Institut Kirchhoff Berlin

MERIEUX NutriSciences

Determination of MOH in edible oils and fats

Institut Kirchhoff Berlin

A strong and high-end scientific know-how

Mineral Oil, Beyond Analytics

MERIEUX NutriSciences

MOSH/MOAH: European-wide recognized reference laboratory

- 2010 we optimized the first commercially available system for MOSH/MOAH analysis and made it routine (up-to-date >200 systems sold worldwide)
- Organisation of two international conferences in Berlin with 120 participants each
- IKB-Method standardized in CEN committee DIN EN 16995:2017
- Training of the European Reference Labs for Food Contact Materials
- Development of best practice method with big confectionary company
- Counselling of Chinese Health Authority on MOSH/MOAH
- 2019: global Infant formula producer recommends IKB for this analysis to all suppliers, and orders training of employees in Berlin

EURL
European Union Reference Laboratory
for Food Contact Materials

MRI Max Rubner-Institut

- Working group consisting of MRI, government and private laboratories in cooparation of a ring trail provider
- Aim: standardise epoxidation, optimisation sensivity (lower LOQ) for mineral oil analysis in fats and oils
- Method provides comparable results to the international standard DIN EN 16995:2017
- Contains additional and partially modified processing steps, specifications for uniform processing of defined product groups
- **Actual**: 3rd round analysis of spiking samples

DGF-Einheitsmethoden	Abteilung C – Fette
	C-VI 22 (19)

Mineralölbestandteile, gesättigte Kohlenwasserstoffe (MOSH) und aromatische Kohlenwasserstoffe (MOAH) mit online gekoppelter LC-GC-FID

Erweiterte Methode für niedrige Bestimmungsgrenzen

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Mineral Oil Fractions - MOH

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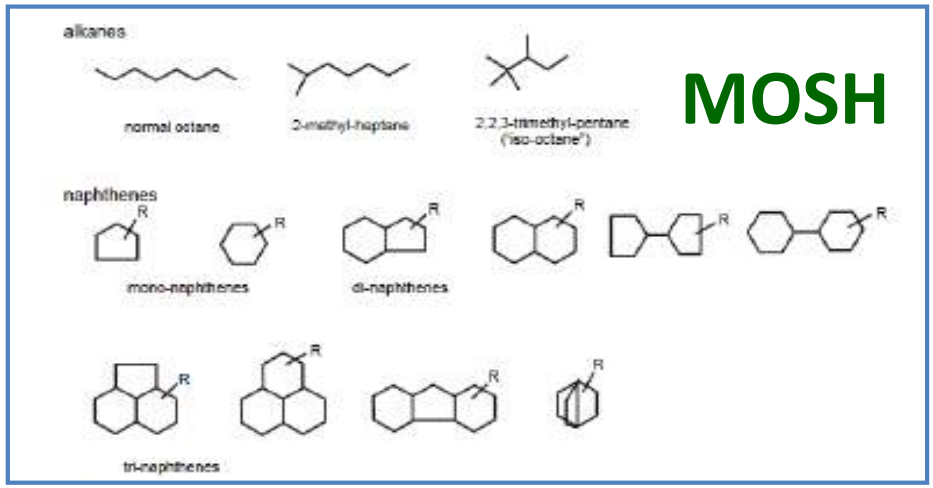
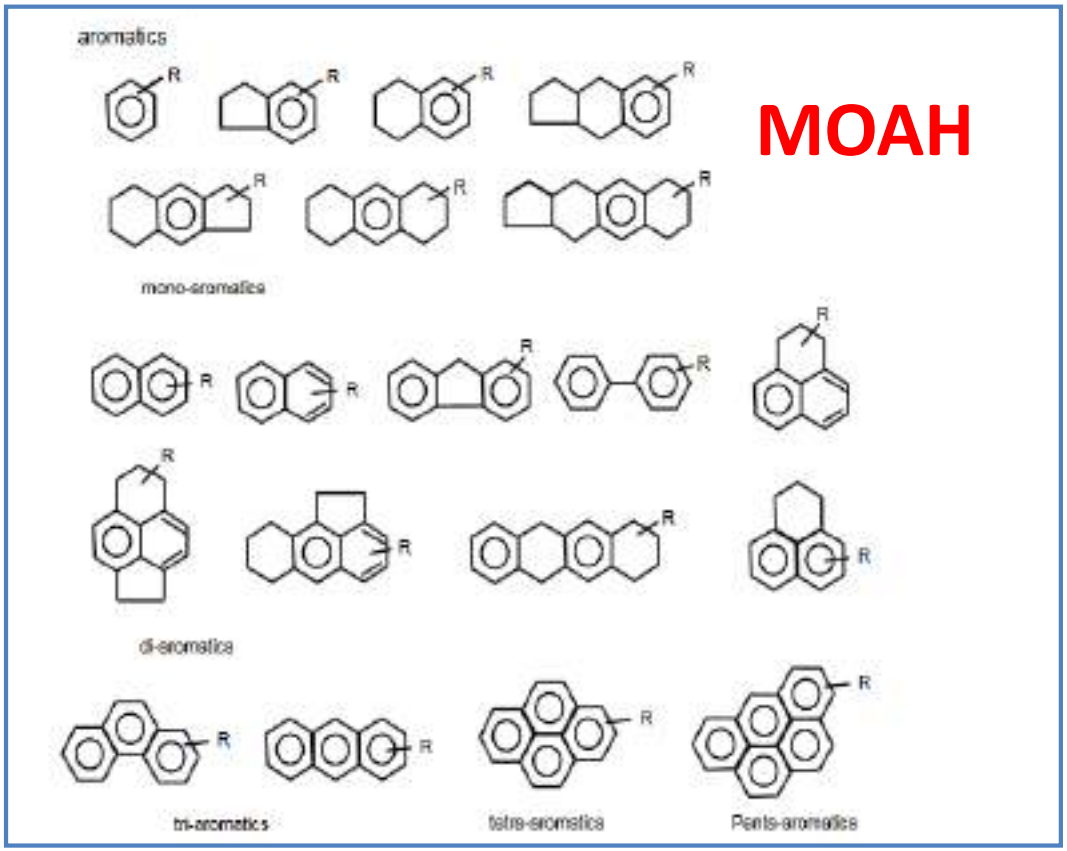


MOSH

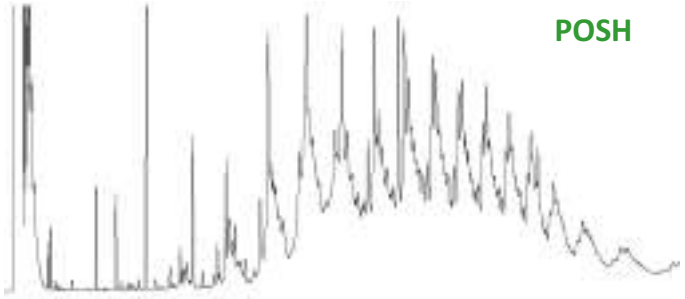


MOAH

MOSH Analogues

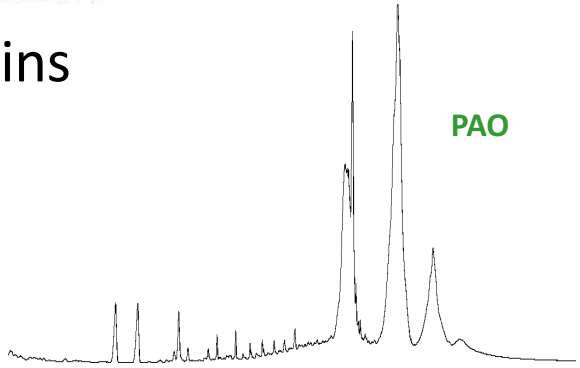
- **POH**

oligomers from polyolefin (eg. PE, PP)



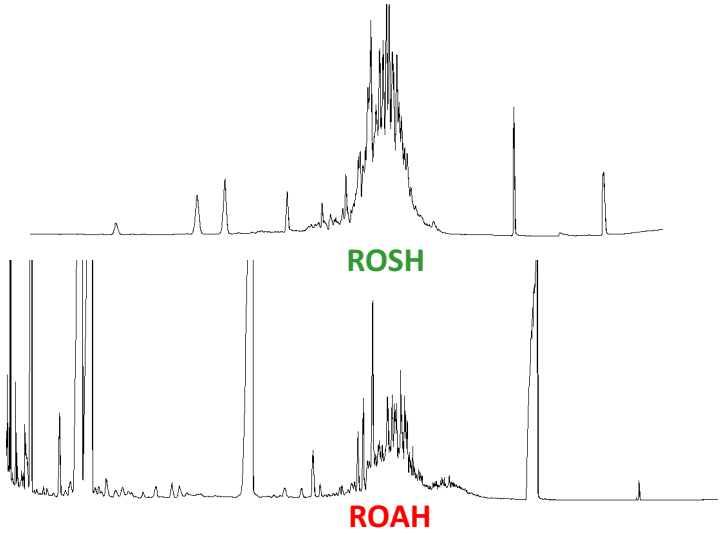
- **PAO**

iso-paraffins with short main- and long sidechains



- **ROH**

resin oligomers from hot melt adhesives



Recommendation (EU) 2017/84 from16.01.2017

17.1.2017 EN Official Journal of the European Union L 12/95

RECOMMENDATIONS

COMMISSION RECOMMENDATION (EU) 2017/84

of 16 January 2017

on the monitoring of mineral oil hydrocarbons in food and in materials and articles intended to come into contact with food

(Text with EEA relevance)

THE EUROPEAN COMMISSION,

Having regard to the Treaty on the Functioning of the European Union, and in particular Article 292 thereof,

Whereas:

(1) Mineral oil hydrocarbons (MOH) are chemical compounds derived mainly from crude oil, but also produced synthetically from coal, natural gas and biomass. MOH can be present in food through environmental contamination, lubricants for machinery used during harvesting and food production, processing aids, food additives and food contact materials. Food grade MOH products are treated in a way that the mineral oil aromatic hydrocarbons (MOAH) content is minimised.

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RECOMMENDATIONS

COMMISSION RECOMMENDATION (EU) 2017/84

of 16 January 2017

Growing relevance of MOH within Europe, USA, Canada and China !

LC-GC-FID method is being globally established.

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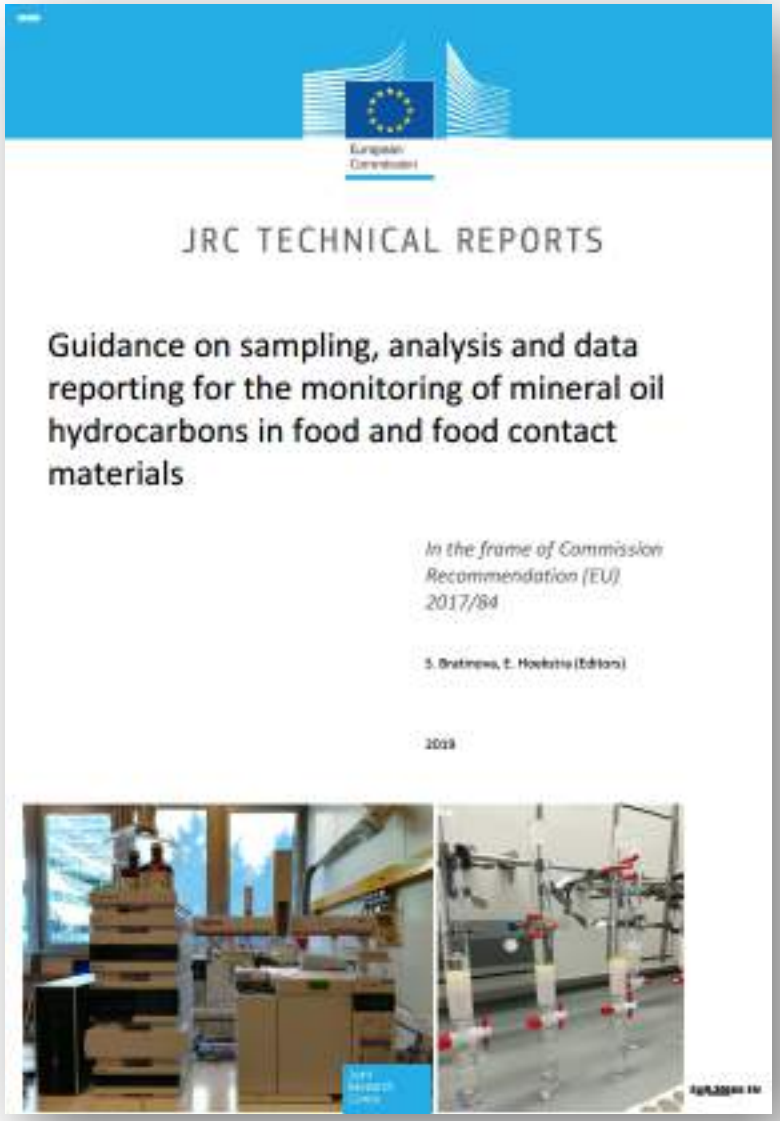
Agreed benchmark levels
(compared to the Belgian Approach and exposure assessment of the NL)

Food category	Benchmark level MOSH	MOSH [mg/kg] C16-C35 Belgium (action threshold)	MOSH [mg/kg] used for exposure ass. NL	Benchmark level MOAH	MOAH [mg/kg] used for exposure ass. NL
Vegetable oils, plant oils (tropical oils excluded)	13	100 (animal and vegetable oils and fats)	43	< LOQ (2)	6,5
Bread, rolls, biscuit, pastry, grains and grain based products,oats, pasta and noodles, rice, breakfast cereals	6	15 (grains and grain based products)	1,8 (bread and rolls) 4,6 (pastry) 1,6 (snacks) 30,4 (pasta, noodles) 1,4 (rice) 1,2 (breakfast cereals) 5,4 (grain based prod.)	< LOQ (0,5 / 1)	0,3 (bread and rolls) 0,7 (pastry) 0,2 (snacks) 1,2 (pasta, noodles) 0,3 (rice) 0,4 (breakfast cereals) 0,8 (grain based prod.)
Confectionary, chocolate	9	20-30 (desserts, sugar, confectionary)	11 (chocolate) 46 (confectionary) 3,6 (sugar) 2,7 (chocolate bunnies) 5,1 (chocolate clauses)	< LOQ (0,5 / 1)	1,7 (chocolate) 6,9 (confectionary) 0,5 (sugar) 0,4 (chocolate bunnies) 0,6 (chocolate clauses)

