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Target and Suspect Analysis with High-Resolution Mass Spectrometry for the Exhaustive Monitoring of PCBs and Pesticides in *Posidonia oceanica* Meadows and Sediments

Marina Astudillo-Pascual [1], Pedro A. Aguilera [1], Antonia Garrido Frenich [2] and Irene Domínguez [2,*]

[1] Department of Biology and Geology, International Campus of Excellence in Marine Science (CEIMAR), University of Almeria, E-04120 Almeria, Spain
[2] Department of Chemistry and Physics, Research Centre for Mediterranean Intensive Agrosystems and Agri-Food Biotechnology (CIAIMBITAL), Agrifood Campus of International Excellence ceiA3, University of Almeria, E-04120 Almeria, Spain
* Correspondence: idominguez@ual.es

Abstract: This study enables the simultaneous monitoring of persistent organics pollutants (POPs) in the relevant marine seagrass *Posidonia oceanica* (L.) Delile (*P. oceanica*), without causing damage and preserving their ecological integrity and their key ecosystem services, and in marine sediments. Two classes of POPs that suppose a current threat to the environmental health status are investigated: polychlorinated biphenyls (PCBs) and pesticides. Comparisons between tissues and sediment compartmentation are studied for the first time. For these purposes, the sediments, *P. oceanica* leaves and, as a novelty, rhizomes, were studied. Samples were analyzed by gas chromatography coupled with high-resolution mass spectrometry (GC-Q-Orbitrap MS) for a comprehensive study. Eco-friendly methods were developed and validated for the determination of 38 POPs, 25 PCBs and 13 priority pesticides. The results showed that, when detected, regulated contaminants were localized mainly in the long-lived rhizomes, and 7 PCBs (the most abundant being PCB 44) and 4 priority pesticides (trifluralin, chlorpyrifos, isodrin and o,p'-DDT) were seen. Additionally, a retrospective analysis (suspect screening) was conducted, exhibiting up to 13 current-use pesticide residues in leaves and rhizomes alike. The results suggest that *P. oceanica* might be acting as a sink to contaminants in coastal areas and that rhizomes, due to their longer lifespan, reflect past and legacy contamination.

Keywords: seagrass; leaves; rhizomes; sediments; organic micropollutants; POPs; GC-HRMS; polychlorinated biphenyls

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1. Introduction

Persistent Organic Pollutants (POPs) represent a serious hazard for living beings. The nature of these mostly synthetic substances, such as their hydrophobic character, elevated bioaccumulation capacity, and long half-life, has facilitated their ubiquitous distribution in the environment. In fact, several remote ecosystems present background levels of POPs, and Background Assessment Concentrations (BACs) have been settled [1,2]. Aiming to control POPs production in industrial and agricultural activities and to reduce their presence in the environment, the Water Framework Directive (WFD) has regulated and cataloged some of them as priority substances [3], while others have been directly banned (Stockholm Convention) [4]. To ensure regulatory compliance and to fulfill the objective of achieving or maintaining the good status of the environment by 2020, the WFD has launched periodic monitoring programs. In the Barcelona Convention, several matrices were proposed for monitoring and considered in the Marine Strategy Framework Directive [5–7]. The proposed matrices were: surficial sediments acting as accumulation zones and POP reservoirs (thus feeding the water column and organisms) and sediment-hosting animals, such as the mussel *Mytilus galloprovincialis* (*M. galloprovincialis*) and the fish *Mullus barbatus* (*M. barbatus*).

The Mediterranean marine phanerogam, *Posidonia oceanica* (L.) Delile (*P. oceanica*), has been long known as one reliable indicator of disturbances in the coast and the presence of marine pollution [8–11]. Additionally, *P. oceanica* meadows are in regression, although they have been acknowledged as priority habitats due to the key ecosystem services provided, such as carbon sequestering [12–14]. It would be of great interest to develop an analytical method that allows for the sustainable monitoring of marine pollutants, such as polychlorinated biphenyls (PCBs) and pesticides, in this matrix. It is imperative to understand if the chemical content within *P. oceanica*, such as organic contaminants, might represent an additional threat to the health status of this relevant seagrass.

A recent study has shown that *P. oceanica* leaves possess a relatively higher residue accumulation than sediments, mussels or fish, which is recently arousing interest in this plant for monitoring programs [15]. However, similar to past studies which focused on other POPs, the extractions present clear disadvantages in terms of the amount of sample used [15], the solvent quantity [16,17] or the duration [18]. Likewise, articles mainly focus on the leaves, tissues with an average life span of about one year, meaning that the substances found reflect current pollution in the marine environment. On the other hand, the rhizomes, tissues that live for decades and are therefore potential providers of information related to legacy or inherited contamination, are often neglected [19,20]. Additionally, the published studies conducted their analysis using gas (GC) or liquid chromatography (LC) coupled with low-resolution mass spectrometry analyzers (LRMS) [15–18,21]. None of these studies have applied the current improvements in high-resolution mass spectrometry (HRMS) analyzers. The HRMS is fundamental to conducting ultrasensitive analysis and retrospective analysis to broaden the search to suspected and/or unknown compounds (even long after analyzing the samples) and is starting to be utilized in environmental studies [22–24]. The application of GC combined with HRMS allows for the exhaustive monitoring (searching for a large number of pollutants simultaneously) of POPs, their metabolites, sources or motion pathways.

Given the aforementioned information, this work aims to: (1) develop and validate eco-friendly methods for the simultaneous extraction of PCBs and pesticides in *P. oceanica* leaves, rhizomes and surficial sediments, using, for the first time, GC combined with HRMS (GC-Q-Orbitrap MS), and (2) analyze the presence and compartmentation of POPs residues, such as PCBs, priority pesticides and current-use pesticides, in the biotic and abiotic matrices, thanks to the combination of target and suspect screenings, which have not been previously addressed.

2. Materials and Methods

2.1. Chemical Reagents

Pesticide residue analysis grade solvents (PAR), such as acetone and ethyl acetate, were obtained from Panreac (Barcelona, Spain) and Riedel-de Haën™, respectively. Additionally, *n*-hexane and dichloromethane (DCM) were purchased from Riedel-de Haën™ and Fluka, respectively. LC/MS-grade water was acquired from Supelco (Darmstadt, Germany).

The employed certified PCB standards were retrieved from Dr. Ehrenstorfer GmbH (Ausgburg, Germany). Pesticide standards, cataloged as priority substances, were obtained from Dr. Ehrenstorfer GmbH, Riedel de Haën (Seelze, Germany) and Fluka (Steinheim, Germany). A second batch of 246 pesticide standards was purchased from Dr. Ehrenstorfer GmbH and Sigma-Aldrich (St. Louis, MO, USA). The isotopically labeled standards (IIS), PCB 28F and hexachlorobenzene-13C, were purchased from Dr. Ehrenstorfer GmbH and Supelco (Bellefonte, PA, USA), respectively. These compounds were used as injection internal standards (IIS) and allowed for the correct normalization and quantification of the detected pollutants. A detailed description of the employed standards and IIS can be found in the supplementary material (*Chemical reagents: Standards* and Table S1).

