Scientific Papers. Series D. Animal Science. Vol. LXVII, No. 1, 2024
ISSN 2285-5750; ISSN CD-ROM 2285-5769; ISSN Online 2393-2260; ISSN-L 2285-5750

VARIATION IN MINERAL OIL HYDROCARBONS CONTENT OF MILK DURING PROCESSING

Mădălina MATEI, Silvia-Ioana PETRESCU, Elena-Iuliana FLOCEA, Dragoș Mihai LĂPUȘNEANU, Daniel SIMEANU, Ioan Mircea POP

"Ion Ionescu de la Brad" Iași University of Life Sciences, 3 Mihail Sadoveanu Alley, Iași, Romania

Corresponding author email: mircea.pop@iuls.ro

Abstract

MOSH/MOAH in a food product may result from unintentional contamination occurring at various levels of the supply chain or migration from packaging. This preliminary research aims to evaluate the differences and variations in contamination levels between raw and processed milk samples to assess the contamination risk associated with milk processing. Using LC-GC-FID, mineral oil hydrocarbons (MOSH; MOAH) were quantified in eight milk samples. A T-test was applied to evaluate the differences in the distribution of carbonic sub-fractions and total levels of MOSH and MOAH contamination (n-C_{10-50}) in experimental samples of unprocessed milk and lyophilized milk. All statistical analyzes were performed using SPSS Statistics 26.0 and GraphPad Prism 9 software packages. The results of the comparative analysis revealed that milk processing operations had a significant impact on the variation of the MOH contamination level in the freeze-dried milk. MOSH and MOAH content varied significantly between initial samples (0.8-8.7 mg/kg MOSH; 0-2.8 mg/kg MOAH) and final samples (4.5-13.1 mg/kg MOSH; 3.2-5.35 mg/kg MOAH). These results are relevant for evaluating the impact of the freeze-drying process on these contaminants.

Key words*: contamination, milk, mineral oil hydrocarbons, processing.*

INTRODUCTION

Mineral oil hydrocarbons (MOH) come from petroleum distillation or are produced synthetically by coal and natural gas extraction (Bratinova & Hoekstra, 2019; Menegoz Ursol et al., 2023). These substances form a major category of contaminants of petrogenic origin, present in the environment as a result of pollution or due to contamination during production processes (Bruhl, 2016; Canavar et al., 2018).

MOH can reach the environment through various means, and their presence can be linked to multiple sources (Srbinovska et al., 2023; Purcaro et al., 2016). The major concerns about MOH contamination stem from the approved use of mineral oils in various products. Certain mineral oils have approved uses, including additives and processing aids in food (Van Heyst et al., 2019; Hoccheger et al., 2021).

Essential technological steps for the packaging and transport of raw materials can play a significant role in MOSH (Mineral Oil Saturated Hydrocarbons) and MOAH (Mineral Oil Aromatic Hydrocarbons) contamination, as these hydrocarbons can be present in paper and cardboard materials (Biedermann & Grob, 2015; Van Heyst et al., 2019; Menegoz Ursol et al., 2023), all with specific food migration properties (EC, 2011).

Currently, only food-grade lubricating mineral oils that are unrefined or partially refined can be used in food processing and related industries (EFSA, 2012; Menegoz Ursol et al., 2022).

The effects and toxicity of MOH are still unclear, but ongoing research shows that MOSH can accumulate in human organs and tissues (Hidalgo Ruiz et al., 2021), and MOAH, due to their structural similarity to PAHs, have carcinogenic potential (Nestola, 2022).

As a result of the non-polar character given by their chemical structure, MOSH and MOAH show a strong affinity for fatty substrates. Amounts of MOH have been found in a variety of foods, including those of animal origin (Foodwatch, 2021).

The occurrence and possible sources of contamination have been discussed in detail in the specialized literature (Moret et al., 2009; Gharbi et al., 2017; Matei & Pop, 2023).

Food safety of milk is an important issue given its essential role in providing nutrients, especially for vulnerable consumer groups (Matei & Pop, 2022; Singh, 2022).

Industries that want to be competitive develop new products for the market (Mierliță et al., 2024). As the industry develops new products, it is essential to consider tailoring them to consumer neuro-perception, balanced with their safety and healthiness. This involves the integration of sensory characteristics according to food safety issues (Lăpușneanu et al., 2021; Ciobanu et al., 2023).

To date, MOH in milk has been addressed mainly in the context of potential risks associated with packaging, and a few studies have reported the presence of mineral oils in infant milk products (Biedermann–Brem et al., 2012; Zhang et al., 2019).

In relation to the environment, MOH represent a new class of contaminants. MOSH and MOAH can be present in food from various sources, such as food packaging or environmental contamination. Because these substances can migrate into food and accumulate in the human body, there are concerns about their potential impact on human health.

The purpose of this preliminary research is to highlight the differences in the level of contamination between raw and processed milk samples, as well as to analyze the variation in contamination levels following processing. This aims to assess the risk of contamination associated with milk processing methods.

