



Time-Dependent Changes in Dioxin Levels in Van Herby Cheese During Brining

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Received Date: Apr, 2026; **Accepted Date:** June, 2026; **Publication Date:** July, 2026.

ABSTRACT

Purpose: Dioxins and dioxin-like compounds are persistent organic pollutants (POPs) that accumulate in foods of animal origin and pose potential risks to human health. This study aimed to compare the levels of polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/Fs), dioxin-like polychlorinated biphenyls (dl-PCBs), and indicator PCBs in fresh and long-term ripened Van herby cheese, and to assess the potential dietary exposure associated with consumption.

Materials and Methods: Eight fresh and eight ripened Van herby cheese samples were purchased from retail markets in Van, Türkiye. Fresh and ripened cheeses were obtained at different sampling periods and therefore represented two independent sample groups rather than paired samples monitored over time. Sample preparation and analysis were carried out in an accredited laboratory using EPA standard methods. PCDD/Fs, dl-PCBs, and indicator PCBs were determined by APGC-MS/MS with isotope dilution and ¹³C-labeled internal standards. Group comparisons were performed using Welch's independent-samples t-test.

Results: Contaminant levels were expressed as WHO toxic equivalency (WHO-TEQ, pg/g wet weight). PCDD/F levels slightly decreased from 0.092 to 0.086 pg WHO-TEQ/g, while dl-PCB levels decreased from 0.065 to 0.056 pg WHO-TEQ/g following long-term brining. A more pronounced reduction was observed in indicator PCBs, which decreased by approximately 47.5% (from 0.80 to 0.42 pg WHO-TEQ/g). The calculated tolerable daily intake (TDI) and tolerable weekly intake (TWI) values were well below the limits established by

Conclusion: Long-term brining does not increase dioxin levels in Van herby cheese and may contribute to a reduction in PCB concentrations. These findings indicate that traditional cheese processing does not pose an additional risk in terms of dioxin exposure and may influence the distribution of lipophilic contaminants.

Keywords: Van herby cheese, Dioxins, Dioxin-like polychlorinated biphenyls, APGC-MS/MS

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Introduction

Dioxins, including polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/Fs), together with dioxin-like polychlorinated biphenyls (dl-PCBs), are persistent organic pollutants (POPs) characterized by high toxicity, environmental persistence, and a strong tendency to bioaccumulate in living organisms World Health Organization. These compounds are mainly formed as by-products of industrial processes and combustion activities and are widely distributed in the environment (1). Due to their lipophilic nature, dioxins accumulate in fatty tissues and biomagnify along the food chain, making foods of animal origin the primary source of human exposure (2, 3).

Among dietary sources, dairy products represent an important route of exposure to dioxins and related compounds, particularly in populations with high consumption of milk and cheese (3). Cheese, in particular, may serve as a reservoir for these contaminants because of its relatively high fat content and concentration during processing (4). The European Food Safety Authority has reported that milk and dairy products contribute significantly to total dietary intake of PCDD/Fs and dl-PCBs, highlighting the importance of continuous monitoring of these contaminants in dairy products.

Traditional food processing methods, including salting (brining) and ripening, can significantly alter the physicochemical properties of cheese, such as moisture content, fat distribution, and matrix structure (5). These changes may influence the measured concentrations of lipophilic contaminants, either through redistribution within the matrix or through apparent concentration effects associated with compositional changes (4). However, despite the well-documented occurrence of dioxins in dairy products, there is limited information regarding the influence of traditional processing techniques-particularly long-term brining-on the behavior of these compounds.

Van herby cheese is a traditional dairy product produced mainly from sheep's milk and characterized by the addition of various aromatic herbs, and it is widely consumed in eastern Türkiye (6). This cheese is typically subjected to prolonged

brining and ripening, often lasting several months to one year, which may affect its chemical composition and contaminant distribution (7). These production and maturation conditions make Van herby cheese a suitable model for evaluating the effects of traditional processing on the distribution of persistent organic pollutants in dairy matrices.