2.2. Study Area and Sampling

In this study, as shown in Figure S1, a total of 28 sampling areas from the Mediterranean Spanish Coast (Almeria, Murcia, Alicante) were selected. The selection was conducted considering their environmental characteristics and potential (point and diffuse) sources of human-driven pollution [25]. Further information on the sampling areas is shown in the supplementary material (Table S2).

Where the sampling area allowed for it, divers manually collected: (1) the five uppermost cm of non-vegetated sediment from the front line of the meadow, (2) one *P. oceanica* specimen and (3) five surficial cm of vegetated sediment inside the meadow. In the areas where *P. oceanica* meadows were not present, such as harbors, two replicates of surficial sediment were taken instead. Seagrass samples were stored in sterile plastic bags, and sediments were stored in plastic containers. Both matrices were kept at low temperatures (from 1 to 4 °C) during transport. On all occasions, *P. oceanica* sampling was carried out without altering the ecological integrity of the meadows and with the permission of the Regional Environmental Administration.

2.3. Sample Pretreatment

P. oceanica was processed as previously described in Astudillo-Pascual et al. [26]. Briefly, sand and salt were removed using distilled water. Afterwards, the samples were divided into two parts: the leaf and rhizome. Note that roots, young leaves, basal sheath and epiphytes were removed and not considered in this study. Later, the samples were stored at −20 °C (48 h) and dried at −50 °C (48 h) in a Thermo Electron Corporation Heto PowerDry LL3000 freeze-dryer (Thermo Fisher Scientific, Bremen, Germany). Subsequently, the samples were homogenized (Mixer Mill MM 200) and stored in desiccators until extraction.

In the case of sediments, these were air-dried at room temperature. During the process, samples were maintained in the dark at low temperatures and covered to avoid aerosol-driven contamination. Dried sediment was then sieved (2 mm stainless steel sieve), and the fine fraction was homogenized using a glass mortar. Samples were stored in the darkness and at room temperature until extraction.

2.4. Extraction Procedure

For the extraction of the organic pollutants from *P. oceanica* tissues, 3 mL of hexane/ethyl acetate 9:1 *v/v*, a solvent previously used in Jebara et al. [15], was added to a 15 mL Falcon tube containing 150 mg of a dry weight (d.w.) powder sample (~1 g wet weight, w.w.). All mixtures were vortexed for 1 min. Afterwards, the samples were homogenized for 2 min using a polytron PT 2100 (Kinematica AG, Lucerne, Switzerland) at room temperature (~25 °C). Subsequently, the samples were centrifuged (5000 rpm, 10 min), and the supernatant was filtered with 0.22 µm nylon filters (LLG, Meckenheim, Germany).

For the sediments, 5 g d.w. samples were placed in a 50 mL Falcon tube and hydrated with 5 mL LC/MS-grade water, followed by the addition of 10 mL of hexane/ethyl acetate 9:1 *v/v*, and they were vortexed for 1 min. The samples were then taken to the ultrasound for 10 min at room temperature and were finally centrifuged at 2700 rpm for 5 min. The resulting supernatant was filtered using 0.22 µm nylon filters.

In all cases, 1 mL of each extract (leaf, rhizome and sediment) was poured into 2 mL vials, spiked with 20 µL IIS mix and analyzed. The PCB 28F and hexachlorobenzene-13C IIS were chosen for the PCB congeners and pesticide analytes, respectively.

2.5. GC-Q-Orbitrap MS Parameters

A GC-Q-Orbitrap system made up of a TriPlus RSH autosampler, a Trace 1300 gas chromatograph and a Q-Exactive Orbitrap mass analyzer (Thermo Fisher Scientific, Bremen, Germany) was used in the present study. The injector was composed of a single taper liner of 78.5 mm × 4 mm ID (Thermo Fisher Scientific), performing hot spitless injections of 1 µL at 280 °C, and 1 min of spitless time. The carrier gas (Helium, 99.999%) flow was set at 1 mL min^{-1}. The GC separation was carried out on a VF-5 ms column of 30 m × 0.25 mm

ID and 0.25 µm. The oven program was set as described in Table S3. Positive electron ionization (EI) with a 50 uA emission current and 70 eV electron energy was chosen for the Q-Exactive Orbitrap mass analyzer. Further information on the analytical conditions and acquisition parameters can be found in the supplementary material (*Analysis of organic contaminants: GC-Q-Orbitrap MS parameters*).

2.6. Method Validation

The final methods, based on polytron extraction for *P. oceanica* and ultrasound-assisted extraction (UAE) for sediments followed by GC-Q-Exactive MS analysis, were validated following the DG SANTE guidelines, with minor modifications [27]. Hence, matrix-matched calibration points spiked with the standard mixture containing the target POPs were prepared at eight different concentrations, ranging from 2 to 2000 µg kg^{-1} in the case of *P. oceanica* tissues and from 0.2 to 200 µg kg^{-1} in the case of sediments. Since *P. oceanica* shoots washed up on the coast and the sediments retrieved from the sea shoreline were used as blanks, the POP signals observed before spiking were subtracted from the spiked blanks.

The parameters investigated were: (1) linearity, represented as the determination coefficient or R^2; (2) trueness in terms of recovery, obtained by spiking blanks at two different levels (VL1 = 20 and VL2 = 400 µg kg^{-1} for the leaf, VL1 = 10 and VL2 = 200 µg kg^{-1} for the rhizome and VL1 = 2 µg kg^{-1} and VL2 = 40 µg kg^{-1} for the sediment) after calculating the recovery values for each of the 38 target analytes; (3) the precision, estimated from the intraday and interday values. The intraday values were estimated at the same levels as the trueness, and the results were expressed as the relative standard deviations (%RSD); as for the interday precision, this was calculated like the intraday precision, but with repeating the process over three different days; (4) the limit of quantification, or LOQ, can be considered as the lowest level of the calibration curve offering suitable recoveries and RSD, but it was also calculated as 10 times the standard deviations obtained for the lowest level of the calibration curve [28]. To obtain all the mentioned parameters, each sample was analyzed in triplicate.

2.7. Analysis of Organic Contaminants: Target and Suspect Screenings

For the target analysis, a database with 38 POPs (25 PCBs and 13 priority pesticides), including their corresponding quantification ions, confirmation ions and retention times, (RT) was generated from the analysis of commercially available standards (Table S4).

The use of the HRMS analyzer operating in full scan mode allowed for conducting a retrospective analysis from the yet-generated raw data file. So, in addition to target analysis, a suspect analysis of additional pesticides of concern was carried out [29].

During the analyses, and to assure the results' reliability, a matrix-matched calibration point and a solvent-matched calibration point were injected as quality controls together with the environmental samples.