Using LC-GC-FID, saturated and aromatic mineral oil hydrocarbons (MOSH; MOAH) were quantified in eight milk samples. A T-test was applied to evaluate the differences in the distribution of MOSH and MOAH structured by carbon chain lenghts and the total level of contamination (n-C_{10-50}) in experimental samples of unprocessed milk and lyophilized milk. All statistical analyses were performed using SPSS Statistics 26.0 and Graph Pad Prism 9 software.

The information available to date and the results of this study can be integrated to develop future models for monitoring food contamination with MOH during processing.

MATERIALS AND METHODS

Samples

As part of the experimental monitoring proposed for the analysis of MOSH/MOAH content, four samples of cow's milk were collected from farms located in the northeastern Romania. Before conducting the actual testing, the milk samples, which were unprocessed and not previously packaged, were analyzed to determine the content of crude fat and dry matter for standardization, compatibility, and accuracy of the determinations.

To highlight the potential influence of processing on the level of sample contamination, the four samples were coded according to the type of processing applied:

A/B/C/D–1: experimental samples of unprocessed milk;

A/B/C/D–2: experimental samples of processed milk (lyophilization).

The processing applied to the experimental samples (2) involved staged lyophilization of frozen samples (-18ºC), at temperatures ranging from -30ºC to -75ºC, with a pressure of 1.65 mbar and a vacuum of 0.200 mbar, over a period of 7 days using a Christ Epsilon 2-4 LSC plus lyophilizer.

During processing, the frozen milk samples were covered with classic, bleached, waxed food paper purchased from a supermarket.

Details regarding the characteristics of the studied samples are included in Table 1.

Table 1. Specifications for experimental milk samples

	Code & sample no.			
	A	**B**	**C**	**D**
DM %	12.3	11.9	13.1	12.5
Fat (% DM)	37.3	35.4	29.6	29.6

DM = dry matter.

MOSH/MOAH analysis

MOH analysis was performed using the advanced coupled LC-GC-FID technique. The determination method was originally developed by Biederman et al. (2009) and later detailed by Bierdemann & Grob (2012), concerning the process of extraction, separation, and quantification of MOH. The current working protocol was adapted and optimized by Moret et al. (2016) and has been successfully applied in more studies, including those conducted by Menegoz Ursol et al. (2022), Srbinovska et al. (2022); Srbinovska et al. (2023). This method has been validated according to the analytical performance criteria set out in the JRC Guideline (Bratinova & Hoekstra, 2019).

Table 2. Infrastructure for MOH analysis

Sample preparation and analysis	Equipment	Performance, integration and quantification	Data collection and processing
n-hexane (≥ 95 %; CAS: 110-54-3) methanol (≥ 99,9 %; CAS: 67-56-1) saturated KOH (CAS: 1310-58-3) metachloroperoxybenzoic acid (70–75 %; CAS: 937-14-4); 200 mg/ mL ethanol	[1]LC-GC 9000*: HPLC Phoenix 9000 coupled to GC model Trace 1310	internal standard (IS) #31070 (150-600 µg/ml in toluene 99 %)	Chromeleon
anhydrous sodium thiosulfate (CAS 7772-98-7) aluminum oxide (CAS: 1344-28-1) sodium sulfate (CAS: 7757-82-6)	HPLC column 25 cm×2.1 mm i.d. with Lichrospher Si 60.5 µm particle size		
Supplier			
Merck Millipore (Massachusetts, SUA) Sigma-Aldrich Supelco Acros Organics, Thermo Fisher Scientific (Waltham, Massachusetts, SUA)	Brechbuhler (Zurich, Elveția) Thermo Fisher Scientific (Waltham, Massachusetts, SUA) DGB (Germany)	Restek, Bellefonte, PA, SUA	Thermo Fisher Scientific (Waltham, Massachusetts, SUA)

[1]configured with a dual channel allowing simultaneous analysis of MOSH and MOAH.

Data distribution was assessed using GraphPad Prism 9 and SPSS Statistics 26.0 software. A T-test was applied to evaluate the differences in the distribution of MOSH and MOAH, structured by carbonic sub-fractions and the total level of contamination (n-C_{10-50}).

RESULTS AND DISCUSSIONS

Milk is highly susceptible to exogenous contamination regardless of its form.

The fat and dry matter content influenced the analytical methods used to detect contaminants. Precision and accuracy of measurements were ensured by adjusting the analytical components based on the fat and dry matter content of the four analyzed milk samples.

Standardization of the samples based on the lipid fraction and dry matter content enable comparability of results across the samples.

MOSH and MOAH tend to accumulate more in the lipids of foods. The distribution of contaminants based on fat content showed varying concentrations depending on the composition of each milk sample.

The levels of MOSH and MOAH varied significantly between experimental samples of raw milk (0.8-8.7 mg/kg MOSH; 0-2.8 mg/kg MOAH) and freeze-dried milk (4.5-13.1 mg/kg MOSH; 3.2-5.35 mg/kg MOAH). Almost all milk samples exceeded the recommended limits set by the Standing Committee for Plants, Animals, Food and Feed, Section for Novel Foods and Toxicological Safety of the Food Chain (ScoPAFF) of the European Commission: 0.5 mg/kg for products with fat content < 4% (samples C, D); 1.0 mg/kg for products with fat content > 4% (samples A, B).

