Land-Based Solutions for Plastics in the Sea

This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101003954

D4.5 1st Report on the presence of additives in the analyzed field samples

Due date of deliverable: 31/05/2024

Actual submission date: DD/MM/YYYY

PROJECT INFORMATION

Project number: 101003954

Project acronym: LABPLAS

Project full title: Land-Based Solutions for Plastics in the Sea

Call: H2020-SC5-2018-2019-2020 submitted for H2020-SC5-2020-2 / 03 Sep 2020

Topic: CE-SC5-30-2020 – Plastics in the environment: understanding the sources, transport, distribution and impacts of plastics pollution

Type of action: RIA – Research and Innovation Action

Starting date: June 1st, 2021

Duration: 48 months

List of participants:

Nº	Participant name	Acronym	Country	Type
1	UNIVERSIDADE DE VIGO	UVI	SPAIN	HES
2	UNIVERSIDADE DA CORUÑA	UDC	SPAIN	HES
3	Bundesanstalt fuer Gewaesserkunde	BfG	GERMANY	RTO
4	LABORATORIO IBERICO INTERNACIONAL DE NANOTECNOLOGIA	INL	PORTUGAL	RTO
5	KATHOLIEKE UNIVERSITEIT LEUVEN	KUL	BELGIUM	HES
6	HELMHOLTZ ZENTRUM FUR OZEANFORSCHUNG KIEL	GEOMAR	GERMANY	RTO
7	NATIONAL OCEANOGRAPHY CENTRE	NOC	UNITED KINGDOM	RTO
8	SORBONNE UNIVERSITE	SU	FRANCE	HES
9	OPEN UNIVERSITEIT NEDERLAND	OUNL	NETHERLANDS	HES
10	LEIBNIZ INSTITUTE FOR BALTIC SEA RESEARCH	IOW	GERMANY	RTO
11	ASSOCIACAO PARA O DESENVOLVIMENTO DO ATLANTIC INTERNATIONAL RESEARCH CENTRE	AC	PORTUGAL	RTO
12	UNIVERSIDADE FEDERAL DO SAO PAULO	UNIFESP	BRAZIL	HES
13	BASF SE	BASF	GERMANY	LE
14	TG ENVIRONMENTAL RESEARCH	ER	UNITED KINGDOM	SME
15	CONTACTICA S.L.	CTA	SPAIN	SME
16	STICHTING EGI	EGI	NETHERLANDS	Non-P
17	STICHTING RADBOUD UNIVERSITEIT	RU	NETHERLANDS	HES
18	UNIVERSIDADE FEDERAL DO PARÁ	UFPA	BRAZIL	HES

DELIVERABLE DETAILS

Document Number:	D4.5
Document Title:	1st Report on the presence of additives in the analyzed field samples
Dissemination level	PU - Public
Period:	PR2
WP:	WP4
Task:	Task 4.4
Status:	Final
Author:	UNIVERSIDADE DA CORUÑA; GEOMAR Helmholtz Centre for Ocean Research Kiel
Recommended citation format	Muniategui-Lorenzo, S.; Beck, A.; Concha-Graña, E.; Moscoso-Pérez, C.; Fernández-González, V.; Nabi, D.; Raupers, B.; Achterberg E. 2024. 1st Report on the presence of additives in the analyzed field samples, Deliverable4.5, LABPLAS Grant Agreement No. 101003954 H2020-SC5-2020-2
Abstract:	This report corresponds to Deliverable 4.5 *1st Report on the presence of additives in the analyzed field sample, resulting from Task 4.4 of the LABPLAS project*. It covers the first results obtained from the analysis of environmental samples from each case study. Plastic-associated additives were identified and quantified on selected hotspot environmental samples in each case study from WP2 in the second year (UDC, GEOMAR). This first report includes the results of water and plastic samples. The results of sediment samples will be included in the final report (D4.6).

Version	Date	Comments
1.0	20/05/2024	Initial version

Disclaimer

The views and opinions expressed in this document reflect only the authors' views, and not necessarily those of the European Commission.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ABBREVIATIONS AND ACRONYMS

Abbreviation / Acronym	Description
MP	Microplastic
REACH	Registration, Evaluation, Authorisation and restriction of CHemicals
ITEX-DHS-GC-MS/MS	In-tube dynamic headspace extraction coupled to gas chromatography, with tandem mass spectrometry detection
SPME-Arrow-GC-MS/MS	Arrow Solid Phase Microextraction coupled to gas chromatography, with tandem mass spectrometry detection
PDMS/DVB	Polydimethylsiloxane/Divinylbenzene
TD/Py-GC-MS	Thermodesorption/ pyrolysis - gas chromatography-mass spectrometry
ICP-MS	Inductively coupled plasma mass spectrometry
SPE	Solid-phase extraction
LCMS	Liquid chromatography-mass spectrometry
HPLC	High-performance liquid chromatography
HPLC-ESI-MS	High-performance liquid chromatography-electrospray ionization mass spectrometry
UHPLC	Ultra high-performance liquid chromatography
LDIR	Laser Direct Infrared
VOCs	Volatile organic compounds
PVC	Polyvinyl chloride
PAHs	Polycyclic aromatic hydrocarbons
WWTP	Wastewater treatment plant
CTD	Conductivity, temperature, and depth sensor
PE	Polyethylene

1 INTRODUCTION

Each year, significant amounts of plastic waste enter the marine environment. These plastics usually include substances added in the manufacturing process to provide the plastic with the desirable characteristics. Plastic additives encompass a wide array of substances (over 400 substances according to the European Chemical Agency [1]) with diverse physicochemical characteristics, serving as plasticizers, fire-retardants, antioxidants, UV filters, or antimicrobials, among others. These substances are present in the polymers and frequently not chemically bonded to them, so they can be easily leached from the plastic to seawater, making them more accessible to organisms [2], and potentially causing harmful effects on the marine environment. Some of these additives are classified as toxic substances and listed in REACH, and therefore, can cause harmful effects on the marine environment. Therefore, is necessary to evaluate their presence in the environment.

To evaluate the levels of plastic additives in the case study areas of WP2 (Mero-Barces River Basin, and Thames-Elbe-North Sea), samples of water, sediments, and plastics from selected sampling points were analyzed, using the analytical methods previously developed.

In this first report, the results obtained from the analysis of water and plastics collected in these areas are included.

2 ANALYTICAL METHODS DEVELOPED

2.1 UDC analytical methods

- For the **analysis of semivolatile organic additives in plastic samples**, a novel method using In-tube dynamic headspace extraction coupled to gas chromatography, with tandem mass spectrometry detection (ITEX-DHS-GC-MS/MS) was optimized and validated (UDC). Figure 1 summarises the developed method which has been published by Concha-Graña et al, 2024 [3].

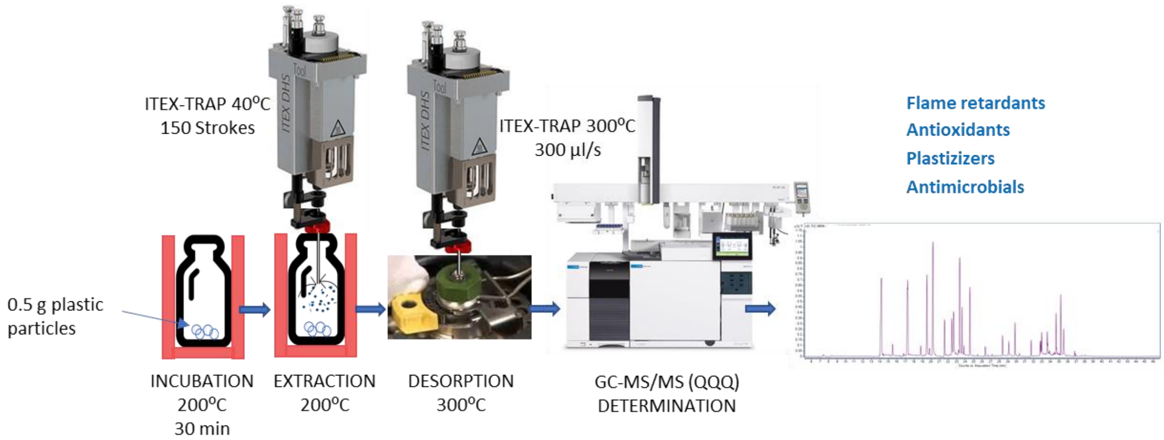

Figure 1. Summary of the ITEX-DHS-GC-MS/MS method developed for the analysis of semivolatile organic additives in plastic.

Several parameters affecting the ITEX-DHS extraction of 47 additives in plastic samples (including phthalates, bisphenols, adipates, citrates, benzophenones, and organophosphorus compounds, among others) were optimized. The use of matrix-matched calibration, together with labeled surrogate standards, minimizes matrix effects, resulting in recoveries between 70 and 128%, with good quantitation limits (below 0.1 µg g−1 for most compounds) and precision (<20%). The method proposed can be applied to any type of polymer, but due to the existence of the matrix effect, calibrates with the adequate matrix should be performed for each polymer. This method represents an effective improvement compared to previous methods because it is fast, solvent-free, fully automated, and provides reliable quantification of additives in plastic samples. See Concha-Graña et al., 2024 for details [3].

- The **analysis of semivolatile organic additives in water samples** was performed using a novel Arrow Solid Phase Microextraction coupled to gas chromatography, with tandem mass spectrometry detection (SPME-Arrow-GC-MS/MS) method optimized and validated [4]. In brief, 20 mL of water was extracted at 40°C for 90 min using a PDMS/DVB 120 µm fibre.