3. Results

3.1. Extraction Procedure Optimization and Validation

Before validation and analysis, an optimization step for each matrix (leaf, rhizome and sediment) was run to improve the efficiency of the extraction procedure. In the case of the *P. oceanica* tissues, five extractions systems were tested (polytron, agitation, UAE, QuEChERS and QuEChERS-UAE) using hexane/ethyl acetate 9:1 *v/v* as the solvent. Detailed information is shown in the supplementary material (*Extraction procedures*). However, for the sediments, a UAE method was tested, which exhibited satisfactory results in past studies, varying the solvent between DCM and hexane/ethyl acetate 9:1 *v/v* [30,31]. To evaluate the performance of each tested extraction strategy, blank samples were spiked with the standard mixtures of PCBs and pesticides at a final concentration of 2 mg kg^{-1}. Later, their corresponding recoveries were compared. The results revealed better recoveries for all the different analytes when using polytron with hexane/ethyl acetate for the leaves

and rhizomes alike (as shown in Table S5), while UAE extraction with hexane/ethyl acetate proved to be more adequate for sediments (Table S6).

Then, the analytical methods were validated according to the parameters described above. The obtained values are shown in Table 1. Briefly, good linearity was obtained in all cases ($R^2 > 0.9728$ for the leaf, $R^2 > 0.9803$ in the case of the rhizome and $R^2 > 0.976$ for the sediment). Additionally, adequate recoveries were achieved (from 80% to 110%) at all concentrations inside their respective linear working ranges, as well as intraday and interday precision values, which showed satisfactory results (RSD < 18% for the leaf and sediments, and RSD < 20% in the case of the rhizome) for most of the analytes in the different matrices. Finally, the observed LOQs parameters ranged from 0.015 to 0.753 and from 0.076 to 5.348 μg kg^{-1} d.w. for PCBs and pesticides in leaves, from 0.009 to 0.534 and from 0.011 to 9.785 μg kg^{-1} d.w. in rhizomes and from 0.001 to 0.093 and from 0.001 to 0.205 μg kg^{-1} d.w. in sediments, showing the excellent sensitivity of the developed methods.

3.2. Application: Occurrence and Compartmentation of POPs

Under the extraction and analytical parameters employed here, several PCBs and pesticides were detected and identified as follows.

3.2.1. Target Analysis: PCBs

The chromatographic conditions employed in this study allowed for an optimal separation of all analytes, excluding the PCB 28 and PCB 31 congeners. Due to their similarities in terms of RT, mass and confirmation ions, it was not possible to differentiate them unequivocally. Therefore, isomers were counted as only one compound and expressed as PCB 28 + 31 [32].

Regarding the samples, no PCB congeners were detected in Almeria or Murcia in any of the matrices (*P. oceanica* or sediments). The PCBs were only located in three sampling sites in the Alicante Region (ALI5, ALI6 and ALI7; Table 2 and Figure S2). As for the distribution among matrices at these three sampling sites, PCB congeners were not detected in the leaves or in non-vegetated sediment, but they were in the rhizomes and vegetated sediment. Rhizomes showed residues of up to seven congeners (PCB 28+31, PCB 52, PCB 44, PCB 81, PCB 77, PCB 153 and PCB 167). Additionally, the ∑7 PCBs recommended by the International Council for the Exploration of the Sea (ICES) (i.e., the sum of PCB 28, PCB 52, PCB 101, PCB 118, PCB 153, PCB 138 and PCB 180) ranged from 2.4 to 14.3 μg kg^{-1} d.w. In rhizomes, PCB 44 and PCB 28 + 31 were usually the most abundant in all sites, as well as PCB 77 at site ALI5. Additionally, at ALI5, where the PCB presence in rhizome was higher, the vegetated sediment (collected during step 3 of the sampling strategy) showed a wider number of PCB congeners (up to 18; Table 2 and Figure S3) but lower concentrations, observing 8.3 μg kg^{-1} d.w. for ∑7 PCBs and 21.5 μg kg^{-1} for ∑PCBs. In sediments, PCB 44 was also the most abundant, followed by PCB 138 and PCB 170 > PCB 157, PCB 28 + 31, PCB 18 and PCB 167 > PCB 101 > PCB 123 and PCB 180 > PCB 128 > PCB 153 > PCB 114 and PCB 105 > PCB 118 > PCB 77 > PCB 81.

Table 1. Validation results for the analytical methods developed in this study for the studied matrices at two concentration levels [a].