Source: R. Helling, „Update on benchmark levels for MOSH/MOAH“, Akademie Fresenius Konferenz 06/2019, Düsseldorf

JCR Guideline



- specific guideline for sampling and analysis
- performance requirements for analysis given
- ensure reliable data for both fractions
- reporting

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JCR Guideline

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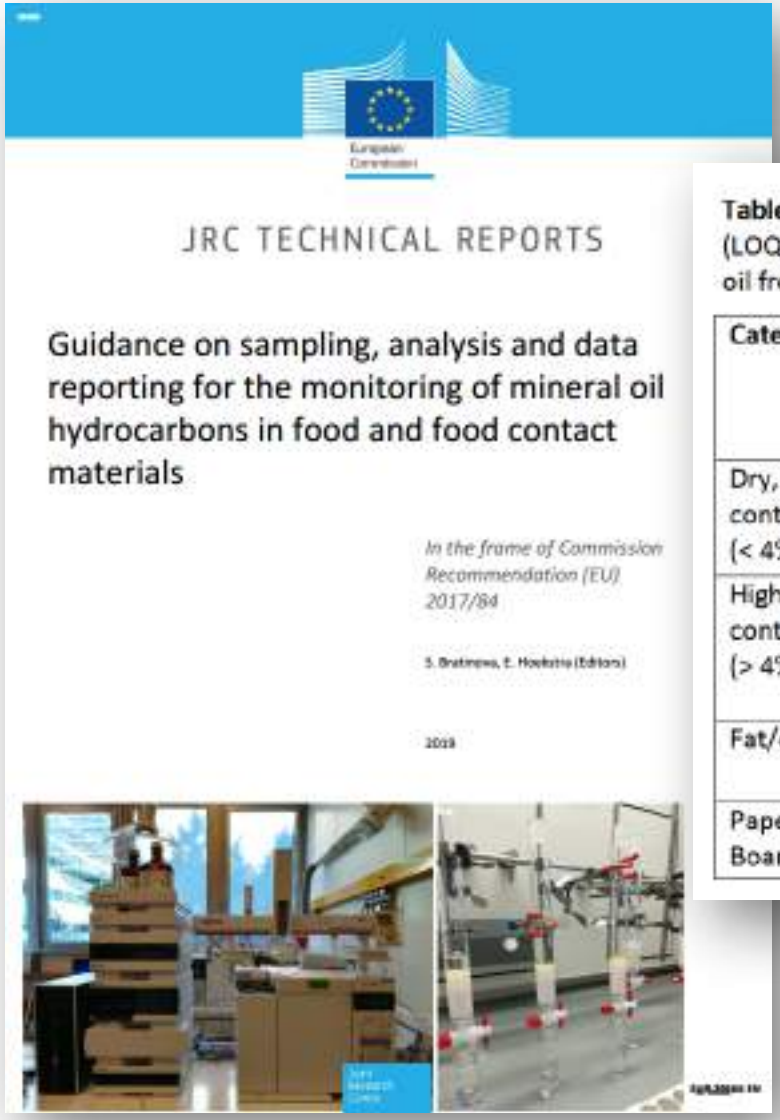
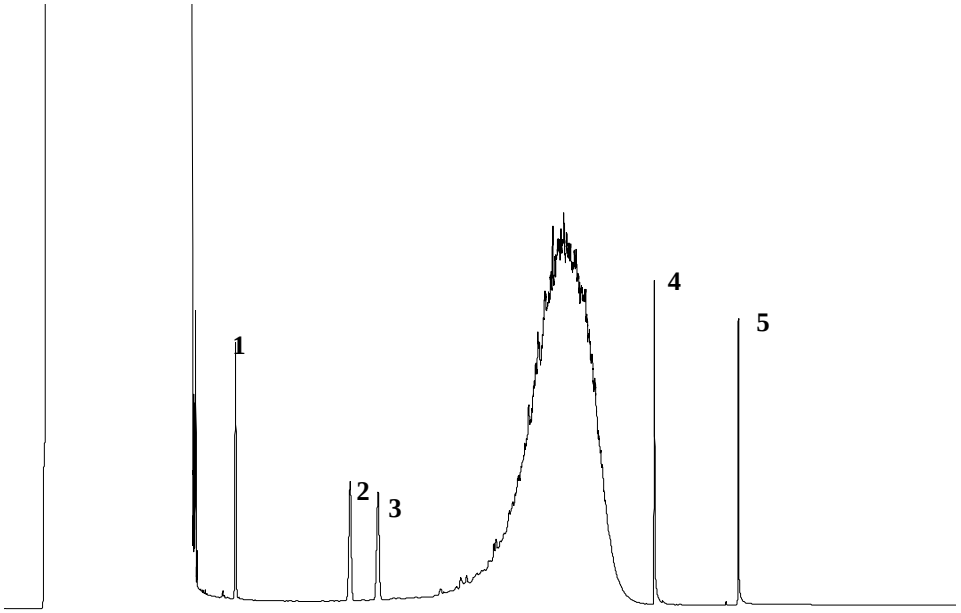


Table II Performance requirements for MOSH and MOAH analysis: maximum LOQ for each C-fraction (LOQ-max), target LOQ for each C-fraction (LOQ-t), acceptable ranges for recovery (R_{rec}) of mineral oil from samples, and intermediate precision

Categories	Associated foods [‡]	LOQ - max [mg/kg]	LOQ -t [mg/kg]	R_{rec} [%]	interme-diate precision [%]
Dry, low-fat content (< 4% fat/oil)	bread and rolls; breakfast cereals; grains for human consumption; pasta, products derived from cereals	0.5	0.1	80 - 110	15
Higher fat/oil content (> 4% fat/oil)	fine bakery ware; confectionery (incl. chocolate) and cocoa; fish meat, fish products (canned fish); oilseeds; pulses; sausages; tree nuts	1	0.2	70 - 120	20
Fat/oils	animal fat (e.g. butter); vegetable oils	2	0.5	70 - 120	20
Paper and Board	Reporting only up to C_{35} (extraction optimised up to C_{35})	10	5	80 - 110	10

Method Principle

- Extraction of sample (adapted to relevant sample matrix)
- Auxiliary methods (clean up, enrichment)
- Isolation of MOSH and MOAH (on-line HPLC on silica gel)
- Separation and quantification by Online HPLC-GC-FID (large volume on-column injection)



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IKB Intermediate Precision, LOQ

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Categories	LOQ	Intermediate precision
low fat (<4%, e.g. rice)	0.2 mg/kg	MOSH 9 % MOAH 12 %
medium fat content (~20%, e.g. chocolate, infant formula)	0.5 mg/kg	MOSH 7 % MOAH 10 %
high fat content (~40%, e.g. vegetable oil)	1 mg/kg	MOSH 7 % MOAH 7 %
paper board	MOSH 0.5 mg/kg MOAH 2 mg/kg	MOSH 11 % MOAH 12 %

Analytical Toolbox

Sample preparation

Extraction with additional ethanol
Wet foods

Saponification
Removal of lipids

Enrichment
Lower detection limit

Selective epoxidation
Removal of olefins by changing their polarity

Activated aluminumoxide
Removal of long-chain n-alkanes

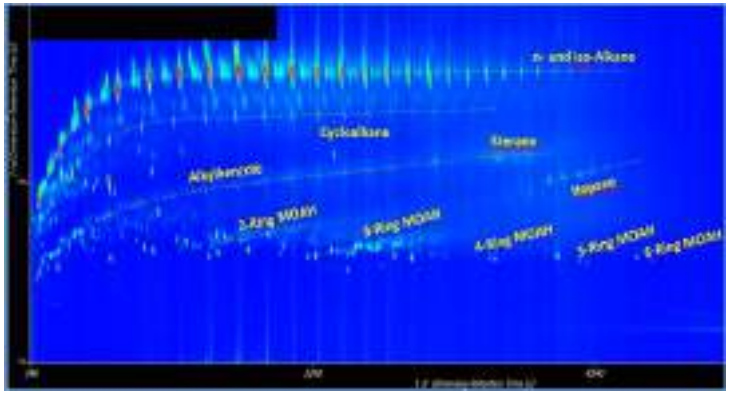
choice depending on

-> matrix
-> interfering substances
-> analytical goal
-> LOQ/LOD

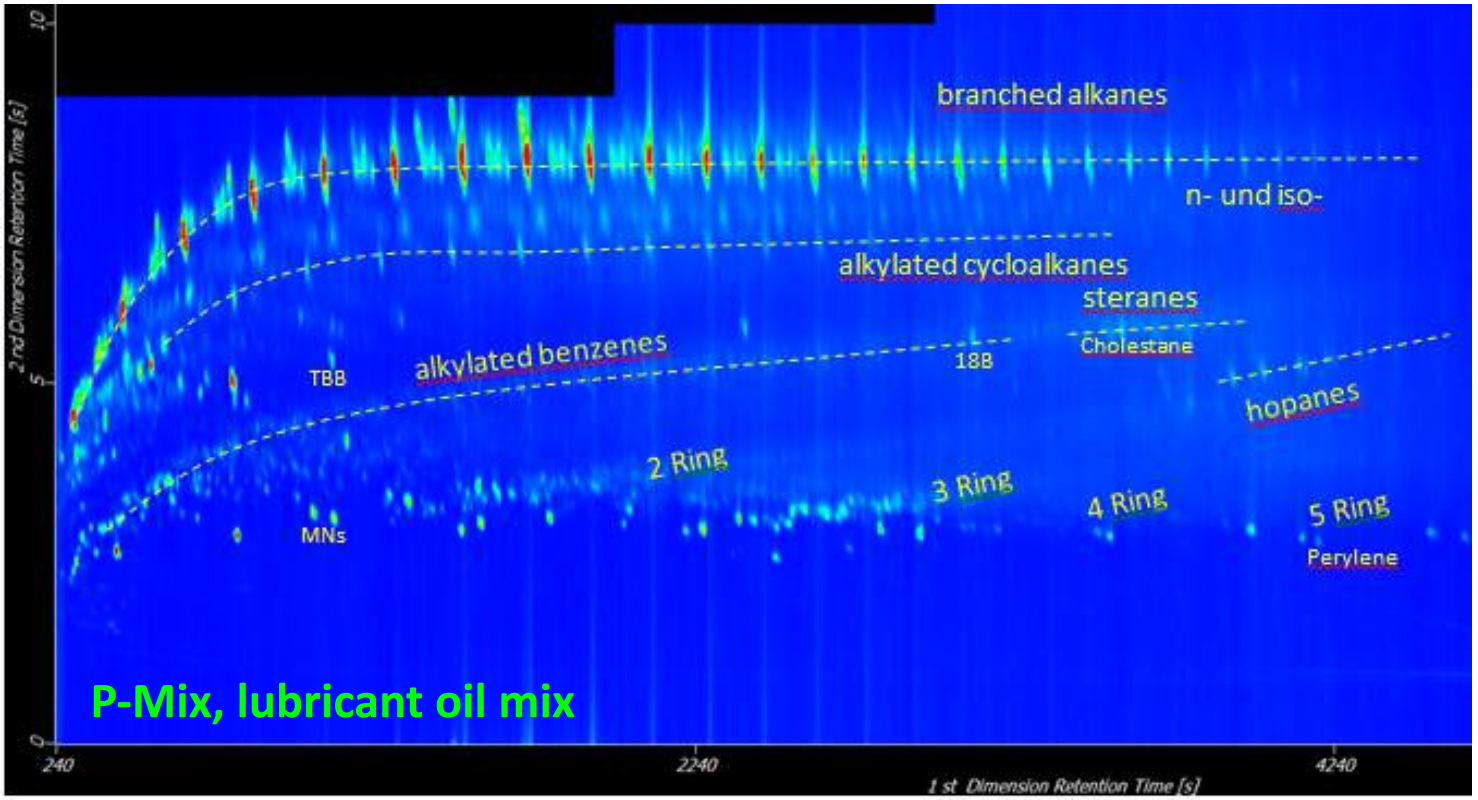
Chromatography

LC-GC-FID
Quantification, established routine method

GCxGC-TOF-MS
verification, characterization, marker for mineral oil origin

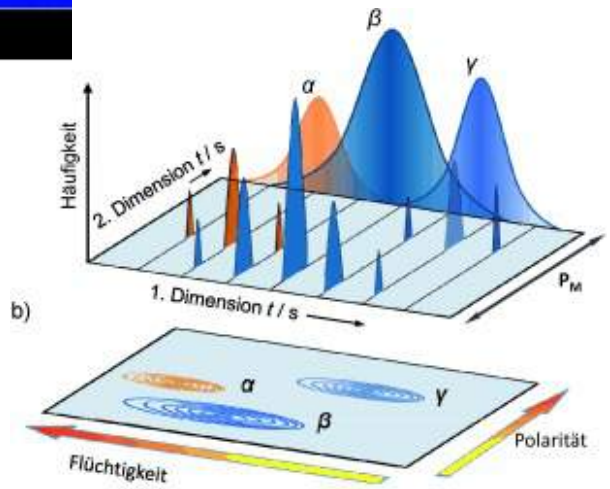


Characterization of MOSH and MOAH with GCxGC-TOF-MS



GCxGC-TOF-MS

- ✓ 2D GCxGC
- ✓ mass selective detector
- ✓ qualitative result
- ✓ characterisation according to substance classes
- ✓ but! separation in single compounds not completely feasible



