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Organochlorine compounds and polycyclic aromatic hydrocarbons in mussels from Ria de Vigo (the Northern Spanish coast). Current levels and long-term trends (2010–2019). Relationship with human pressures

N. Carro *, J. Cobas, I. García, M. Ignacio, A. Mouteira, M. Miranda, L. Picado

Instituto Tecnolóxico para o Control do Medio Mariño de Galicia, INTECMAR, Consellería do Mar. Xunta de Galicia, Peirao de Vilaxoán s/n. 36611, Vilagarcía de Arousa, Spain

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ABSTRACT

OCs (organochlorine compounds), such as PCBs (polychlorinated biphenyls) and OCPs (organochlorine pesticides), and PAHs (polycyclic aromatic hydrocarbons) were determined in wild and raft mussels collected in several stations from Ría de Vigo during the period 2010–2019. The concentration levels of ΣPCBs ΣDDs (sum of pp'-DDE and pp'-DDD metabolites) and ΣPAHs ranged from 22.1, 2.63 and 7.55 ng g^{-1} to 58.0, 6.89 and 25.2 ng g^{-1} dry weight, respectively. The concentrations of isomers of PCBs in mussel were in the order hexachlorobiphenyls > pentachlorobiphenyls > tetrachlorobiphenyls > trichlorobiphenyls. PCBs 153 and 138 were the most abundant in all samples. Pp'-DDE was the predominant metabolite (70.8% of ΣDDs). The main PAHs in decreasing order of abundance were CHR > BbF > BaA > IcdP. Three biological parameters: lipid content, shell length and condition index have also been investigated. Univariate techniques confirmed that levels of many compounds presented significant relation ($p<0.05$) with two of the biological parameters (shell length and condition index). In the case of lipid content, only PCB180 congener was significantly related. Multivariate techniques of data exploration such as Principal Component Analysis (PCA) showed that spatial distribution of PCBs, OCPs and PAHs levels appeared in the studied samples and was justified by local human pressures. Temporal trends (linear regressions) showed a decrease, mainly of OCs levels, along the period 2010–2019.

1. Introduction

OCs (organochlorine compounds), such as PCBs and OCPs are considered as pollutants of special concern in the aquatic environment because of their high capacity of bioaccumulation and biomagnification in marine organisms (Convention, 2001). PCBs (polychlorinated biphenyls) were used in electrical equipments such as transformers and capacitors, in industrial products like hydraulic fluids, in paints and plastics as plasticizers, and so on. PCBs are related to reproductive problems, endocrine disruption and immunodeficiency in marine organisms (Ross et al., 2013). OCPs (organochlorine pesticides) are synthetic products used widely in agricultural activity from the 1920s through 1970s. They have been related to liver and kidney cancer and endocrine and reproductive diseases in organism. The production of PCBs and OCPs was banned in most of developed countries since 1979 and 1970, respectively (UNEP, 2003). Polycyclic aromatic hydrocarbons (PAHs) are originated from natural and anthropogenic sources, some accidental such as oil spills or forest fires, and others intentional, as incomplete combustion of organic materials in human activities (Baumard et al., 1998). PAHs have been considered as possible carcinogenic and teratogenic in marine organisms (Burgess, 2009). However PAHs levels decrease in the food web due to their fast metabolism in higher organisms.

PCBs and OCPs have been classified as POPs (persistent organic pollutants). They are persistent, and have long environmental half-lives and long-range atmospheric transport (Luna-Acosta et al., 2015). Those properties are the reasons why their presence is justified in remote areas. Some compounds of these families, such as hexachlorobenzene (HCB) from OCPs and naphthalene, anthracene and fluoranthene from PAHs, are listed as priority pollutants by the Water Framework Directive (WFD; Directive 2013/39/EU).

Bivalve molluscs, mainly mussels and oysters, have been considered as bioindicators in order to assess spatial distribution

* Corresponding author.
E-mail address: ncarro@intecmar.gal (N. Carro).