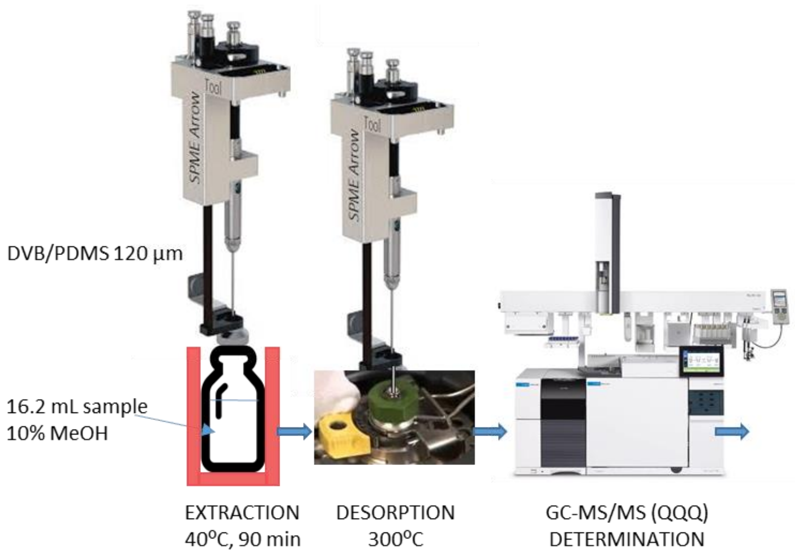

Figure 2. Summary of the SPME-Arrow-GC-MS/MS method for the analysis of plastic additives from water samples.

Univariate and multivariate approaches were used to optimize the parameters: desorption time, percentage of organic modifier, split vent time and flow, extraction time, and temperature. Other parameters were fixed previous to the optimization (fiber, incubation time, desorption temperature, sample salinity…). The quantitation limits were between 0.0017µg L^{-1} (DiBP) and 81 µg L^{-1} (TCEP). Recoveries were between 70 and 130% for all the analytes except vitamin E (57%) with good precision (<20% relative standard deviation).

The optimized method supposes an improvement to the typical ones, regarding analytical performance characteristics, and green analytical chemistry principles.

- The **analysis of volatile organic compounds (VOC)** in plastic samples was carried out by ITEX-DHS-GC-MS with an incubation temperature of 60ºC.
- **Non-target additives analysis** was performed by dual shot pyrolysis analysis (TD/Py), coupled to gas chromatography with mass spectrometry detection (TD/Py-GC-MS). The dual shot includes a thermodesorption step at 300ºC for the analysis of additives, followed by pyrolysis at 600ºC for the identification of polymers. The identification of additives was done using the F-Search library (Frontier Laboratories Ltd.).

- The **analysis of metals** was performed using microwave-assisted extraction and inductively coupled plasma mass spectrometry (ICP-MS) determination (Element XR, Thermo-Finningan). For plastic samples, 0.1 g was digested in a microwave oven with HNO3, HCl, and H2O2, and analyzed by ICP-MS. The metals analyzed were As, Ba, Ca, Cd, Co, Cr, Cu, Fe, Hg, Mg, Mn, Pb, Sb, Sn, and Zn.
- Development of **analytical methods for additives in sediment and biota** are ongoing and the results will be included in D4.6.

The full list of target compounds (n=122), including, semivolatile plastic additives (54), volatile organic compounds (53), and metals (15) is in Appendix 1.

2.2 GEOMAR analytical methods

- Development of **high-resolution UHPLC-ESI-MS method for additives quantification in water samples**

- Additive preconcentration

A solid-phase extraction (SPE) protocol adapted from Gledhill et al. (2019) was used to preconcentrate dissolved additives from seawater. In brief, Chromabond Easy cartridges (3 mL, 200 mg, 85 µm, Macherey-Nagel) were conditioned with LCMS-grade acetonitrile. Seawater samples (1 L) were filled into infusion bags and extracted through the columns by vacuum. A glass fiber filter (GF/C, 1.2 µm) was used to remove particles and prevent clogging of the cartridge. The SPE cartridges were then rinsed of salt and residual particles with ultrapure water (Milli-Q) and dried under vacuum for 90 min. Chemicals retained in the cartridges were eluted into glass HPLC vials with LCMS grade acetonitrile, and 100 µl ultrapure water was added as “keeper”. Extracts were concentrated to 50 µL using a centrifugal evaporator. Concentrated extracts were brought to 200 µl with 20% acetonitrile and transferred to conical 300 µl HPLC vials for analysis. Samples were stored at – 20°C until analysis.

- Analysis

Samples were analyzed using a newly developed method for plastic-associated chemicals (n = 84) by HPLC-ESI-MS. The UHPLC (Thermo Vanquish) was equipped with a C-18 column (Agilent Zorbax RRHD Eclipse Plus C18, 95 Å, 2.1 mm x 150 mm, 1.8 µm) with a guard column of the same material. The same chromatographic method was used for positive and negative modes. Samples were separated in 27 min with a gradient of Milli-Q water (A) and methanol (B), both with modifiers 2 mM ammonium acetate and 0.1% formic acid. A was held at 95% for 1 min, reduced to 10% over 7 min, and further reduced to 1% in the following 10 min. At this point, conditions were kept for 5 min before the starting conditions of 95% A were restored over 2 min and stabilized for a further 2 min. The injection volume was 10 µL and the flow rate was 0.2 mL min-1. The UHPLC was coupled to an Exploris 120 Orbitrap high-resolution mass spectrometer (Thermo). Full scan mass resolution was set to 120,000, with a scan range of 80-1200. Tandem mass spectrometry was performed in data-dependent mode using a Top 6 inclusion, applying a dynamic exclusion for 8 sec and an apex detection of 30% of the window. MassHunter software v10 was used for data acquisition and evaluation.

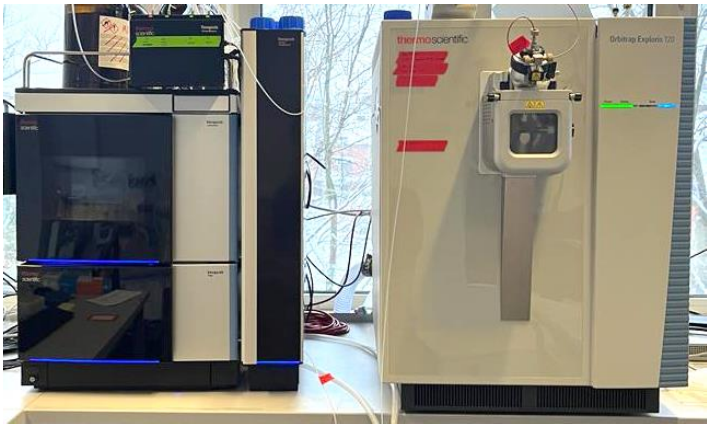

Figure 3 :Exact mass orbitrap UHPLC-ESI/APCI-MS.

- Development of **solvent extraction method for additives in net-collected microplastics** is ongoing and the results will be included in D4.6.
- The development of an **online SPE method for dissolved additives** will adapt the current manual SPE method described above and is ongoing. The results will be included in D4.6.

3 ANALYSIS OF SAMPLES FROM THE MERO-BARCÉS RIVER BASIN (UDC)

3.1 Plastic samples

Macro and meso plastics were collected in the Mero-Barces River basin. Different plastic items were collected from the shoreline, including bags, plastic bottles, wrappers, lollipop sticks, and caps, mainly. Samples were ground to 1 mm and homogenized to obtain a pool sample (sample ID095_LBP_Cecebre). See D2.3 for details.

The pooled sample was analyzed by Laser Direct Infrared (LDIR) Imaging and also by pyrolysis-gas chromatography mass spectrometry, and both techniques confirmed polyethylene (PE) as the main polymer in the pooled sample.

3.1.1 Quantitative analysis of semivolatile organic additives

Quantitative analysis of semivolatile organic additives was performed using the ITEX-DHS-GC-MS/MS method optimized and validated. Five replicates of 0.5 g of each of the pooled samples were analyzed.

Plasticisers are clearly the main contributors, at relatively high concentration.

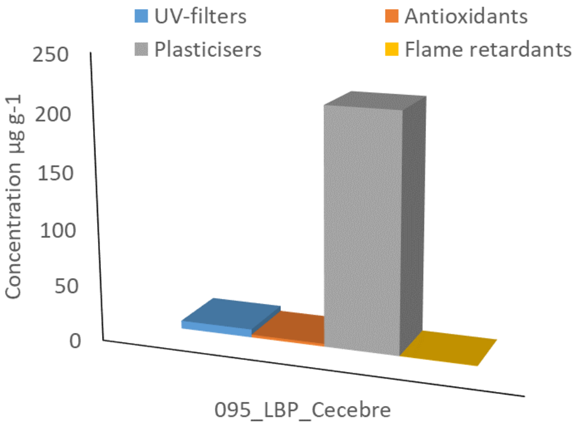

Figure 4. Plastic additives grouped by families detected in plastic sample ID095_LBP_Cecebre.