The MOSH/MOAH concentrations measured in lyophilized milk (A_2; B_2; C_2; D_2) were significantly higher than those in liquid form (A_1; B_1; C_1; D_1) within the same category.

The test results indicate significant differences in MOH contamination levels. A T-test was applied to evaluate the differences in MOSH and MOAH distribution based on carbonic sub-fractions and total contamination levels (n-C_{10-50}) between raw milk and freeze-dried milk.

Table 3 presents the results of MOSH distribution based on carbon sub-fractions and total contamination levels (n-C_{10-50}) in experimental samples of unprocessed milk, aiding in the understanding of MOSH contamination levels in unprocessed samples. The Skewness coefficient for n-C_{10-16}, indicates a slight left skewness of the data, but not significant, while for the total level of contamination (n-C_{10-50}) it suggests a positive skewness, indicating values both lower and higher than the mean (3.73) in the dataset.

Table 4 shows the results of MOSH distribution structured based on carbon chain lenghts and total contamination levels (n-C_{10-50}) in experimental samples of freeze-dried milk.

For n-C_{10-16}, the Skewness coefficient of -0.75 indicates a slight left skewness in the distribution, suggesting that there are several values lower than the mean of 0.53.

Table 3. Distribution of MOSH mg kg^{-1} structured by carbonic sub-fractions and the total level of contamination (n-C_{10-50}) in experimental samples of unprocessed milk (A, B, C, D – 1)

	n-C_{10-16}	n-C_{16-20}	n-C_{20-25}	n-C_{25-35}	n-C_{35-40}	n-C_{40-50}	n-C_{10-50}
Mean	0.58	0.83	0.70	1.30	0.23	0.10	**3.73**
Standard Error	0.22	0.25	0.50	1.00	0.13	0.07	**1.73**
Median	0.60	0.85	0.25	0.35	0.10	0.05	**2.70**
Standard Deviation	0.44	0.51	1.00	2.00	0.25	0.14	**3.46**
Sample Variance	0.20	0.26	1.01	4.01	0.06	0.02	**11.97**
Kurtosis	(4.77)	(0.10)	3.87	3.97	4.00	1.50	**2.58**
Skewness	(0.13)	(0.26)	1.96	1.99	2.00	1.41	**1.52**
Range	0.90	1.20	2.10	4.10	0.50	0.30	**7.90**
Minimum	0.10	0.20	0.10	0.20	0.10	0	**0.80**
Maximum	1.00	1.40	2.20	4.30	0.60	0.30	**8.70**
Sum	2.30	3.30	2.80	5.20	0.90	0.40	**14.90**
Count	4.00	4.00	4.00	4.00	4.00	4.00	**4.00**
Confidence Level (95.0%)	0.70	0.80	1.60	3.19	0.40	0.23	**5.51**

For the total contamination (n-C_{10-50}), values in dataset vary below or above the mean of 8.83. Figure 1 and the results in Table 5 indicate significant differences between raw milk and freeze-dried milk ($p \leq .015$) for the following carbon sub-fractions: n-C_{10-16}, n-C_{16-20}, n-C_{20-25}, n-C_{25-35}, n-C_{35-40} and n-C_{40-50}.

Table 4. Distribution of MOSH mg kg^{-1} structured by carbonic sub-fractions and the total level of contamination (n-C_{10-50}) in experimental samples of freeze-dried milk (A, B, C, D – 2)

	n-C_{10-16}	n-C_{16-20}	n-C_{20-25}	n-C_{25-35}	n-C_{35-40}	n-C_{40-50}	n-C_{10-50}
Mean	0.53	4.28	2.18	1.35	0.30	0.20	8.83
Standard Error	0.09	0.88	0.33	0.33	0.09	0.07	1.76
Median	0.55	4.30	2.15	1.40	0.30	0.15	8.85
Standard Deviation	0.17	1.76	0.66	0.66	0.18	0.14	3.51
Sample Variance	0.03	3.09	0.43	0.43	0.03	0.02	12.34
Kurtosis	0.34	1.44	1.41	1.61	(3.30)	1.50	1.46
Skewness	(0.75)	(0.08)	0.23	(0.45)	0.00	1.41	(0.04)
Range	0.40	4.30	1.60	1.60	0.40	0.30	8.60
Minimum	0.30	2.10	1.40	0.50	0.10	0.10	4.50
Maximum	0.70	6.40	3.00	2.10	0.50	0.40	13.10
Sum	2.10	17.10	8.70	5.40	1.20	0.80	35.30
Count	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Confidence Level (95.0 %)	0.27	2.80	1.04	1.04	0.29	0.23	5.59

Significant differences ($p = .002$) were observed for the total contamination level (n-C_{10-50}) of MOSH between raw milk (1) and freeze-dried milk (2). Freeze-dried milk (2) exhibited significantly higher contamination levels compared to raw milk (1), particularly evident in the values of n-C_{10-50}.

The effect size, Cohen's d was estimated at 0.3117 with a confidence interval ranging from 0.604 and 2.883, indicating significant results for n-C_{10-16}. For the total contamination level (n-C_{10-50}) of MOSH in raw milk and freeze-dried milk, Cohen's d was calculated as 4.2250, and with Hedges correction, it was 4.7568.