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Contamination Sources

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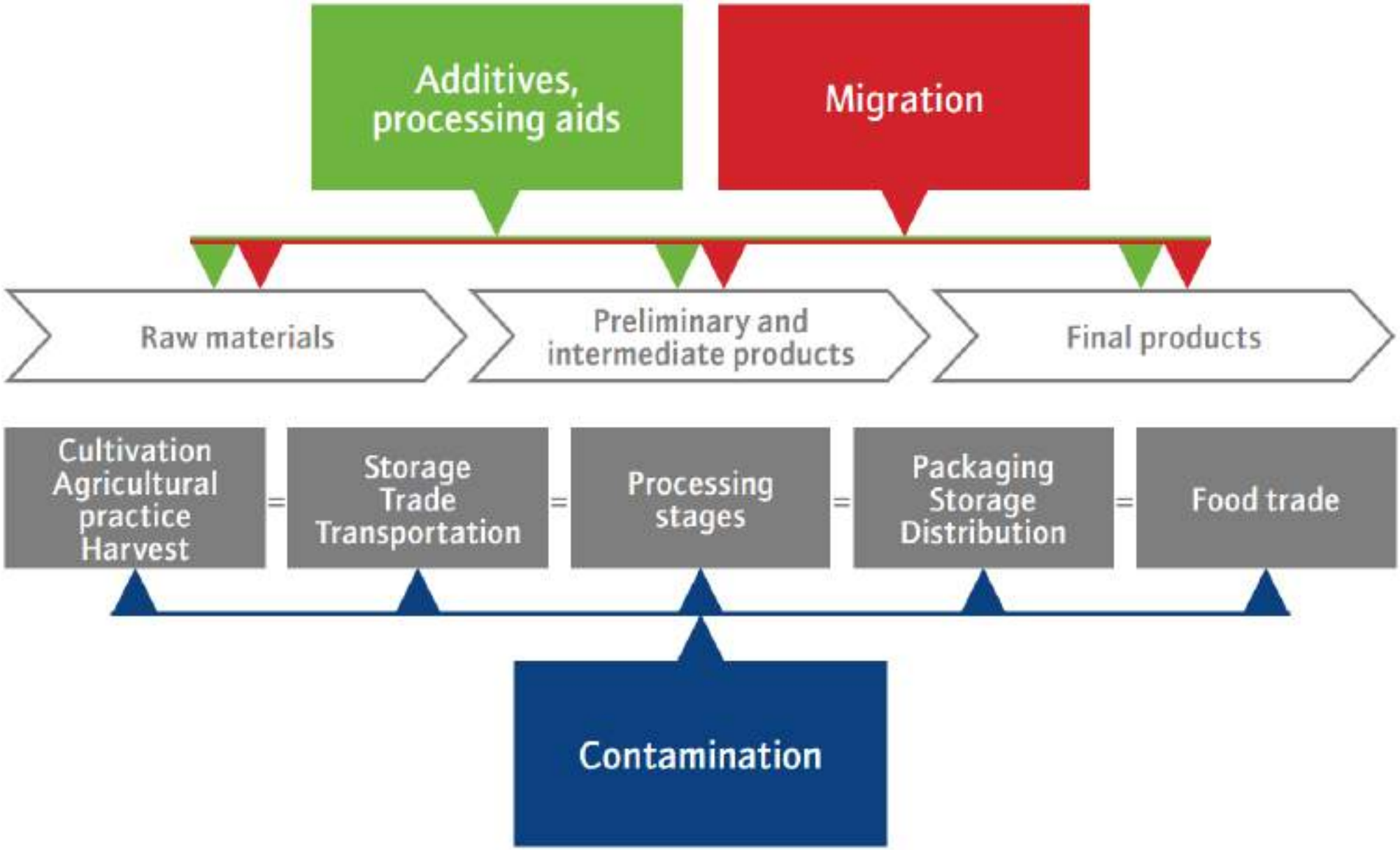
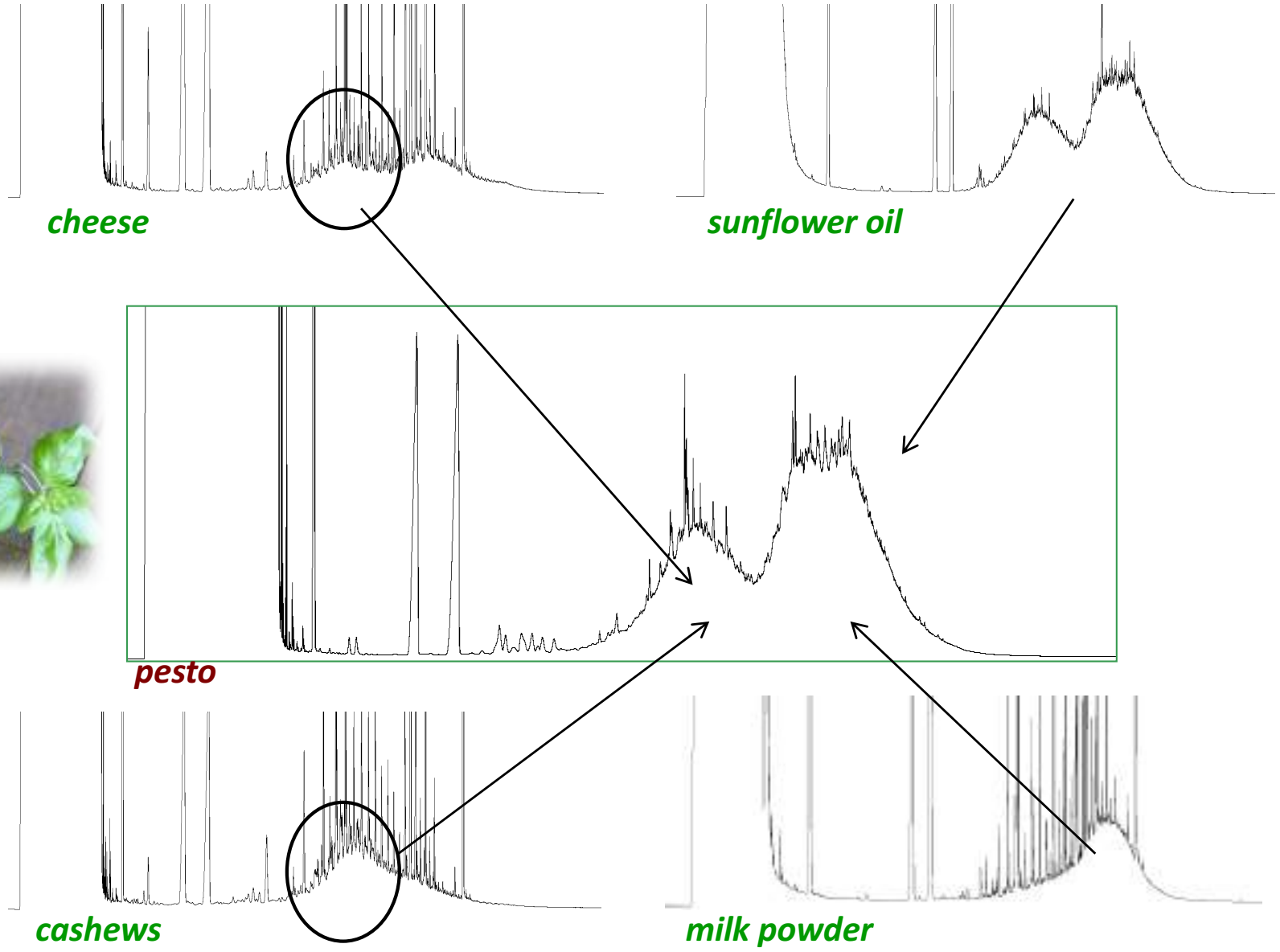
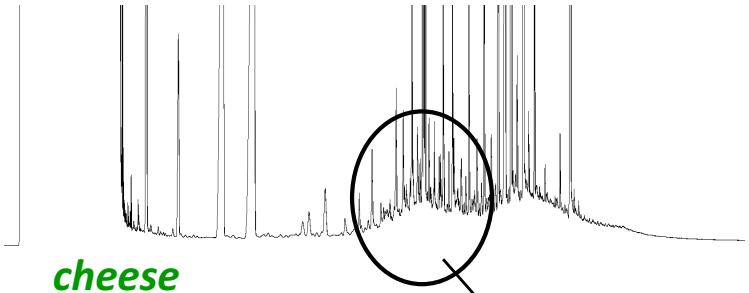


source: BLL, „Toolbox for Preventing the Transfer of Undesired Mineral Oil Hydrocarbons into Food“, 2017