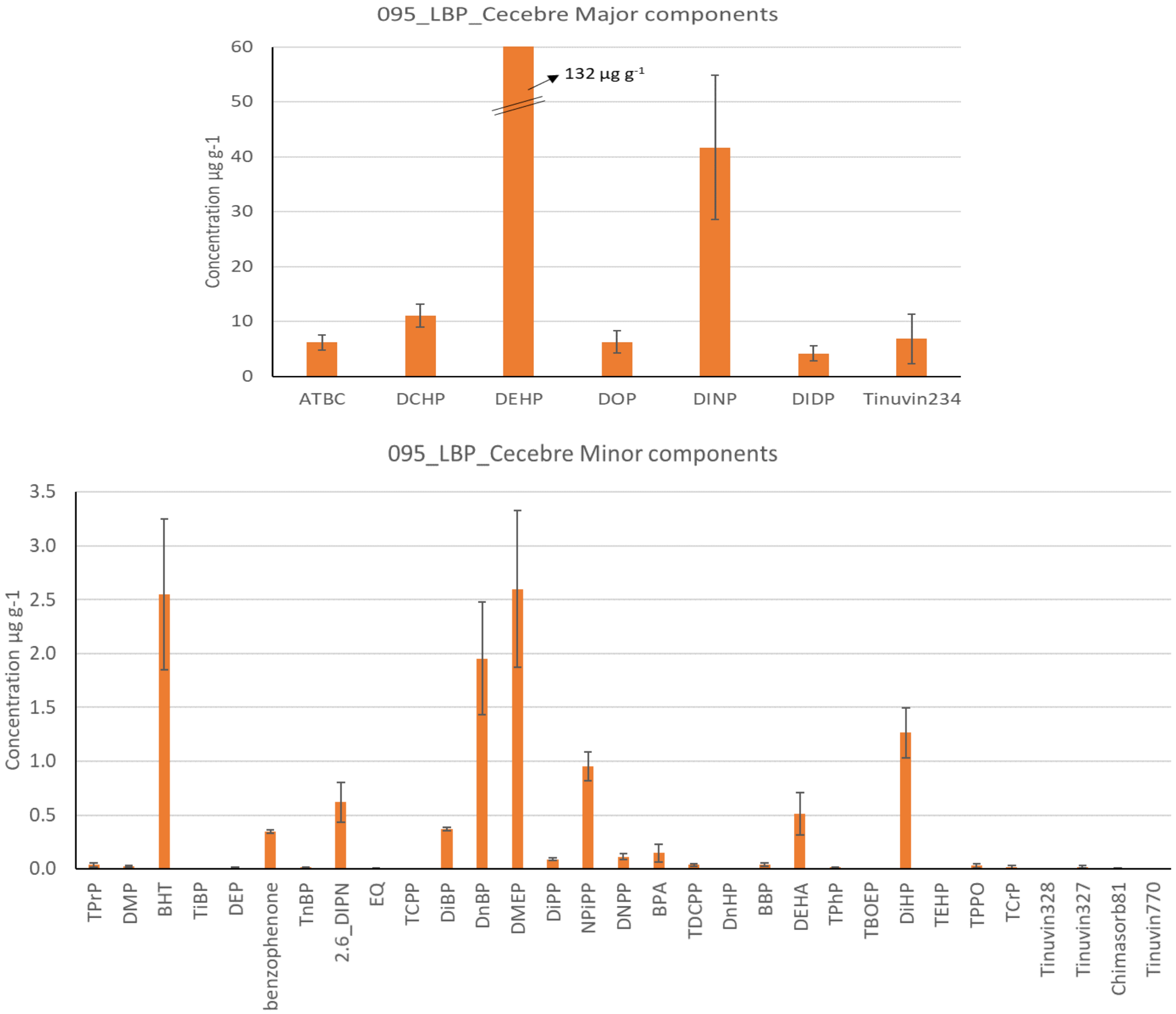

Figure 5. Individual additives quantified in plastic sample ID095_LBP_Cecebre.

Considering the individual contaminants, the most abundant additives were phthalates (DEHP, DINP, DCHP, DIDP, DOP, and others) but also ATBC (plasticizer), BHT (antioxidant), and Tinuvin 234 (UV filter).

Other additives (minor components) were detected at low concentrations (lower than 3 µg g^{-1}).

3.1.2 Quantitative analysis of volatile organic compounds

The analysis of VOCs includes 52 compounds. Only 11 compounds were found above the quantitation limit but all of them were detected at low concentrations (below 0.2 µg g^{-1}), being trichloromethane (chloroform) the most abundant (about 0.13 µg g^{-1}), followed by dichloromethane (about 0.025 µg g^{-1}).

Trichloromethane and dichloromethane are used in the production of plastics (PVC and polycarbonate respectively).

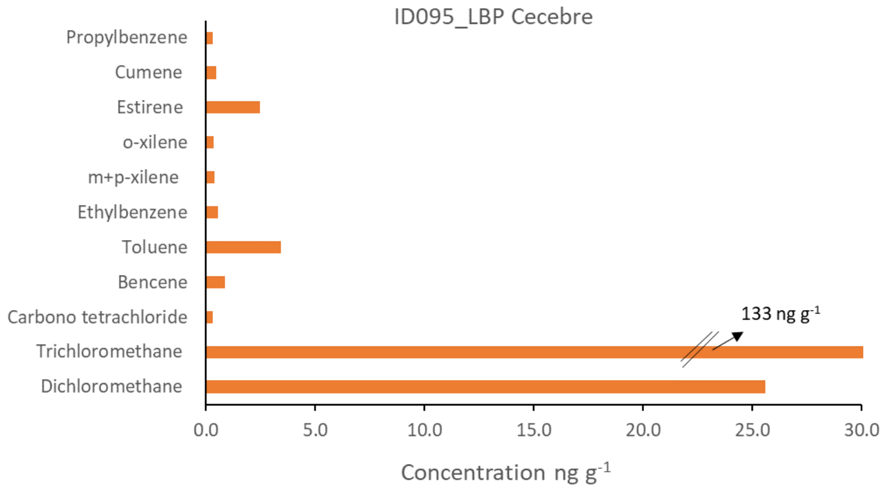

Figure 6. VOCs detected in plastic sample ID095_LBP_Cecebre.

3.1.3 Quantitative analysis of metals

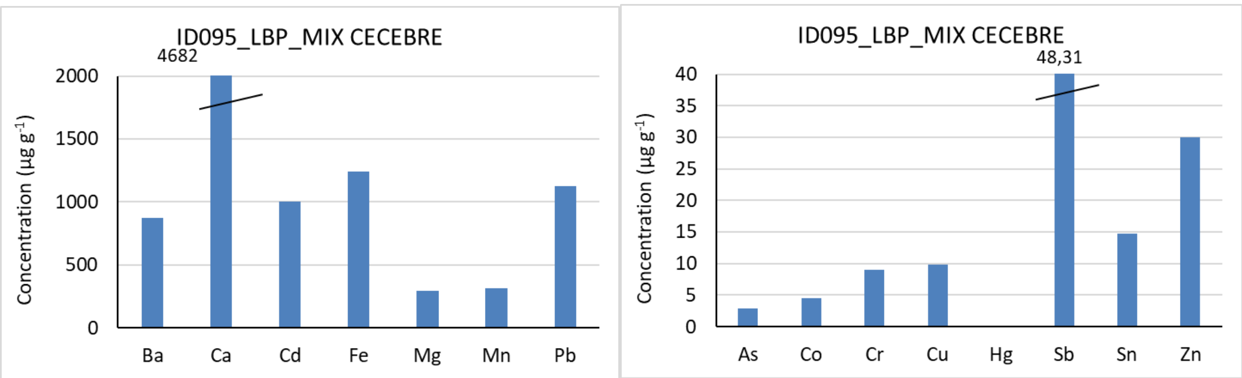

Figure 7. Metals detected in plastic sample ID095_LBP_Cecebre.

ID095_LBP_Cecebre contains high concentrations of some metals, being Ca and Fe the most abundant, whereas Hg was not detected. The concentration of Ba, Cd (used as PVC stabilizers and Ba is also used as a filler), and Mn (additive to improve the mechanical, thermal, and fire resistance properties of certain types of plastics) found in the Cecebre sample was higher than the detected in Kiel sample.

3.1.4 Qualitative analysis of organic non-target compounds

A qualitative analysis of non-target compounds in the mixture was performed by dual-shot pyrolysis, using the thermodesorption step (heated until 300ºC). Only compounds with a library spectra match above 85% were considered (Table 1).

Table 1. Non-target compounds detected in ID095_LBP_Cecebre sample (TD/Py-GC-MS).

Compound	**Library spectra match (%)**
Naphthalene	97
Benzoic acid	88
3-Methylcyclopentene	97
Phthalic anhydride	99
Biphenyl	98
1-Chlorohexadecane	90
4-tert-Octylphenol	99
Triethyl citrate	95
3-Butene-1,3-diyldibenzene (styrene dimer) [C=C(Ph)-C-C-Ph]	96
4-Phenylcyclohexene	92
Anthracene	97
Palmitic acid	98
5-Hexene-1,3,5-triyltribenzene (styrene trimer)	94
Di-(2-ethylhexyl) phthalate	95
Squalene	94

Some cyclic hydrocarbons and some PAHs were detected. Those compounds are mainly organic pollutants that were retained by the polymer. Some plastic constituents and additives were also detected: phthalates, alkylphenols, squalene, benzoic acid.

