


## Article

# Presence and Distribution of Sun-Blocking Agents in the Danube River: Implications for Aquatic Ecosystem Health

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**Abstract:** Organic sun-blocking agents are recognized as emerging pollutants of the aquatic environment. The consequences and effects of these harmful substances on aquatic organisms are not fully understood, particularly in the context of Romania. This study examines the presence and environmental hazards posed by ten UV filters found in the Danube River. Six of the ten targeted contaminants were detected in surface water samples: 2-hydroxy-4-methoxy-benzophenone (BP-3), 2,4-dihydroxy benzophenone (BP-1), 2,3,4-trihydroxy benzophenone (234HBP), 2,2'-dihydroxy-4-methoxy-benzophenone (BP-8), 2-hydroxy-4-methoxy-4'-methyl-benzophenone (BP-10), and benzyl salicylate (BS). In sediment samples, seven of the ten compounds were identified: BS, BP-3, 4-hydroxybenzophenone (4HBP), BP-1, 234HBP, 2,2',4,4'-tetrahydroxybenzophenone (BP-2), and BP-10, with different frequencies. The correlation coefficients showed strong relationships between benzophenone derivatives in surface water samples and a common contamination source for BP-3 and BS in sediment samples. In the assessment, the risk quotient (RQ) for BP-1 in fish was determined to be 1.81. Similarly, RQ values for BP-3 were calculated as 1.24 for *Daphnia magna* and 57 for fish, suggesting that these UV filters could potentially present substantial hazards to the aquatic ecosystem. The study findings indicate that additional research is needed to investigate the presence and potential risks of organic sun-blocking agents in surface waters, with a particular emphasis on densely populated areas in Romania.



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**Keywords:** aquatic environment; endocrine disruptors; occurrence monitoring; risk quotients; UV-filters

## 1. Introduction

Over the last few years, awareness of the risks posed by solar radiation to human health has led to a growing demand for cosmetics and personal care products containing organic UV filters (UVFs). These compounds are used to absorb ultraviolet A (UVA) and ultraviolet B (UVB) radiation, protecting the skin, lips, and hair from damage [1]. However, the presence of UVFs in aquatic ecosystems, such as rivers and lakes, raises concerns about their potential impact on aquatic organisms. Limited studies have been conducted on the presence of UVFs in surface water and sediment samples in Romania, prompting the need for a comprehensive investigation along the Danube River.

Organic sun-blocking agents are active compounds used in creams and cosmetics to prevent skin, lip, and hair damage from sunlight exposure, but they are also used in various other industrial products, such as plastics, adhesives, rubber, textile, and polymers, to improve their stability against UV light. Because each of these compounds has a limited spectral absorption band, they are usually combined. A maximum of 45 UVFs are currently allowed in cosmetic products, with maximum additive limits ranging from 2 to 20% [2,3].

As per the European Union Cosmetics Directive, the register of commercial cosmetic products includes 28 UV filters approved for use as active elements in cosmetics [4].

These UV filters encompass benzophenones, such as 2-hydroxy-4-methoxy-benzophenone (BP-3), 2,4-dihydroxybenzophenone (BP-1), 2,2',4,4'-tetrahydroxybenzophenone (BP-2), 2,2'-dihydroxy-4-methoxy-benzophenone (BP-8), 2-hydroxy-4-methoxy-4'-methyl-benzophenone (BP-10), 4-hydroxybenzophenone (4HBP), and 2,3,4-trihydroxybenzophenone (234HBP); derivatives of salicylate include ethyl hexyl salicylate (EHS), benzyl salicylate (BS), and homosalate (HS), which are some of the most common UVFs used in commercial products [5,6]. UVFs can enter the aquatic environment through multiple pathways, including indirect routes like the release of effluents from wastewater treatment plants into natural water bodies, as well as direct routes from recreational activities. Various studies conducted globally have documented the presence of UVFs in rivers, lakes, seas, oceans, and sediments at varying concentration levels, ranging from nanograms per liter (ng/L) to micrograms per liter ( $\mu\text{g/L}$ ) in water matrices and nanograms per gram (ng/g) in sediments [7–14].

The widespread use of these products has raised concerns about the side effects of organic UV filters. Many studies have shown that both benzophenone and salicylate derivatives may have endocrine-disrupting properties [15–18], with benzophenone derivatives generating complex hormonal activities in organisms [13,17].