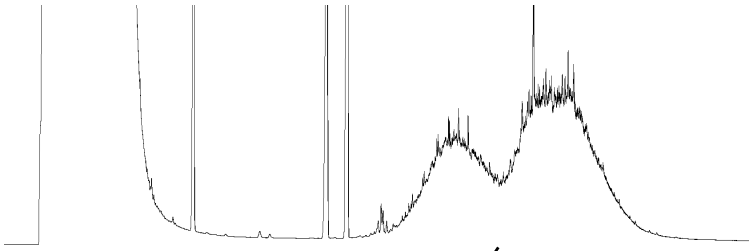
Stage Control



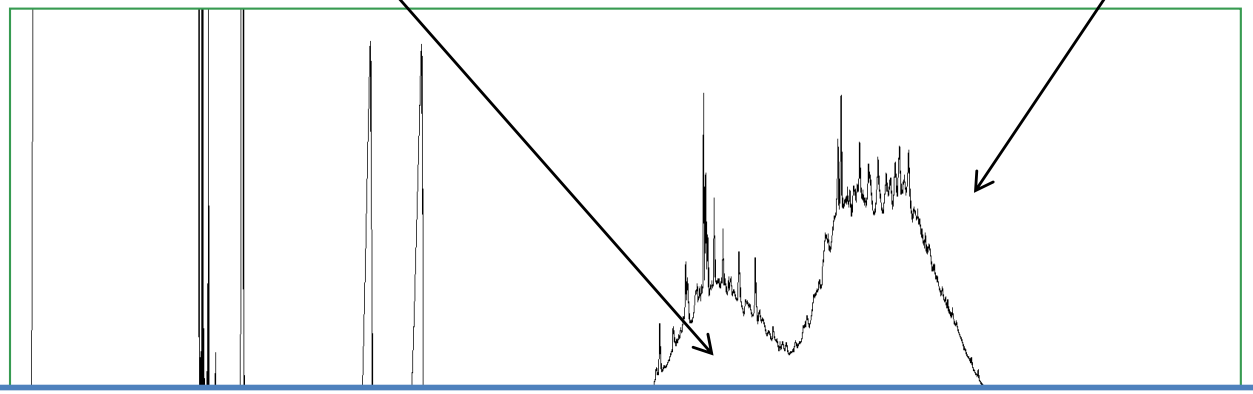
Stage Control



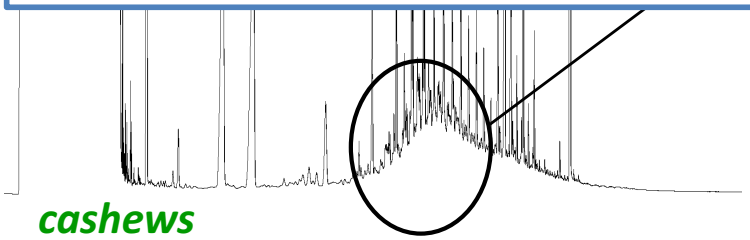
cheese



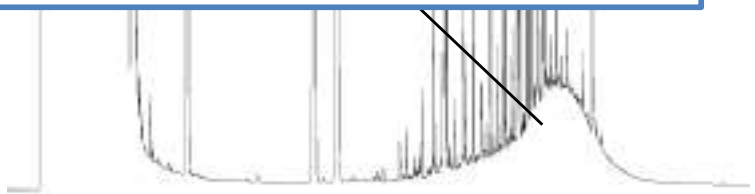
sunflower oil



Multiple sources of contamination with different, overlapping distributions

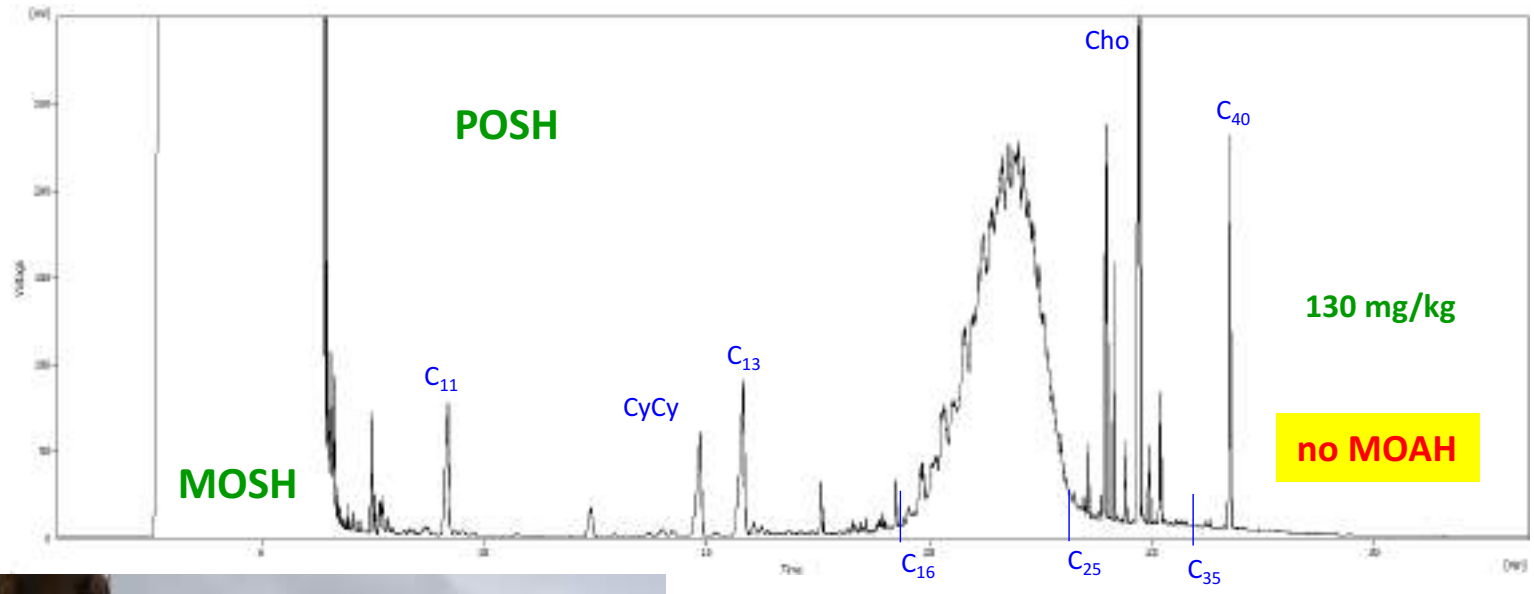


cashews



milk powder

crude soy oil



usage of anti dust agents

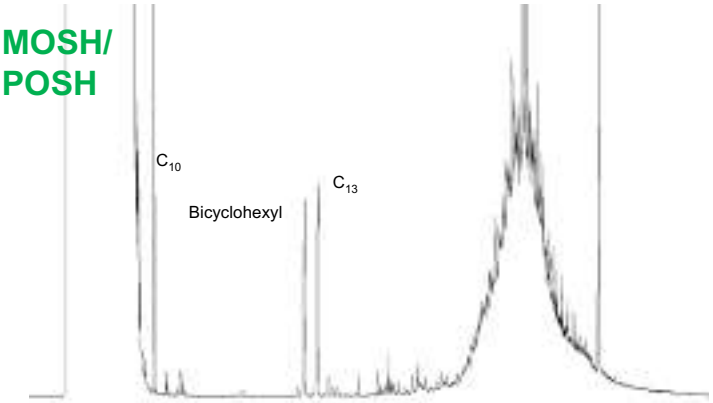
pictures: creative commons

Production

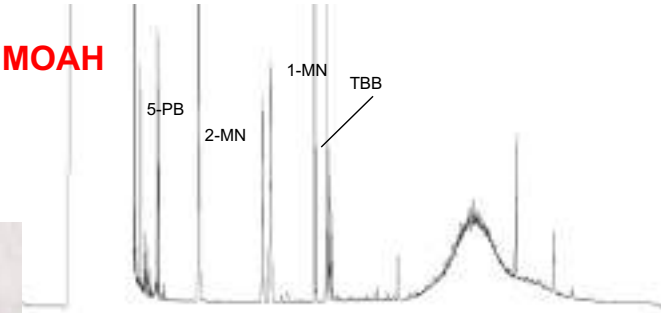
Contamination Source Lubricants

Almond oil

MOSH/ POSH

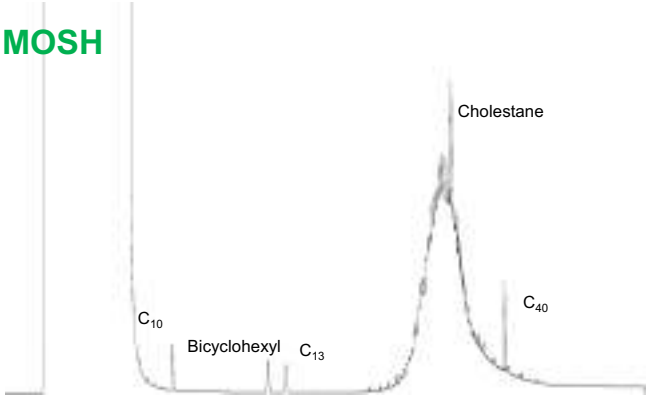


MOAH

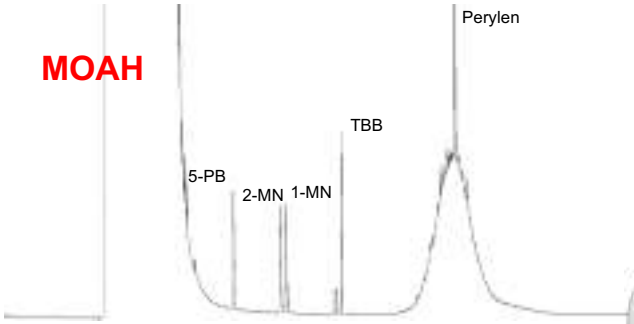


Lubricant

MOSH



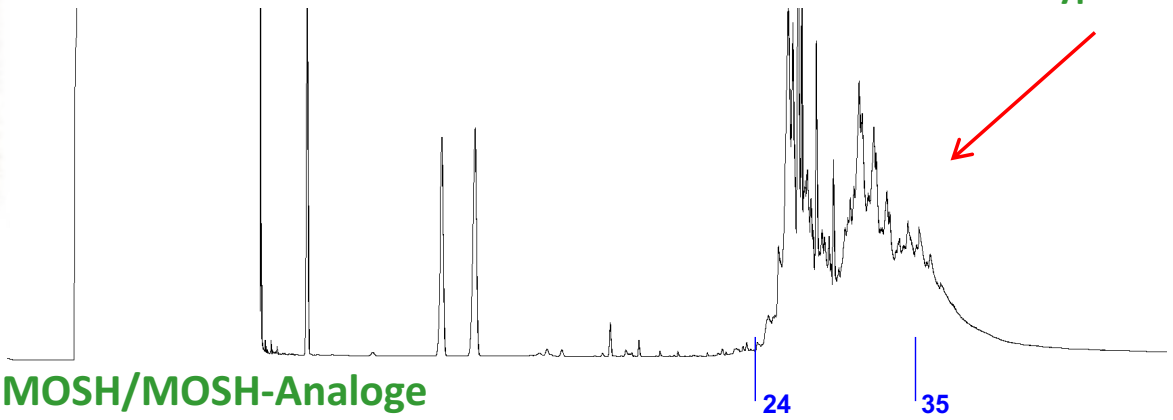
MOAH



Production

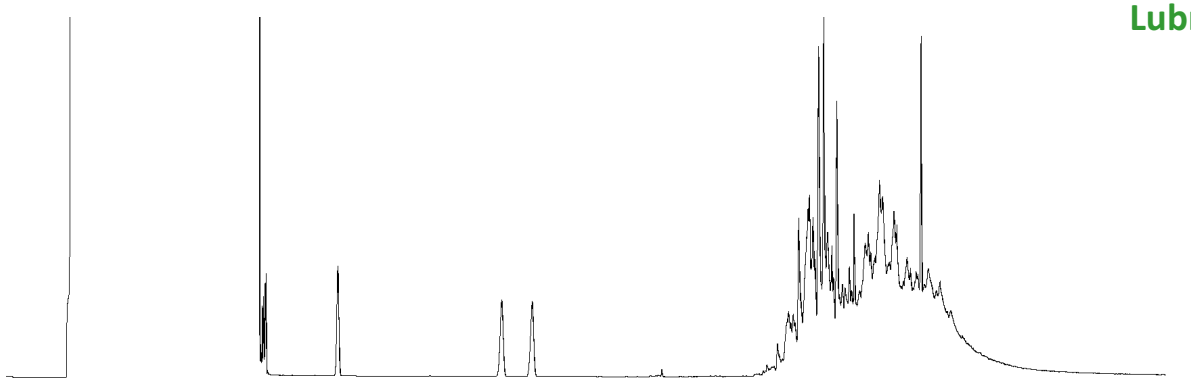
Contamination Source Lubricants

Mixed oil



MOSH/MOSH-Analoge

Lubricant



MOSH/MOSH-Analoge

-> contamination by lubricant

Food Processing

Lubricating Oils

> 240 lubricating oils

Food Processing

Lubricating Oils

> 240 lubricating oils

Mineral oil products

- higher molecular mass
- n-alkanes were removed
- Gaussian distribution: depicts distillation

Poly-alpha-Olefines (PAOs)

- Oligomers of synthetic olefins or cracking products
- synthetic lubricating oils
- food-grade oils
- adhesives

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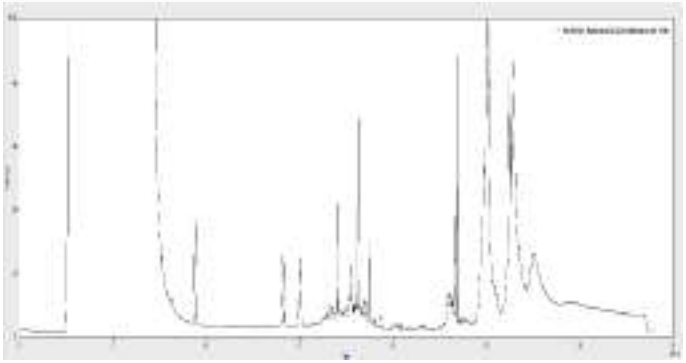

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Lubricating Oils

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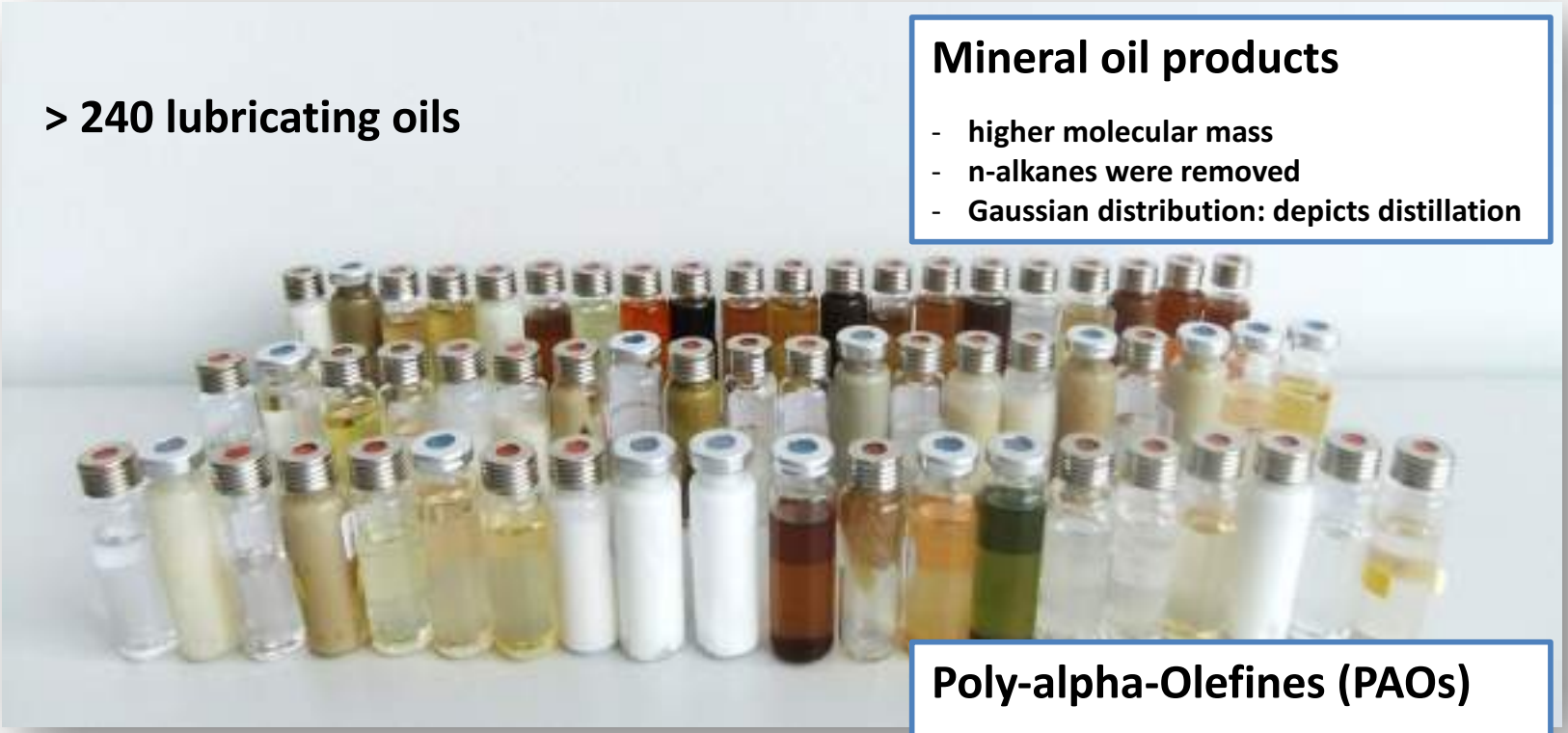

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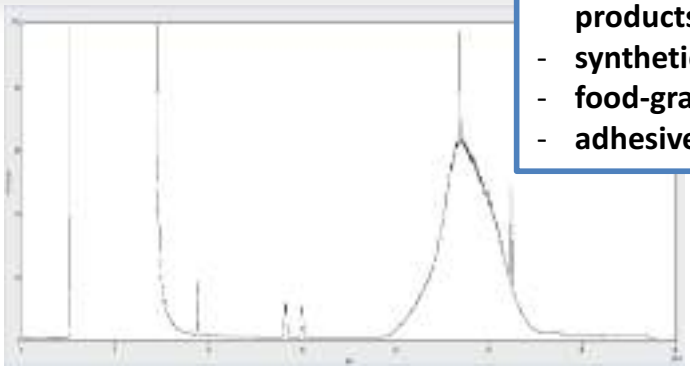

