEYWORDS

Matériaux au contact des denrées alimentaires – Papiers / Cartons recyclés – Hydrocarbures d'huile minérale (MOH) – Hydrocarbures saturés d'huile minérale (MOSH) - Hydrocarbures aromatiques d'huile minérale (MOAH)

Food contact materials – Recycled paper and board – Mineral Oil Hydrocarbons (MOH) – Mineral Oil Saturated Hydrocarbons (MOSH) – Mineral Oil Aromatic Hydrocarbons (MOAH)

REFERENCES

Achten, C. and J. T. Andersson (2015). Overview of Polycyclic Aromatic Compounds (PAC). Polycycl Aromat Compd **35**(2-4): 177-186

Adam, F., F. Bertoncini, D. Thiebaut, S. Esnault, D. Espinat and M. C. Hennion (2007). Towards comprehensive hydrocarbons analysis of middle distillates by LC-GCxGC. J Chromatogr Sci 45(10): 643-649.

Adenuga, D. Carrillo, J. C. McKee, R. H. (2014). The sub-chronic oral toxicity of dearomatized hydrocarbon solvents in Sprague-Dawley rats. Regul Toxicol Pharmacol 70(3). 659-672.

Ávila, B., Pereira, Gomes and Azevedo (2011). Chemical characterization of aromatic compounds in extra heavy gas oil by comprehensive two-dimensional gas chromatography coupled to time-of-flight mass spectrometry. Journal of Chromatography A 1218(21): 3208-3216.

Barp, L. (2014). Mineral oil hydrocarbons: development/optimization if analytical methods, investigation of migration from food packaging into semolina and egg pasta and occurrence in human tissues. University of Udine.

Barp, L., Kornauth, C., Wuerger, T., Rudas, M., Biedermann, M., Reiner, A., Concin, N., Grob, K. (2014b). Mineral oil in human tissues, Part I: concentrations and molecular mass distributions. Food and Chemical Toxicology 72 (2014) 312–321.

Barp, L., M. Suman, F. Lambertini and S. Moret (2015). Migration of selected hydrocarbon contaminants into dry semolina and egg pasta packed in direct contact with virgin paperboard and polypropylene film. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 32(9): 1542-1551.

Barp, L., Biedermann, M., Grob, K., Blas-Y-Estrada, F., Nygaard, U.-C., Alexander, J., Cravedi, J.-P (2017). 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 575 (2017) 1263 –1278.

BfR (2012). Determination of hydrocarbons from mineral oil (MOSH & MOAH) or plastics (POSH & PAO) in packaging materials and dry foodstuffs by solid phase extraction and GC-FID. BfR.

Brooks TM, Priston RA, Wright AS, Watson WP (1995). Evaluation of modified bacterial mutagenicity assays for the genotoxicity testing of mineral oils. Mutagenesis, 10: 409–415.

Biedermann M, Fiselier K, Grob K. (2009). Aromatic hydrocarbons of mineral oil origin in foods: method for determining the total concentration and first results. J Agric Food Chem. 2009

Biedermann, M. and K. Grob (2010). Is recycled newspaper suitable for food contact materials? Technical grade mineral oils from printing inks. European Food Research and Technology 230(5): 785-796.

Biedermann, M., J.-E. Ingenhoff, M. Barbanera, D. Garbini and K. Grob (2011a). Migration of Mineral Oil into Noodles from Recycled Fibres in the Paperboard Box and the Corrugated Board Transport Box as well as from Printing Inks: A Case Study. Packaging Technology and Science 24(5): 281-290.

Biedermann, M., Y. Uematsu and K. Grob (2011b). Mineral oil contents in paper and board recycled to paperboard for food packaging. Packaging Technology and Science 24(2): 61-73.

Biedermann-Brem, S., N. Kasprick, T. Simat and K. Grob (2012). Migration of polyolefin oligomeric saturated hydrocarbons (POSH) into food. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 29(3): 449-460.

Biedermann, M. and K. Grob (2012a). "On-line coupled high performance liquid chromatography-gas chromatography for the analysis of contamination by mineral oil. Part 1: method of analysis." J Chromatogr A 1255: 56-75.