The best-known compound in this class, BP-3, exhibits both antiestrogenic and antiandrogenic activities and also moderate estrogenic activities [19,20]. Moreover, BP-3 is transformed into different (bio)degradation products in aquatic environments, such as BP-1, BP-2, and 234HBP. While BP-1 showed stronger estrogenic activity than BP-3, BP-2, and 234HBP showed lower activity compared to the parent compound [17].

To the best of our understanding, information regarding the presence of sun-blocking agents in surface water and sediment samples in Romania remains limited. Recent studies identified BP-1, BP-8, and 4HBP as the predominant UVFs in five major rivers in Romania (Bahlui, Siret, Ialomita, Somes, and Dambovita), with concentrations reaching up to 206 ng/L, 108 ng/L, and 159 ng/L, respectively. These findings have raised significant concerns about the potential risks posed to aquatic species [9]. Given the negative properties of these organic contaminants and the potential effects they can have on aquatic organisms, the objective of this study was to establish the concentration levels at which these compounds are found along the largest river in Romania, the Danube River, and their repercussions on aquatic ecosystems. Given the environmental risks associated with UVFs and the lack of extensive data on their presence in aquatic ecosystems in Romania, there is a clear need for a thorough study to enhance environmental stewardship both in Romania and globally.

In this study, we aim to establish the concentration levels of UVFs in the Danube River and assess their ecological implications on aquatic ecosystems. The identification of UVFs in surface water and sediment samples will provide valuable insights into the environmental risks associated with these compounds. By conducting an ecological risk assessment, we can better understand the potential effects of UVFs on aquatic organisms and improve environmental stewardship practices in Romania and globally. This study introduces a novel approach to investigating organic UVFs in the Danube River, focusing on the utilization of a rapid and sensitive LC-MS/MS method. The method allows for a quick elution time of only 12 min, enabling efficient analysis of UVF compounds in surface water and sediment samples. The method's sensitivity is demonstrated by the low limits of quantification (LOQs), ranging from 0.1 to 0.69 ng/L for surface water samples and 0.21 to 0.79 ng/g dry weight for sediment samples. These results showcase the advancement in analytical techniques employed in this study, providing detailed insights into the occurrence and levels of UVF contaminants in the Danube River ecosystem. The combination of the fast elution time and high sensitivity of the LC-MS/MS method contributes significantly to the innovation and robustness of this research, offering valuable contributions to the field of environmental monitoring and risk assessment related to UVFs in aquatic environments.

## 2. Materials and Methods

### 2.1. Study Area and Sample Collection

Surface water and sediment samples were collected along the Danube River from five sampling points in September 2020: Bazias (S1, N 44.8166, E 21.3887), Eselnita (S2, N 44.6817, E 22.3559), Gruia (S3, N 44.2630, E 22.6894), Zimnicea (S4, N 43.6437, E 25.3325), and Giurgiu (S5, N 43.8862, E 25.9907). During the sampling process, 2 L of surface water was gathered from specific locations using clean glass containers. These containers were first rinsed with ultra-pure water and then rinsed with the sample water before obtaining the samples. Sediment samples were collected using a stainless-steel device, and the top 10 cm of sediment was specifically targeted for sampling. The samples were then placed in glass jars (previously cleaned with acetone) and stored at  $-4\text{ }^{\circ}\text{C}$  until sample preparation.

### 2.2. Chemicals

BP-3 (2-hydroxy-4-methoxy-benzophenone), 4HBP (4-hydroxybenzophenone), 234HBP (2,3,4-trihydroxybenzophenone), BP-1 (2,4-dihydroxybenzophenone), BP-2 (2,2',4,4'-tetrahydroxybenzophenone), BP-8 (2,2'-dihydroxy-4-methoxy-benzophenone), BP-10 (2-hydroxy-4-methoxy-4'-methyl-benzophenone), BS (benzyl salicylate), ES (ethylhexyl salicylate), and HS (homosalate) were provided by Sigma-Aldrich (Darmstadt, Germany). Benzophenone-13C (13C-BP) was also sourced from Sigma-Aldrich (Darmstadt, Germany). Additional details regarding the physicochemical characteristics can be found in Tables S1 and S2. Methanol and formic acid (FA) were purchased from Merck (Darmstadt, Germany). High-quality water was generated on site using a water purification system (Millipore Milli-Q, Merck-Millipore, Darmstadt, Germany). Strata C18-U<sup>®</sup> (500 mg, 6 mL) solid-phase extraction (SPE) cartridges were acquired from Phenomenex (Torrance, CA, USA), and Silicagel was procured from Sigma Aldrich (Darmstadt, Germany).