The present study aimed to investigate the effect of brining duration on the levels of PCDD/Fs, dl-PCBs, and indicator PCBs in Van herby cheese. By comparing fresh and long-term brined samples, this study seeks to evaluate whether extended maturation influences the apparent concentrations of these contaminants and to contribute to a better understanding of processing-related changes in lipophilic pollutant behavior in dairy products.

Materials and Methods

Herby Cheese Samples

In this study, a total of 8 fresh Van herby cheese were taken January 2024 and 8 ripened Van herby cheese were taken October 2024 from Van retail markets in Van province. The age of the fresh cheeses was approx. 1 months and the age of the ripened cheeses was approx. 9 months. All cheese samples were stored at -20 °C until analysis.

Solvents, Standards and Quantification

All solvents (toluene, isooctane, dichloromethane, n-hexane, cyclohexane, 2-propanol and sodium sulfate) were purchased from Merck (Darmstadt, Germany). The standards EDF-7999, EDF-8999, EC-4986, EC-4987, EC-5179, EC-4187, EC-4188, EC-4058, ED-911, ED-996 and ED-907, all obtained from Cambridge Isotope Laboratories (CIL, USA), were used in the analysis. Dioxins (PCDD/PCDFs) were quantitatively determined using a 7-point calibration curve with a range of 0.02 to 20 pg/μL, and non-ortho, mono-ortho, and indicator PCBs were determined using an 8-point calibration curve with a range of 0.10 to 50 pg/μL. For PCDD/PCDF and dl-PCB analysis, ¹³C-labeled analogues of the target congeners were added to standards and samples prior to analysis. The mass spectrometer measured both isotopically labeled (¹³C) and unlabeled (¹²C) mass-to-charge (m/z) values of the analytes, and concentrations were determined based on the ratios of these two signals.

Extraction, Clean-up, and APGC-MS/MS Analysis Conditions

Analysis of PCDD/Fs and dl-PCBs in cheese samples was conducted at an accredited laboratory approved by the Republic of Türkiye Ministry of Agriculture and Forestry. Composite sampling was used: cheeses were homogenized, and 12.861 g (wet weight) of fresh herby cheese homogenate (total lipid content: 14.54%) and 11.005 g (wet weight) of ripened herby cheese (total lipid content: 12.54%) were analyzed as a representative sample. Analyses of PCDD/F, dl-PCB, and indicator PCB congeners were conducted using APGC-MS/MS with isotope dilution, employing ^{13}C -labeled internal standards. The analysis of PCDD/Fs was carried out using EPA Method 1613 Revision B, which employs isotope dilution combined with high-resolution gas chromatography and high-resolution mass spectrometry (8) to detect tetra- to octa-chlorinated dioxins and furans. EPA Method 1668B (9) was applied for the determination of PCB congeners, including non-ortho, mono-ortho, and indicator PCBs. A DB-5MS GC column ($250\ \mu\text{m} \times 0.25\ \mu\text{m} \times 60\ \text{m}$) was employed. High-purity helium gas (99.999%) was used. For the first fraction, injections were performed in Pulsed Splitless mode at $280\ ^\circ\text{C}$ using a Back-Inlet system. The oven temperature program included ramps from $100\ ^\circ\text{C}$ to $315\ ^\circ\text{C}$ with specific hold times to ensure optimal separation. Ionization settings included a corona pin current of $2.5\ \mu\text{A}$, cone voltage of $40\ \text{V}$, and gas flow of $230\ \text{L/hour}$. For the second fraction, a Cool On-Column inlet was used at $280\ ^\circ\text{C}$ (Ramped Temperature mode), with the oven initially at $110\ ^\circ\text{C}$, ramping to $300\ ^\circ\text{C}$ in programmed steps. The cone gas flow was maintained at $200\ \text{L/hour}$ to ensure consistent ionization conditions.