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and temporal trend of marine systems and to identity potential sources of contamination. In the last decades, many coastal monitoring programmes have been established by using mussels as sentinel, Mussel Watch Programmes (Ifremer, 2006; Bellas et al., 2011; OSPAR, 2012, 2015). Mussels accumulate contaminants from marine environment giving an integrative measure of levels in seawater. Levels of contaminants found in mussel also allow knowing if governmental measures reduce their load on marine systems. On the other hand, seafood control is also crucial because it is known that sea products contribute around 15% to intake of total human protein (FAO, 2006). The content of PCBs and PAHs has been regulated in fishery products by European Union (EU, 2011a,b)

The Ría de Vigo (NW, Spain), the most southern estuarine bay of Galician Rías, is densely populated, Vigo town has one of the most important fishing ports in Europe. In addition to the important fishing and tourism activities, the shellfish culture contributes significantly to the socio-economic developing of zone. The culture of mussel cultivation in rafts, where they are hung from ropes, is a traditional practice in Galicia. During the 10–12 months period, mussel grows until commercial sizes (Pérez-Camacho et al., 2013). In the last decade, the mussel harvest in Spain is in a range between 170.000 tons and 240,000 tons. For example, in 2018 the mussel harvest was estimated in 273.600 tons and a total value of 133.2 million Euros. Galician production represents 97% of the total national mussel (Apromar, 2019). With very limited exceptions, wild mussel is not marketable in Galicia but it is a good indicator of the quality of the marine environment.

The intense maritime traffic, commercial activities, discharges of domestic, industrial and agricultural wastes become a great concern due to intentional or unintentional release of chemicals compounds into the sea, increasing the human pressure on the Rías. Along the Ría de Vigo, several studies have been performed dealing with temporal trends and spatial distribution of environmental pollutants, such as PCBs, OCPs and PAHs (Alvarez-Piñeiro et al., 1995; Carro et al., 2004; Viñas et al., 2009; Carro et al., 2010; Bellas et al., 2011; Carro et al., 2014).

This research represents a part of Galician (N.W., Spain) monitoring programme, ten years, focused on priority pollutants in wild and raft mussels from estuarine bays. The principal aim of this work is to know contamination by PCBs, OCPs and PAHs in the Ría de Vigo for the period 2010–2019, relating these concentrations to biological parameters of mussel (lipid content, shell length and condition index) and investigating the status and temporal trends, and later their relationship with the human pressures.

2. Experimental

2.1. Reagents and standards

Solvents used were supplied by Merck (Darmstadt, Germany), Carlo Erba (Milan, Italy), Sigma-Aldrich (St. Louis, MO, USA) and VWR Prolabo (Fontenay-sous-Bois, France). The different sorbents employed were purchased from Merck, Agilent Technologies (Santa Clara, CA,USA), Sigma-Aldrich and Supelco (Bellefonte, PA, USA).

Analytical reagent grade PCBs (IUPAC numbers: 31, 28, 52, 101, 118, 153, 105, 138, 156 and 180), OCPs (pp′-DDE, pp′-DDD and HCB) were purchased from Dr. Ehrenstorfer (Augsburg, Germany). PAHs in acetonitrile (ACN) (SRM 1647e) were supplied by NIST (Gaithersburg, MD, USA). Eight compounds of this mixture are considered according to scientific opinion of European Commission on PAHs (EFSA, 2008), benz[*a*]anthracene (BaA), chrysene (CHR), benzo[*b*]fluoranthene (BbF), benzo[*k*]fluoranthene (BkF), benzo[*a*]pyrene (BaP), dibenz[*a,h*]anthracene (DBahA), benzo[*ghi*]perylene (BghiP) and indeno[1,2,3-*cd*]pyrene (IcdP).