### 2.3. Sample Preparation

All surface water samples were initially filtered to remove large particles using qualitative filter paper. Following this, 200 mL of every surface water sample was combined with 1 mL of 50  $\mu\text{g/L}$  internal standard  $^{13}\text{C}$ -BP. The solid-phase extraction (SPE) procedure followed a previously reported method [9]. Prior to passing the surface water samples through the cartridges, Strata-C18 SPE cartridges were pre-conditioned with 10 mL of methanol and 10 mL of ultra-pure water. After eluting the samples, the cartridges were washed with 5 mL of deionized water and subsequently dried for 30 min under vacuum. The organic UVF compounds were then eluted using two rounds of 10 mL methanol into 40 mL conical tubes. The extracted samples were evaporated until dry, then reconstituted in 1 mL of methanol and placed into sample vials for LC-MS/MS analysis. For the sediment samples, approximately 0.5 g of freeze-dried and homogenized sediment was spiked with 50  $\mu\text{g/L}$  of internal standard  $^{13}\text{C}$ -BP in a 25 mL glass tube, and 10 mL of methanol was added to the mixture, followed by ultrasonication for 30 min at room temperature. After ultrasonication, the sample was centrifuged at 3000 rpm for 10 min. The ultrasonic-assisted extraction process was repeated twice, and the resulting organic extracts were combined. The organic mixture was subsequently purified on silica gel, transferred into 40 mL conical tubes, evaporated using a nitrogen stream, and reconstituted in 1 mL of methanol prior to LC-MS/MS analysis.

### 2.4. LC-MS/MS Analysis

The analysis was performed utilizing an LC-MS/MS system comprising a 1260-liquid chromatograph connected to a 6410 triple-quadrupole mass spectrometer, both produced by Agilent in Germany. The instrument was furnished with an electrospray ionization (ESI) source and employed a C18 chromatographic column (specifically, a Luna C18 column:  $150 \times 2.0\text{ mm}$ , 3.0 mm, from Phenomenex) that was operated at a temperature of  $30\text{ }^{\circ}\text{C}$ . The mobile phase employed in the analysis consisted of water with 0.15% formic acid (A) and methanol (B). The separation of compounds was achieved using a flow rate of 0.2 mL/min

in a gradient elution program. The solvent gradient was initiated with 55% of solvent B and was increased to 95% B over 3 min, where it was held constant for an additional 9 min. Following a 12 min interval, the mobile-phase composition reverted to 55% B and remained under the original conditions for a duration of 10 min. The operational parameters of the MS detector and ESI source can be found in Table S3.

### 2.5. Total Organic Carbon (TOC) Analysis

TOC analysis was conducted using a TOC-LCPN instrument featuring an infrared detector obtained from Shimadzu in Japan. The TOC values were determined by combusting the samples at 720 °C in a synthetic air flow. Each sample was measured in duplicate.

### 2.6. Environmental Risk Assessment

Risk quotients (RQs) for each UVFs were evaluated according to similar studies reported in the literature [21–23] and the European Guide (CVMP, 2016) using Equations (1) and (2):

$$RQ = MEC/PNEC \quad (1)$$

$$PNEC = NOEC/AF \quad (2)$$

The calculation of risk quotients (RQs) involved dividing the Measured Environmental Concentrations (MECs) identified in this investigation by the Predicted-No-Effect Concentrations (PNECs). PNECs, which are derived by dividing the No-Effect Concentrations (NOECs) by a standard Assessment Factor (AF), were utilized in this process. An Assessment Factor (AF) of 100 was determined to demonstrate the chronic toxicity of the specific compounds under investigation. Risk quotient (RQ) categorization was determined using a risk-ranking system, with classifications including “Unlikely to pose risk” for  $RQ < 0.01$ ; “Low risk” for  $0.01 < RQ < 0.1$ ; “Medium risk” for  $0.1 < RQ < 1$ ; and “High risk” for  $RQ > 1$  [24].