Statistical analysis

Data are presented as mean $\pm$ standard deviation (SD). Because fresh and ripened cheeses were obtained from different retail sample groups, between-group comparisons were performed using Welch's independent-samples t-test. Statistical significance was accepted at $p < 0.05$. Analyses were conducted for total PCDD/Fs, total dl-PCBs, and total indicator PCBs on both wet-weight and lipid-normalized bases. Indicator PCBs were evaluated as mass concentrations (ng/g wet weight

and ng/g fat) because toxic equivalency factors are not assigned to indicator PCB congeners.

Results

The concentrations of PCDD/Fs, dioxin-like PCBs (dl-PCBs), and indicator PCBs in fresh and ripened Van herby cheese samples are presented in Table 1 and Table 2. Changes of this parameters based on wet weight and fat content and Comparison of this parameters with EU limits are presented in Table 3 and Table 4.

On a wet weight basis, total PCDD/F levels showed a slight decrease from 0.092 to $0.086\ \text{pg WHO-TEQ/g}$, corresponding to a reduction of approximately 6.5%. Individual congener analysis revealed minor variations between fresh and ripened samples, with no consistent increasing or decreasing trend observed across all congeners. Similarly, dl-PCB levels decreased from 0.065 to $0.056\ \text{pg WHO-TEQ/g}$ (13.8%). Among individual dl-PCB congeners, PCB 126 contributed the most to the total TEQ in both sample groups, while slight fluctuations were observed in other congeners. A more pronounced decrease was observed in indicator PCB levels. The total indicator PCB concentration decreased from $0.80\ \text{ng/g wet weight}$ in fresh cheese to $0.42\ \text{ng/g wet weight}$ in ripened cheese, corresponding to a reduction of approximately 47.3%. This reduction was consistent across most indicator PCB congeners, particularly PCB 118, PCB 105, and PCB 156, which exhibited noticeable decreases following long-term.

To better interpret these findings, concentrations were further evaluated on a lipid-normalized basis. The fat content decreased from 14.54% in fresh cheese to 12.54% in aged cheese. After normalization, PCDD/F levels increased slightly from 0.633 to $0.686\ \text{pg WHO-TEQ/g fat}$, while dl-PCB levels remained essentially unchanged (0.447 vs. $0.447\ \text{pg WHO-TEQ/g fat}$). In contrast, indicator PCB levels remained substantially lower in ripened samples, decreasing from 5.50 to $3.35\ \text{ng /g fat}$ (39%). Overall, ripening did not result in an increase in dioxin (PCDD/F) levels. Instead, a slight decrease was observed for PCDD/Fs and dl-PCBs, while a substantial reduction was evident for indicator PCBs.

Table 1. PCDD/F levels in fresh and ripened Van Herby Cheese (Otlı peynir) (N=8).