Biedermann, M. and K. Grob (2012b). On-line coupled high performance liquid chromatography-gas chromatography for the analysis of contamination by mineral oil. Part 2: migration from paperboard into dry foods: interpretation of chromatograms. J Chromatogr A 1255: 76-99.

Biedermann, M., J.-E. Ingenhoff, G. Dima, M. Zurfluh, S. Biedermann-Brem, L. Richter, T. Simat, A. Harling and K. Grob (2013a). Migration of mineral oil from printed paperboard into dry foods: survey of the German market. Part II: advancement of migration during storage. European Food Research and Technology 236(3): 459-472.

Biedermann, M., J. E. Ingenhoff, M. Zurfluh, L. Richter, T. Simat, A. Harling, W. Altkofer, R. Helling and K. Grob (2013b). Migration of mineral oil, photoinitiators and plasticisers from recycled paperboard into dry foods: a study under controlled conditions. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 30(5): 885-898.

Biedermann-Brem, S. and K. Grob (2014). Barriers Against the Migration from Recycled Paperboard into Food: Measuring Efficiency by Surrogate Components. Packaging Technology and Science 27(9): 713-726.

Biedermann, M. and K. Grob (2015). Comprehensive two-dimensional gas chromatography for characterizing mineral oils in foods and distinguishing them from synthetic hydrocarbons. J Chromatogr A 1375: 146-153.

Biedermann, M., Barp, L., Kornauth, C., Würger, T., Rudas, M., Reiner, A., Concin, N., Grob, K., (2015b) Mineral oil in human tissues, Part II: Characterization of the accumulated hydrocarbons by comprehensive two-dimensional gas chromatography. Science of the Total Environment 506-507 (2015) 644-655.

IARC (1984). Polynuclear aromatic hydrocarbons, Part 2, carbon blacks, mineral oils (lubricant base oils and derived products) and some nitroarenes. IARC Monogr Eval Carcinog Risk Chem Hum, 33: 1–222.

IARC (1987). Overall evaluations of carcinogenicity: an updating of IARC Monographs volumes 1 to 42. IARC Monogr Eval Carcinog Risks Hum Suppl, 7: 1–440.

IARC (2012). Monographs 100F. Mineral oil, untreated or mildly treated

Cnubben NHP, van Stee LLP, 2010. Single dose pharmacokinetic study of P15 white oil in two rat strains. TNO Quality of Life Report V8503 (unpublished study report).

EFSA (2005). Opinion of the Scientific Panel on food additives, flavourings, processing aids and materials in contact with food (AFC) on mineral oils in jute and sisal bags. EFSA Journal 3(1)

EFSA (2008). Polycyclic Aromatic Hydrocarbons in Food - Scientific Opinion of the Panel on Contaminants in the Food Chain. EFSA Journal 6(8): 724

EFSA (2012). Scientific Opinion on Mineral Oil Hydrocarbons in Food. EFSA Panel on Contaminants in the Food Chain (CONTAM). Parma, Italy, EFSA. EFSA Journal 2012;10(6):2704.

EFSA (2012b). Scientific Opinion on Exploring options for providing advice about possible human health risks based on the concept of Threshold of Toxicological Concern (TTC). EFSA Journal 2012;10(7):2750 [103 pp.] doi:10.2903/j.efsa.2012.2750. Available online: www.efsa.europa.eu/efsajournal

EFSA (2013a). Scientific Opinion on the re-evaluation of microcrystalline wax (E 905) as a food additive. EFSA Journal 11(4)

EFSA (2013b). Scientific opinion on the safety assessment of medium viscosity white mineral oils with a kinematic viscosity between 8.5 –11 mm²/s at 100°C for the proposed uses as a food additive. EFSA Panel on Food additives and Nutrient Sources added to Food (ANS). Parma, Italy, EFSA. EFSA Journal 2013;11(1):3073.

Fiselier, K. and K. Grob (2009). Determination of mineral oil paraffins in foods by on-line HPLC–GC–FID: lowered detection limit; contamination of sunflower seeds and oils. European Food Research and Technology 229(4): 679-688.