### 2.7. Quality Assurance and Quality Control

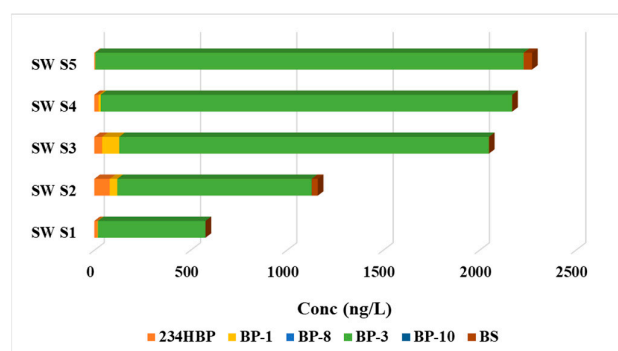
The effectiveness of the LC-MS/MS technique was examined through an evaluation of its linearity, sensitivity, accuracy, recovery, and the impact of matrix effects on both surface water and sediment samples. The mass spectrometer response was assessed across a concentration range of 0.1–100 µg/L by employing a labeled standard at a concentration of 50 µg/L. This testing resulted in linear regression data with correlation coefficients greater than 0.998 for all targeted compounds. Accuracy was determined by comparing the average values of the determined concentration with the spiked concentration. The method sensitivity was determined by quantifying the limit of detection (LOD) and limit of quantification (LOQ) values, which were calculated utilizing signal-to-noise ratios (S/N) of 3× and 10×, respectively. Analyte recovery was calculated considering the entire analytical process. Matrix effects were assessed to understand how components of the co-extracted matrix could influence the ionization of the analyte in the electrospray ionization source, potentially leading to signal suppression or enhancement. Information on accuracy, limit of detection (LOD), limit of quantification (LOQ), recovery rates, and matrix effects for surface water and sediment samples is available in Tables S4 and S5 [25].

## 3. Results

### 3.1. Occurrence of Sun-Blocking Agents in Surface Water

In the surface water samples analyzed, six organic UV filters (UVFs) were detected. The concentrations of each organic compound at various sampling points are detailed in Table S6, while the total amount of organic UVFs in the surface water samples is depicted in Figure 1. Among the contaminants identified, BP-3, BP-1, and 234HBP were the most frequently detected, while BP-8 and BS were only found in samples collected from two out of the five sampling locations. Notably, BP-3 exhibited the highest concentrations, ranging from 558 to 2220 ng/L. This high prevalence of BP-3 is attributed

to its widespread use in personal care products for sun protection [26]. Additionally, elevated concentrations of the degradation products 234HBP and BP-1 were observed, reaching up to 80 ng/L and 89 ng/L, respectively. Furthermore, the salicylate derivative BS was detected in samples SW S2 and SW S5 at concentrations of 32 ng/L and 44 ng/L, respectively. The cumulative concentration of organic UV filters in the surface water samples ranged from 780 to 2270 ng/L, displaying a consistent increase downstream from SW S1 to SW S5 (Table S8). Comparisons with previous studies conducted in Romania revealed lower concentrations of BP-3, ranging from 3.0 to 52 ng/L, while similar levels were reported for 234HBP and BP-1 in major rivers [10]. To assess the pollution status of the Danube River accurately, concentrations of organic UVFs were compared to those reported in significant rivers worldwide.



**Figure 1.** The total amount of UVFs determined in surface water samples.

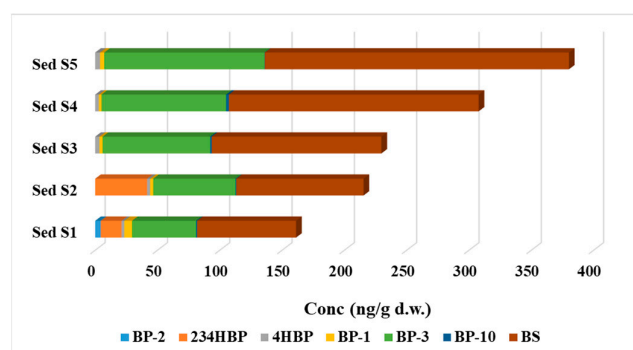
It is worth noting that the samples were collected in September, marking the end of the sunbathing season for European tourists. This timing is significant as it captures the potential impact of UV filters used during the peak sun exposure period in the region. The detection of UVFs in the samples collected at this time provides insight into the persistence of these compounds in the aquatic environment, even after the sunbathing season has concluded. This information is valuable for understanding the environmental implications of UV filter contamination in the Danube River, particularly following periods of heightened sun exposure and UV filter usage by tourists in the area.