The 95 % confidence interval for Cohen's d ranged from 0.433 to 2.493, and for Hedges correction, it ranged from 0.385 to 2.214. These values suggest a significant and consistent difference in the effect of MOSH across various carbon chain lengths.

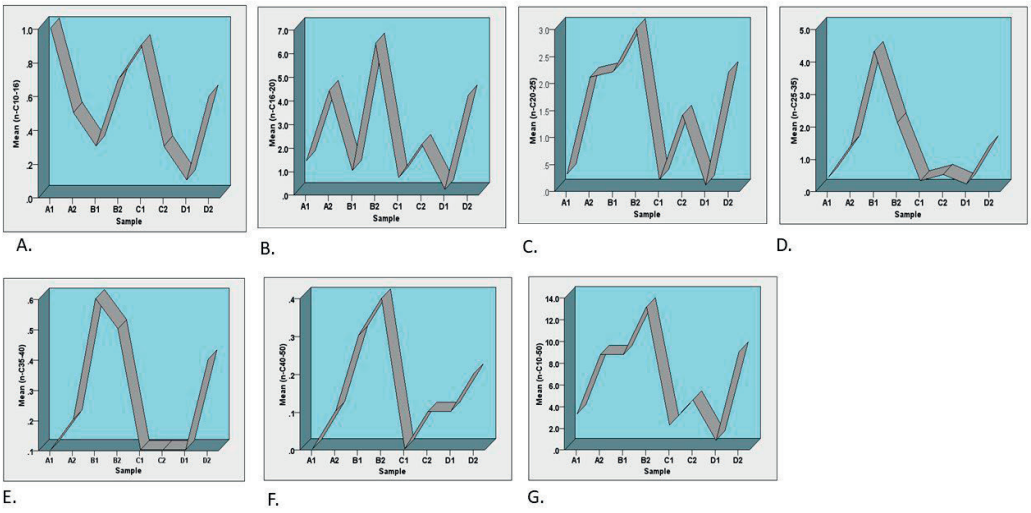

Figure 1. Distribution of structured MOSH averages at different carbon sub-fractions A (n-C_{10-16}), B (n-C_{16-20}), C (n-C_{20-25}), D (n-C_{25-35}), E (n-C_{35-40}), F (n-C_{40-50}) and total contamination level G (n-C_{10-50}) in experimental raw milk samples (A_1, B_1, C_1, D_1) and lyophilized milk samples (A_2, B_2, C_2, D_2)

Table 3. Differences in the distribution of MOSH mg kg^{-1} structured by carbonic sub-fractions and the total level of contamination (n-C_{10-50}) in experimental unprocessed milk samples (1) and freeze-dried milk samples (2)

	t	Df	Significance		Mean Difference	95 % Confidence Interval of the Difference	
			One-Sided *p*	Two-Sided *p*		Lower	Upper
n-C_{10-16}	4.991	7	<.001	.002	.5500	.289	.811
n-C_{16-20}	3.280	7	.007	.013	2.5500	.712	4.388
n-C_{20-25}	3.656	7	.004	.008	1.4375	.508	2.367
n-C_{25-35}	2.717	7	.015	.030	1.3250	.172	2.478
n-C_{35-40}	3.594	7	.004	.009	.2625	.090	.435
n-C_{40-50}	3.000	7	.010	.020	.1500	.032	.268
n-C_{10-50}	4.201	7	.002	.004	6.2750	2.743	9.807

T-test, significance level *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

Table 4. Cohen's d and Hedges' correction results for evaluating the effect of MOSH mg kg^{-1} between experimental samples of unprocessed milk (1) and lyophilized milk (2), with 95 % confidence intervals for carbonic sub-fractions n -C_{10-16}, n-C_{16-20}, n-C_{20-25}, n-C_{25-35}, n-C_{35-40}, n-C_{40-50} and total contamination level (n-C_{10-50})

		Standardizer[a]	Point Estimate	95 % Confidence Interval	
				Lower	Upper
n-C_{10-16}	Cohen's d	.3117	1.765	.604	2.883
	Hedges' correction	.3509	1.567	.537	2.561
n-C_{16-20}	Cohen's d	2.1987	1.160	.224	2.051
	Hedges' correction	2.4754	1.030	.199	1.821
n-C_{20-25}	Cohen's d	1.1122	1.292	.311	2.229
	Hedges' correction	1.2522	1.148	.276	1.980
n-C_{25-35}	Cohen's d	1.3792	.961	.088	1.789
	Hedges' correction	1.5528	.853	.078	1.589
n-C_{35-40}	Cohen's d	.2066	1.271	.296	2.199
	Hedges' correction	.2326	1.129	.263	1.954
n-C_{40-50}	Cohen's d	.1414	1.061	.157	1.920
	Hedges' correction	.1592	.942	.139	1.705
n-C_{10-50}	Cohen's d	4.2250	1.485	.433	2.493
	Hedges' correction	4.7568	1.319	.385	2.214

a. The denominator used in estimating the effect sizes. Cohen's d uses the sample standard deviation. Hedges' correction uses the sample standard deviation, plus a correction factor.