2.2. Apparatus

PCBs and OCPs (OCs) were quantitatively analysed by a gas chromatograph using an electron capture detector (ECD-Perkin-Elmer Autosystem, Waltham, MA, USA). A TRB-5 (Teknokroma, Spain) 5% diphenyldimethyl siloxane capillary column (60 m × 0.20 mm i.d. × 0.4 μm phase thickness) was used. The chromatographic conditions were: the column temperature programme was 90 °C (3 min) to 215 °C (40 min) at 30 °C/min and to 275 °C (30 min) at 5 °C/min. The injector temperature (splitless mode, 1.8 min) was 270 °C. The ECD temperature was 365 °C. Carrier gas was hydrogen (Air Liquide, Spain) The confirmation of OCs was performed using a gas chromatograph coupled to an ion trap mass spectrometer (Varian 3800 coupled to Varian Saturn 2000) with a wave-board for MS-MS analysis. The system was also equipped with a Varian 8200 CX autosampler (Varian, Walnut Creek, LS, USA). The capillary column and experimental conditions were identical to those described before. Carrier gas was helium (Air Liquid, Spain). Automatic gain control (AGC) was used to optimize the sensitivity, 5000 was the selected value for the mode of the electron impact ionization (EI).

PAHs were analysed by an Alliance 2695 HPLC system equipped with a 2475 fluorescence detector (Waters, Milford, MA, USA). The analytical column was a Waters PAH C_{18} 250 mm × 4.6 mm × 5 μm. PAHs were separated using a binary solvent system of acetonitrile and water at 1.0 mL min^{-1}. The gradient elution programme was as follows: initial condition, 50% ACN for 2 min, then linear ramps to 60% ACN within 14 min, 80% ACN within 21 min and 100% ACN within 12 min with holding for 5 min. The excitation/emission wavelengths pairs (nm) were: 275/420 for BaA and CHR; 290/430 for BbF, BkF and BaP; 284/400 for DahA and BghiP; and 286/500 for IcdP.

2.3. Samples

Wild mussels (WM) (*Mytilus galloprovincialis*) were collected once a year (February–March) from two stations in the Ría de Vigo (N.W. Spain) with codes WM 1 and WM 3, and twice a year (February–March and September–October) from two stations WM 2 and WM 4. Raft mussels (RM) (*Mytilus galloprovincialis*) placed on the rope at 1, 5 and 10 m of depth were collected once a year (May) from four stations (polygons) in the Ría de Vigo with codes RM 1, RM 2, RM, 3 and RM 4 and twice a year (May and October–November) from RM 5. All samples were sampled in the period from 2010 to 2019 (See Fig. 1a and b).

Wild mussels were collected manually in low tide. Analyses of mussel were performed in pools of 30 individuals, in the case of raft mussel ten individuals by each depth, 1, 5 and 10 m. After removing the shell, mussel was stored at −30 °C before being lyophilisated for a week in a Labconco Freezone 12 Plus freeze-drier (Kansas City, USA). After lyophilization, samples were homogenized using a mixer mill with zirconium oxide balls and stored at room temperature until analysis.

2.4. Procedures

For OCs determination, 5 g of lyophilized sample were extracted with 150 mL of dichloromethane–pentane (1:1 v/v) using a Soxhlet device for 8 h. The extract was concentrated to 1 mL. An aliquot of the extract was used to determine gravimetrically the lipid content. Extracts were initially cleaned using 6% deactivated Alumina column chromatography. The two fractions (PCBs and OCPs) were further separated on a 1% deactivated Silica column. PCBs fraction was eluted with isooctane and OCPs fraction with a mixture of isooctane-diethyl ether (8.5:1.5 v/v). The internal