Fiselier, K., F. Grundbock, K. Schon, O. Kappenstein, K. Pfaff, C. Hutzler, A. Luch and K. Grob (2013). Development of a manual method for the determination of mineral oil in foods and paperboard. J Chromatogr A 1271(1): 192-200.

Gilman JPW & Vesselinovitch SD (1955). Cutting oils and squamous-cell carcinoma. II. An experimental study of the carcinogenicity of two types of cutting oils. Br J Ind Med, 12: 244–248.

Gradiski D, Vinot J, Zissu D *et al.* (1983). The carcinogenic effect of a series of petroleum-derived oils on the skin of mice. Environ Res, 32: 258–268.

Granella M and Clonfero E, 1991. The mutagenic activity and polycyclic aromatic hydrocarbon content of mineral oils. International Archives of Occupational and Environmental Health, 63, 149-153

Granella M, Ballarin C, Nardini B, Marchioro M, Clonfero E. Mutagenicity and contents of polycyclic aromatic hydrocarbons in new high-viscosity naphthenic oils and used and recycled mineral oils. Mutat Res. 1995 Jun;343(2-3):145-50.

Grimmer G, Dettbarn G, Brune H *et al.* (1982). Quantification of the carcinogenic effect of polycyclic aromatic hydrocarbons in used engine oil by topical application onto the skin of mice. Int Arch Occup Environ Health, 50: 95–100.

Grob, K., M. Lanfranchi, J. Egli and A. Artho (1991). Determination of food contamination by mineral oil from jute sacks using coupled LC-GC. J Assoc Off Anal Chem 74(3): 506-512.
Grob, K., M. Huber, U. Boderius and M. Bronz (1997). Mineral oil material in canned foods. Food Addit Contam 14(1): 83-88.

Grob, K. (2014). Update on recycled paperboard and its compliance for food contact. Journal für Verbraucherschutz und Lebensmittelsicherheit 9(3): 213-219.

Guazzotti, V., S. Limbo, L. Piergiovanni, R. Fengler, D. Fiedler and L. Gruber (2015). A study into the potential barrier properties against mineral oils of starch-based coatings on paperboard for food packaging. Food Packaging and Shelf Life 3: 9-18.

Ingram AJ and Phillips JC, 1993. The dermal bioavailability of radiolabelled benzo[a]pyrene from acetone or from oils of differing viscosity, assessed by DNA and protein binding. Journal of Applied Toxicology, 13, 25-32.

Ingram AJ, Phillips JC and Davies S, 2000. DNA adducts produced by oils, oil fractions and polycyclic aromatic hydrocarbons in relation to repair processes and skin carcinogenesis. Journal of Applied Toxicology, 20, 165-174.

INRS, 2012. Estimation du potentiel cancérogène des huiles minérales. ND 2356-227-12.

JECFA (2013). Compendium of food additive specifications. 77th meeting of the Joint FAO/WHO Expert Committee on Food additives. Monograph 14.
Jepsen JR, Stoyanov S, Unger M *et al.* (1977). Cutting fluids and their effects on the skin of mice. Acta pathol microbiol scand, 85: 731–738.

Kane ML, Ladov EN, Holdsworth CE, Weaver NK (1984). Toxicological characteristics of refinery streams used to manufacture lubricating oils. Am J Ind Med, 5: 183–200.

Kimber I, and Carillos J-C (2016). Oral exposure to mineral oils: Is there an association with immune perturbation and autoimmunity? Toxicology 2016 344-346: 19-25.

Lommatzsch, M., L. Richter, S. Biedermann-Brem, M. Biedermann, K. Grob and T. J. Simat (2016). Functional barriers or adsorbent to reduce the migration of mineral oil hydrocarbons from recycled cardboard into dry food. European Food Research and Technology: 1-7.

Lorenzini, R., K. Fiselier, M. Biedermann, M. Barbanera, I. Braschi and K. Grob (2010). Saturated and aromatic mineral oil hydrocarbons from paperboard food packaging: estimation of long-term migration from contents in the paperboard and data on boxes from the market. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 27(12): 1765-1774.