The levels of BP-3 detected in this research were significantly elevated compared to concentrations documented in previous studies conducted in Thailand (maximum 116 ng/L) [27], Spain (maximum 37.8 ng/L) [21], the Czech Republic (maximum 67 ng/L) [28], and Taiwan (maximum 15.4 ng/L) [29]. However, they were lower in comparison to concentrations measured in China (maximum  $5.01 \times 10^3$  ng/L) [30]. The difference in concentrations emphasizes the varying levels of organic UV filters in different regions globally and emphasizes the importance of ongoing monitoring and evaluation of these pollutants in aquatic settings.

### 3.2. Occurrence of Sun-Blocking Agents in Sediment

In the sediment samples analyzed, BS and BP-3 were the predominant organic UV filters (UVFs), with concentrations ranging from 79 to 244 ng/g dry weight (d.w.) for BS and 52 to 129 ng/g d.w. for BP-3 (Table S7). The escalating contamination levels of UV filters detected along the Danube River from the entry point in Romania to the discharge point into the Black Sea suggest a continuous buildup of these pollutants throughout the Romanian territory along the river course (Figure 2). Notably, significant concentrations of 234HBP were detected in samples from the first two locations, with values of 17 and 42 ng/g d.w., respectively. The presence of 234HBP in sediment samples may be attributed to the environmental degradation of BP-3, highlighting the potential transformation of BP-3 into degradation products in the aquatic environment. This finding is corroborated by the lower levels of BP-3 in comparison to 234HBP in water samples, indicating the potential for BP-3 to break down more readily in aquatic settings. The lack

of 234HBP in subsequent samples could be due to the dilution effects resulting from the overall volume of the river.



**Figure 2.** The total amount of UVFs determined in sediment samples.

Significantly, BP-2 was solely detected in the initial sediment sample, whereas the concentrations of 4HBP and BP-1 remained consistent throughout the Danube River, measuring below 6.5 ng/g d.w. BP-8, HS, and ES were not evident in any of the sediment samples examined. Comparisons with studies conducted globally revealed that the concentrations of BP-3 in the Danube River sediment samples were higher than those reported in Hawaii (nd—4.43 ng/g d.w.) [31], China (nd—4.0 ng/g d.w.) [32], and Spain (<LOD—33 ng/g d.w.) [33]. While BS and ES were not observed in the sediment samples of the Danube River, they have been detected in sediments from various locations, such as Hawaii (ranging from not detected to 38.84 ng/g d.w. for HS and not detected to 13.3 ng/g d.w. for ES) [31], China (with concentrations of up to 10.7 ng/g d.w. for HS and up to 13.7 ng/g d.w. for ES) [32], and Spain (reaching levels of up to 149 ng/g d.w. for HS and up to 609 ng/g d.w. for ES) [33]. These findings highlight the variability in UVF concentrations across different regions and emphasize the importance of ongoing monitoring and assessment of these contaminants in sediment environments.

### 3.3. Distribution Pattern of Sun-Blocking Agents in Surface Water and Sediment

Distinct patterns of organic UV filter (UVFs) contamination were identified through the examination of surface water and sediment samples. Surface water samples were primarily contaminated by BP-3, 234HBP, and BP-1, while sediment samples were dominated by more hydrophobic UVFs with higher partition coefficients, such as BS and BP-3. The differences in the types of contaminants found in surface water and sediment indicate varying sources and behavior of UVFs in aquatic environments.

To investigate the connections between individual UVFs in surface water and sediment samples, a Pearson correlation analysis was performed. Positive correlations were found to be significant between BP-3 and 234HBP, BP-1 and 234HBP, and BP-10 and BP-8 in surface water samples (Table 1). Conversely, significant negative correlations were found between BP-3 and BP-10, and 4HBP and BS. These correlations suggest potential degradation pathways or transformation processes among the UVFs in surface water, indicating the influence of biotic and abiotic factors on the behavior and interactions of these compounds.