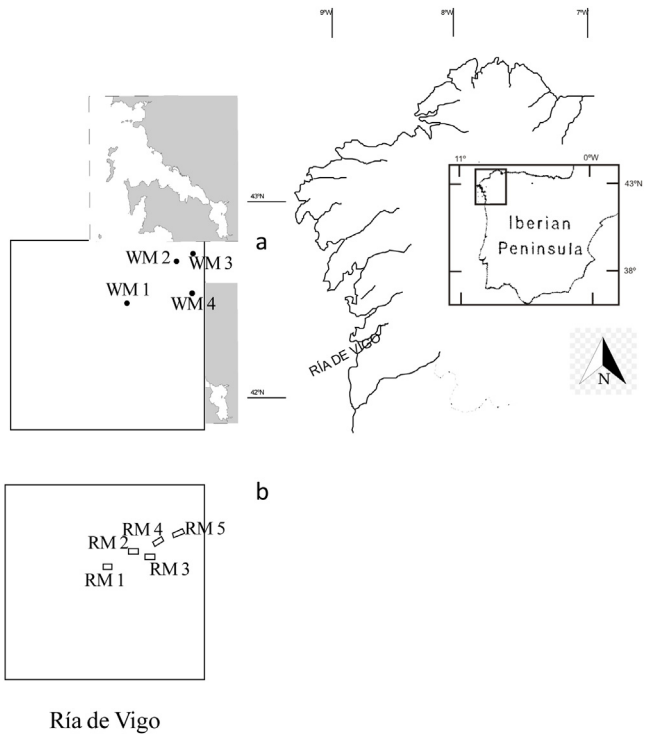

Fig. 1. a: Map of sampling points of wild mussel coming from Ría de Vigo. b: Map of sampling points of raft mussel from Ría de Vigo.

standard PCB 155 was added to each fraction prior to gas chromatographic analysis (González-Quijano and Fumega, 1996; Carro et al., 2010).

For PAHs, 1.2 g of freeze-dried sample ground with 2.5 g of Hydromatrix® was extracted using an ASE 200 System (Dionex, CA, USA) (2000 psi, 100 °C oven, 5 min. static time, n-hexane-acetone, 60% flush volume, 150 psi for 45 s nitrogen purge). The extract was concentrated to 3 mL before the purification on 10% deactivated Alumina column chromatography. After solvent exchange with acetonitrile, the extract were made up to 2 mL, filtered through a 0.20 μm PTFE filter and injected into the HPLC-FLD system (Cobas-Sáenz, 2008).

Condition index (CI) was calculated on a total of 30 individuals per sample taking account shell and body weights of each sample. The CI is the quantitative relationship of the soft tissue weight to its overall weight (the weight of soft tissue + shell).

2.5. Quality assurance

For OCs, accuracy of determination method was periodically checked by analysing spiked mussel or certified reference material (freeze-dried matrix) supplied by National Institute of Standards and Technology (NIST 2974a) (Gaithersburg, MD, USA). The recoveries for PCBs and OCPs were determined from six replicate analyses of spiked mussels and ranged from 60 to 115%. The precision expressed as relative standard deviation (RSD) was between 6 and 20% for PCBs and between 3 and 20% for OCPs. The limits of detection (LODs) were 0.01 ng g^{-1} for all organochlorine compounds in term of freeze-dried samples. Blank samples were analysed to check for interferences or contamination from solvents and glassware.

For PAH, the quality control activities imply the analysis of blanks, duplicate samples and spiked samples. The whole analytical method performance is also checked periodically using the certified reference material NIST 2974a. PAHs recoveries ranged from 70 to 110% and the reproducibility was below 20% RSD. The LODs referred to freeze-dried sample ranges between 0.12 and 0.35 ng g^{-1}.

For both OCs and PAHs, the limits of detection were calculated as the compound concentration giving a signal-to-noise ratio of three.

The laboratory participates annually in international intercomparison exercises organized by the Quality Assurance of Information for Marine Environmental Monitoring in Europe (QUASIMEME) and is accredited according to the ISO/IEC 17025 standard for the organochlorine compounds and polycyclic aromatic hydrocarbons analysis.