	Fresh Van Herby Cheese					Ripened Van Herby Cheese					
	Congeners	Concentration (pg/g wet weight)	LOD	LOQ	WHO- TEF	TEQ (pg/g wet weight)	Concentration (pg/g wet weight)	LOD	LOQ	WHO- TEF	TEQ (pg/g wet weight)
Polychlorinated dibenzo-p-dioxins (PCDDs)	2,3,7,8-TCDF	0.0101±0.0014*	0.00043	0.0012	0.1	0.00101	0.0232±0.0033	0.00051	0.0014	0.1	0.00232
	1,2,3,7,8-PeCDF	0.0018±0.0003	0.00067	0.0018	0.03	0.00005	0.0023±0.0004	0.00087	0.0023	0.03	0.00007
	2,3,4,7,8-PeCDF	0.0251±0.0027	0.00053	0.0014	0.3	0.00753	0.0166±0.0018	0.00074	0.0020	0.3	0.00498
	1,2,3,4,7,8-HxCDF	0.0248±0.0036	0.00052	0.0014	0.1	0.00248	0.0295±0.0043	0.00068	0.0018	0.1	0.00295
	1,2,3,6,7,8-HxCDF	0.0015±0.0002	0.00057	0.0015	0.1	0.00015	0.0225±0.0036	0.00072	0.0019	0.1	0.00225
	2,3,4,6,7,8-HxCDF	0.0015±0.0002	0.00055	0.0015	0.1	0.00015	0.0085±0.0008	0.00075	0.0020	0.1	0.00085
	1,2,3,7,8,9-HxCDF	0.0359±0.0046	0.00077	0.0021	0.1	0.00359	0.0024±0.0003	0.00089	0.0024	0.1	0.00024
	1,2,3,4,6,7,8-HpCDF	0.0293±0.0029	0.00064	0.0017	0.01	0.00029	0.0316±0.0031	0.00068	0.0018	0.01	0.00032
	1,2,3,4,7,8,9-HpCDF	0.0020±0.0005	0.00075	0.0020	0.01	0.00002	0.0030±0.0007	0.00112	0.0030	0.01	0.00003
OCDF	0.0109±0.0013	0.00096	0.0026	0.0003	0.00000	0.0028±0.0003	0.00105	0.0028	0.0003	0.00000	
Polychlorinated dibenzofurans (PCDFs)	2,3,7,8-TCDD	0.0387±0.0049	0.00040	0.0011	1	0.03870	0.0526±0.0067	0.00065	0.0017	1	0.05260
	1,2,3,7,8-PeCDD	0.0266±0.0058	0.00058	0.0016	1	0.02660	0.0019±0.0004	0.00071	0.0019	1	0.00189
	1,2,3,4,7,8-HxCDD	0.0960±0.0124	0.00074	0.0020	0.1	0.00960	0.1730±0.0224	0.00064	0.0017	0.1	0.01730
	1,2,3,6,7,8-HxCDD	0.0123±0.0017	0.00052	0.0014	0.1	0.00123	0.0021±0.0003	0.00077	0.0021	0.1	0.00021
	1,2,3,7,8,9-HxCDD	0.0061±0.0011	0.00051	0.0014	0.1	0.00061	0.0020±0.0003	0.00076	0.0020	0.1	0.00020
Polychlorinated dibenzofurans (PCDFs)	1,2,3,4,6,7,8-HpCDD	0.0394±0.0078	0.00120	0.0032	0.01	0.00039	0.0037±0.0007	0.0014	0.0037	0.01	0.00004
	OCDD	0.0685±0.0042	0.00115	0.0031	0.0003	0.00002	0.1100±0.0068	0.001	0.0027	0.0003	0.00003
	Σ PCDD/F pg WHO-TEQ/g wet weight	0.092 ± 0.013					0.086±0.012				

*: Mean ± standard deviation; TCDF: Tetrachlorodibenzofuran; PeCDF: Pentachlorodibenzofuran; HxCDF: Hexachlorodibenzofuran; HpCDF: Heptachlorodibenzofuran; OCDF: Octachlorodibenzofuran; PeCDD: Pentachlorodibenzo-p-dioxin; HxCDD: Hexachlorodibenzo-p-dioxin; HpCDD: Heptachlorodibenzo-p-dioxin; OCDD: Octachlorodibenzo-p-dioxin. LOD: Limit of Detection; LOQ: Limit of Quantification; TEF: Toxic Equivalency Factor; TEQ=Σ(Concentration_i × TEF_i).

Table 2. Mono-ortho and non-ortho dioxin-like polychlorinated biphenyls (dl-PCBs) and indicator PCBs levels in fresh and ripened Van Herby Cheese (Otlı peynir) (N=8). |