In the analysis of sediment samples, a robust negative correlation was detected between 234HBP and 4HBP, while notable positive correlations were identified between BP-3 and 4HBP, BP-3 and BS, and BS and 4HBP (Table 2). These correlations imply potential degradation relationships between these compounds, with 4HBP possibly acting as a degradation product for BP-3, 234HBP, and BS. The close relationship between BP-3 and BS in sediment samples may also suggest a common source of contamination for these UVFs in the sediment environment.

**Table 1.** Pearson correlation coefficients of UVFs in surface water samples.

		SW ( <i>n</i> = 5)					
Analytes		4HBP	BP-1	BP-8	BP-3	BP-10	BS
234HBP	Pearson Corr.	0.205	0.900	−0.112	0.867	0.100	−0.224
	<i>p</i> -value	0.7406	0.037	0.858	0.052	0.873	0.718
4HBP	Pearson Corr.		0.564	0.459	−0.051	0.154	−0.860
	<i>p</i> -value		0.322	0.437	0.935	0.805	0.061
BP-1	Pearson Corr.			0.112	−0.800	0.200	−0.447
	<i>p</i> -value			0.858	0.624	0.747	0.450
BP-8	Pearson Corr.				−0.671	0.894	−0.625
	<i>p</i> -value				0.215	0.041	0.260
BP-3	Pearson Corr.					−0.800	0.447
	<i>p</i> -value					0.044	0.450
BP-10	Pearson Corr.						−0.335
	<i>p</i> -value						0.417

Note: the green color indicates the strong correlations calculated between the compounds.

**Table 2.** Pearson correlation coefficients of UVFs in sediment samples.

		SED ( <i>n</i> = 5)				
Analytes		4HBP	BP-1	BP-3	BP-10	BS
234THBP	Pearson Corr.	−0.783	0.112	0.619	−0.335	−0.783
	<i>p</i> -value	0.017	0.858	0.118	0.581	0.118
4HBP	Pearson Corr.		−0.100	0.900	−0.200	0.900
	<i>p</i> -value		0.873	0.037	0.747	0.037
BP-1	Pearson Corr.			−0.300	−0.500	−0.300
	<i>p</i> -value			0.624	0.391	0.624
BP-3	Pearson Corr.				−0.100	1.000
	<i>p</i> -value				0.873	0.000
BP-10	Pearson Corr.					−0.100
	<i>p</i> -value					0.873

Note: the green color indicates the strong correlations calculated between the compounds.

Overall, the distinct correlations observed in surface water and sediment samples indicate different processes governing the behavior and fate of UVFs in these environments. The lack of significant correlations for some UVFs further underscores the presence of distinct contamination sources for these compounds. Continued monitoring and research are essential to better understand the sources, fate, and potential risks associated with UVFs in surface water and sediment environments [11].

### 3.4. Correlation with TOC

The total organic carbon (TOC) content in surface water and sediment samples was determined to range from 2.4 to 3.8 mg/L and from 36 to 212 mg/kg, respectively (Table S8). To investigate the relationship between TOC content and the total amount of organic UV filters (UVFs) present in the samples, Pearson correlation analysis was conducted. Surprisingly, no significant correlation was observed between TOC content and the UVF amount in both surface water and sediment samples (Table S9). This finding is consistent with similar results reported in other studies, such as Apel et al., (2018) [34].

The lack of correlation between TOC content and the UVF amount may be attributed to several factors. First, UVFs can partition through different phases in aquatic ecosystems, including water and sediment [34]. This behavior may result in the independent distribution of UVFs and TOC, leading to a lack of correlation between the two parameters. Additionally, the relatively low concentrations of UVFs compared to other unknown organic compounds present in the samples could contribute to the absence of a significant correlation. These unknown compounds may also play a substantial role in the TOC content, overshadowing the contribution of UVFs.

The complexity of the aquatic environment, including the presence of various organic compounds and the dynamic interactions between different phases, can further complicate the relationship between TOC content and the UVF amount. The partitioning behavior of UVFs and their interactions with other organic compounds may vary based on factors such as environmental conditions and the specific characteristics of the contaminants.