3.2 Water samples

1st sampling: July 2022
2nd sampling: February 2023
3rd sampling: May 2023
4th sampling: July 2023
5th sampling: September 2023
6th sampling: December 2023

Mero-Barcés (MB) sampling sites:
MB1, Mero River (Cecebre reservoir bridge 1)
MB2, Barcés River (Cecebre reservoir bridge 2)
MB3, Mero River upstream (rural, urban influence)
MB4, Reservoir (cleaned spot)
MB5, Reservoir
MB6, Water Treatment Plant Dam A Telva
MB7, Reservoir exit

Figure 8. Mero-Barces River Basin sampling points (water).

Six sampling campaigns were performed seasonally between summer 2022 and winter 2023-2024. Water samples were collected in amber bottles and stored refrigerated until analysis without any filtration or acidification.

3.2.1 Quantitative analysis of semivolatile organic additives

Quantitative analysis of semivolatile organic additives in water samples was performed using the SPME-Arrow-GC-MS/MS method optimized and validated. The volume of the sample analyzed was 16.2 mL.

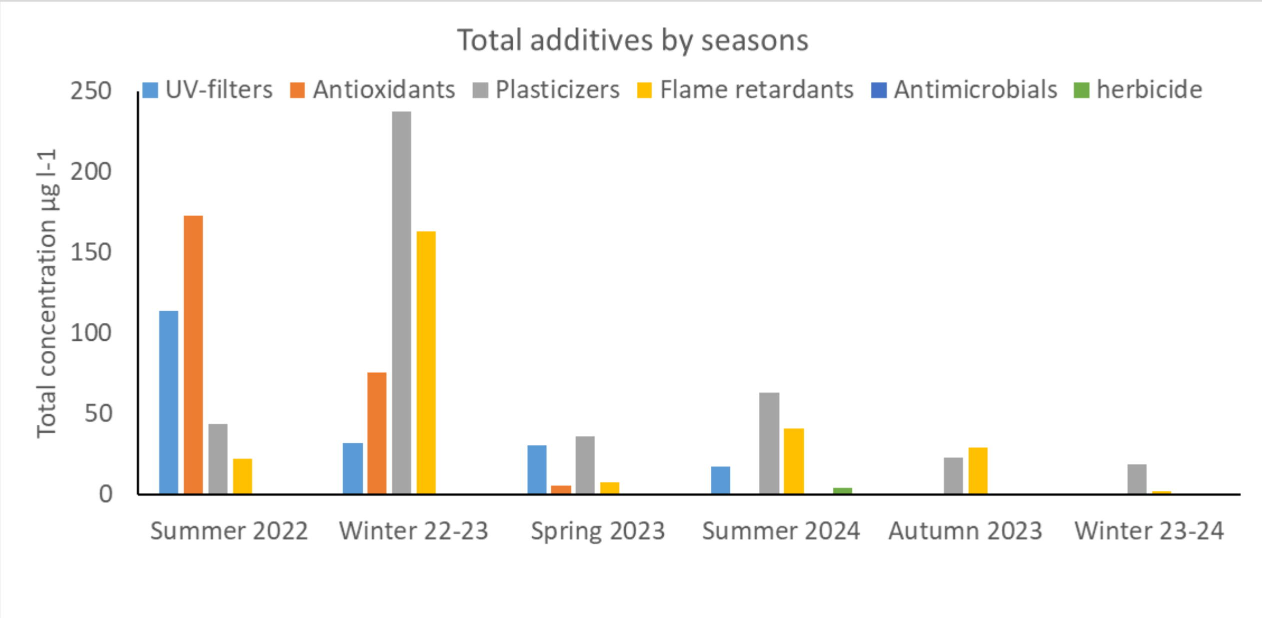

Figure 9. Plastic additives grouped by families and season detected in water samples.

As can be seen as a summary in Figure 10, the total amount of additives decreased in time, with higher concentrations observed in summer 2022 and winter 2022, and the concentration decreased since spring 2023. The additive families profile was variable, depending on the season, but plasticizers were the most abundant in all campaigns except in the 1st sampling event (when antioxidants were the most abundant).

Comparing the incidence of plastic additives in the 7 sampling points, to detect possible hot spots, we observed that there are no clear hot spots, with variation over sampling seasons, and differences in the additives family considered.

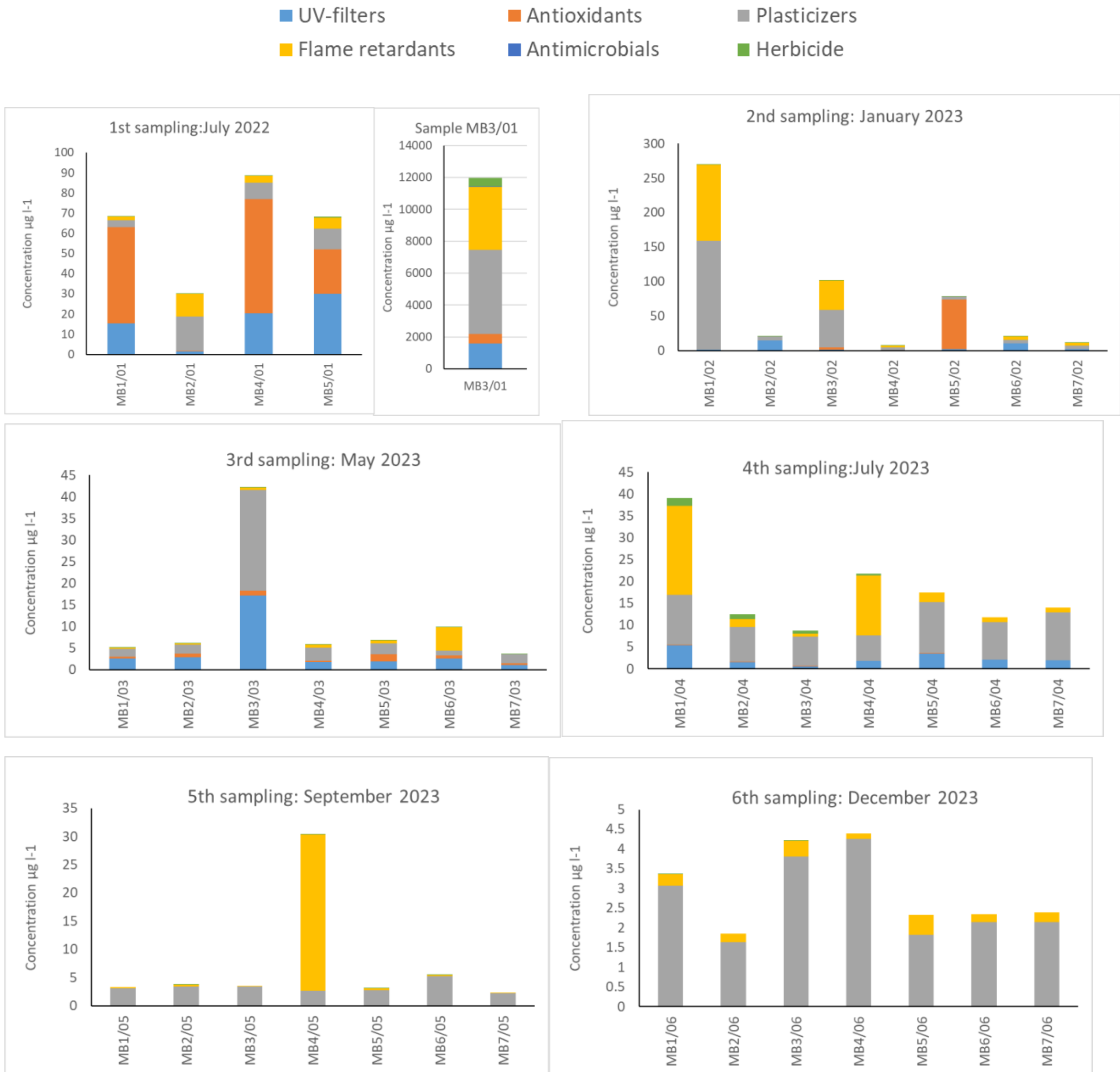

Figure 10. Plastic additives in water samples from Mero-Barcés

A high concentration of all plastic additives families was measured in the MB3 sampling point in the first campaign. This point is located near to a WWTP. Considering this high result was not found in any of the next samplings, these high values can be due to a punctual discharge by overflowing of the plant.

Regarding the individual additives, some of the compounds included in the method were not detected in any sample above the quantification limit: TBHQ, EQ, BP, TCEP, DPrP, BPF, DiPP, Triclosan, NPiPP, DNPP, BPA, ATBC, DnHP, BBP, TPhP, TBOEP, DHNA, DnOP, DTDA, Tinuvin 770, Irgafos 168, Irganox 1076, 6-PPD and 6-PPDQ (the last two compounds, tire markers, were included in the method since the 4th sampling campaign).