Cohen's d is a standard measure of effect size, and Hedges' correction is a modification of Cohen's d designed to account for potential overestimation in small sample sizes. Both measures are valuable for interpreting the magnitude of differences between groups in research (Table 4).

Table 5 shows the results of MOAH distribution structured by carbon sub-fractions and total contamination level (n-C_{10-50}) in experimental unprocessed milk samples, aiding in the understanding of MOAH contamination levels in these samples. For n-C_{10-16}, the Skewness coefficient of 2.00 indicates a significant right skewness in the data distribution, suggesting a trend towards higher values in the dataset. The total contamination level (n-C_{10-50}) exhibits positive skewness in the distribution, indicating that the dataset contains values lower and higher than the mean (0.70).

Table 5. Distribution of MOAH mg kg^{-1} structured by carbonic sub-fractions and the total level of contamination (n-C_{10-50}) in experimental samples of unprocessed milk

	n-C_{10-16}	**n-C_{16-25}**	**n-C_{25-35}**	**n-C_{35-50}**	**n-C_{10-50}**
Mean	0.15	0.43	0.13	0	0.70
Standard Error	0.15	0.43	0.13	0	0.70
Median	0	0	0	0	0
Standard Deviation	0.30	0.85	0.25	0	1.40
Sample Variance	0.09	0.72	0.06	0	1.96
Kurtosis	4.00	4.00	4.00	0	4.00
Skewness	2.00	2.00	2.00	0	2.00
Range	0.60	1.70	0.50	0	2.80
Minimum	0	0	0	0	0
Maximum	0.60	1.70	0.50	0	2.80
Sum	0.60	1.70	0.50	0	2.80
Count	4.00	4.00	4.00	4.00	4.00
Confidence Level (95.0 %)	0.48	1.35	0.40	0	2.23

The results of the distribution of MOAH (mg/kg) structured by the total contamination level (n-C_{10-50}) in experimental samples of freeze-dried milk (2) indicate a positive skewness in the distribution to the right (Table 6). This suggests that there are values in the dataset higher than average (4.25).

The distribution of MOAH structured by carbonic sub-fractions and the total contamination level (n-C_{10-50}) in experimental samples of unprocessed milk (1) and freeze-dried milk (2), did not reveal significant differences for n-C_{10-16} and n-C_{35-50} carbon sub-fraction ($p = 0.175$) (Figure 2).

Significant differences ($p = 0.008$) were observed between unprocessed milk and freeze-dried milk for the carbon sub-fractions n-C_{16-25}, n-C_{25-35} and n-C_{10-50} (Table 7).

Table 6. Results of the distribution of MOAH mg kg^{-1} structured by carbonic sub-fractions and total level of contamination (n-C_{10-50}) in experimental samples of freeze-dried milk

	n-C_{10-16}	**n-C_{16-25}**	**n-C_{25-35}**	**n-C_{35-50}**	**n-C_{10-50}**
Mean	0	2.96	1.28	0.01	4.25
Standard Error	0	0.40	0.08	0.01	0.45
Median	0	2.90	1.30	0	4.23
Standard Deviation	0	0.80	0.15	0.03	0.90
Sample Variance	0	0.64	0.02	0.00	0.80
Kurtosis	0	(0.92)	(3.90)	4.00	0.31
Skewness	0	0.38	(0.37)	2.00	0.15
Range	0	1.85	0.30	0.05	2.15
Minimum	0	2.10	1.10	0	3.20
Maximum	0	3.95	1.40	0.05	5.35
Sum	0	11.85	5.10	0.05	17.00
Count	4.00	4.00	4.00	4.00	4.00
Confidence Level (95.0 %)	0	1.27	0.24	0.04	1.43

Table 7. Differences in the distribution of structured MOAH by carbon sub-fractions and the total contamination level (n-C_{10-50}) in experimental samples of unprocessed milk and freeze-dried milk

	t	df	Significance		Mean Difference	95 % Confidence Interval of the Difference	
			One-Sided *p*	Two-Sided *p*		Lower	Upper
n-C_{10-16}	1.000	7	.175	.351	.0750	-.102	.252
n-C_{16-25}	3.078	7	.009	.018	1.69375	.3927	2.9948
n-C_{25-35}	3.076	7	.009	.018	.7000	.162	1.238
n-C_{35-50}	1.000	7	.175	.351	.00625	-.0085	.0210
n-C_{10-50}	3.200	7	.008	.015	2.47500	.6461	4.3039

T-test, significance level *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

Table 8. Cohen's d and Hedges' correction results for evaluating the effect of MOAH mg kg^{-1} between raw milk and experimental freeze-dried milk samples; 95 % confidence intervals for n-C_{10-16} carbon sub-fractions, n-C_{16-20}, n-C_{20-25}, n-C_{25-35}, n-C_{35-40}, n-C_{40-50} and total n-C_{10-50} contamination level

		Standardizer[a]	Point Estimate	95 % Confidence Interval	
				Lower	Upper
n-C_{10-16}	Cohen's d	.2121	.354	-.374	1.058
	Hedges' correction	.2388	.314	-.333	.940
n-C_{16-25}	Cohen's d	1.55619	1.088	.176	1.956
	Hedges' correction	1.75207	.967	.156	1.737
n-C_{25-35}	Cohen's d	.6437	1.088	.175	1.955
	Hedges' correction	.7247	.966	.156	1.736
n-C_{35-50}	Cohen's d	.01768	.354	-.374	1.058
	Hedges' correction	.01990	.314	-.333	.940
n-C_{10-50}	Cohen's d	2.18763	1.131	.205	2.013
	Hedges' correction	2.46298	1.005	.182	1.788

[a]The denominator used in estimating the effect sizes. Cohen's d uses the sample standard deviation. Hedges' correction uses the sample standard deviation, plus a **correction** factor.