Compounds	LEAF							RHIZOME							SEDIMENT					
	LOQ (µg kg^{-1})	Linear Working Range (µg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)		LOQ (µg kg^{-1})	Linear Working Range (µg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)		LOQ (µg kg^{-1})	Linear Working Range (µg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)	
				VL1	VL2	VL1	VL2				VL1	VL2	VL1	VL2				VL1	VL2	VL1	VL2
PCBs																					
PCB 18	0.266	10–1000	0.9922	98(7)	108(10)	18	15	0.168	10–400	0.9986	97(9)	98(3)	17	6	0.017	1–200	0.9977	104(9)	98(3)	11	7
PCB 28+31	0.516	20–2000	0.9978	104(5)	99(0)	6	17	0.378	10–400	0.9988	93(3)	99(8)	1	7	0.023	1–200	0.9989	99(2)	108(3)	7	5
PCB 52	0.404	10–400	0.9966	110(5)	99(1)	17	13	0.378	10–400	0.9979	101(5)	100(5)	4	3	0.013	1–200	0.9991	111(4)	100(1)	5	3
PCB 44	0.015	20–2000	0.9998	99(0)	100(0)	8	17	0.127	10–1000	0.9982	105(4)	99(2)	5	6	0.009	1–200	0.9986	118(2)	98(3)	3	3
PCB 66	0.151	20–2000	0.9993	120(4)	101(1)	17	9	0.534	10–200	0.9930	115(17)	95(8)	18	5	0.027	1–200	0.9984	108(8)	98(4)	15	8
PCB 101	0.753	20–2000	0.9998	99(2)	100(0)	18	17	0.210	10–400	0.9894	94(11)	106(10)	2	4	0.040	1–200	0.9951	114(9)	97(8)	13	9
PCB 81	0.485	10–1000	0.9997	99(5)	99(2)	10	15	0.061	10–1000	0.9982	80(3)	99(2)	8	4	0.004	0.2–200	0.9926	102(7)	108(9)	10	10
PCB 77	0.018	10–2000	0.9997	106(7)	100(1)	8	14	0.162	10–200	0.9916	106(7)	101(1)	9	3	0.030	1–200	0.9964	94(10)	105(7)	12	10
PCB 123	0.363	20–1000	0.9996	104(1)	99(2)	8	15	0.009	10–400	0.9987	93(1)	95(13)	12	2	0.015	1–40	0.9975	103(7)	100(9)	8	9
PCB 118	0.121	20–1000	0.9979	101(2)	100(1)	5	14	0.037	10–400	0.9986	99(2)	91(8)	14	3	0.004	0.2–200	0.9926	102(5)	93(10)	7	6
PCB 114	0.032	10–400	0.9991	101(2)	102(3)	5	6	0.074	10–400	0.9984	98(8)	94(10)	18	9	0.046	1–200	0.9965	118(11)	96(7)	17	8
PCB 153	0.368	20–400	0.9951	100(6)	100(3)	11	18	0.035	10–400	0.9988	93(2)	99(10)	11	3	0.093	2–200	0.9989	97(10)	100(1)	15	5
PCB 105	0.608	20–1000	0.9973	97(4)	101(1)	10	18	0.075	10–400	0.9987	101(3)	98(10)	14	11	0.006	2–200	0.9936	102(13)	95(9)	15	9
PCB 138	0.267	10–1000	0.9970	99(0)	100(2)	3	14	0.261	10–1000	0.9803	93(15)	97(4)	10	5	0.004	1–200	0.9984	118(2)	99(2)	2	1
PCB 126	0.136	20–2000	0.9998	94(2)	100(0)	4	14	0.021	10–2000	0.9996	107(1)	100(1)	16	12	0.016	2–40	0.9760	85(5)	109(15)	15	7
PCB 128	0.022	20–400	0.9908	107(1)	101(1)	11	12	0.121	10–400	0.9979	99(12)	99(8)	1	3	0.019	2–200	0.9987	90(12)	100(5)	13	9
PCB 167	0.023	10–1000	0.9982	102(1)	100(1)	4	11	0.130	10–400	0.9988	92(7)	99(10)	3	1	0.046	1–200	0.9987	113(10)	100(1)	15	8
PCB 156	0.177	20–2000	0.9985	101(1)	109(2)	4	14	0.050	10–2000	0.9994	80(3)	100(1)	5	10	0.010	1–200	0.9990	85(7)	99(2)	18	7
PCB 157	0.265	20–1000	0.9934	100(1)	102(1)	6	15	0.035	10–400	0.9988	86(2)	99(10)	2	4	0.024	2–200	0.9995	84(4)	100(1)	18	10
PCB 180	0.206	20–1000	0.9965	108(9)	100(0)	9	17	0.182	10–400	0.9976	88(16)	98(4)	18	8	0.028	2–200	0.9997	101(16)	100(3)	18	5
PCB 169	0.171	10–1000	0.9977	100(1)	100(1)	10	15	0.052	10–2000	0.9996	102(1)	100(0)	13	8	0.048	2–200	0.9984	115(7)	101(0)	10	5
PCB 170	0.253	20–1000	0.9976	95(1)	99(2)	8	14	0.057	10–400	0.9987	91(4)	99(9)	5	4	0.001	1–200	0.9981	115(1)	99(3)	12	10
PCB 189	0.253	20–1000	0.9978	99(8)	100(1)	15	15	0.094	10–2000	0.9991	98(8)	101(2)	9	12	0.017	2–200	0.9981	82(6)	103(4)	10	10
PCB 194	0.078	20–2000	0.9986	97(5)	99(0)	5	16	0.069	10–2000	0.9993	90(8)	100(1)	10	5	0.070	2–40	0.9801	101(14)	102(7)	17	5
PCB 206	0.212	20–1000	0.9986	83(8)	99(2)	14	14	0.018	10–1000	0.9992	120(1)	100(1)	10	5	0.005	1- 40	0.9948	81(9)	98(5)	13	9

Table 1. *Cont.*

Compounds	LEAF							RHIZOME							SEDIMENT					
	LOQ (μg kg^{-1})	Linear Working Range (μg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)		LOQ (μg kg^{-1})	Linear Working Range (μg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)		LOQ (μg kg^{-1})	Linear Working Range (μg kg^{-1})	Linearity (R^2)	Recovery R (%) [b]		Inter-Day Precision (RSD%)	
				VL1	VL2	VL1	VL2				VL1	VL2	VL1	VL2				VL1	VL2	VL1	VL2
Pesticides																					
Pentachloro-benzene	0.286	2–1000	0.9993	85(17)	102(3)	18	5	0.070	2–400	0.9996	99(3)	100(1)	13	10	0.001	0.2–40	0.9995	104(0)	100(1)	5	2
Trifluralin	0.123	10–1000	0.9998	100(2)	102(0)	6	3	0.305	10–400	0.9980	106(5)	99(7)	7	2	0.050	1–40	0.9950	117(4)	101(1)	7	4
Hexachloro-benzene	1.131	10–2000	0.9985	97(10)	98(2)	5	18	0.359	10–200	0.9969	120(8)	101(0)	3	8	0.205	2–40	0.9966	114(10)	99(2)	4	5
Simazine	0.550	20–1000	0.9991	101(4)	102(2)	10	3	0.011	10–200	0.9978	119(8)	107(1)	5	2	0.019	2–40	0.9875	91(1)	103(6)	4	9
Atrazine	0.376	10–2000	0.9994	102(6)	102(2)	5	2	0.777	10–400	0.9981	108(8)	94(12)	4	10	0.047	1–40	0.9981	102(4)	101(4)	5	11
Chlorpyrifos	0.076	10–2000	0.9974	101(2)	97(4)	6	3	0.158	10–400	0.9863	96(5)	109(12)	3	5	0.103	2–40	0.9940	87(9)	101(3)	10	16
Aldrin	0.216	20–1000	0.9993	85(5)	101(1)	2	1	0.635	10–2000	0.9997	112(3)	99(2)	4	6	0.025	2–40	0.9958	81(4)	100(2)	6	9
Isodrin	0.756	10–2000	0.9976	97(15)	100(2)	5	1	0.221	10–1000	0.9997	90(5)	99(2)	20	10	0.098	2–100	0.9991	100(6)	100(2)	9	9
Dieldrin	0.126	20–1000	0.9959	82(3)	101(3)	4	5	0.621	10–200	1.0000	108(13)	101(3)	9	6	0.002	1–100	0.9989	87(1)	99(1)	14	6
Endrin	0.946	10–400	0.9805	89(1)	104(2)	9	10	1.819	10–200	0.9964	107(11)	99(1)	12	6	0.002	1–100	0.9873	87(1)	100(2)	13	4
o,p′-DDT	5.348	20–1000	0.9882	92(16)	100(1)	18	15	9.785	10–400	0.9991	115(15)	110(6)	17	10	0.130	1–40	0.9973	91(9)	102(3)	11	9
p,p′-DDD	0.482	10–400	0.9923	110(12)	106(9)	15	10	1.982	10–200	0.9991	107(13)	100(1)	16	8	0.048	1–40	0.9923	92(12)	94(9)	13	6
p,p′-DDT	4.725	10–400	0.9982	118(15)	100(9)	15	13	0.419	10–400	0.9996	110(13)	100(6)	19	8	0.015	2–100	0.9996	112(4)	99(2)	8	9

[a] The two validation levels (VL1 and VL2) correspond to 20 and 400 μg kg^{-1} for the leaf, 10 and 200 μg kg^{-1} for the rhizome and 2 and 40 μg kg^{-1} for the sediments. [b] Intra-day precision values (or repeatability, expressed as % RSD) are given in brackets (n = 3).