	Fresh Van Herby Cheese					Ripened Van Herby Cheese				
Congeners	Concentration (pg/g wet weight)	LOD	LOQ	WHO-TEF	TEQ (pg/g wet weight)	Concentration (pg/g wet weight)	LOD	LOQ	WHO-TEF	TEQ (pg/g wet weight)
Mono-ortho dl-PCBs	PCB 123	8.19±1.31	3.07	8.19	0.00003	0.00025	0.00662	0.0177	0.00003	0.00028
	PCB 118	70.10±11.21	2.91	7.77	0.00003	0.00210	0.0074	0.0197	0.00003	0.00091
	PCB 114	9.43±1.43	3.54	9.43	0.00003	0.00028	0.00736	0.0196	0.00003	0.00024
	PCB 105	29.70±5.08	2.8	7.46	0.00003	0.00089	0.00764	0.0204	0.00003	0.00061
	PCB 167	11.40±2.16	1.34	4.26	0.00003	0.00034	0.00478	0.0127	0.00003	0.00022
	PCB 156	25.80±4.15	1.6	4.92	0.00003	0.00077	0.00467	0.0125	0.00003	0.00052
	PCB 157	6.19±1.02	1.84	3.62	0.00003	0.00019	0.00463	0.0123	0.00003	0.00023
	PCB 189	6.87±1.41	2.36	6.28	0.00003	0.00021	0.00426	0.0114	0.00003	0.00028
	PCB 81	0.12±0.038	0.00333	0.00887	0.0003	0.00004	0.000155	0.000413	0.0003	0.00000
Non-ortho dl-PCBs	PCB 77	0.31±0.093	0.00312	0.00833	0.0001	0.00003	0.000151	0.000403	0.0001	0.00003
	PCB 126	0.57±0.108	0.000768	0.00205	0.1	0.05690	0.00136	0.00363	0.1	0.04820
	PCB 169	0.11±0.013	0.000496	0.00132	0.03	0.00333	0.000112	0.000299	0.03	0.00474
	Σ PCDD/F + dl-PCB pg WHO-TEQ/g wet weight		0.065 ±0.005				0.056 ±0.005			
Congeners	Fresh Van Herby Cheese					Ripened Van Herby Cheese				
	Concentration (pg/g wet weight)	LOD	LOQ	Mass Concentration (ng/g wet weight)	Concentration (pg/g wet weight)	LOD	LOQ	Mass Concentration (ng/g wet weight)		
Indicator PCBs	PCB 28	47.9±13.17	3.38	9.01	0.0479	58.6±16.12	8.06	21.5	0.0586	
	PCB 52	44.6±8.61	1.39	3.71	0.0446	39.8±7.68	9.52	25.4	0.0398	
	PCB 101	79.4±22.07	1.91	5.09	0.0794	16.1±4.48	1.41	3.77	0.0161	
	PCB 138	199±37.61	1.34	3.58	0.276	37.8±7.14	2.06	5.48	0.0378	
	PCB 153	276±50.78	0.98	2.61	0.199	157±28.88	1.79	4.77	0.157	
	PCB 180	154±30.49	1.31	3.48	0.154	113±22.37	1.79	4.76	0.113	
Σ Indicator PCB ng/g wet weight		0.80 ±0.07				0.42 ±0.037				

Values are mean ± standard deviation. dl-PCBs were expressed as WHO-TEQ using WHO-TEF values. Indicator PCBs do not have WHO-TEF values and therefore were not expressed as TEQ. Sum values are expressed as mass concentrations.

Table 3. Change of PCDD/Fs, dioxin-like PCBs (dl-PCBs), and indicator PCBs concentrations of in fresh and ripened Van herby cheese samples based on wet weight and fat content.

Parameter	Fresh cheese	Ripened cheese	Change	p value
Σ PCDD/Fs (pg WHO-TEQ/g ww)	0.092 $\pm$ 0.013	0.086 $\pm$ 0.012	-6.5%	0.354
Σ PCDD/F+dl-PCB(pg WHO-TEQ/g ww)	0.065 $\pm$ 0.005	0.056 $\pm$ 0.005	-13.8%	0.003
Σ indicator PCBs (ng/g ww)	0.801 $\pm$ 0.070	0.422 $\pm$ 0.037	-47.3%	<0.001
Σ PCDD/Fs (pg WHO-TEQ/g fat)	0.633 $\pm$ 0.089	0.686 $\pm$ 0.096	+8.4%	0.272
Σ PCDD/F+dl-PCB (pg WHO-TEQ/g fat)	0.447 $\pm$ 0.034	0.447 $\pm$ 0.040	0.0%	1.000
Σ indicator PCBs (ng/g fat)	5.50 $\pm$ 0.48	3.35 $\pm$ 0.30	-39.1%	<0.001

Table 4. Comparison of PCDD/Fs, dioxin-like PCBs (dl-PCBs), and indicator PCBs concentrations in fresh and ripened Van herby cheese samples with EU limits.