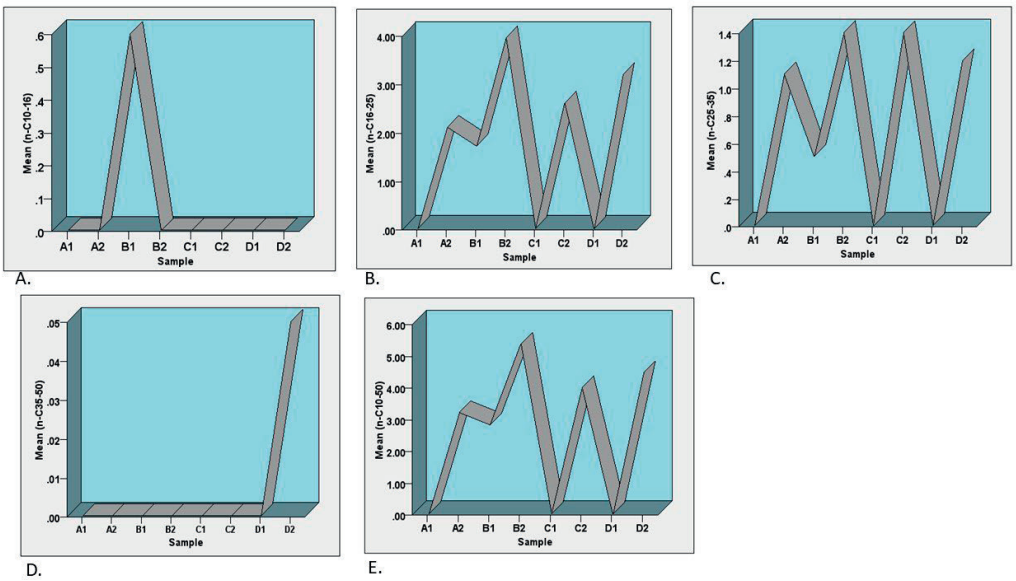

Figure 2. Distribution of structured MOAH media at different carbon sub-fractions A (n-C_{10-16}), B (n-C_{16-25}), C (n-C_{25-35}), D (n-C_{35-50}) and total contamination level E (n-C_{10-50}) in experimental raw milk samples (A_1, B_1, C_1, D_1) and freeze-dried milk samples (A_2, B_2, C_2, D_2)

For the total contamination level (n-C_{10-50}) of MOAH in raw milk (1) and freeze-dried milk (2), the point estimate of Cohen's d is 2.18763, and for the Hedges correction it is 2.46298. The 95 % confidence interval, for Cohen's d ranges from 0.205 to 2.013, and for the Hedges correction it ranges from 0.182 to 1.788 (Table 8). These results highlight a significant trend in the MOAH effect across the carbon fraction length scale, as indicated by the point estimates and confidence intervals for Cohen's d and Hedges correction.

This study underscores the potential for food contamination with MOH during food processing, particularly in the context of packaging. Different components of food contact material (FCM) can migrate into food, particularly following volatilization under the influence of temperature (Groh et al., 2021; Sonego et al., 2023).

Waxed paper (paraffin paper) which consists of cellulose and silicon, is an example of FCM that poses contaminations risks due to its chemical composition and production processes (Fellows, 2022). Previous research (Lorenzini et al., 2010), has shown significant migration of MOH from FCM to food products during processing.

Studies have also highlighted the transfer of MOH from packaging materials to food (Pack et al., 2020; Conchione et al., 2020; Pan Jing et al., 2021; Fengler & Gruber, 2022). However, concrete scientific guidance on this matter is lacking in current legislation, leaving the possibility and risk of MOH contamination through packaging a subject of debate. Due to their higher toxicity (EFSA et al., 2019), MOAH remains a priority for monitoring contaminants in the food chain.

CONCLUSIONS

The difference in contamination levels between raw and processed milk samples, and the variation in contamination levels following processing, were evaluated to assess the contamination risk associated with different milk processing methods. A T-test was applied to assess the differences in the distribution of MOSH and MOAH, structured by carbon sub-fractions and the total contamination level (n-C_{10-50}), in experimental samples of unprocessed milk (1) and lyophilized milk (2).

Milk is highly susceptible to exogenous contamination regardless of its form. The MOSH/MOAH concentrations measured in lyophilized milk (A_2; B_2; C_2; D_2) showed significantly higher values than liquid samples from the same category (A_1; B_1; C_1; D_1). These test results are essential for evaluating the impact of the freeze-drying process on these contaminants.