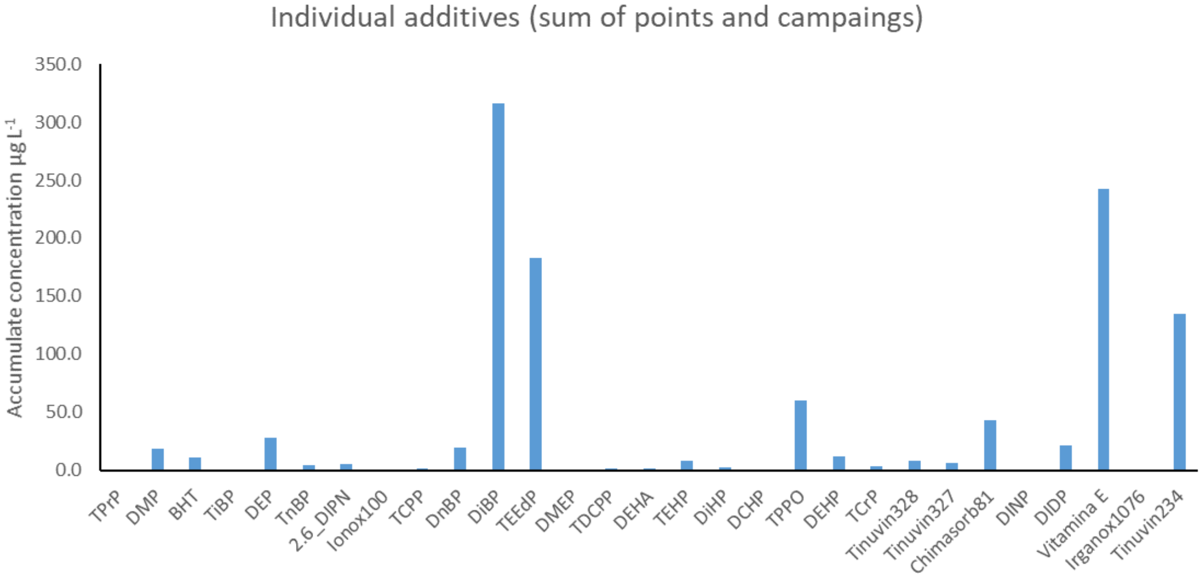

Figure 11. Individual additives detected in water samples (sum of sampling points and sampling campaigns).

DiBP, TEEdP, TTPO, vitamin E, and Tinuvin 234 are the main additives found in most samples. DEHP was not measured in any sample at levels above the environmental quality standards established by the DIRECTIVE 2013/39/EU for inland surface waters (1.3 µg l^{-1}).

Sediment samples were also collected from hot spots in the Mero-Barces river basin. Analysis of the samples is ongoing and the results will be included in D4.6.

4 ANALYSIS OF SAMPLES FROM THAMES-ELBE AND NORTH SEA

4.1 Plastic samples (UDC analysis)

Macro and meso plastics were collected in the Thames-Elbe-North Sea area. Plastics collected included taps, polystyrene, foam rubber and butts. Samples were ground to 1 mm and homogenized to obtain a pool sample (sample ID093_LBP_Mix Kiel). See D2.3 for details.

The pooled sample was analyzed by LDIR Imaging and also by pyrolysis-gas chromatography mass spectrometry, and both techniques confirmed polypropylene as the main polymer in the pooled sample.

4.1.1 Quantitative analysis of semivolatile organic additives

As in the case of the Cecebre sample, five replicates of 0.5 g of each of the pooled samples were analyzed by ITEX-DHS-GC-MS/MS.

As well as the Cecebre sample, plasticizers are the main contributors, but at a concentration 10 times lower than the Cecebre sample. UV filters were the second family of additives, present at a concentration similar to the level found in the Cecebre sample.

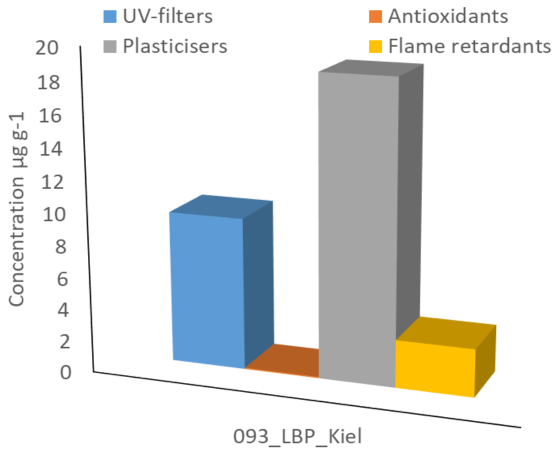

Figure 12. Additives grouped by families detected in plastic sample ID093_LBP_Mix Kiel.

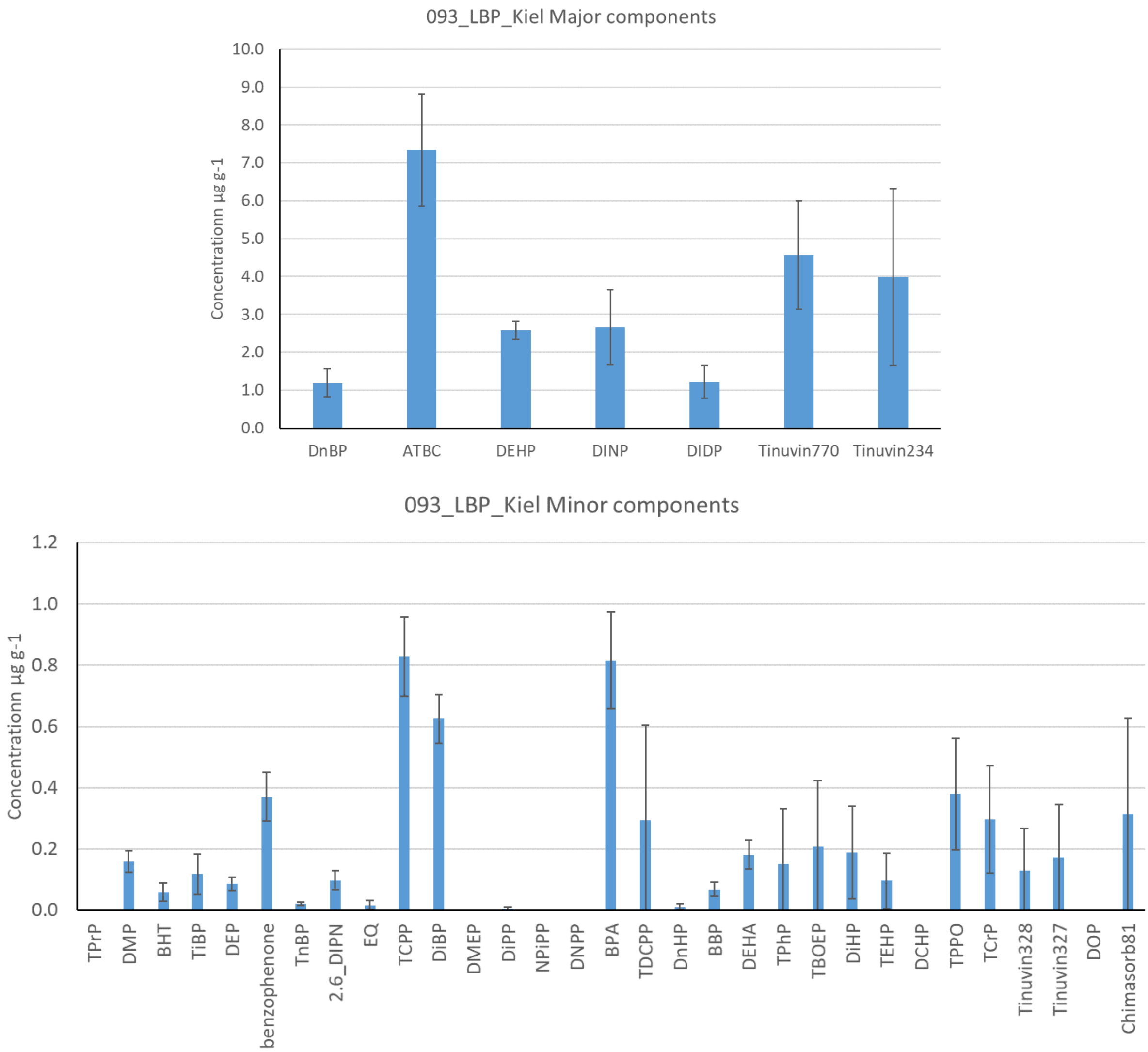

Figure 13. Individual additives cuantified in plastic sample ID093_LBP_Mix Kiel.

Considering the individual contaminants, the most abundant additive was ATBC (plasticizer), at a concentration similar to the one detected in the Cecebre sample. Also, some phthalates (DEHP, DINP, DIDP, DnBP, and others) and some Tinuvin (UV filter) were detected at relatively high concentrations (<10 µg g^{-1}), but lower than in Cecebre (<60 µg g^{-1}). Most additives were detected at concentrations lower than 1 µg g^{-1}.

4.1.2 Quantitative analysis of volatile organic compounds

In the analysis of 52 VOCs, 15 compounds were found above the quantitation limit but all of them were detected at low concentrations (below 0.2 µg g^{-1}). As well as in the Cecebre sample, trichloromethane (chloroform) was the most abundant (about 0.14 µg g^{-1}), followed by naphthalene, and styrene.