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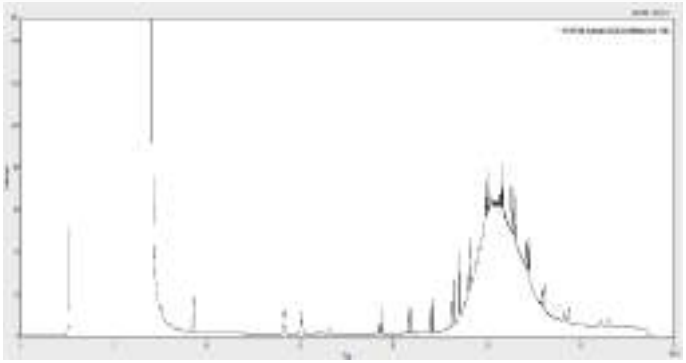

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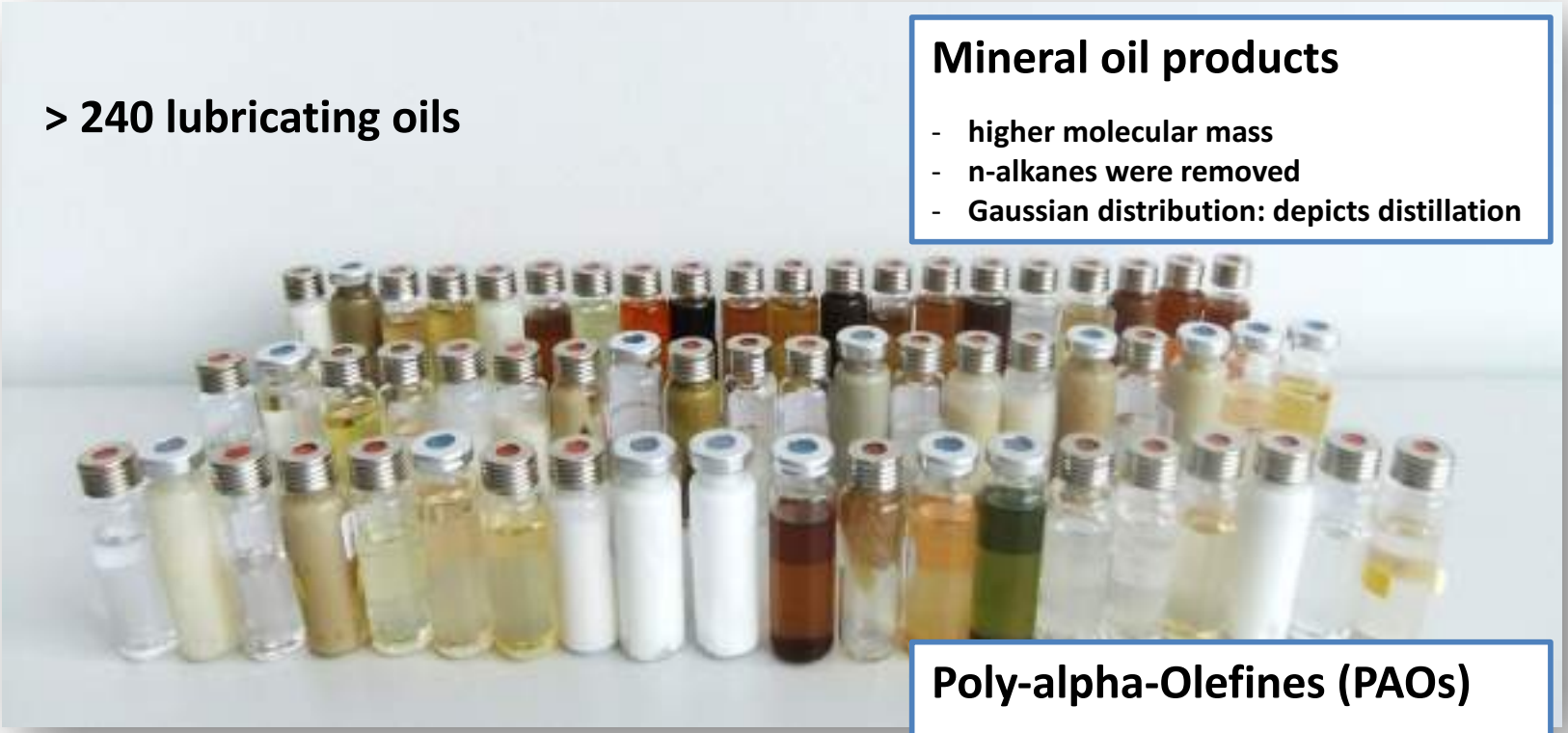

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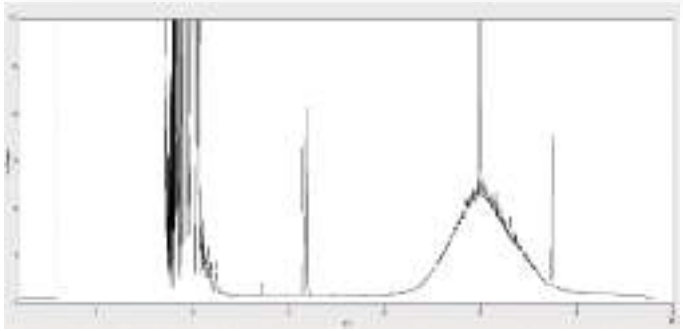

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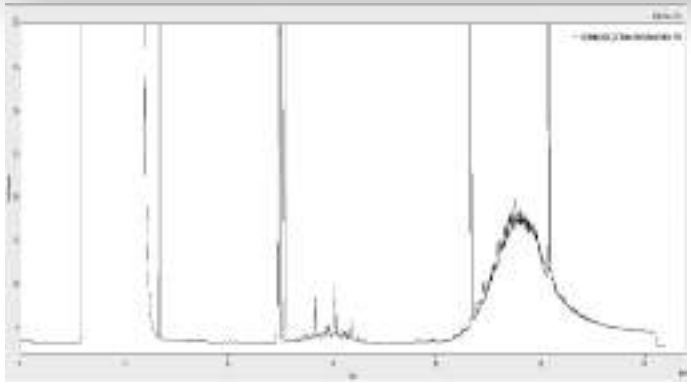

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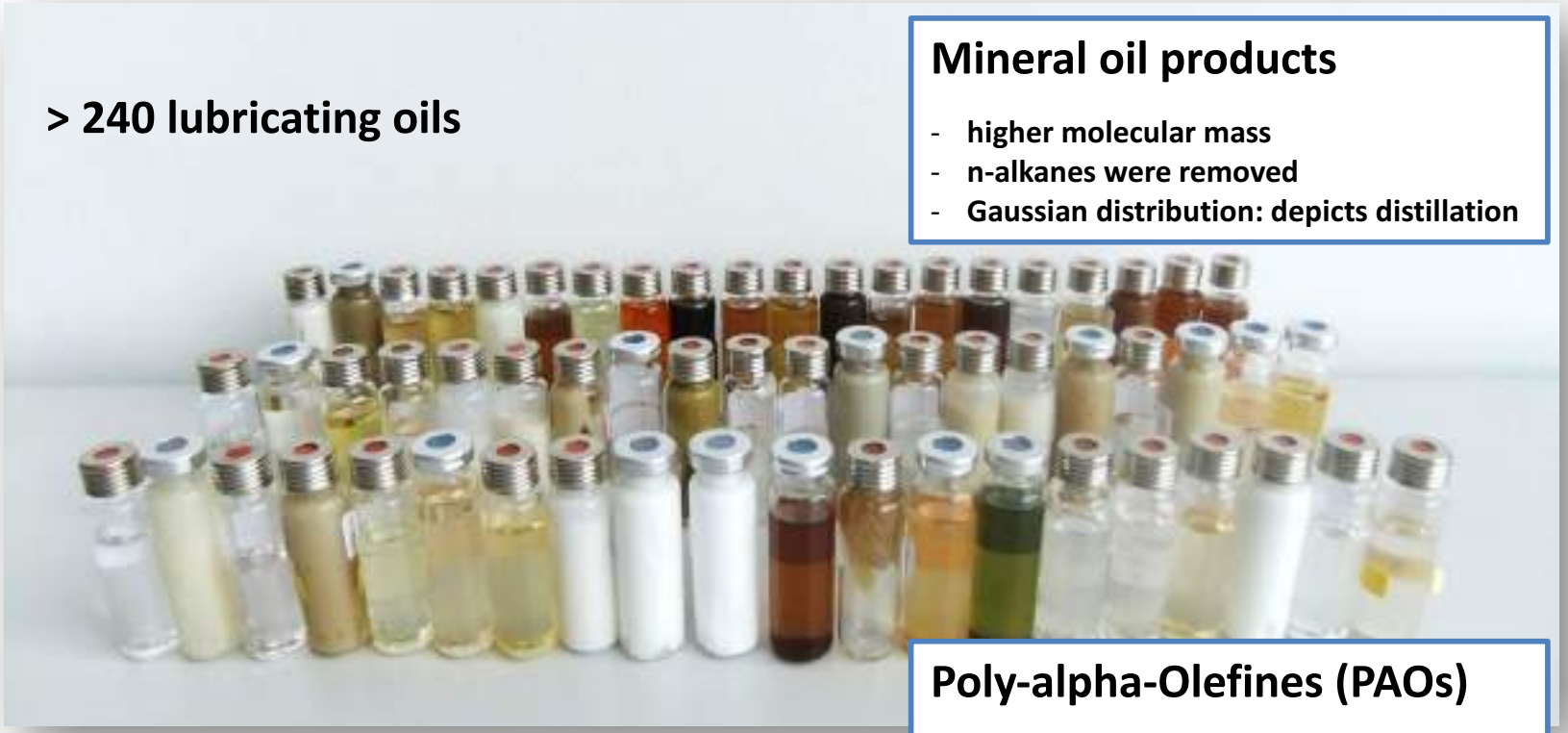

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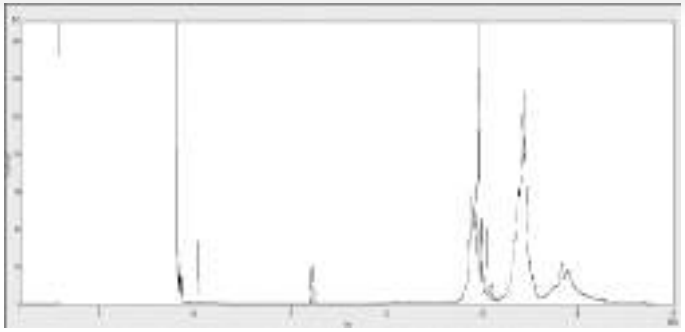

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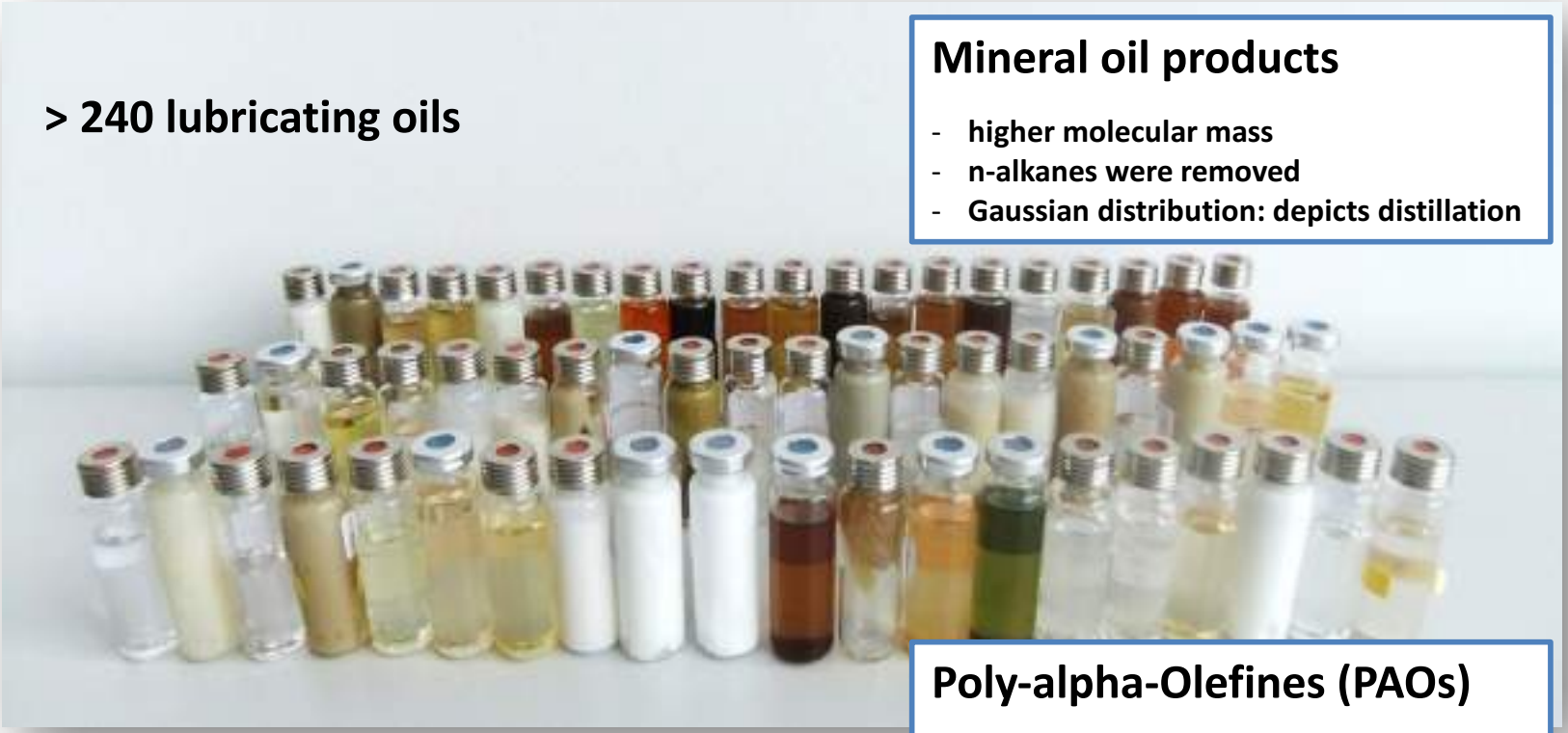