This study highlights the potential for food contamination with MOH during food processing. Various components of FCM can migrate into food products, particularly following volatilization under the temperature.

The findings indicate that milk processing operations significantly influenced the variation in contamination levels of freeze-dried milk. However, precise identification of the causes of contamination is challenging due to milk's increased susceptibility to contamination and the involvement of multiple factors.

ACKNOWLEDGEMENTS

The methodology, instrumentation conditions, and materials used in this study were established by the Food Chemistry group at the University of Udine (Prof. Sabrina Moret, PhD, PhD Chiara Conchione, PhD Erica Moret). This research is a collaboration between Control, Expertise, and Services Department from Iași University of Life Sciences, Romania (IULS), and Agrifood, Environmental, and Animal Sciences Department from University of Udine, Italy.

REFERENCES

Biedermann, M., & Grob, K. (2009). How “white” was the mineral oil in the contaminated Ukrainian sunflower oils? *European Journal of Lipid Science and Technology*, *111*, 313–319.

Biedermann, M., & Grob, K. (2012). 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. *Journal of Chromatography A, 1255*, 76–99.

Biedermann, M., & Grob, K. (2015). Comprehensive two-dimensional gas chromatography for characterizing mineral oils in foods and distinguishing them from synthetic hydrocarbons. *Journal of Chromatography A, 1375*, 146–153.

Biedermann-Brem, S., Kasprick, N., Simat, T., & Grob, K. (2012). Migration of polyolefin oligomeric saturated hydrocarbons (POSH) into food. *Food*

Additives and Contaminants: Part A Chemistry, Analysis, Control, Exposure and Risk Assessment, 29, 449–460.

Bratinova, S., & Hoekstra, S. (2019). Guidance on sampling, analysis and data reporting for the monitoring of mineral oil hydrocarbons in food and food contact materials. *OP European Union*, Luxemburg, JRC115694.

Brühl, L. (2016). Occurrence, determination, and assessment of mineral oils in oilseeds and vegetable oils. *European Journal of Lipid Science and Technology, 118*, 361–372.

Canavar, Ö., Kappenstein, O., & Luch, A. (2018). The analysis of saturated and aromatic mineral oil hydrocarbons in dry foods and from recycled paperboard packages by online HPLC-GC-FID. *Food Additives & Contaminants Part A, 35*, 2471-2481.

Ciobanu, M.M., Manoliu, D.R., Ciobotaru, M.C., Anchidin, B.G., Matei, M., Munteanu, M., Frunză, G., Murariu, O.C., Flocea, E.-I., & Boișteanu, P.-C. (2023). The Influence of Sensory Characteristics of Game Meat on Consumer Neuroperception: A Narrative Review. *Foods, 12*, 1341.

Conchione, C., Picon, C., Bortolomeazzi, R., & Moret, S. (2020). Hydrocarbon contaminants in pizza boxes from the Italian market. Food Packaging and Shelf Life, 25, 100535.

European Commission (EC) Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food. OJEU 2011, 10, 1–89.

European Food Safety Authority (EFSA), Arcella, D., Baert, K., & Binaglia M. (2019). 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), *EFSA Supporting Publications*, *16*.

European Food Safety Authority, (2012). EFSA Scientific opinion on mineral oil hydrocarbons in food. *EFSA Journal*, *10*, 1–185.

Fellows, P.J. (2022). Food Processing Technology, Principles and Practice, Chapter 23 – Packaging. *Woodhead Publishing Series Food Science & Nutrition Technology*, 633–709.

Fengler, R, & Gruber, L. (2022). Migration of mineral oil hydrocarbons from contaminated paperboard into the food simulants Tenax and Sorb-Star – A comparison. *Packaging Technology and Science*, 1–18.

Foodwatch International tests of various food products for their contamination by mineral oil hydrocarbons MOSH/MOAH. Project-report 2023. https://www.foodwatch.org/fileadmin/INT/mineral_oil/documents/2021-12-03_technical__minoil__project_report.pdf

Gharbi, I., Moret, S., Chaari, O., Issaoui, M., Conte, L.S., Lucci, P., & Hammami, M. (2017). Evaluation of hydrocarbon contaminants in olives and virgin olive oils from Tunisia. *Food Control*, *75*, 160–166.

Groh, K.J., Geueke, B., Martin, O., Maffini, M., & Muncke, J. (2021). Overview of intentionally used food contact chemicals and their hazards. Environment International, 150.

Hochegger, A., Moret, S., Geurt, L., Gude, T., Meitner, E., Mertens, B., O'Hagan, S., Poças, F., Simat, T., & Purcaro, G. (2021). Mineral oil risk assessment: knowledge gaps and roadmap. Outcome of a multi-stakeholders workshop. *Trends in Food Science & Technology*, 113, 151–166.

Lăpușneanu, D.M., Pop, I.M., Pop, C., Radu–Rusu, C.G., Musca, A., & Zaharia, R. (2021). Study on analysis of biological hazards associated with compound feed producing in relation on food safety. *Scientific Papers. Series D. Animal Science, LXIV*, 175–181.