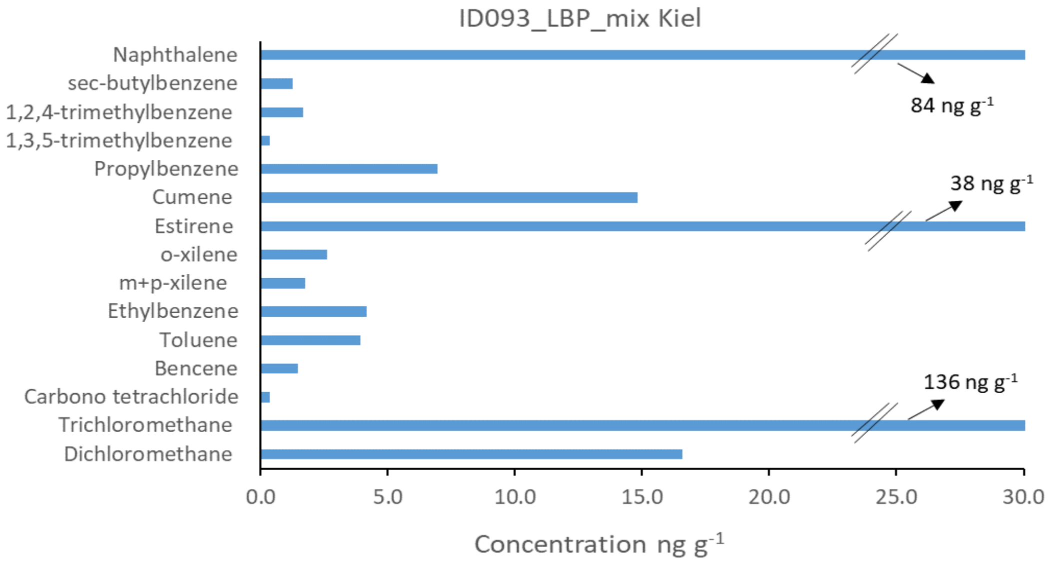

Figure 14. Volatile organic compounds quantified in plastic sample ID093_LBP_Mix Kiel.

The presence of naphthalene can possibly come from the butts included in this pool sample.

4.1.3 Quantitative analysis of metals

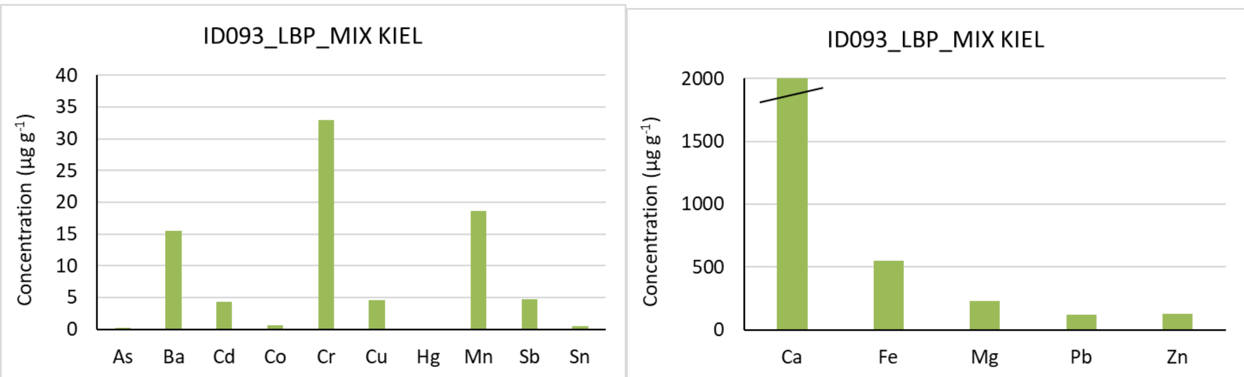

Figure 15. Metals detected in plastic sample ID093_LBP_Mix Kiel.

ID093_LBP_Mix Kiel pool plastic sample contains a lower concentration of metals than ID095_LBP_Cecebre. Ca and Fe were the elements found in the highest concentrations and Hg was not detected. As in the Cecebre sample, high concentrations of Ba, Cd (used as PVC stabilizers and Ba is also used as a filler), and Mn (additive to improve the mechanical, thermal, and fire resistance properties of certain types of plastics) were found.

4.1.4 Qualitative analysis of organic non-target compounds

A qualitative analysis of non-target compounds in the plastic mixture was performed by dual-shot pyrolysis (TD-GC-MS), using the thermodesorption step (heated until 300ºC). Only compounds with a library spectra match above 85% were considered (Table 2).

Table 2. Non-target compounds detected in ID093_LBP_Mix Kiel sample (TD/Py-GC-MS).

Detected Compounds	Library spectra match (%)
Styrene	85%
2,3-Dimethyl-4-ethylpyrrole	65%
2,4,6,8-Tetramethyl-1-undecene (isotactic)	95%
2,4,6,8-Tetramethyl-1-undecene (syndiotactic)	93%
2,4,6,8-Tetramethyl-1-undecene (syndiotactic)	94%
2,4,6,8,10-Pentamethyl-1-tridecene (isotactic)	83%
2,4,6,8,10-Pentamethyl-1-tridecene (isotactic)	77%
2,4,6,8,10-Pentamethyl-1-tridecene (isotactic)	80%
4-tert-Octylphenol	99%
1,3-Diphenylpropane [C(Ph)-C-C-Ph]	82%
Triethyl citrate	89%
3-Butene-1,3-diyldibenzene (styrene dimer) [C=C(Ph)-C-C-Ph]	97%
2,4,6,8,10,12-Hexamethyl-1-pentadecene (isotactic)	79%
5-Hexene-1,3,5-triyltribenzene (styrene trimer)	98%
Squalene	97%
2,4,6,8,10,12,14,16,18,20,22,24,26-Tridecamethyl-1,26-heptacosadiene	82%
2,4,6,8,10,12,14,16,18,20,22,24,26,28-tetradecamethyl-1,28-nonacosadiene (isotactic)	85%
2,4,6,8,10,12,14,16,18,20,22,24,26,28,30-Pentadecamethyl-1,30-hentriacontadiene	91%

The amount of sample analyzed with this technique is very small (less than 1 mg), and due to the characteristics of the pool simple, some differences in the compounds detected by this technique can be observed when different aliquots are analyzed. Also, differences with the compounds detected by the other techniques used can be obtained. For example, naphthalene was quantified in the VOCs analyses of this sample, but not detected in the non-target TD-GC-MS analysis.

4.2 North Sea Water samples (GEOMAR)

4.2.1 Sample collection

Water samples were collected from the North Sea along a transect of stations between the Thames and Elbe Rivers. Four stations were sampled during winter 2023 (cruise AL586, 4-12 Feb) and five stations were sampled in summer 2023 (cruise AL596, 26 June – 02 July). Samples were collected using a Niskin bottle-rosette and extracted on board as described above. The rosette was equipped with sensors for conductivity, temperature, and depth (CTD). The sensor data from every CTD cast was collected on the downcast so that the measured water column was not disturbed. During AL586, samples were collected below the seawater surface (~1 m depth), mid-water column (variable depth, depending on the physical structure e.g., temperature and salinity), and approximately 2 m above the seafloor. During Al596, samples were collected only from surface and deep water.

4.2.2 Preliminary Results

Not all of the target compounds were measurable in the field samples, and Table 3 lists the additives that were detectible (n=18). The full list of target compounds (n=84) is in Appendix 1.

Table 3. Plastic additive compounds detected in the North Sea field samples and their description

Application	Full Name	CAS	Abbreviations
Antioxidant and stabilizer	diphenylamine	122-39-4	DPA
Epoxy resin component	Bisphenol A diglycidyl ether	1675-54-3	BADGE
Catalyst	benzyldimethylamine	203-149-1	BDMA
PFAS	Nonafluoro-1-butane sulfonic acid	375-73-5	NfBSA
PFAS	Perfluoroheptanoic acid	375-85-9	PFHpA
PFAS	Perfluorooctanoic acid	335-67-1	PFOA
PFAS	Perfluorooctane sulfonic acid	1763-23-1	PFOS
PFAS	Perfluorononanoic acid	375-95-1	PFNA
PFAS	Tricosafluordodecanoic acid	307-55-1	TFDCA
Plasticizers/Flame retardants	tri-o-tolyl phosphate	78-30-8	TOTP
Plasticizers	dihexyl phthalate	84-75-3	DHP
Plasticizers	diisoheptyl phthalate	71888-89-6	DIHP
UV absorber	Tinuvin 770	52829-07-9	TIN770
UV absorber	Tinuvin P	2440-22-4	TINP
UV absorber	UV-531	1843-05-6	UV531
UV absorber	UV-350	36437-37-3	UV350
UV absorber	UV-328	25973-55-1	UV328
UV absorber	2,2',4,4'-tetrahydroxybenzophenone	131-55-5	THBP

Most of the target compounds were detected at levels between 0.1 and a few ng/L (figure 16). Two UV-absorbing compounds, Tinuvin 770 and Tinuvin P, were found at much higher levels of several ng/L and up to 260 ng/L, respectively. This contrasts with the results from the Mero-Barces system, where Tinuvin 770 was not detected (see Section 3.1.1). Tinuvin 770 was only detected in the North Sea in the winter samples. In contrast, Tinuvin P was lower and intermittently detected in the winter samples, but at high concentrations and found in all the summer samples. The PFAS class of compounds was most frequently detected, and four PFAS were found in every sample (NfBSA, PFHpA, PFOA, PFOS). Plasticizers were the most infrequently detected compounds, found in only 5-14% of the samples.