The lack of correlation between TOC content and the UVF amount highlights the intricate nature of organic compound distribution and dynamics in aquatic systems. Additional research and thorough investigations are required to clarify the factors that impact the occurrence and behavior of UVFs concerning TOC content in both surface water and sediment samples.

### 3.5. Potential Environmental Risk for Individual UVFs in Surface Water

The evaluation of the environmental risks associated with specific organic UV filters (UVFs) by utilizing short-term toxicity data and maximum concentration levels found in surface water samples indicated diverse levels of risk for aquatic organisms. The estimated risk quotients (RQs) were calculated for two aquatic species (fish and *Daphnia magna*), based on the maximum UVF concentrations detected in the studied area. The results, as detailed in Table 3, indicated that 234HBP and BS pose minimal risk to *Daphnia magna*, with RQ values below levels of concern. These findings are consistent with previous studies, which also reported low ecological risk associated with these compounds [11].

**Table 3.** The environmental risk (ER) assessment of organic UV filters in surface water.

Analytes	Toxicity	Species	NOEC (ng/L)	MEC (ng/L)	RQ	ER
234THBP	Chronic	<i>Daphnia magna</i>	29,400,000 <sup>b</sup>	80	0.0003	It is unlikely to pose a risk
4HBP	Chronic	-	-	2	-	-
BP-1	Chronic	Fish	4919 <sup>a</sup>	89	1.81	High risk
BP-3	Chronic	<i>Daphnia magna</i>	180,000 <sup>b</sup>	2224	1.24	High risk
	Chronic	Fish	3900 <sup>b</sup>	2224	57.0	High risk
BP-10	Chronic	-	-	1	-	-
BS	Chronic	<i>Daphnia magna</i>	894,000 <sup>b</sup>	43	0.005	It is unlikely to pose a risk

Note: <sup>a</sup> [35]; <sup>b</sup> <https://echa.europa.eu> (accessed on 11 February 2021).

In contrast, the RQ values for BP-1 and BP-3 in the studied area indicated a potential environmental risk for aquatic species. Specifically, RQ values for BP-1 in fish and BP-3 in *Daphnia magna* exceeded a value of 1, suggesting a moderate to high risk for these species. Of particular concern was BP-3, with an RQ value for fish exceeding 50, indicating a significant risk to this aquatic species. Similar findings regarding the ecological risk posed by BP-3 have been reported in the literature [22].

The findings of the environmental risk assessment emphasize the significance of assessing the potential effects of UVFs on aquatic ecosystems. The high RQ values for BP-1 and BP-3 highlight the need for further monitoring and research to better understand the ecological implications of these compounds. Mitigation measures may be necessary to reduce the risk posed by these UVFs to aquatic species and overall ecosystem health.

The results of the risk assessment highlight the crucial need to take into account both the concentration levels of UVFs in the environment and their toxicological impacts on aquatic organisms. Continued research and monitoring efforts are essential to assess and mitigate the potential risks associated with UVF contamination in aquatic ecosystems.

## 4. Discussion

The results of this study shed light on the presence of six organic UV filters (UVFs) in surface water and sediment samples collected along the Danube River in Romania. The detection of BP-3, BP-1, and 234HBP in the surface water samples at elevated concentrations indicates widespread contamination of UVFs in the aquatic environment. The dominance of BP-3 in surface water samples can be attributed to its common use in personal care



products, highlighting the potential sources of contamination along the river course. The significant concentrations of 234HBP and BP-1 suggest the environmental degradation of BP-3, leading to the formation of degradation products in the aquatic ecosystem. The detection of BS in specific surface water samples underscores the presence of salicylate derivatives in the river, with potential effects on aquatic organisms.

In sediment samples, the predominance of BS and BP-3 indicates the accumulation of more hydrophobic UVFs with higher partition coefficients compared to water samples. The escalating contamination levels of UV filters along the Danube River from Romania to the Black Sea suggest a continuous buildup of pollutants throughout the river course. The presence of 234HBP and BP-2 in sediment samples further highlights the transformation of parent compounds into degradation products in the sediment environment. The differences in the types of contaminants found in surface water and sediment samples indicate varying sources and behaviors of UVFs in aquatic environments.