Lorenzini, R., M. Biedermann, K. Grob, D. Garbini, M. Barbanera and I. Braschi (2013). Migration kinetics of mineral oil hydrocarbons from recycled paperboard to dry food: monitoring of two real cases. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 30(4): 760-770.

McKee RH, Daughtrey WC, Freeman JJ *et al.* (1989). The dermal carcinogenic potential of unrefined and hydrotreated lubricating oils. J Appl Toxicol, 9: 265–270.

OECD (Organisation for Economic Co-Operation and Development), 2004. High Production Volume Screening Information Dataset (OECD SIDS) on 1,2,3,4-tetrahydronaphthalene.

Pivnenko, K., M. E. Olsson, R. Gotze, E. Eriksson and T. F. Astrup (2016). Quantification of chemical contaminants in the paper and board fractions of municipal solid waste. Waste Manag 51: 43-54.

Saffiotti U & Shubik P (1963). Studies on promoting action in skin carcinogenesis. National Cancer Institute Monograph, 10: 489–507

Shoda T, Toyoda K, Uneyama C, Takada K and Takahashi M, 1997. Lack of carcinogenicity of medium-viscosity liquid paraffin given in the diet to F344 rats. Food and Chemical Toxicology, 35, 1181-1190.

Shubik P & Saffiotti U (1955). The carcinogenic and promoting action of low boiling catalytically cracked oils. Acta Unio Int Contra Cancrum, 11: 707–711

Tarnow P, Hutzler C, Grabiger S, Schön K, Tralau T et Luch A. (2016). Estrogenic Activity of Mineral Oil Aromatic Hydrocarbons Used in Printing Inks. PLoS One 11(1): 1-15.

Van Stee LLP, 2011. Validation of an analytical method for the quantitative determination of P15 white oil in rat and human whole blood. TNO Triskelion report V8477 (unpublished study report).

Vollmer, A., M. Biedermann, F. Grundböck, J.-E. Ingenhoff, S. Biedermann-Brem, W. Altkofer and K. Grob (2011). Migration of mineral oil from printed paperboard into dry foods: survey of the German market. European Food Research and Technology 232(1): 175-182.

Weng, N., S. Wan, H. Wang, S. Zhang, G. Zhu, J. Liu, D. Cai and Y. Yang (2015). Insight into unresolved complex mixtures of aromatic hydrocarbons in heavy oil via two-dimensional gas chromatography coupled with time-of-flight mass spectrometry analysis. Journal of Chromatography A 1398: 94-107.

Zoccali, M., Barp, L., Beccaria, M., Sciarrone, D., Purcaro, G., Mondello, L. (2016). Improvement of mineral oil saturated and aromatic determination in edible oil by liquid-liquid-gas chromatography with dual detection. J. Sep. Sci., 39: 623-631.

ANNEXES

Annex 1: Presentation of participants

PREAMBLE: The expert members of the Expert Committees and Working Groups or designated rapporteurs are all appointed in a personal capacity, *intuitu personae*, and do not represent their parent organisation.

WORKING GROUP AND EXPERT COMMITTEE

The work that is the subject of this opinion was monitored and adopted by the following WG and CES:

WG on "Assessment of substances and processes subject to authorisation" (2015-2018)

Chairman

Mr Claude ATGIE – Professor at Bordeaux 1 University – Food toxicology.

Members

Mr Fabien BOLLE – Department Head at the Scientific Institute of Public Health (Belgium) – expertise in chemical risks related to food contact materials.

Mr Jalloul BOUAJILA – Lecturer at Paul Sabatier University (Toulouse) – expertise in pharmacology, analytical methodology.

Mr Nicolas CABATON – Researcher at INRA (Toulouse) – expertise in toxicology, food contact materials.

Ms Marie-Christine CHAGNON – Professor at AgroSup (Dijon) – expertise in toxicology, food contact materials.

Mr Dany CHEVALIER – Lecturer at Lille 2 University – expertise in toxicology, xenobiotics, contaminants.

Ms Véronique COMA – Lecturer at Bordeaux University – expertise in biochemistry, biological products.