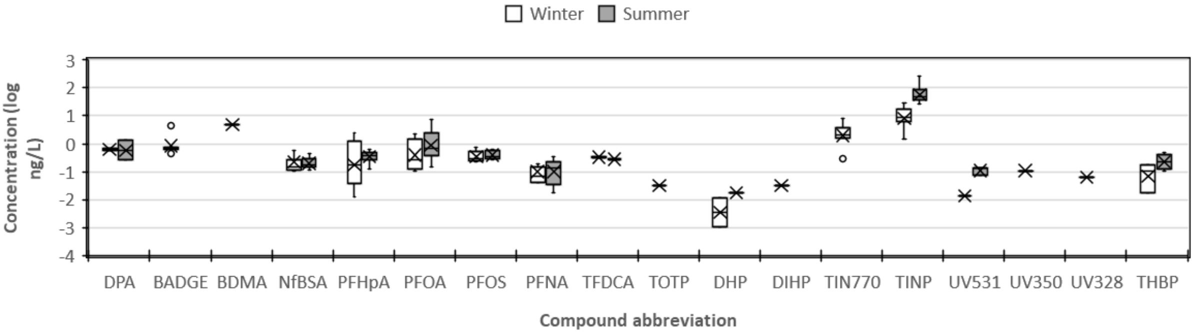

Figure 16. Box and whisker plot of additive concentrations across all stations in summer and winter sampling seasons. Note that the concentrations are on a log scale. Refer to Table 3 for the compound name abbreviations.

In general, concentrations were higher toward the eastern side of the sampling region, near the Elbe estuary (Figures 17 and 18). Tinuvin 770 was an exception to this, and was higher at stations in the western region, although TIN770 was only found in the winter season. Excluding TINP, the most commonly detected compounds were in the PFAS class and were markedly higher toward the Elbe estuary. The antioxidant DPA was found during both seasons in the central or eastern region, but only in deep water. The epoxy resin component BADGE contributed substantially to the total (non-TINP) additive inventory, but only during winter.

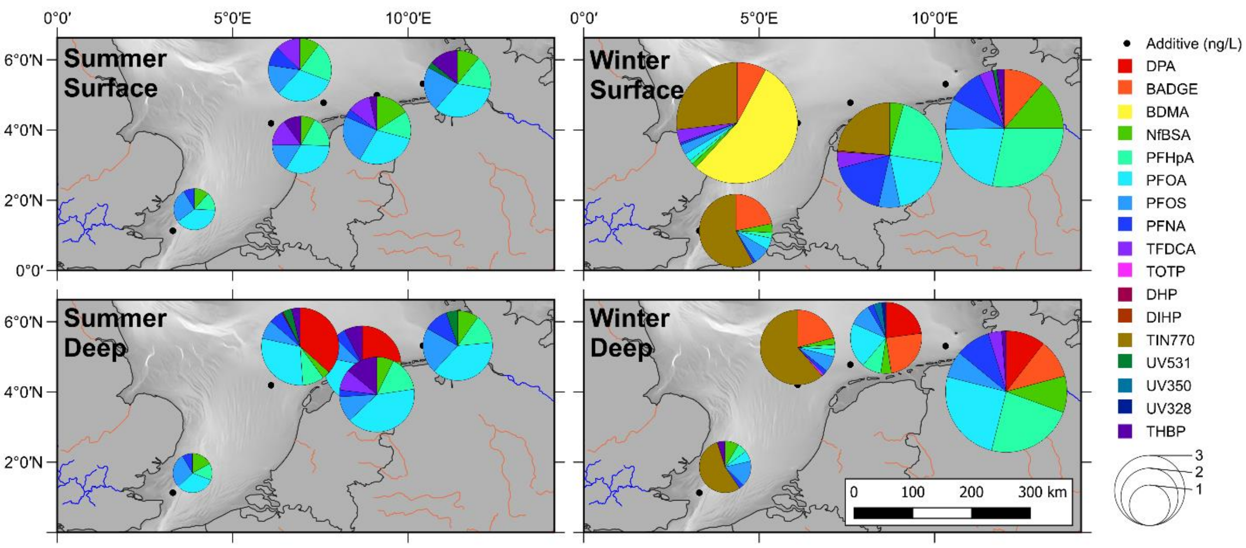

Figure 17. Spatial distribution of detected additives in surface (upper panels) and deep water (lower panels) in summer (left) and winter (right) seasons. Colors indicate the relative proportion of different additives, and the circle size shows the sum concentration of the shown additives. Black dots indicate the exact sampling locations; five stations were sampled in summer and four in winter.

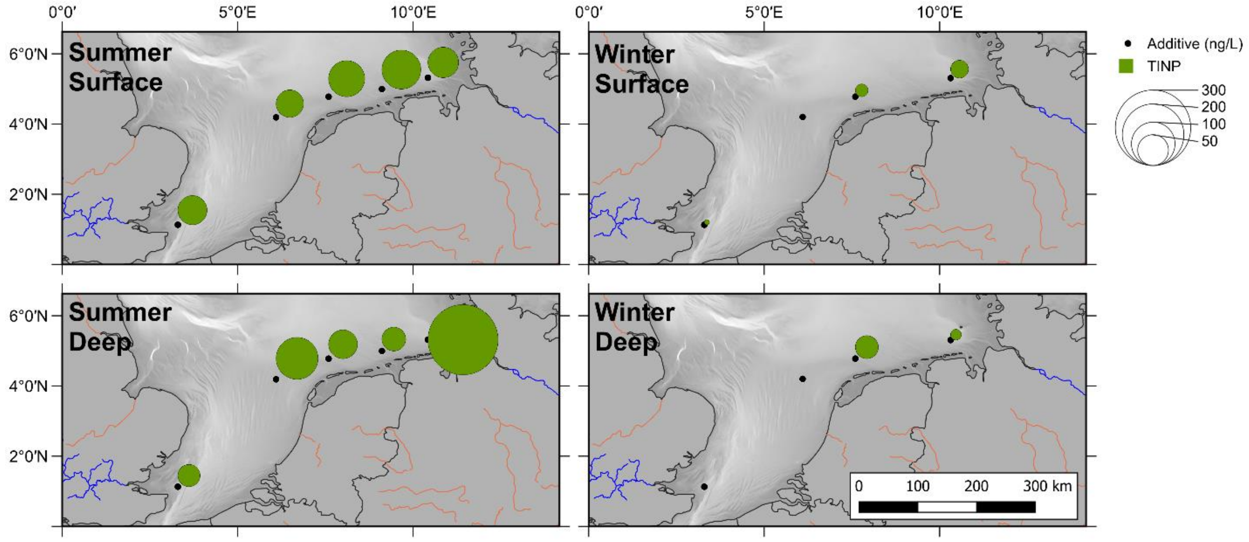

Figure 18. The spatial distribution of Tinuvin P is shown separately from other compounds due to the much higher concentrations of TINP. Symbols as in Fig. 17.

4.3 Thames and Elbe Water samples (UDC)

Water samples of the hot spots in the Thames and Elbe rivers were collected, including some samples at different depths to study the effect of depth in the presence of additives. These samples were analyzed and the results are being processed and will be included in D4.6.

5 REFERENCES

[1] ECHA, UE inventory, (2016). https://echa.europa.eu/es/information-on-chemicals/ec-inventory (21/12/2023).

[2] K. Gunaalan, E. Fabbri, M. Capolupo, The hidden threat of plastic leachates: A critical review on their impacts on aquatic organisms, Water Res. 184 (2020) 116170. https://doi.org/10.1016/J.WATRES.2020.116170.

[3] Concha-Graña, E.; Moscoso Pérez, C.M.; Fernández-González, V.; López-Mahía, P.; Muniategui-Lorenzo, S.; 2024. A green approach for the automatic quantitative analysis of additives from plastic samples using In-Tube Extraction Dynamic Headspace sampling technique coupled to GC-MS/MS. Anal Chim Acta 1302: 3424. https://doi.org/10.1016/j.aca.2024.342487

[4] Concha-Graña, E.; Fernández-González, V.; Moscoso-Pérez, C.M.; López-Mahía, P.;Muniategui-Lorenzo, Soledad; 2022. Arrow solid-phase microextraction-gas chromatography-tandem mass spectrometry for the analysis of plastic additives from seawater. Poster presented in MICRO2022.

6 ANNEXES

Appendix 1. Full list of target additives in GC-MS/MS and volatile organic compounds analytical methods (UDC).

ACRONYM	NAME	CAS
UV FILTERS		
Tinuvin 328	2-(3,5-Di-tert-amyl-2-OH-phenyl)benzotriazole	25973-55-1
Benzophenone (BP)	diphenylmethanone	119-61-9
Tinuvin 234	2-(2H-Benzotriazol-2-yl)-4,6-bis(1-methyl-1-phenylethyl)phenol	70321-86-7
Tinuvin 327	2,4-Di-tert-butyl-6-(5-chloro-2H-benzotriazol-2-yl)phenol	3864-99-1
Tinuvin 770	bis(2,2,6,6-tetramethylpiperidin-4-yl) decanedioate	52829-07-9
Chimasorb 81	2-Hydroxy-4-n-octoxybenzophenone	1843-05-6
ANTIOXIDANTS		
Irganox 1076	Octadecyl-3-(3.5-di-tert-butyl-4-hydroxyphenyl)propionate	2082-79-3
TBHQ	t-butyl hydroquinone	1948-33-0
EQ	ethoxyquin	91-53-2
Irgafos 168	Tris(2,4-di-tert-butylphenyl)phosphite	31570-04-4
BHT	Butilated hydroxytoluene	128-37-0
Ionox100	2,6-di-tert-butyl-4-hydroxymethyl-phenol	88-26-6
Vitamina E	α-tocopherol	59-02-9
6-PPD	N-(1,3-Dimethylbutyl)-N'-Phenyl-P-Phenylenediamine	793-24-8
6-PPD-Q	2-anilino-5-(4-methylpentan-2-ylamino)cyclohexa-2,5-diene-1,4-dione	2754428-18-5
PLASTICIZERS		
BBP	Butyl benzyl phthalate	85-68-7
DEHP	Bis(2-ethylhexyl) phthalate	117-81-7
DnBP	Di-n-butyl phthalate	84-74-2
DEP	Diethyl phthalate	84-66-2
DMP	Dimethyl phthalate	131-11-3
DOP	Di-n-octyl phthalate	117-84-0
DMEP	Bis(2-methoxyethyl) phthalate	117-82-8
DPrP	Di-n-propylphthalate	131-16-8
DiBP	Diisobutyl phthalate	84-69-5
DnBP	Di-n-butyl phthalate	84-74-2
DiPP	Diisopentyl phthalate	605-50-5
DnHP	Di-n-hexyl phthalate	84-75-3
DNPP	Dipentyl phthalate	131-18-0
DCHP	Di-cyclo-hexylphtalate	84-61-7
DINP	Di-isononylphthalate	28553-12-0
DIDP	Diisodecyl phthalate	26761-40-0
NPiPP	N-pentyl-isopentyl phthalate	84777-06-0
BPA	Bisphenol A	80-05-7
BPF	Bisphenol F	620-92-8
DEHA	Di(2-ethylhexyl)adipate	103-23-1