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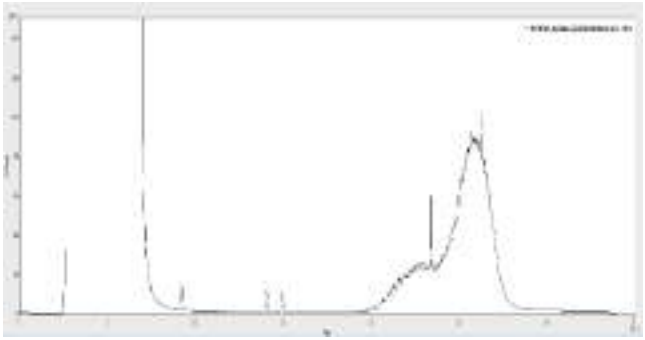
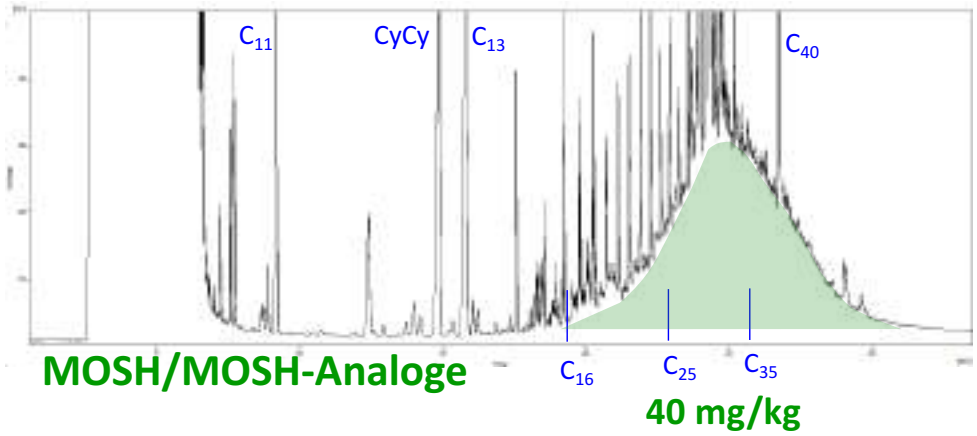
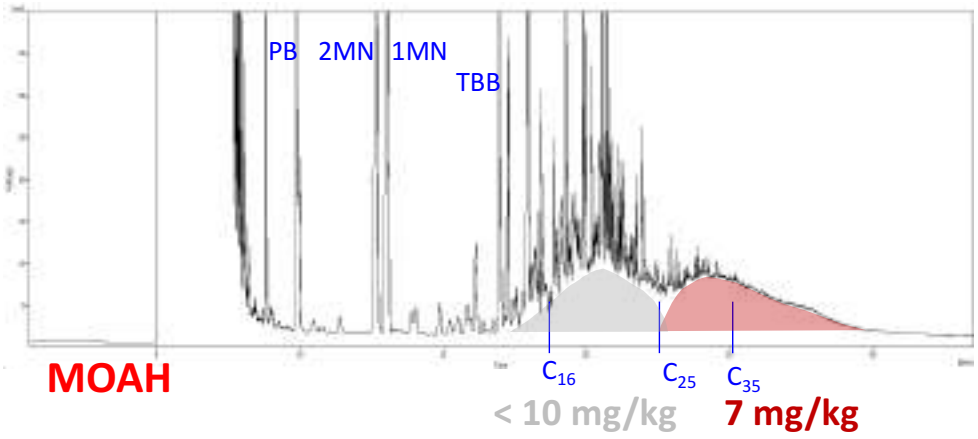


crude coconut oil



MOSH/MOSH-Analoge

40 mg/kg



MOAH

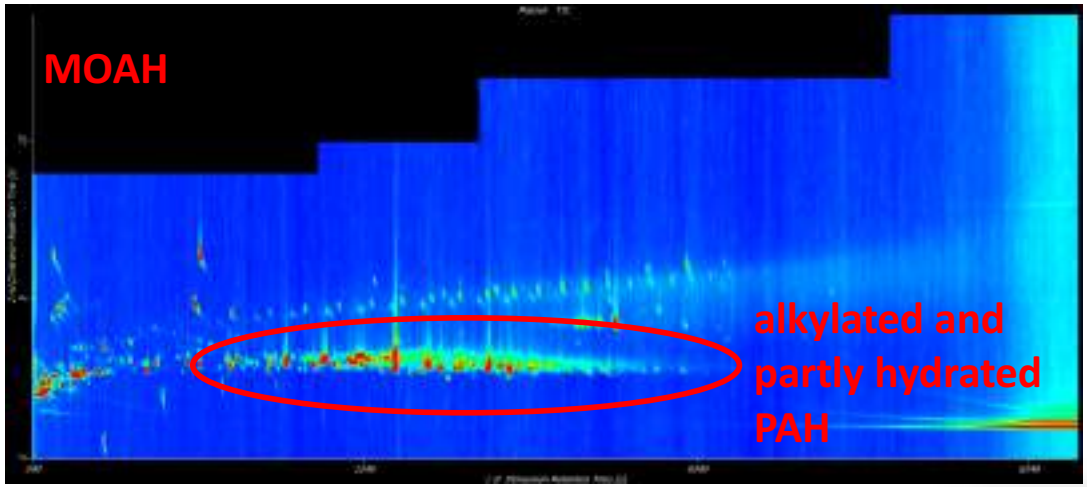
< 10 mg/kg

7 mg/kg

picture: creative commons

photo and copyright by Jeroen Hellingman

crude coconut oil



picture: creative commons

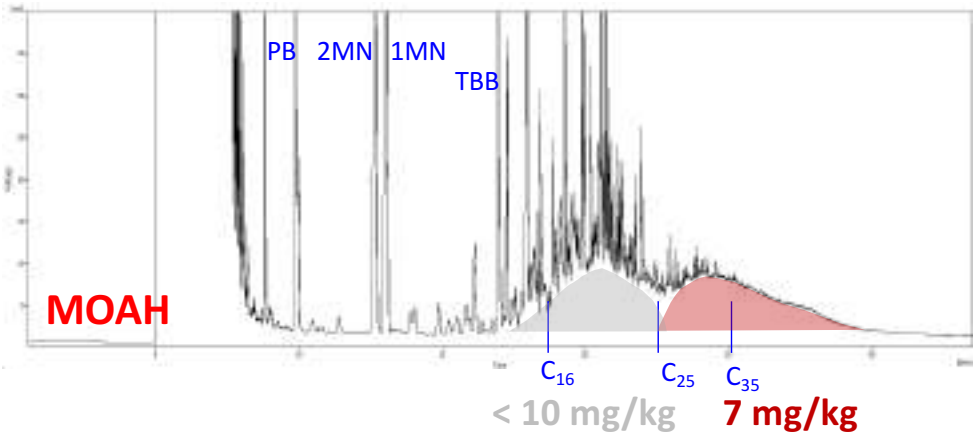
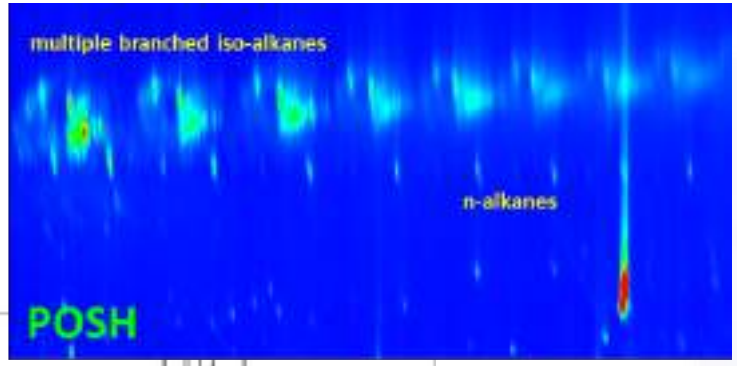
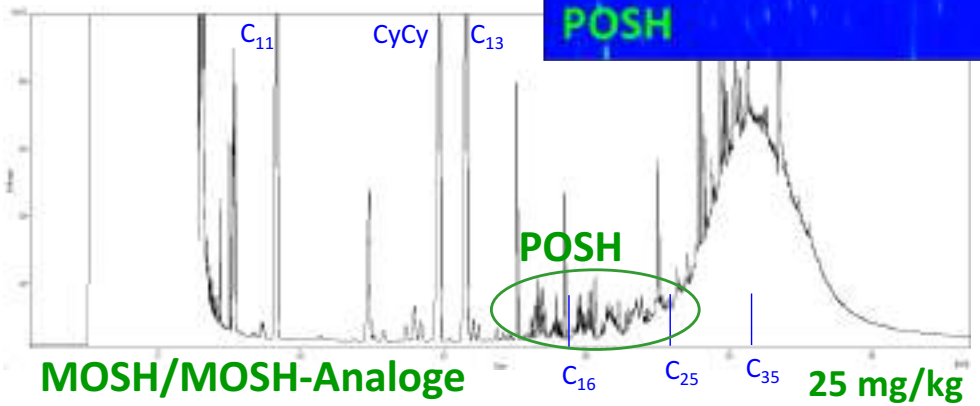