2.6. Statistical analyses

Data analysis was carried out using Minitab 16 statistical Software. Statistical analysis considers as quantitative variables the levels of PCBs, OCPs and PAHs congeners and biological parameters (lipid content, shell length and condition index). The concentration of PCBs, OCPs and PAHs was expressed in ng g^{-1} dry wet, lipid content and condition index in % and shell length in mm. Qualitative variables were culture type (Wild or Raft), sampling location (Stations), sampling timepoints, situation in the Ría de Vigo (North seashore and South seashore; Inner, Middle and Outer). The outer comprises the area near the mouth of the Ría with greater depth and where RM1 station is situated. From here the estuary converges towards the head. The middle extends to Rande strait, this part is especially important because of the large number of sampling stations (WM1, RM2, RM3 and RM5) and the urban pressures of Vigo town. The inner goes from Rande strait to the head of the Ría, WM2, WM3, WM4 and RM4 stations are located here.

All data was log-transformed to achieve a normal distribution. That distribution was analysed by Kolmogorov–Smirnov test.

The relationship between quantitative and qualitative variables was assessed by one-way analysis of variance ANOVA. When ANOVA F-test was significant post hoc comparisons were assessed by Tukey's test. Possible correlations between variables were also checked by the Pearson product-moment coefficients (r). A significance level of $p < 0.05$ was used for all analyses.

Principal component analysis (PCA) and temporal trends analysis were performed to check differences in priority pollutants levels among stations, sampling timepoints, position in the Ría in order to determine the spatial and temporal distributions of contaminants. The limit of detection value was assigned to levels reported as "not detected". "Not detected" compounds contributed to the sum of congeners as limit of detection value.

3. Results and discussion

3.1. Biological parameters

Biological parameters, lipid content (% dry weight), shell length (mm) and condition index (%), of wild and raft mussels collected from Galicia coast during the period 2010–2019 are shown in Table S1.

Only shell length and condition index presented a statistically significant difference between stations. This occurred in both wild and raft mussel ($p < 0.05$). In wild mussel, shell length increased from WM1 to WM4, from the middle to inner area. In the case of raft mussel the opposite happened, shell length decreased from RM1 to RM5, being logical because RM1 is located in the mouth of estuary where there is a bigger oceanic water renewal that is nutrient rich. Growth of mussels from inshore rafts was smaller than mussels from rafts placed in the mouth of the Rías which illustrates food limitation at inshore sites (Navarro et al., 1991).

Table 1
The mean levels of organochlorine compounds, Σ PCBs (sum of PCBs 31, 28, 52, 101, 118, 153, 105, 138,156 and 180), Σ DDs (sum of pp'-DDE and pp′-DDD) and Σ PAHs (sum of Ba, BbF, BkF, BaP, DBahA and IcdP) expressed in ng g^{-1} dw (dry weight) in wild mussel from four sampling points and in raft mussel from five polygons in Ría de Vigo during the period 2010–2019. Mean and standard deviation (SD) of all years.