Mr Luc FILLAUDEAU – Researcher at INSA (Toulouse) – expertise in manufacturing process engineering.

Mr Angel GIL-IZQUIERDO – Researcher for the National Research Council (Spain) – expertise in metabolism, plant extracts.

Ms Florence LACOSTE – Head of the analysis & expert appraisal department at ITERG (Pessac) – expertise in chemistry, analytical methods.

Mr Claude LAMBRE – Retired – expertise in toxicology, immunotoxicology.

Mr Michel LAURENTIE – Research Director, Department Head at ANSES in Fougères – expertise in toxicokinetics, toxicodynamics, statistics.

Mr Jean-Michel MAIXENT – Professor at Poitiers University – expertise in pharmacological trials, toxicology.

Ms Anne PLATEL – Lecturer at Institut Pasteur in Lille – expertise in toxicology, genotoxicity, QSAR.

Mr Philippe SAILLARD – CTCPA expert (Bourg-En-Bresse) – expertise in food packaging, toxicology, impact of processes, food contact materials.

Mr Patrick SAUVEGRAIN – Engineer at the LNE (Trappes) – expertise in methodological trials, chemistry, food contact materials.

Mr François ZUBER – Scientific Director at the CTCPA (Avignon) – expertise in industrial processes, industrial systems.

CES on "Assessment of physical-chemical risks in food" (2015-2018)

Chairman

Mr Cyril FEIDT – University Professor – expertise in the transfer of contaminants

Members

Mr Claude ATGIE – University Professor – expertise in toxicology

Mr Pierre-Marie BADOT – University Professor – expertise in the transfer of contaminants

Mr Jacques BELEGAUD – Honorary Professor – expertise in toxicology

Ms Valérie CAMEL – University Professor – expertise in analytical chemistry

Ms Martine CLAUW – University Professor – expertise in toxicology

Mr Guillaume DUFLOS – Laboratory Manager – expertise in analytical chemistry

Ms Camille DUMAT – University Professor – expertise in analytical chemistry

Mr Jérôme GAY-QUEHEILLARD – University Lecturer – expertise in digestive impacts and metabolism

Mr Thierry GUERIN – Research Director – expertise in analytical chemistry

Ms Nicole HAGEN-PICARD – University Professor – expertise in toxicology

Ms Laila LAKHAL – Engineer, Project Coordinator – expertise in toxicology

Mr Claude LAMBRE – Retired – expertise in toxicology

Mr Bruno LE BIZEC – University Professor – expertise in analytical chemistry

Ms Raphaële LE GARREC – University Lecturer – expertise in toxicology

Mr Eric MARCHIONI – University Professor – expertise in analytical chemistry

Mr César MATTEI – University Lecturer – expertise in toxicology

Ms Sakina MHAOUTY-KODJA – Research Director – expertise in toxicology

Mr Fabrice NESSLANY – Laboratory Director – expertise in toxicology

Mr Alain-Claude ROUDOT – University Professor – expertise in mathematical modelling

Ms Karine TACK – Laboratory Manager – expertise in analytical chemistry

Ms Paule VASSEUR – Professor Emeritus – expertise in toxicology

Mr Eric VERDON – Laboratory Manager – expertise in analytical chemistry

Mr Jean-Paul VERNOUX – Professor Emeritus – expertise in toxicology

RAPPORTEURS

Ms Florence LACOSTE– Department Manager – expertise in chemistry, analytical methods

Mr Claude LAMBRE – Retired – expertise in toxicology, immunotoxicology

Mr Bruno LE BIZEC – University Professor – expertise in analytical chemistry

Mr Fabrice NESSLANY – Laboratory Director – expertise in toxicology

Mr Philippe SAILLARD – Expert – expertise in food packaging, toxicology, impact of processes, food contact materials.