photo and copyright by Jeroen Hellingman

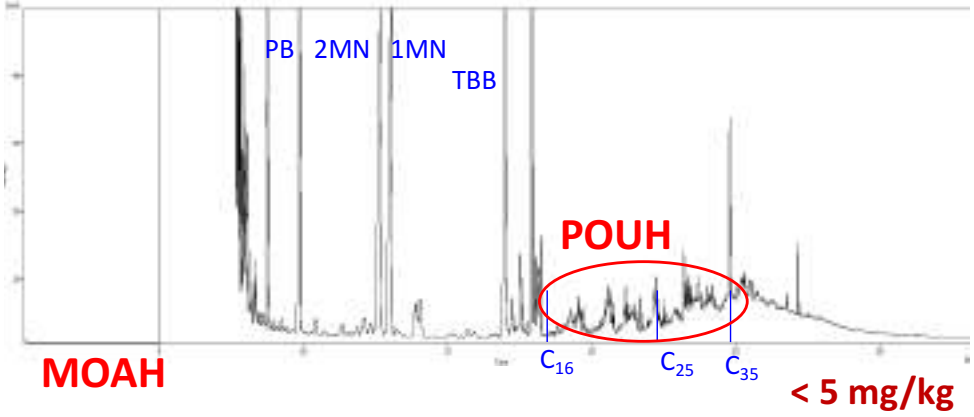
Transport and Packaging

raffinated palm oil





MOSH/MOSH-Analoge **25 mg/kg**

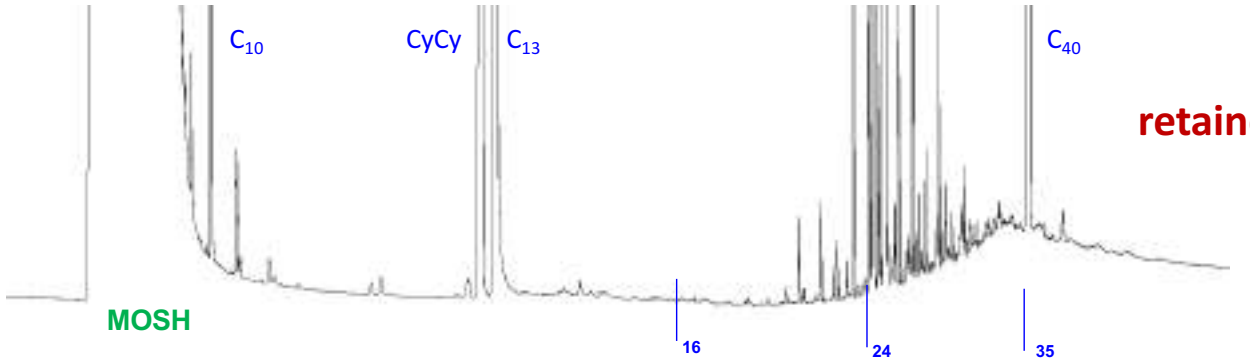


MOAH **< 5 mg/kg**

PP sample containers

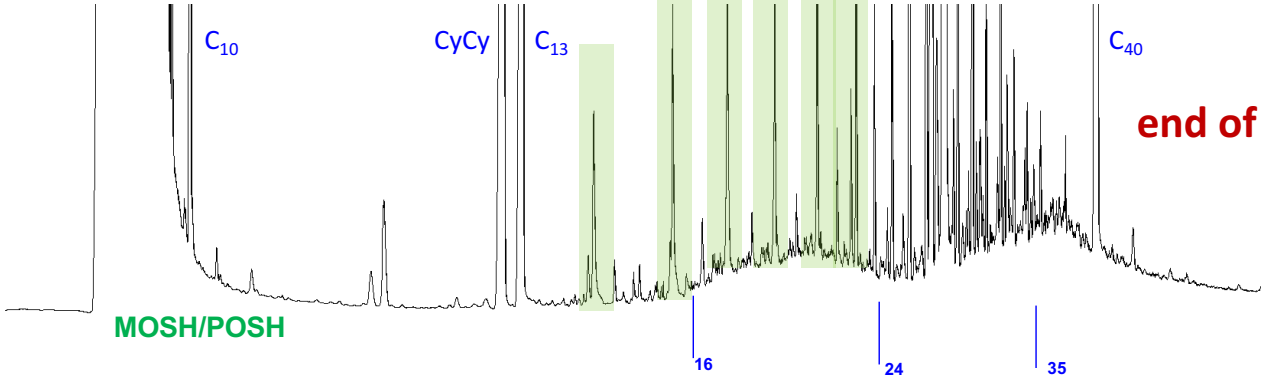
pictures: creative commons

Packaging and Storage



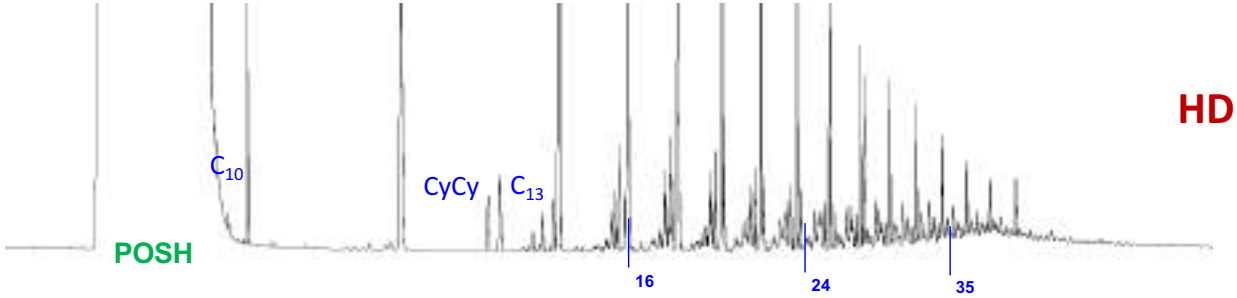
MOSH

retained sample



MOSH/POSH

end of shelf life



POSH

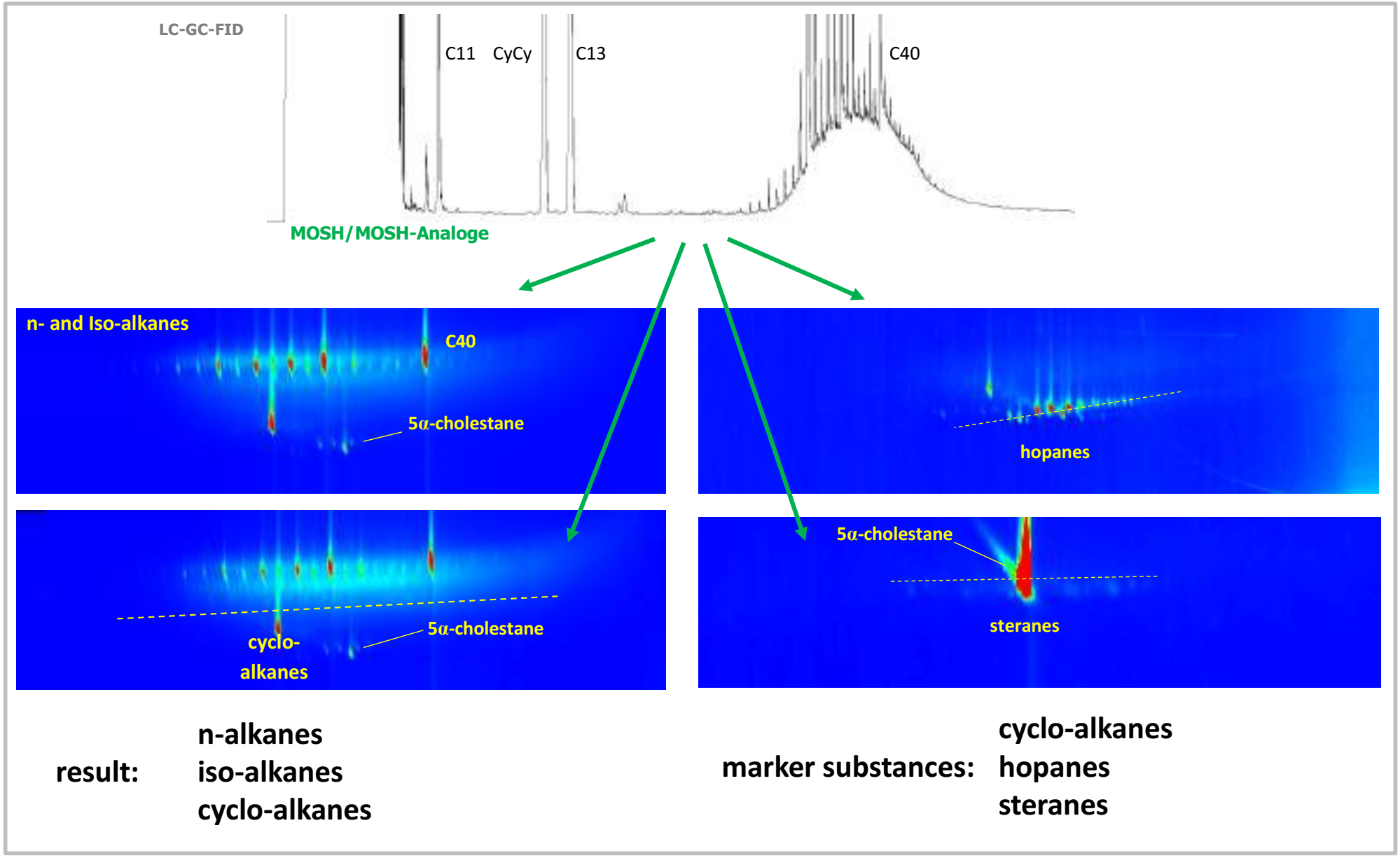
HDPE

Institut Kirchhoff Berlin

Palm oil

Hopane as Marker for Fossil MOH

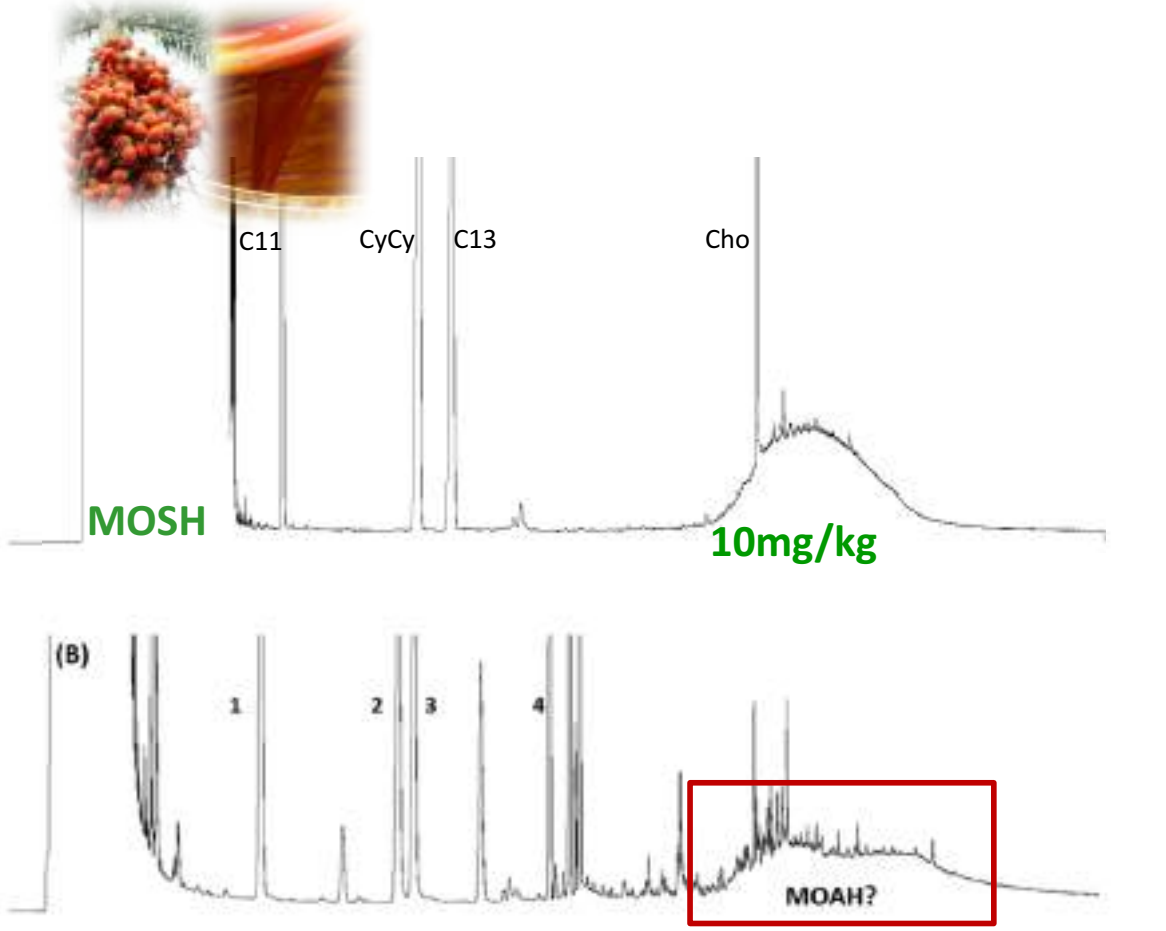
MERIEUX NutriSciences



result: n-alkanes, iso-alkanes, cyclo-alkanes

marker substances: cyclo-alkanes, hopanes, steranes

Characterization / Differentiation of MOAH to Biogenic Components



HPLC-GC-FID-Chromatogramm der MOAH in „Palmöl": (A) vor Aufreinigung, (B) nach Aufreinigung mittels Epoxidierung; 1 - 5PB, 2 - 1-MN, 3 - 2-MN, 4 – TBB, 5 - Squalen, 6 - Carotinoide

- in case of disturbances due to matrix components, an additional purification step is necessary
- after epoxidation a "hump" remains for certain samples
- no MOAH, but not epoxidized, biogenic substances

-> "Hump" is assigned to the MOAH by laboratories (false-positive)

-> significant consequences for raw material suppliers and food companies

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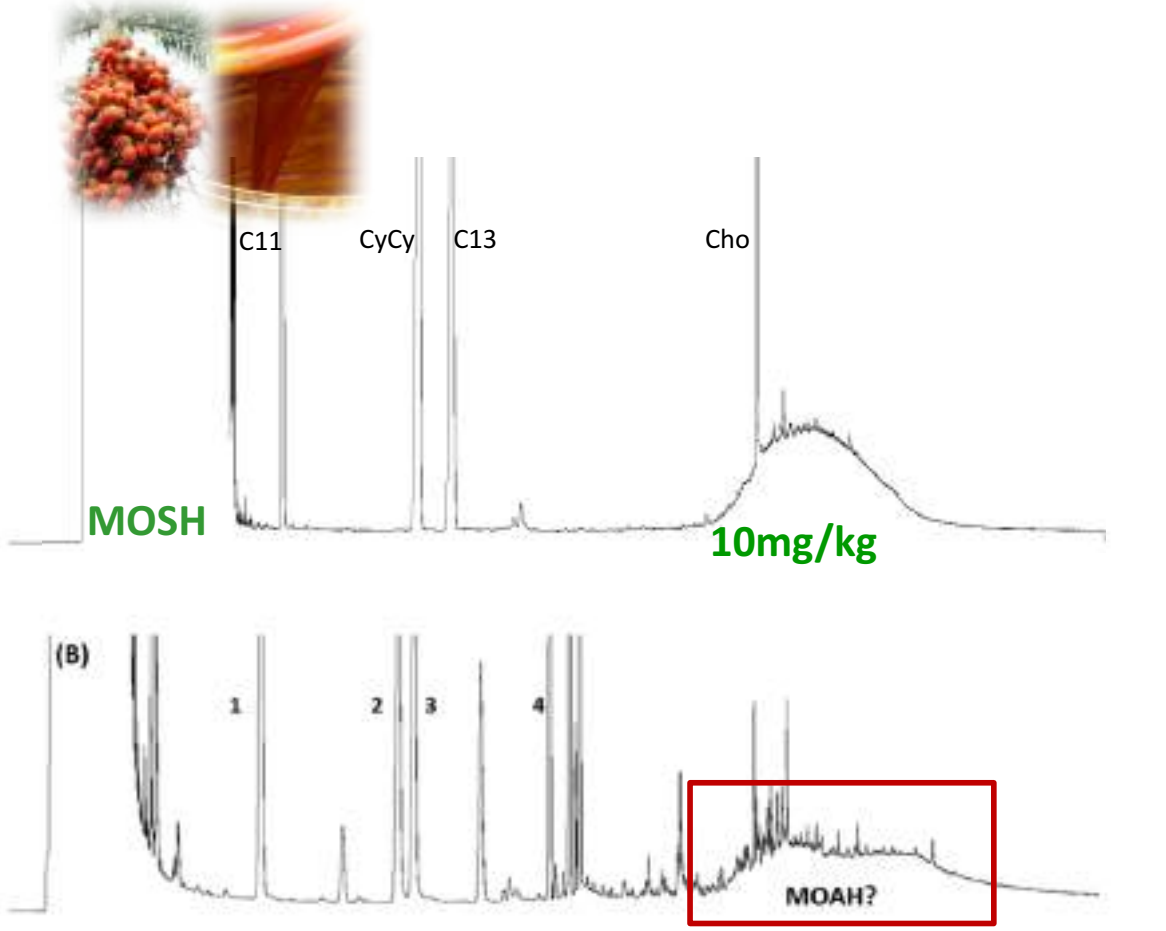