DIBA	Diisobutyl adipate	84-69-5
DHNA	Di(n-heptyl, n-nonyl) adipate	68515-75-3
ATBC	Acetyltributyl citrate	77-90-7
DTDA	Di(tridecyl)adipate	16958-92-2
FLAME RETARDANTS		
TDCPP	1,3-Dichloro-2-propanol phosphate	13674-87-8
TEEdP	Tetraethyl ethylene diphosphonate	995-32-4
TEHP	Tri(2-ethylhexyl) phosphate	78-42-2
TiBP	Tri-iso-butyl phosphate	126-71-6
TnBP	Tri-n-butyl phosphate	126-73-8
TCrP	Tri-m-tolyl phosphate	563-04-2
TPhP	Triphenyl phosphate	115-86-6
TPPO	Triphenylphosphine oxide	791-28-6
TPrP	Tripropyl phosphate	513-08-6
TBOEP	Tris (2-butoxyethyl) phosphate	78-51-3
TCEP	Tris(2-chloroethyl) phosphate	115-96-8
ΣTCPP	Tris(chloropropyl) phosphate, mixture of isomers	13674-84-5
ANTIMICROBIALS		
Triclosan		3380-34-5
Lawsona	1,4-naphthalenedione,2-hydroxy	83-72-7
HERBICIDE		
2,6-DIPN	2,6-Di-isopropyl naftaleno	24157-81-1

Volatile organic compounds	**CAS**
1,1 dichloroethene	75-35-4
Dichloromethane	75-09-2
cis 1,2-dichloroethene	156-59-2
1,1-dichloroethane	75-34-3
2,2-dichloropropane	594-20-7
trans 1,2-dichloroethene	156-60-5
Bromochloromethane	74-97-5
Trichloromethane	67-66-3
1,1,1-trichloroethane	71-55-6
Carbon tetrachloride	56-23-5
1,1-dichloropropene	563-58-6
Benzene	71-43-2
1,2-dichloroethane	107-06-2
Trichloroethene	79-01-6
1,2-dichloropropane	78-87-5
Dibromomethane	74-95-3
Bromodichloromethane	75-27-4
cis1,3-dichloropropene	10061-01-5

Toluene	108-88-3
trans 1,3-dichloropropene	10061-02-6
1,1,2-trichloroethane	79-00-5
Tetrachloroethene	127-18-4
1,3-dichloropropane	142-28-9
Dibromochloromethane	124-48-1
1,1-dibromoethano	557-91-5
Chlorobenzene	108-90-7
1,1,1,2-tetrachloroethane	79-34-5
Ethybenzene	100-41-4
m+p-xylene	108-38-3 /106-42-3
o-xylene	95-47-6
Styrene	100-42-5
Bromoform	75-25-2
Cumene	98-82-8
Bromobenzene	108-86-1
1,1,2,2-tetrachloroethane	79-34-5
1,2,3 trichloropropane	96-18-4
Propylbenzene	103-65-1
2-chlorotoluene	95-49-8
4-chlorotoluene	106-43-4
1,3,5- trimethylbenzene	108-67-8
Terc-butylbenzene	98-06-6
1,2,4-trimethylbenzene	95-63-6
sec- butylbenzene	135-98-8
1,3-dichlorobenzene	541-73-1
p-isopropyltoluene	99-87-6
1,4-dichlorobenzene	106-46-7
1,2-dichlorobenzene	95-50-1
n-butylbenzene	104-51-8
1,2-dibromo-3-chloropropane	96-12-8
1,2,4-trichlorobenzene	120-82-1
Hexachloro-1.3-butadiene	87-68-3
Naphthalene	91-20-3
1,2,3-trichlorobenzene	87-61-6

Appendix 2. Full list of target additives in HPLC-ESI-MS analytical method (GEOMAR).

#	Chemical Name	Abbreviation
1	bis(2-dimethylaminoethyl)(methyl)amine	DMAEA
2	2-((2-AMINOETHYL)AMINO)ETHANOL	AAET
3	m-phenylenediamine	MPDA
4	3-AMINOMETHYL-3,5,5- TRIMETHYLCYCLOHEXYLAMINE	ATCYH
5	Tinuvin RS	TinRS
6	4-methyl-m-phenylenediamine	MPD
7	Biphenyl phosphate	DPP
8	4,4-methylenedianiline	MDA
9	benzyldimethylamine	BDMA
10	N,N-diethylaniline	DEA
11	1,3,5-tris(oxiranylmethyl)-1,3,5-triazine-2,4,6(1H,3H,5H)-trione	TOTM
12	CAPROLACTAM	CaP
13	Benzoic Acid	BAD
14	Perfluorobutyric acid	PFBA
15	Dibenzylamin	DZ
16	Bisphenol S	BPS
17	Nonafluoro-1-butane sulfonic acid	NfBSA
18	toluene diisocyanate	TDI
19	Tinuvin 770	TIN770
20	Triethylphosphate	TEP
21	Dimethoxyethyl phthalate	DMEP
22	2,2',4,4'-tetrahydroxybenzophenone	THBP
23	tris(2-chloroethyl) phosphate	TCEP
24	Bisphenol F	BPF
25	2-mercaptobenzothiazole	MBT
26	Undecafluorohexanoic acid	UFA
27	Perfluoroheptanoic acid	PFHpA
28	diethyl phthalate	DEP
29	Bisphenol A	BPA
30	toluene-2,6-di-isocyanate	TD126
31	1,5-naphthylene diisocyanate	NDI
32	Perfluorooctanoic acid	PFOA
33	2,4-dihydroxybenzophenone	DHBP
34	diallyl phthalate	DPT
35	Tripropylphosphate	TriPP
36	Perfluorooctane sulfonic acid	PFOS
37	Benzophenone	Bzq
38	Perfluorononanoic acid	PFNA
39	diphenylamine	DPA
40	Di-n-propyl-phthalate	DPP
41	Bisphenol A diglycidyl ether	BADGE
42	Tris(1,3-dichlorisopropyl)phosphate	TDCPP
43	Bisphenylphthalat	BIP

44	triphenyl phosphate (TPP)	TPP
45	Oxybenzone	OBZ
46	2-Pyrrolidinone, 1-octyl-	NOP
47	Perfluoroundecanoic acid	PFUnA
48	Tricosafluordodecanoic acid	TFDCA
49	benzyl butyl phthalate	BBP
50	phenyl-beta-naphthylamine	PBN
51	dibutyl phthalate	DBP
52	diisobutyl phthalate	DIBP
53	2,2',6,6'-tetrabromobisphenol A	TBBPA
54	triclosan	TCS
55	tri-o-tolyl phosphate	TOTP
56	Tinuvin P	TINP
57	(Z)-octadec-9-enylamine	ODA
58	dicyclohexyl phthalate	DCHP
59	Butylated Hydroxytoluene	BHT
60	N-dodecyl-2-pyrrolidone	ND2P
61	Bis(2,4-di-tert-butylphenyl) phosphat/Bis[2,4-bis(2-methyl-2-propanyl)phenyl] hydrogen phosphate	DBPP
62	Butyl-(2-ethylhexyl)-phthalate	BEHP
63	dihexyl phthalate	DHP
64	oleamide	OLE
65	Irganox 1330	I1330
66	diisoheptyl phthalate	DIHP
67	Cyasorb UV-531/Octabenzone	UV531
68	bis(2-ethylhexyl) phthalate	DEHP
69	dioctyl phthalate	DOP
70	Tinuvin 1577	TIN1577
71	UV-350	UV350
72	diisononyl phthalate (DINP)	DINP
73	UV-320	UV320
74	diundecyl phthalate	DUP
75	Tinuvin 326	TIN326
76	Tinuvin 234/2-(2H-Benzotriazol-2-yl)-4,6-bis(1-methyl-1-phenylethyl)phenol	TIN234
77	Di(2-ethylhexyl)tetrabromophthalate	DTP
78	di-isodecyl phthalate	DIDP
79	Irganox 1076	I1076
80	UV-328	UV328
81	Diisononylcyclohexane-1,2-dicarboxylate/Diisononyl hexahydrophthalate	DINCH
82	UV-327	UV327
83	Irgafos 168	I168
84	Irganox 1010	I1010