Lorenzini, R., Fiselier, K., Biedermann, M., Barbanera, M., Braschi, I., & Grob, K. (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 Additives & Contaminants Part A Chemistry Analysis Control Exposure Risk Assessment, 27*, 1765–1774.

Matei, M., & Pop I.M. (2022). Monitoring of dairy farms to assess the potential level of pollution of animal feed and animal production, *Scientific Papers. Series D. Animal Science, LXV* (2), 129–136.

Matei, M., & Pop, I.M. (2023). Mineral oil hydrocarbons (MOH) analysis in animal feed: a characterization based on modern pollution. *Scientific Papers. Series D. Animal Science, LXVI* (1), 113–122.

Menegoz Ursol, L., Conchione, C., Peroni, D., Carretta, A., & Moret, S. (2023). A study on the impact of harvesting operations on the mineral oil contamination of olive oils. *Food Chemistry, 406*.

Menegoz Ursol, L., Conchione, C., Srbinovska, A., & Moret, S. (2022). Optimization and validation of microwave assisted saponification (MAS) followed by epoxidation for high-sensitivity determination of mineral oil aromatic hydrocarbons (MOAH) in extra virgin olive oil. *Food Chemistry, 370*, 130966.

Mierliță, D., Teușdea A.C., Matei, M., Pascal, C., Simeanu, D., & Pop, I.M. (2024). Effect of Dietary Incorporation of Hemp Seeds Alone or with Dried Fruit Pomace on Laying Hens' Performance and on Lipid Composition and Oxidation Status of Egg Yolks, *Animals, 14*, 750.

Moret, S., Populin, T., & Conte, L.S. (2009). La contaminazione degli oli vegetali con oli minerali. *Rivista Italiana delle Sostanze Grasse, LXXXVI*, 3–14.

Moret, S., Scolaro, M., Barp, L., Purcaro, G., & Conte, L.S. (2016). Microwave assisted saponification (MAS) followed by on-line liquid chromatography (LC)-gas chromatography (GC) for high-throughput and high-sensitivity determination of mineral oil in different cereal-based foodstuffs. *Food Chemistry, 196*, 50–57.

Nestola, M. (2022). Automated workflow utilizing saponification and improved epoxidation for the sensitive determination of mineral oil saturated and aromatic hydrocarbons in edible oils and fats. *Journal of Chromatography A, 1682*, 463523.

Pack, E.C., Jang, D.Y., Cha, M.G., Koo, Y.J., Kim, H.S., Yu, H.H., Park, S.C., Kim, Y.S., Lim, K.M., Lee, S.H., & Choi, D.W. (2020). Potential for short-term migration of mineral oil hydrocarbons from coated and uncoated food contact paper and board into a fatty

food simulant. *Food Additives & Contaminants Part A, 37*, 858–868.

Pan, J.J., Chen, Y.F., Zheng, J.G., Hu, C., Li, D., & Zhong, H.N. (2021). Migration of mineral oil hydrocarbons from food contact papers into food simulants and extraction from their raw materials. *Food Additives & Contaminants Part A, 38*, 870–880.

Purcaro, G., Barp, L., & Moret, S. (2016). Determination of hydrocarbon contamination in foods. A review. *Journal of Analytical Methods, 8*, 5755–5772.

Ruiz, J.L.H., Arrebola Liebanas, J., Martinez Vidal, J.L., Garrido Frenich, A., & Romero Gonzalez, R., (2021). Offline solid-phase extraction and separation of mineral oil saturated hydrocarbons and mineral oil aromatic hydrocarbons in edible oils, and analysis via GC with a flame ionization detector. *Foods, 10*, 1–12.

Singh, R. (2022). The consequences of Pollution on Livestock and Livestock Products, Environment & Livestock, https://www.pashudhanpraharee.com/the-consequences-of-pollution-on-livestock-and-livestock-products/, Martie 2023.

Sonego, E., Di Filippo, P., Riccardi, C., Pomata, D., Banno, A., Simonetti, G., & Buiarelli, F. (2023). Occurrence and migration study of chemicals from baking paper and aluminium foil. *Food Chemistry, 409*.

Srbinovska, A., Conchione, C., Celaj, F., Menegoz Ursol, L., & Moret, S. (2022). High sensitivity determination of mineral oils and olefin oligomers in cocoa powder and related packaging: Method validation and market survey. *Food Chemistry, 396*.

Srbinovska, A., Gasparotto, L., Conchione, C., Menegoz Ursol, L., Lambertini, F., Suman, M., & Moret, S. (2023). Mineral oil contamination in basil pesto from the Italian market: Ingredient contribution and market survey. *Journal of Food Composition and Analysis, 115*, 104914.

Van Heyst, A., Goscinny, S., Bel, S., Vandevijvere, S., Mertens, B., Elskens, M., & Van Hoeck, E. (2019). Dietary exposure of the Belgian population to mineral oil. *Food Additives & Contaminants* Part A, 37, 1–13.

Zhang, S., Liu, L., Li, B., Xie, Y., Ouyang, J., & Wu, Y. (2019). Concentrations of migrated mineral oil/polyolefin oligomeric saturated hydrocarbons (MOSH/POSH) in Chinese commercial milk powder products. *Food Additives & Contaminants Part A, 36*, 1261–1272.