Table 2. Occurrence of PCBs (μg kg^{-1} d.w.) in the rhizomes of the marine plant *P. oceanica* and vegetated sediments (V-sed). Note that only the sampling areas where contamination was detected have been included in the table.

Matrix	Site	PCB 18	PCB 28+31	PCB 52	PCB 44	PCB 101	PCB 81	PCB 77	PCB 123	PCB 118	PCB 114	PCB 153	PCB 105	PCB 138	PCB 128	PCB 167	PCB 157	PCB 180	PCB 170	ΣPCBs	∑7 PCBs
Rhizome	ALI5	–	10.7	1.6	11.2	–	9.7	11.1	–	–	–	2.0	–	–	–	8.7	–	–	–	55.0	14.3
	ALI6	–	1.8	0.6	1.9	–	–	1.3	–	–	–	–	–	–	–	–	–	–	–	5.6	2.4
	ALI7	–	2.6	0.6	1.6	–	1.1	1.6	–	–	–	–	–	–	–	–	–	–	–	7.5	3.2
V-sed	ALI5	1.4	1.5	1.1	1.7	1.2	0.8	0.9	1.1	0.9	0.9	0.9	0.9	1.6	1.0	1.4	1.5	1.1	1.5	21.5	8.3

In this study the PCB 28 congener eluted together with its isomer PCB 31, and both were counted as only one compound; thus, the ∑7 PCBs value might be slightly overrated. (n.s.): not specified or not studied; (–): analyte not found or detected.

3.2.2. Target Analysis: Priority Pesticides

Regulated pesticides were not detected in any of the matrices (*P. oceanica* or sediments) from Almeria or Murcia. Instead, the priority pesticides were detected at the same stations of the Alicante Region where the PCBs were observed: ALI5, ALI6 and ALI7. In these stations, only rhizomes reflected the presence of these analytes; they were not seen in leaves and vegetated or non-vegetated sediment. The detected pesticides in the rhizomes were trifluralin, chlorpyrifos, isodrin and o,p'-DDT (Table 3). Some examples of extracted ion chromatograms are shown in Figure S4. The sum of all the priority pesticides considered in this study (ΣPesticides) ranged from 1.0 to 9.0 μg kg^{-1} d.w; the greater concentration was seen again at ALI5, followed by ALI6 and ALI7 (Figure S2).

Table 3. Occurrence of the priority pesticides (μg kg^{-1} d.w.) in the rhizomes of *P. oceanica*. Note that only the sampling areas where contamination was detected have been included in the table.

Matrix	Site	Trifluralin	Chlorpyrifos	Isodrin	o,p´-DDT	ΣPesticides
Rhizome	ALI5	3.9	3.2	1.9	*	9.0
	ALI6	2.0	1.3	1.7	–	5.0
	ALI7	–	–	1.0	–	1.0

(–) analyte not found or detected. (*) < LOQ.

3.2.3. Suspect Analysis: Current-Use Pesticides

Since the target screening of priority pesticides revealed the presence of four banned analytes in rhizomes (trifluralin, chlorpyrifos, isodrin and o,p´-DDT), and considering the historical records of agricultural activities on the Mediterranean Spanish coast, a supplementary retrospective analysis was conducted, aiming at an additional 246 pesticides of concern. From this wide array of compounds, several were considered emerging pesticides. As a result, several pesticides were tentatively identified in nine sampling areas of Almeria and seven from Alicante, while none were observed in Murcia (Table 4). In total, 16 sampling stations out of the 28 sampled showed these compounds in any of their matrices. The pesticide presence was major in the biotic compartment, i.e., leaves (revealing six compounds: 1,4-dimethyl naphthalene, 2-phenylphenol, terbutryn, tetraconazole, piperonylbutoxide and difenoconazole) and rhizomes (showing seven compounds: 1,4-dimethyl naphthalene, 2,4,6-trichlorophenol, lindane, pyrimethanil, penconazole, fludioxonil and fenbuconazole). Only the prallethrin insecticide was encountered in the abiotic compartment, i.e., vegetated and non-vegetated sediments. This analyte was detected at the RM6,

C2 and V1 from Almeria and at the ALI1, ALI2, ALI3 and ALI7 from Alicante. Examples of extracted ion chromatograms are shown in Figure 1.

These tentatively identified structures were confirmed using analytical standards. Matching RTs, confirmation ions and the comparison of the spectra between real samples and the spiked blank extracts allowed for their confirmation (Table S7) [33,34]. For quantification, a standard addition procedure was followed, and concentrations were estimated [35]. The registered summatory values for leaves ranged from <LOQ to 366.50 μg kg^{-1} d.w., with the maximum at CG3 in Almeria (ΣCurrent-use pesticides; Figure S5). In the case of the rhizomes, concentrations varied from 0.13 to 47.15 μg kg^{-1} d.w.; higher concentrations were seen at ALM3 and CG4 and also in Almeria. On the other hand, sediments from several areas exhibited the prallethrin insecticide. The ΣCurrent-use pesticide concentrations ranged from 5.28 to 7.53 μg kg^{-1} d.w. in Almeria and from 4.27 to 6.01 μg kg^{-1} d.w. in Alicante, whereas non-vegetated sediments reflected 5.52–6.90 and 4.23–7.31 μg kg^{-1} d.w., respectively (Table 4 and Figure S5).