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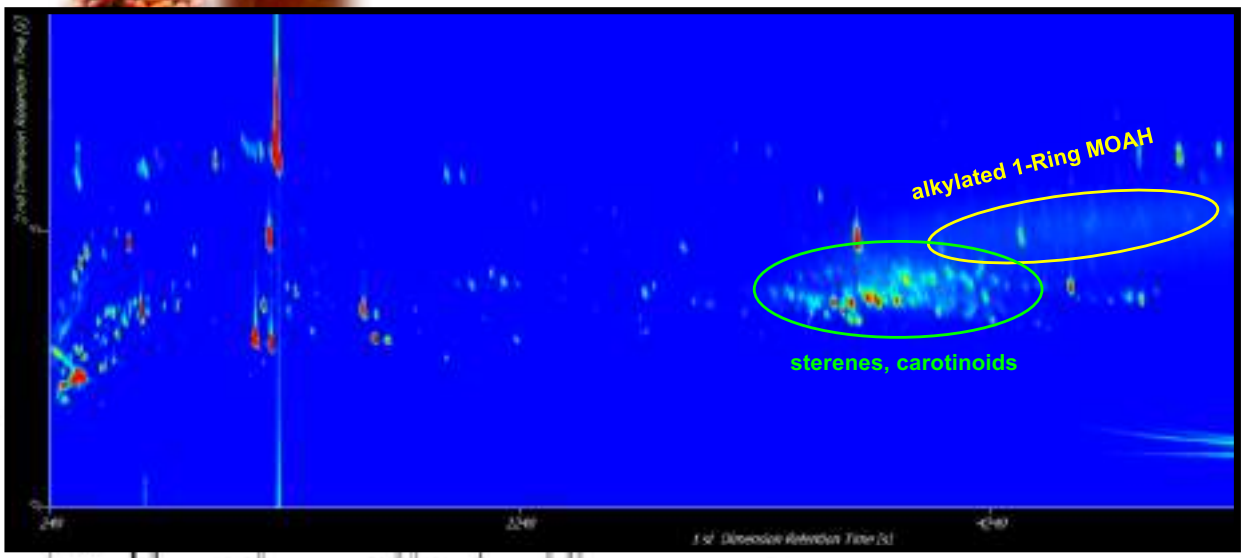

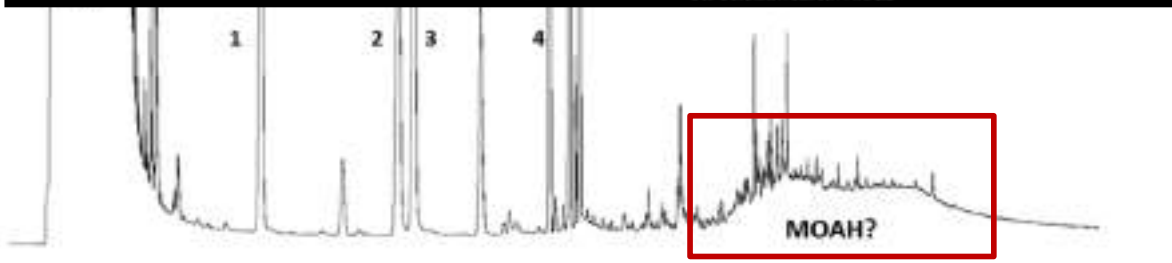

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Separation of polynuclear MOAH (≥3 ring) from the 1- and 2-ring systems

- According to the BfR contamination of food with MOAH should be avoided (potentially cancerogenic)
- EFSA: carcinogenic potential correlates with increasing number of aromatic ring systems

EFSA Journal 2012; 10(6): 2704

"MOAH with three or more, non- or simple alkylated, aromatic rings may be mutagenic and carcinogenic and therefore of potential concern."

J Agric Food Chem 2018 Jul 11;66(27):6968-6974

"MOAH of at least 3 (conjugated) aromatic rings may include genotoxic constitutents.

For this reason, it seems important to distinguish between MOAH of 1-2 and more aromatic rings."

-> reaction with DA complex chromatography

-> separation of the condensed aromatics regardless of the degree of alkylation only by number of aromatic rings

-> for the first time differentiated, quantitative statement about the composition of MOAH possible

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efsa European Food Safety Authority Supporting Publications

TECHNICAL REPORT

APPROVED: 15 November 2019

doi:10.2903/sp.efsa.2019.EN-1741

Rapid risk assessment on the possible risk for public health due to the contamination of infant formula and follow-on formula by mineral oil aromatic hydrocarbons (MOAH)

European Food Safety Authority (EFSA),
Davide Arcella, Katleen Baert, Marco Binaglia

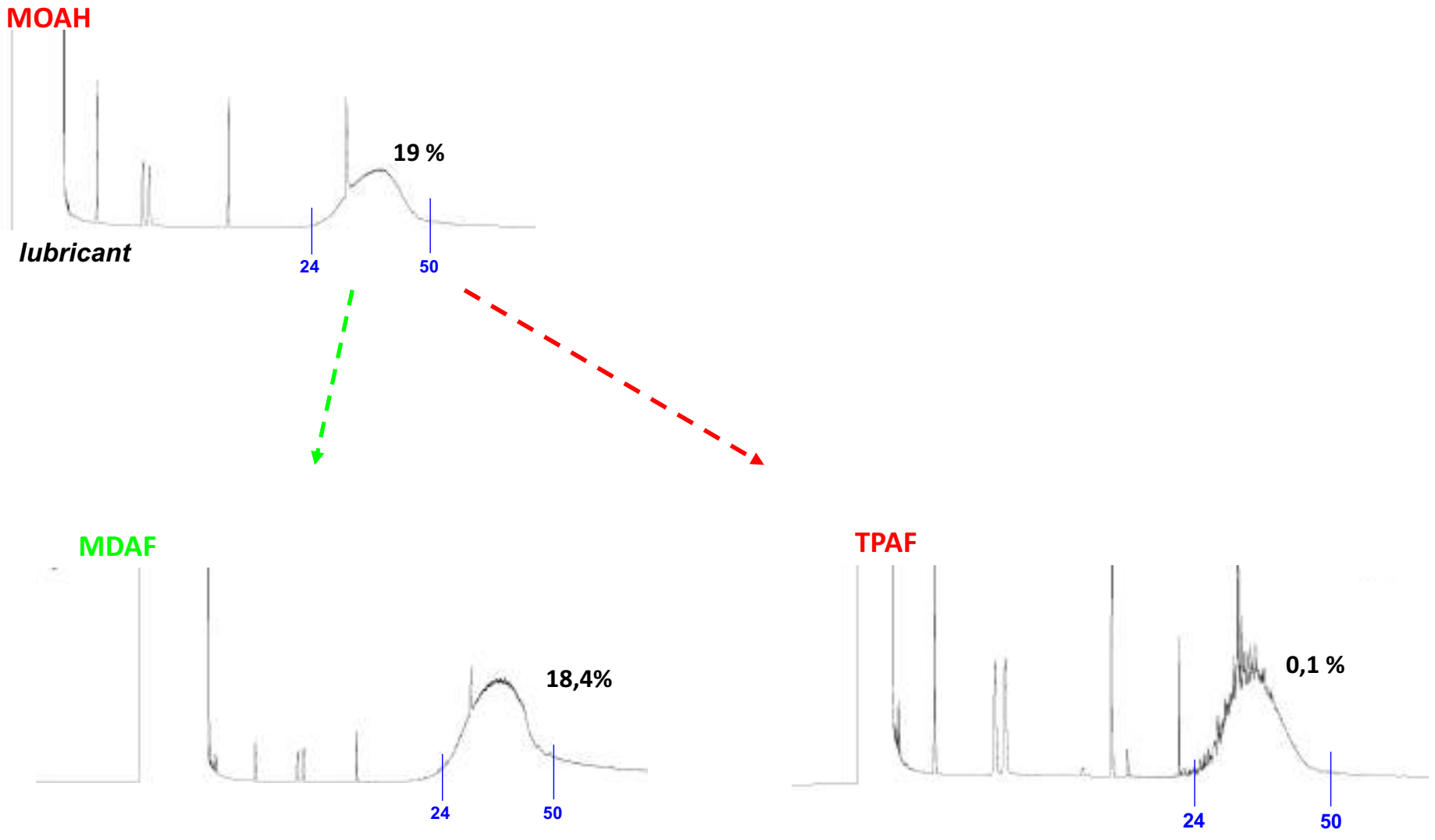
Conclusions

- Contamination of infant and follow-on formula, and more in general of food, with MOAH can originate from different sources either via the environment, during industrial food manufacturing and processing, or via transfer from food contact materials.
- 3-7 PAC are MOAH components of main concern for their genotoxic and carcinogenic nature.
- The likelihood of the presence of the genotoxic and carcinogenic 3-7 PAC in the MOAH detected in food depends on the contamination sources.

Recommendations

- Analytical methods to identify 3-7 PAC should be routinely applied when MOAH are detected in food.

Separation of polynuclear MOAH (≥3 ring) from the 1- and 2-ring systems



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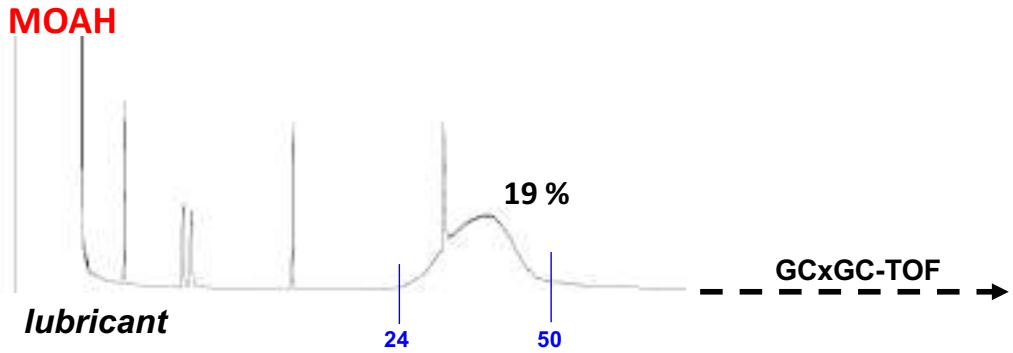

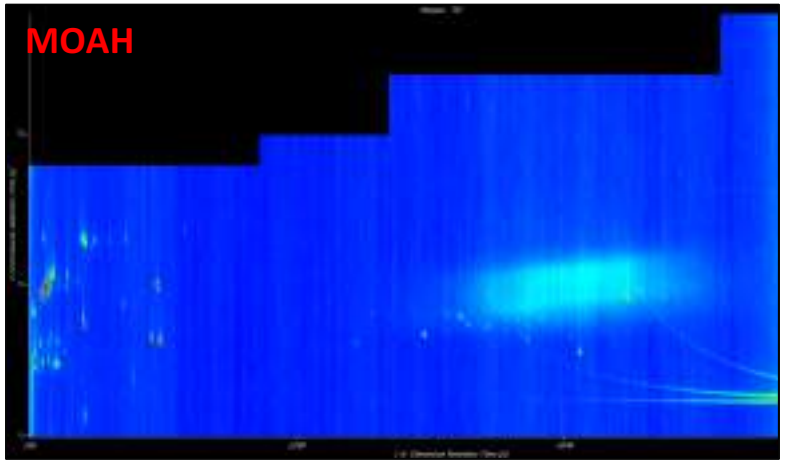

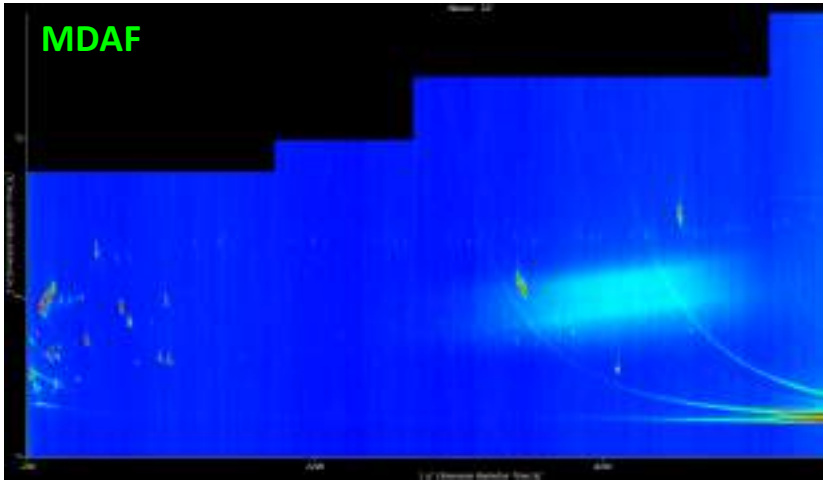

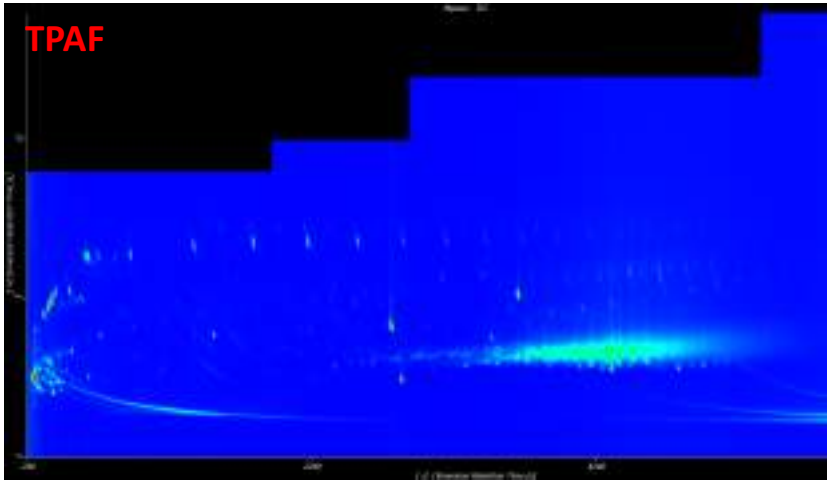

Thank you for your kind attention!

Knowing
what is inside…

Institut Kirchhoff Berlin GmbH
Oudenarder Straße 16 / Carrée Seestraße
13347 Berlin
Tel.: +49 (0) 30/457 98 93-0
www.institut-kirchhoff.de MXNS@institut-kirchhoff.de