Analyte	EU maximum level	Fresh cheese (lipid-normalized)	Ripened cheese (lipid-normalized)	Interpretation
Σ PCDD/Fs	1.75 pg WHO-TEQ/g fat	0.633 pg WHO-TEQ/g fat (36.2% of limit)	0.686 pg WHO-TEQ/g fat (39.2% of limit)	Below limit
Σ PCDD/Fs + dl-PCBs	2.00 pg WHO-TEQ/g fat	1.080 pg WHO-TEQ/g fat (54.0% of limit)	1.133 pg WHO-TEQ/g fat (56.7% of limit)	Below limit
Σ Indicator PCBs	40 ng/g fat	5.50 ng/g fat (13.8% of limit)	3.35 ng/g fat (8.4% of limit)	Below limit

These findings suggest that prolonged brining may influence PCB levels more markedly than PCDD/F concentrations in Van herby cheese.

Table 4 clearly shows that PCDD/Fs, dioxin-like PCBs (dl-PCBs), and indicator PCBs concentrations in fresh and ripened Van herby cheese samples are significantly below the maximum legal limits set by EU regulations, which supports the conclusion that Van Herby cheeses do not pose a significant risk in terms of PCDD/F and dl-PCB contamination.

Tolerable Daily Intake (TDI) was determined by taking the detected concentration of 0.065 ± 0.005 pg WHO-TEQ per gram for fresh van herby cheese, 0.056 ± 0.005 pg WHO-TEQ per gram for ripened van herby cheese and multiplying it by the average daily intake of cheese at 53.7 grams, and then dividing the result by the standard adult body weight of 70 kilograms (11,12) (For fresh van herby cheese $\text{TDI} = 0.065 \times 53.7/70 = 0.05$ pg WHO-TEQ/kg bw/day and for ripened van herby cheese $\text{TDI} = 0.056 \times 53.7/70 = 0.043$ pg WHO-TEQ/kg bw/day). This corresponds to ~17.2% and ~14.8% of the EFSA (2018) TDI limit of 0.29 pg WHO-TEQ/kg bw/day, indicating safe

exposure levels. Tolerable Weekly Intake (TWI) is the amount that can be safely consumed weekly. Using weekly cheese consumption of 214.8 g ($53.7 \text{ g/day} \times 7$), TWI was calculated as $0.065 \times 214.8/70 = 0.2$ pg WHO-TEQ/kg bw/week for fresh van herby cheese and $0.056 \times 214.8/70 = 0.17$ pg WHO-TEQ/kg bw/week for ripened van herby cheese. This is about 10% and 8.5% of the EFSA TWI limit of 2 pgWHO-TEQ/kg bw/week.

Discussion

The present study evaluated the effect of long-term brining on the levels of PCDD/Fs, dioxin-like PCBs (dl-PCBs), and indicator PCBs in Van herby cheese. The results clearly demonstrate that prolonged brining does not increase PCDD/F concentrations, while a marked reduction is observed in PCB levels, particularly indicator PCBs. Also, the levels of PCDD/Fs, PCDD/Fs + dl-PCBs, and indicator PCBs in van herby cheese samples were well below regulatory limits (MRL, TDI, TWI), indicating negligible risk to public health through consumption. The relative stability of PCDD/F levels observed in this study can be explained by their strong lipophilic character and high chemical stability.

PCDD/Fs are highly lipophilic, chemically stable, and strongly associated with lipid fractions in food matrices, resulting in limited mobility during food processing (World Health Organization). Numerous studies have reported that PCDD/F levels in dairy products are determined by environmental contamination and feed sources rather than post-production processing (4, 13, 14). Therefore, the absence of a significant increase in PCDD/F levels during prolonged brining supports the notion that traditional processing methods do not substantially alter these contaminants.