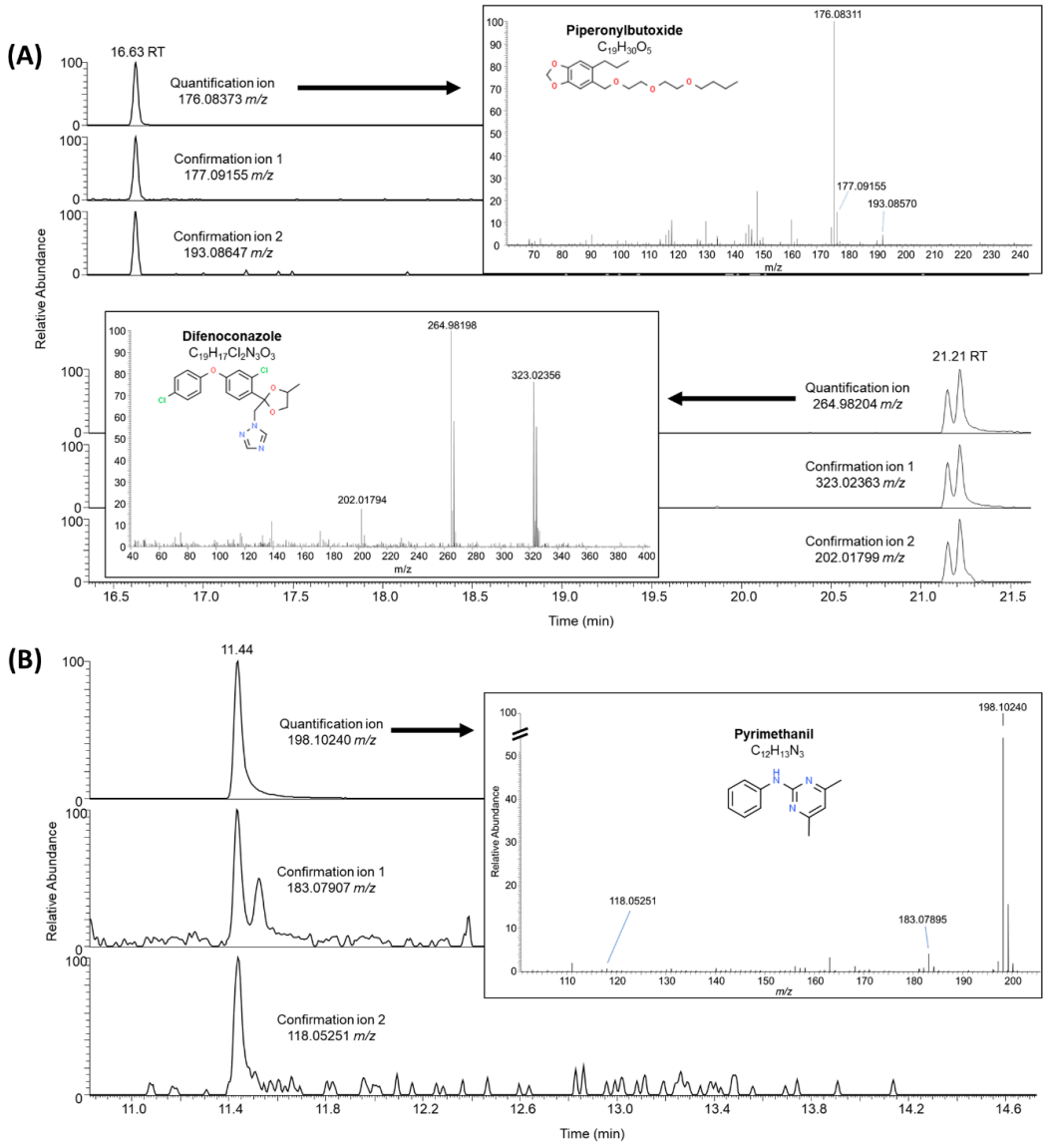

Figure 1. Extracted ion chromatograms and mass spectrum of some of the detected current-use pesticides: (**A**) piperonylbutoxide and difenoconazole in leaves at CG3, (**B**) pyrimethanil in the rhizome at station ALM3.

Table 4. Current-use pesticides (μg kg^{-1} d.w.) identified in this study in *P. oceanica*, non-vegetated sediments (NV-sed) and vegetated sediments (V-sed).

		Almeria Region												Alicante Region						
Matrix	**Compound**	**EE2**	**EE4**	**RM4**	**RM6**	**ALM1**	**ALM3**	**V1**	**V2**	**CG2**	**CG3**	**CG4**	**C2**	**ALI1**	**ALI2**	**ALI3**	**ALI7**	**ALI5**	**ALI6**	**ALI4**
Leaf	1,4-Dimethyl naphthalene	3.22	8.97	–	n.s.	–	–	n.s.	28.96	–	16.87	–	–	–	–	–	10.02	14.31	16.32	9.88
	2-Phenylphenol	–	–	2.89	n.s.	–	–	n.s.	–	–		–	–	–	–	11.03	8.43	9.18	7.16	9.6
	Terbutryn	–	–	–	n.s.	–	–	n.s.	–	–	–	–	–	0.21	0.11	–	–	–	–	–
	Tetraconazole	–	–	–	n.s.	–	–	n.s.	–	–	–	–	–	–	–	–	1.12	–	–	–
	Piperonylbutoxide	–	–	–	n.s.	–	–	n.s.	–	–	84.39	–	–	–	–	–	–	–	–	–
	Difenoconazole	–	–	–	n.s.	–	–	n.s.	–	–	265.24	–	–	–	–	–	10.05	–	–	–
	Mean regional values						82.11										15.35			
Rhizome	2,4,6-trichlorophenol	–	–	–	n.s.	–	0.88	n.s.	–	–	–	0.36	–	–	–	–	–	–	–	–
	1,4-Dimethyl naphthalene	–	–	–	n.s.	–	5.33	n.s.	–	–	–	4.46	–	–	–	–	–	–	–	–
	Lindane	–	–	–	n.s.	–	–	n.s.	–	–	–	–	–	–	–	–	0.22	0.51	0.11	–
	Pyrimethanil	–	–	–	n.s.	–	40.66	n.s.	–	–	–	29.29	–		0.13	–	–	–	–	–
	Penconazole	–	–	–	n.s.	–	0.28	n.s.	–	–	–	0.13	–	–	–	–	–	–	–	–
	Fludioxonil	–	–	–	n.s.	–	–	n.s.	–	–	–	–	–	–	–	–	–	–	2.85	7.93
	Fenbuconazole	–	6.16	8.73	n.s.	6.59	–	n.s.	–	6.84	7.11	–	–	4.08	–	–	3.15	–	–	–
	Mean regional values						16.69										3.63			
NV-Sed	Prallethrin	–	–	–	5.78	–	–	7.45; 5.52	n.s.	–	–	–	6.9	4.23	n.s.	n.s.	7.31	–	–	–
V-Sed		–	–	–	n.s.	–	–	n.s.	n.s.	–	–	–	5.28	5.01	6.01; 4.27	7.53; –	5.74	–	–	–

–: analyte not found. n.s.: not sampled.

4. Discussion

4.1. Extraction Procedure

The developed methods showed suitability for PCBs and priority pesticide detection in the different matrices (*P. oceanica* tissues and sediment). As a novelty, the premises of the sample-friendly techniques and the green chemistry were followed, i.e., diminishing the amount of the sample (~1 g w.w. for leaves and rhizomes) and solvent required [36]. This is of special interest when studying living organisms playing key roles in the environment. Therefore, this method would allow for conducting periodical analysis without compromising the meadows' ecosystem services. For minor interference with the meadow's health, sampling programs should be conducted during summer. In this period, leaves reach the maximum maturity and height, gathering chemical information for about 1 year.

Additionally, in all observed articles, extraction methods were time-consuming. In contrast, the extraction methods employed in this study proved to be faster (<15 min) compared to the others, such as the Soxhlet extraction (24 h; Table S8).

4.2. Target Analysis: PCBs

For the first time, the PCB congeners 44, 81 and 167 were detected in *P. oceanica*. In fact, PCB 44, the most abundant compound in rhizomes and sediments, was not considered before in *P. oceanica*-related studies. It is also important to highlight that, under our experimental conditions, PCBs were only noticed in three sampling areas from the Alicante Region (ALI5, ALI6 and ALI7). Here, all rhizomes showed PCB residues, and only ALI5 reflected contamination in the vegetated sediment. This distinction between vegetated sediment and non-vegetated sediment was also seen in other seagrass beds from Florida [37].