ΣPCBs (ng g^{-1})	Wild				Raft				
	WM1	WM2	WM3	WM4	RM1	RM2	RM 3	RM4	RM5
2010	33.4	57.5	44.4	64.7	24.7	50.3	50.3	44.4	59.1
2011	73.6	61.1	52.9	59.3	83.5	40.5	172	95.4	50.1
2012	62.6	62.5	44.4	64.1	10.7	34.9	51.2	61.2	59.1
2013	50.0	52.8	39.8	69.2	19.6	35.4	39.1	40.4	50.7
2014	44.2	72.1	46.9	68.8	19.7	26.5	23.2	34.9	58.5
2015	42.6	36.5	43.5	70.0	13.3	26.4	47.5	35.4	35.0
2016	38.0	44.8	34.3	49.6	13.9	23.9	29.2	21.6	32.7
2017	33.1	43.7	32.8	42.5	13.6	23.7	36.9	27.3	43.4
2018	34.5	54.0	34.7	49.6	11.9	18.0	22.4	20.7	36.4
2019	24.1	48.8	15.6	42.5	10.1	22.4	33.2	28.5	26.9
Mean	43.6	53.4	38.9	58.0	22.1	30.2	50.5	41.0	45.2
SD	14.9	10.5	10.3	11.0	22.1	9.87	43.9	22.5	12.0
ΣDDs (ng g^{-1})	Wild				Raft				
	WM1	WM2	WM3	WM4	RM1	RM2	RM 3	RM4	RM5
2010	28.9	3.98	4.28	2.37	3.23	0.09	1.17	2.14	7.72
2011	17.0	21.1	3.96	5.88	2.31	2.58	24.3	7.01	3.58
2012	4.77	6.50	4.32	5.92	5.03	3.73	2.64	3.65	4.12
2013	2.54	2.83	4.53	3.44	2.42	3.38	2.75	3.69	4.70
2014	2.45	3.53	4.56	3.11	1.95	4.35	4.24	4.09	3.90
2015	3.94	4.02	5.76	6.82	2.19	2.43	3.16	2.62	3.21
2016	2.97	3.50	2.66	3.38	3.05	1.32	2.18	2.00	2.42
2017	3.37	3.75	2.90	8.75	2.42	4.06	3.37	2.27	3.55
2018	1.87	3.60	2.83	3.58	3.19	1.50	0.94	0.91	0.95
2019	1.10	3.54	2.16	3.30	0.54	3.98	1.31	4.74	4.03
Mean	6.89	5.63	3.80	4.66	2.63	2.74	4.61	3.31	3.82
SD	8.96	5.50	1.12	2.06	1.15	1.41	7.01	1.73	1.73
ΣPAHs (ng g^{-1})	Wild				Raft				
	WM1	WM2	WM3	WM4	RM1	RM2	RM 3	RM4	RM5
2010	33.7	30.5	15.8	40.1	14.1	29.9	39.7	42.8	57.6
2011	33.3	23.4	19.4	27.7	16.8	27.6	40.4	35.9	25.9
2012	40.7	19.5	16.4	25.0	4.94	14.6	21.1	32.9	21.9
2013	10.4	29.6	17.1	32.4	7.68	11.1	28.5	25.8	16.6
2014	16.7	15.4	13.8	19.5	6.09	9.7	17.0	17.2	12.5
2015	26.3	29.6	10.9	19.9	–	–	30.9	37.3	24.2
2016	20.1	25.6	22.5	22.6	4.92	12.7	21.0	17.1	12.6
2017	21.3	18.0	16.1	20.0	4.84	11.8	18.0	18.4	11.5
2018	23.3	33.8	19.1	26.2	3.56	5.76	11.1	9.07	23.2
2019	21.3	25.2	15.8	17.1	5.01	13.0	19.1	15.2	15.1
Mean	24.7	25.1	16.7	25.0	7.55	15.1	24.7	25.2	22.1
SD	8.99	6.01	3.10	6.99	4.67	8.13	9.85	11.4	13.6

– Data not presented.

The condition index is an indicator of biological status of mussel. In relation to wild mussel, WM2 and WM4 had the highest values of CI (35.6 and 37.4%, respectively), these stations were placed in the most internal area of the Ría. Raft mussels had CI values above 40% confirming that nutrient availability was higher than in wild mussels because the cultivated mussels are permanently submerged in water with a regular available food.

For lipid content, statistical analysis indicated no significant difference among the sampling stations, for wild mussel the highest value was in samples from WM1 (6.86%) and for raft mussel in samples from RM1 (8.29%), the most external stations (increased availability food).

There were no significant annual differences of the three biological parameters studied (condition index, lipid and shell length) ($p > 0.05$) when all samples were considered in data analysis. However when only raft mussel samples were examined, shell length varied over the years (ANOVA, $p = 0.018$), it can be observed in SDs of Table S1, in wild mussel the RSDs (relative standard deviations) are around 3%–6% and in raft mussel around 7%–9% .