In contrast, a more pronounced decrease was observed in indicator PCB levels, suggesting that these compounds may be more susceptible to redistribution or loss during brining and ripening processes. Although PCBs are also lipophilic, their physicochemical properties differ from those of PCDD/Fs, particularly in terms of volatility and molecular mobility (1). PCBs exhibit slightly higher mobility compared to PCDD/Fs due to differences in molecular structure and physicochemical behavior, which may facilitate their migration within the food matrix or into surrounding media such as brine (15). This may facilitate their partial migration within the cheese matrix or into the surrounding brine solution over time. Moreover, studies on food contamination have shown that PCB concentrations can vary depending on processing conditions, including fat redistribution and matrix interactions (16-18). The fat content of cheese decreased from 14.54% in fresh samples to 12.54% in ripened samples. Considering the lipophilic nature of dioxins and PCBs, a reduction in fat content would typically be expected to result in an apparent increase in lipid-normalized contaminant concentrations. However, in the present study, a decrease—particularly in indicator PCB levels—was observed despite the reduction in fat content. This suggests that the observed decline cannot be solely attributed to compositional changes and may reflect actual redistribution or loss of PCB compounds during the brining and ripening process.

The moderate decrease observed in dl-PCB levels further supports the hypothesis that PCB congeners may be more affected by processing conditions than PCDD/Fs. It is also noteworthy that PCB 126 contributed substantially to the total TEQ in both fresh and aged samples, which

is consistent with previous reports highlighting its dominant role in dl-PCB toxicity profiles (15, 19).

The findings of this study have important implications for food safety and risk assessment. From a food safety perspective, the results suggest that the absence of an increase in PCDD/F levels suggests that long-term brining does not increase dioxin exposure risk through consumption of Van herby cheese. Moreover, the observed reduction in PCB levels may indicate a potential mitigating effect of traditional processing methods on certain contaminants. This highlights the importance of controlling contamination sources at the agricultural level rather than relying on processing methods for contaminant reduction.

Conclusion

The present study demonstrates that long-term brining does not lead to an increase in PCDD/F levels in Van herby cheese, while a noticeable reduction is observed in PCB concentrations, particularly indicator PCBs. These findings suggest that traditional brining and ripening processes do not pose an additional risk in terms of dioxin exposure. Furthermore, the detected contaminant levels were significantly below the regulatory limits established by the European Union, and the calculated TDI and TWI values indicate no significant risk to human health from the consumption of Van herby cheese.

Overall, this study provides important insights into the behavior of persistent organic pollutants during traditional cheese processing and highlights the need for further studies focusing on lipid-normalized assessments and compositional changes.

This study has some limitations. First, fresh and ripened cheeses were purchased from retail markets at different times and did not represent paired samples from the same production batches; therefore, the findings should not be interpreted as direct longitudinal changes within the same cheese. Second, brine samples were not analyzed, so migration of contaminants from cheese into brine could not be confirmed. Third, the sample size was limited to eight samples per group, and data were obtained from a geographically restricted retail market setting. Fourth, potential differences in milk source, fat

content, herb composition, production practices, and storage conditions may have contributed to between-group variation. These limitations were considered when interpreting the results, and the conclusions were revised to avoid causal statements beyond the data.

Declaration

Ethical Approval Statement

This study does not involve human participants or experimental animals. Therefore, ethical approval is not required in accordance with national and institutional guidelines.

Author Contributions Statement

NAK: Conceptualization, Laboratory analysis coordination, data curation, writing review and editing, statistical analysis

UMY: Conceptualization, methodology, laboratory analysis coordination, data curation, statistical analysis, risk assessment, writing original draft preparation, writing review and editing, project administration

NAT: Laboratory analysis coordination

MB: Sample collection and field coordination

FA: Laboratory analysis coordination

All authors have read and approved the final version of the manuscript.

Funding Statement

The authors declare that no financial support or funding was received for this study.

Conflicts of Interest Statement

The authors declare that there are no conflicts of interest regarding this study.

Declaration on the Use of AI

The authors declare that no artificial intelligence (AI)-assisted technologies were used in the design, analysis, or writing of this manuscript.

Acknowledgement

None.

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