Clear compartmentation was spotted at ALI5, where PCB congeners showed a matrix-specific distribution based on their n-octanol/water partition coefficient (log K_{ow}). The PCBs with a low and high log K_{ow} were seen in the sediments (Table 2 and Table S1). On the contrary, only PCBs with a log K_{ow} lower than 7 were noticed in the rhizomes, except for PCB 167 (7.5 log K_{ow}), implying that the physicochemical properties of the PCBs might be influencing their fate and bioaccumulation, as observed in mangroves ecosystems [38].

For a primary evaluation of the Alicante PCB concentrations and their potentially harmful effects on the marine environment, the results were compared to the BAC, to the Effects Range–Low (ERL, i.e., concentrations in the sediments associated with biological effects) and the Environmental Assessment Criteria limits (EAC; below such concentration on marine species, chronic effects are not expected to occur). For instance, the ∑7 PCBs in sediments from ALI5 were above background levels or BAC but did not reach ERL (11.5 μg kg^{-1} d.w.). On the other hand, rhizomes were compared to internationally recognized bioindicators, the mussel *M. galloprovincialis* and the fish *M. barbatus*. In general, several PCB congeners were above BAC, even exceeding EAC for mussels in the cases of the congeners PCB 28+31 and PCB 52 (Table S9). Note that no BAC or EAC for *P. oceanica* has been described yet, and, therefore, PCB concentrations in the rhizomes cannot be truly evaluated or classified as low, moderate or high.

In comparison to other studies, the ∑PCB values encountered in the rhizomes were similar to those reported in the leaves of the Central Eastern Tunisia Coast or inside the interval when considering ∑PCB7 [15]. Note that, to our knowledge, no other study delved into PCBs in *P. oceanica* rhizomes; thus, the values were compared to the only study conducted in leaves. Individual congener concentrations were generally greater in rhizomes than in fish and mussels from the Mediterranean Spanish coast and Tunisia but were surpassed by the values observed in the Adriatic Sea (Table S10) [15,39].

As for the ALI5 vegetated sediments, both ∑7 PCBs and ∑PCBs were of several orders of magnitude greater than those detected in the Central Eastern Tunisia Coast [15] and higher than those in other locations from the Mediterranean Spanish coast [40] and in the Rosseta Estuary in Egypt [41]. The values encountered in this study were surpassed by certain concentrations detected in the River Mouth Fiumicino Canal [42] or the Port of

Trieste in Italy (Table S10) [43]. The results obtained in the Alicante and Murcia regions were in line with previous studies conducted in these areas, where PCB congeners were not detected, or the levels were below EAC limits [44]. Likewise, the non-detection of PCBs in the sampling areas belonging to the Almeria region concurs with the results obtained in past published information on marine sediments [30,31] or on monitoring programs in sediment and mussels, where very low levels for the ∑7 PCBs were observed (<2.5 μg kg^{-1} d.w.) [45].

4.3. Target Analysis: Priority Pesticides

The three priority pesticides detected and quantified in *P. oceanica* have been reported in this study according to the published data: trifluralin, chlorpyrifos and isodrin. Likewise, the analytes were observed only in rhizomes, and none were observed in *P. oceanica* leaves or sediments. On the other side, the priority pesticide levels detected in rhizomes cannot be assessed since a specific EAC for the *P. oceanica* marine plant or matrix is lacking, as well as for other marine vascular plants or biota. However, the estimated chlorpyrifos concentrations reported for *P. oceanica* rhizomes surpassed the predicted no-effect concentration of 0.032 μg kg^{-1} d.w. established for marine sediment by the WFD, which gives an approximated idea of the magnitude [40].

Similar to the PCBs case, since no other studies have reported pesticides in rhizomes, the obtained data have been compared to those described for leaves, mussels, fish and other surficial marine sediments. The ∑Pesticides found in rhizomes from the Alicante Region were always of several orders of magnitude smaller than those observed in leaves and fish from the Central-Eastern Tunisia Coast, such as *Sparus aurata* and *Sarpa Salpa* (Table S11) [15]. Here, the presented values were also lower than those found in mussels along the Adriatic Sea coast [46]. Comparing the rhizomes to the sediments from other areas, ALI6 and ALI7 showed similar values to those encountered in the Rosseta Estuary in Egypt [41] but slightly higher values than those reported in Tunisia sediments, the River Mouth Fiumicino Canal or concentrations found along the Mediterranean Spanish Coast, as can be observed in Table S11 [15,40,42].

The findings reported here are supported by previous studies. For instance, in Alicante and Almeria, regions that are well known for their elevated intensive agricultural activities, no priority pesticides were detected during the initial evaluation of the Marine Strategy [44,45]. The agreement between the results presented here and the published literature is also attained in the case of the Murcia Region; according to past studies, no priority pesticides were found at concentrations higher than the LOQ at the exterior of the Mar Menor Lagoon in 2009. Nonetheless, two priority pesticides were detected later in autumn of 2010 (chlorpyrifos and simazine), and one was detected in spring of 2010 (chlorpyrifos), suggesting a potential seasonal factor altering the priority pesticides' presence at this location [47].

4.4. Suspect Analysis: Current-Use Pesticides

An unprecedented analysis aiming at current-use pesticides of concern has been conducted in *P. oceanica*. This analysis was also applied to surface sediments. Several areas from Alicante and Almeria showed some sort of pesticide residue. Overall higher mean values were observed in the eastern sampling areas of Almeria (Table 4). Additionally, a wider distribution was observed in terms of matrices compared to PCBs or the priority pesticides, being detected in leaves, rhizomes and sediments. However, these analytes gathered preferably in the biotic compartment, where they seemed to follow a tissue-dependent distribution, except for 1,4-dimethyl naphthalene, which was detected in both matrices. This preference for the biotic compartment concurs with the published information about the elevated bioaccumulation efficiency of *P. oceanica* over the sediment [15,18].

Leaves' concentrations were especially remarkable at CG3, followed by V2, ALI7, ALI5, ALI6 and ALI4 (Figure S5, Table 4). The plant growth regulator 1,4-dimethyl naphthalene found at V2 can be ascribed to the closeness to the Almanzora watercourse, surrounded by

agricultural activities. At the four Alicante sampling stations, 1,4-dimethyl naphthalene and 2-phenylphenol (fungicide) were detected, as well as difenoconazole and tetraconazole (fungicides) in ALI7. Several activities occur in these four sampling areas, which might be acting as potential contamination sources. For instance, the relatively urbanized areas are influenced by seasonal tourism or the closeness to watercourses and breakwaters (at ALI4 and ALI6), as well as urban stormwater runoff or septic tanks (at ALI6 and ALI5). Additionally, ALI4, ALI5 and ALI6 have a long record of non-irrigated and irrigated agriculture in their surroundings.