The Pearson correlation analysis revealed significant positive and negative correlations among UVFs in surface water and sediment samples, suggesting potential degradation pathways and transformation processes among these compounds. The lack of correlation between total organic carbon (TOC) content and the UVF amount in both surface water and sediment samples can be attributed to the complex interactions between different organic compounds in aquatic systems, influencing the distribution and fate of UVFs.

The environmental risk assessment based on short-term toxicity data and maximum concentration levels of UVFs in surface water samples indicated varying levels of risk for aquatic organisms. While 234HBP and BS posed minimal risk to *Daphnia magna*, BP-1 and BP-3 showed a potential environmental risk, particularly for fish and *Daphnia magna*. The high risk quotients (RQs) for BP-1 and BP-3 underscore the need for continued monitoring and research to mitigate the ecological impact of these compounds on aquatic ecosystems.

This study highlights the importance of assessing the presence, distribution, and ecological implications of UVFs in aquatic environments. The findings underscore the need for ongoing monitoring and evaluation of UVF contamination to protect aquatic species and ecosystem health. Mitigation measures may be necessary to reduce the risks posed by UVFs and safeguard water quality in the Danube River and beyond. Further research is essential to elucidate the complex interactions between UVFs and other organic compounds in aquatic systems and to mitigate potential threats to aquatic ecosystems.

## 5. Conclusions

This study provides valuable insights into the presence and distribution of UV filters (UVFs) in the Danube River, highlighting the extensive contamination of these compounds in both surface water and sediment samples. The concentration range observed in the analyzed samples varied between n.d. (not detected) and 2224 ng/L in surface water samples, and between n.d. and 244 ng/g d.w. in sediment samples. The most commonly detected compounds in all surface water samples were 234HBP, BP-1, and BP-3, with BP-3 being found at levels of up to 2244 ng/L. In sediment samples, BP-3 and BS were the predominant compounds identified in all samples, with BP-3 reaching levels of up to 129 ng/g d.w. and BS up to 244 ng/g d.w. The prevalence of UVFs, such as 234HBP, BP-1, BP-3, and BS, underscores the potential environmental risks associated with these compounds in this aquatic ecosystem.

The correlation analysis conducted in this study suggests potential common sources and biodegradation pathways for UVFs in the dynamic aquatic environment of the Danube River. The observed correlations between different UVFs indicate complex interactions and transformation processes among these compounds, emphasizing the interconnected nature of UVF contamination in the study area.

The environmental risk assessment revealed varying risk levels for different UVFs, with compounds like BP-1 and BP-3 posing significant risks to aquatic species. These findings underscore the importance of considering both the presence of UVFs and their toxicological effects on organisms when assessing environmental risks.

While this study provides valuable insights, it is essential to acknowledge its limitations. This study focused on a specific region of the Danube River and a limited set of UVFs, so the findings may not be generalizable to other regions or compounds. Additionally, the short-term toxicity data used for the risk assessment may not fully capture the long-term effects of UVFs on aquatic ecosystems.

Moving forward, future studies should expand the scope of research to encompass a broader range of UVFs and locations along the Danube River. Investigating the sources, fate, and impacts of UVFs in more detail will inform effective management strategies and conservation efforts. Continued monitoring and research are crucial to mitigate potential risks and safeguard the health of aquatic ecosystems in the Danube River and beyond.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/w16070945/s1>, Table S1: Physico-chemical properties; Table S2: Adsorption Classifications (Estimating Physical/Chemical and Environmental Fate Properties with EPI Suite™. Sustainable Futures/Pollution Prevention (P2) Framework Manual. EPA-748-B12-001. U.S. Environmental Protection Agency, OCSPP); Table S3: The LC-MS/MS parameters for the 10 UV filters and mass-labeled standard detection and quantification; Table S4: Quantitation and confirmation information of analytes in surface water; Table S5: Quantitation and confirmation information of analytes in sediment; Table S6: Concentration levels (ng/L) of the 10 UV filters in surface water samples; Table S7: Concentration levels (ng/g s.u.) of the 10 UV filters in sediment samples; Table S8: The total amount of UVFs and TOC concentration levels; Table S9: Pearson correlation calculated between UVFs and TOC amount.

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