ANSES PARTICIPATION

Scientific coordination

Mr Gilles RIVIÈRE – Deputy Head of the Food Risk Assessment Unit – ANSES

Mr Sébastien GORECKI – Scientific Project Leader – ANSES

Mr Bruno TESTE – Scientific Project Leader – ANSES

Administrative and secretarial assistance

Ms Angélique LAURENT – ANSES

Annex 2: Technical specifications of the additive E 905 (extract from Regulation (EU) No 231/2012)

E 905 MICROCRYSTALLINE WAX

Synonyms	Petroleum wax; Hydrocarbon wax; Fischer-Tropsch wax; Synthetic wax; Synthetic paraffin
Definition	Refined mixtures of solid, saturated hydrocarbons, obtained from petroleum or synthetic feedstocks
Description	White to amber, odourless wax
Identification	
Solubility	Insoluble in water, very slightly soluble in ethanol
Refractive index	$[n]_D^{100}$ 1,434-1,448 Alternative $[n]_D^{120}$ 1,426-1,440
Purity	
Molecular weight	Average not less than 500
Viscosity	Not less than 1.1×10^{-5} m^2s^{-1} at 100 °C Alternative: Not less than 0.8×10^{-5} m^2s^{-1} at 120 °C, if solid at 100 °C
Residue on ignition	Not more than 0.1 %
Carbon number at 5 % distillation point	Not more than 5 % of molecules with carbon number less than 25
Colour	Passes test
Sulphur	Not more than 0.4 wt %
Arsenic	Not more than 3 mg/kg
Lead	Not more than 3 mg/kg
Polycyclic aromatic compounds	Benzo(a)pyrene no more than 50 µg/kg

Annex 3: Authorised uses of mineral oils as processing aids in France (extract from the Ministerial Order of 19 October 2006)

Processing aid	Category of the processing aid	Foodstuff	Conditions of use / Functions	Maximum residual dose
Paraffin oil	Mould release agent	Spaghetti Pasta for soups obtained by recycling broken strands.	At the dose strictly necessary	20 mg/kg
Paraffin oil	Mould release agent	Fine bakery products Cheeses, with the exception of those benefiting from a designation of origin (AOC).	At the dose strictly necessary	Technically unavoidable dose
Mineral oils of high molecular weight	Mould release agent	Tuile biscuits.	At the dose strictly necessary to achieve the desired effect. The mineral oil is used in a mixture with beeswax.	Residual content lower than 2 g/kg for the mineral oil. Technically unavoidable dose for the beeswax.

Annex 4: Classification of mineral oils according to their refining process (IARC, 1984)

Class 1. Vacuum distillate oils. These may have undergone subsequent finishing stages, such as caustic neutralisation, dewaxing, clay treatment and/or mild hydrotreatment. They have not been acid treated or solvent extracted.

Class 2. Acid-treated oils. These may have undergone subsequent finishing stages, such as caustic neutralisation, dewaxing, clay treatment and/or mild hydrotreatment. They have not been solvent extracted.

Class 3. Solvent-refined oils. These may have undergone subsequent finishing stages, such as dewaxing, clay treatment and/or mild hydrotreatment.

Class 4. Hydro-treated oils.

Class 5. White oils and petrolatums suitable for food and/or medical use (generally produced by severe refining of naphthenic or paraffinic distillates to eliminate the unsaturated compounds).

Class 6. Aromatic oils:
  - 6.1 Solvent extracts
  - 6.2 Catalytically cracked oils

Class 7. Miscellaneous materials:
  - 7.1 Formulated products
  - 7.2 Used oils

Class 8. Petroleum-derived materials not otherwise classified (not sufficiently described to permit assignment to other classes).

Annex 5: PAHs (polycyclic aromatic hydrocarbons), PACs (polycyclic aromatic compounds) and their 2- or 3-ring derivatives for which toxicological data are available (*from Achten et al., 2015*)