In the case of the rhizomes, ALM3 and CG4 gathered the greatest concentrations, represented by the pyrimethanil fungicide. Here, the potential sources also consist of seasonal watercourses and different agricultural activities. For instance, at ALM3, greenhouses and irrigated extensive crops are predominate, whereas in CG4, non-irrigated crops are found.

As for the sediment, several areas of Almeria (RM6, C2 and V1) and Alicante (ALI1, ALI2, ALI3 and ALI7), usually close to seasonal watercourses or crops, presented signals of the prallethrin insecticide commonly employed in household products for mosquitoes' control. At these stations, no pattern or differentiation was identified between the non-vegetated and vegetated sediment.

The current-use pesticide concentrations measured inside the MPA were unexpected. Part of the contamination may originate in external sources, similar to what occurred in Santa Maria, another MPA from the Mediterranean Sea which presented a surprisingly elevated amount of microplastics [48]. It is well known that POPs may undergo long-range transport through ocean currents or atmospheric movements [49], affecting places that were meant to be pristine.

In summary, neither PCBs nor priority pesticides were detected in the leaves. Considering that *P. oceanica* regenerates its leaves periodically (~1 year) and the greater accumulation capacity (compared to the sediments) [15], the non-detection in the leaves or in the relatively long-lived rhizomes (~30 years) might indicate their absence (or concentrations below LODs) in the near water column. This would correlate with the decline in their synthesis and usage since their prohibition. Therefore, rhizomes might have been sequestering coastal POPs for years. Its monitoring would be of great interest in delving into near-past chemical events. Nonetheless, considering the key role of *P. oceanica* in carbon sequestration and other ecosystem services, rhizome monitoring should be performed only when the background or baseline information of a coastal environment is lacking.

Additionally, in other studies, *P. oceanica* meadows have been seen to affect the coastal water fluxes and act as a barrier, enhancing deposition and trapping organic and inorganic particles, even plastic debris, for years [50,51]. Therefore, in the case of the PCBs at ALI5, their presence in vegetated sediments and rhizomes alike could be due to the *P. oceanica* canopies preventing land-based POPs (adsorbed to particles, organic matter or microplastics) from entering into marine dynamics [52–54]. This would explain, to an extent, their non-detection in the non-vegetated surficial sediments from shallow waters, which, without the *P. oceanica* protection, are exposed to marine motion, such as currents and waves, and, therefore, to a stronger resuspension and remobilization [55]. Contrarily, the current-use pesticides employed nowadays and probably in the water column are found in leaves, rhizomes, non-vegetated sediment and those covered by the meadow.

5. Conclusions

Novel, sustainable and eco-friendly methods were optimized and validated to simultaneously detect PCBs and priority pesticides in the protected *P. oceanica* (rhizomes and leaves) and surficial sediments. These practical techniques allow for a reliable extraction without compromising the organism's integrity. This leaves open the possibility to conduct sustainable monitoring programs in this interesting and unique matrix. Additionally, for the first time, an ultra-sensitive analysis using the advantages provided by the GC combined with HRMS (sensitivity, selectivity and retrospective analyses) was conducted, saving time and resources.

These methods allowed for the satisfactory monitoring of several pollutants in marine samples from near-shore environments of the Mediterranean Spanish Coast. Overall, PCB contamination appeared preferentially in the rhizomes, followed by the sediments. For the first time, the PCB congeners 44, 81 and 167 were detected in *P. oceanica*. The values reflected the concentrations of PCBs in rhizomes exceeding the BAC for biota, even surpassing the EAC for mussels in some cases. Likewise, priority pesticides were also found in the rhizome of one sampling area of Alicante, detecting, for the first time, trifluralin, chlorpyrifos (which surpassed the predicted no-effect concentration for sediments) and isodrin.

An additional screening of current-use pesticides was conducted in leaves, rhizomes and sediments. To our knowledge, such screening was not performed before in *P. oceanica* and permitted the identification of another 13 compounds in the study areas of Almeria and Alicante. The current-use pesticides of concern presented a wider distribution compared to the banned or regulated compounds (PCBs and priority pesticides), likely due to the current usage in agriculture activities.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/chemosensors10120531/s1, Table S1: The target PCBs, pesticides and IIS included in this study with their corresponding CAS number and n-octanol/water partition coefficient (log K_{ow}); Table S2: Sampling areas description, from west to east orientation. Non-vegetated sediment (▒, n = 23), leaves (◊, n = 21), rhizome (■, n = 20), vegetated sediment (▓, n = 20); Table S3: Oven program; Table S4: Retention time and *m/z* ions selected for the quantification and confirmation of the PCB and pesticide compounds in the present study and the IIS; Table S5: Minimum and maximum recoveries (%) per class of POP regarding the extraction procedures in *P. oceanica* leaves and rhizomes; Table S6: Minimum and maximum recoveries (%) per class of POP regarding the extraction solvent employed during the UAE of surficial sediments; Table S7: Current-use pesticides identified in this study and their corresponding RT, molecular weight, log K_{ow} and quantification and confirmation ions; Table S8: Summary of the known POPs extraction methods in *P. oceanica*. PLE: Pressurized Liquid Extraction; Table S9: PCBs concentration in rhizomes (μg kg^{-1} w.w.) compared to BAC and EAC for the fish (*M. barbatus*) and mussels (*M. galloprovincialis*) used by OSPAR and the Marine Strategy Framework Directive (MSFD). Wet weight values were obtained by considering the moisture content (%) of each sample; Table S10: PCBs (μg kg^{-1} d.w.) concentrations detected in the Mediterranean Sea[a,b,c,d]. Only the sampling areas where contamination was detected have been included in the table; Table S11: Priority pesticide concentrations (μg kg^{-1} d.w.) detected in the biota and sediments from other studies; Figure S1. Maps with the three regions belonging to the Mediterranean Spanish coast surveyed in this study: (A) Almeria region, (B) Murcia and (C) Alicante: stations ALI1, ALI2, ALI3, ALI4, ALI5, ALI6 and ALI7. Figure made by the author using Ocean Data View (Schlitzer, R., Ocean Data View, https://odv.awi.de, 2021); Figure S2: Location of ALI5, ALI6 and ALI7 sampling areas from Alicante and their corresponding ΣPCBs and ΣPriority Pesticides values (μg kg^{-1}) in rhizomes (RHIZ) and vegetated sediment (V-SED). Images retrieved from Google (©Images 2022 CNES, NOAA, U.S. Navy. NGA, GEBCO, TerraMetrics, Landsat, Copernicus); Figure S3: Extracted ion chromatogram and mass spectrum of PCB 153, 138, 128, 167 and 157 isomers detected in the vegetated sediment at ALI6; Figure S4: Extracted ion chromatogram of trifluralin, chlorpyrifos and isodrin detected in the rhizomes at ALI5; Figure S5: ΣCurrent-use pesticides of concern (μg kg^{-1}) seen in the different matrices of the aquatic plant *P. oceanica*, vegetated sediments (V-sed) and non-vegetated sediments (NV-sed) among different sampling stations.

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