	Name	CAS number	No. of aromatic rings	Chemical formula	Evidence of carcinogenicity
O-PACs	Benzofuran	271-89-6	2	C8H6O	+
	Dibenzofuran	132-64-9	3	C12H8O	+
	Xanthene	92-83-1	3	C13H10O	+
	Xanthenone	90-47-1	3	C13H8O2	
N-PACs	Inol	120-72-9	2	C8H7N	+
	Quinoline	91-22-5	2	C9H7N	+
	2-Methylquinoline	91-63-4	2	C10H9N	-
	Methylquinoline	612-58-8	2	C10H9N	I
	4-Methylquinoline	491-35-0	2	C10H9N	I
	6-Methylquinoline	91-62-3	2	C10H9N	I
	7-Methylquinoline	612-60-2	2	C10H9N	I
	8-Methylquinoline	611-32-5	2	C10H9N	I
	Isoquinoline	119-65-3	2	C9H7N	I
	3-Methylisoquinoline	1125-80-0	2	C10H9N	-
	6-Methylisoquinoline	42398-73-2	2	C10H9N	I
	7-Methylisoquinoline	54004-38-5	2	C10H9N	I
	8-Methylisoquinoline	62882-00-2	2	C10H9N	I
	2,4-Dimethylquinoline	1198-37-4	2	C11H11N	-
	2,6-Dimethylquinoline	877-43-0	2	C11H11N	-
	2,7-Dimethylquinoline	93-37-8	2	C11H11N	-
	Benzo[f]quinoline	85-02-9	3	C13H9N	I
	Benzo[h]quinoline	230-27-3	3	C13H9N	I
	Phenanthridine	229-87-8	3	C13H9N	I
	Carbazole	86-74-8	3	C12H9N	+
	Acridine	260-94-6	3	C13H9N	I
	Acridone	578-95-0	3	C13H9NO	I
	Phenanthridone	1015-89-0	3	C13H9NO	I
	8-Hydroxyquinoline	148-24-3	2	C9H7NO	I
Nitro-PACs	1-Nitronaphthalene	86-57-7	2	C10H7NO2	
	2-Nitronaphthalene	581-89-5	2	C10H7NO2	I
	5-Nitroacenaphthene	602-87-9	3	C12H9NO2	I
	2-Nitrofluorene	607-57-8	3	C13H9NO2	I
Amino-PACs	1-Aminonaphthalene	134-32-7	2	C10H9N	I
	2-Aminonaphthalene	91-59-8	2	C10H9N	I
S-PACs	Benzothiophene	95-15-8	2	C8H6S	-
	2,2'-Naphthylbenzothiophene	17164-77-1	2	C18H12S	I
	Dibenzothiophene	132-65-0	3	C12H8S	-
	1-Methyldibenzothiophene	7372-88-5	3	C13H10S	-
	2-Methyldibenzothiophene	20928-02-3	3	C13H10S	-
	3-Methyldibenzothiophene	16587-52-3	3	C13H10S	-
	4-Methyldibenzothiophene	7372-88-5	3	C13H10S	-
	4,6-Dimethyldibenzothiophene	1207-12-1	3	C14H12S	-

Sufficient and limited evidence (+); inadequate evidence for evaluation (0); at least one carcinogenic/mutagenic/cell toxic/EROD proof available (I)

Annex 6: Summary of the literature relating to barrier materials

Many studies have focused on the effectiveness of various barriers. First of all, it appears that polyethylene bags do not constitute an effective barrier to the migration of MOHs and that internal polypropylene bags limit the migration of MOHs but for a period limited to a few months (Lorenzini *et al.* 2013, Barp *et al.* 2015). Bags made from bi-oriented polypropylene (BOPP) and polypropylene coated with an acrylate layer have however demonstrated high efficiency at limiting the migration of MOHs into dry food during long-duration storage at room temperature (Biedermann *et al.* 2013b). These same studies indicate that bags consisting of a double layer of polyethylene (PE) and poly(terephthalate ethylene) (PET) appear to be a hermetic barrier, because no migration of mineral oil compounds from paper and cardboard packaging was observed above the threshold of detection. Nevertheless, these studies should be repeated on a large number of foods in order to confirm the total absence of migration related to the presence of these barriers. In addition, the application of various coatings acting as barriers (PET, acrylate, polyamide, *etc.*) directly to the paper and cardboard packaging is also a solution proposed to limit the migration of contaminants (Grob 2014, Lommatzsch *et al.* 2016). Other barriers, in particular starch-based ones, are currently being studied (Guazzotti *et al.* 2015). Lastly, the integration of activated carbon as an adsorbent in paper and cardboard packaging has also been considered (Lommatzsch *et al.* 2016).