3.2. Chemical levels

The levels and mean levels in the case of two sampling timepoints of PCBs (ΣPCBs, sum of PCBs 31, 28, 52, 101, 118, 153, 105, 138, 156 and 180), OCPs (ΣDDs, sum of pp′-DDE and pp′-DDD) and PAHs (ΣPAHs, sum of BaA, BbF, BkF, BaP, DBahA and IcdP) in wild (WM) and raft mussel (RM) tissues during 2010–2019 are listed in Table 1. All levels found in the studied samples for legacy compounds (PCBs and PAHs) were below the maximum limits established by European Union for human consumption (EU, 2011a,b). In Table S2 levels of congeners of PCBs, (PCBs 31, 28, 52, 101, 118, 153, 105, 138,156 and 180), OCPs (pp'-DDE, pp′-DDD and HCB) and PAHs (Ba, BbF, BkF, BaP, DBahA and IcdP) expressed in ng g^{-1} dw (dry weight) in wild mussel (WM) and raft mussel (RM) from sampling points in Ría de Vigo for the period 2010–2019 are shown.

3.2.1. PCBs levels and profile

The levels of ΣPCBs in studied samples ranged from 10.1 ng g^{-1} dw (RM1 station in 2019) to 172 ng g^{-1} dw (RM3 station in 2011). These values were consistent with those found in other aquatic systems, such as in mussel (*Mytilus galloprovincialis*) from

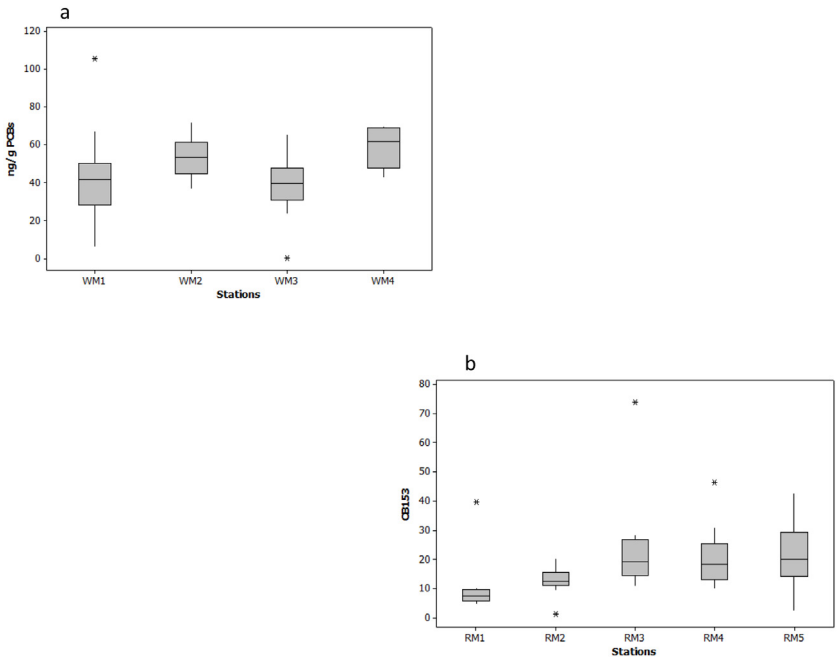

Fig. 2. a: Average levels (ng/g dw) for ΣPCBs in wild mussel coming from Ría de Vigo. b: Average levels of PCB 153 in raft mussel coming from Ría de Vigo.

Adriatic sea where PCBs levels covered from 3.7 ng g^{-1} dw to 114 ng g^{-1} dw (Bajt et al., 2019), oysters coming from Xiamen Island (China) with levels from ND (not detected) to 243 ng g^{-1} dw and from Minjiang Estuary with concentrations from ND to 6.78 ng g^{-1} dw (Chen et al., 2002) or mussels from River Ebro and Peñiscola (Spain) where PCBs levels ranged from 9.3 to 17.6 ng g^{-1} and from 7.6 to 15.7 ng g^{-1} ww (on wet weight basis), respectively (Campillo et al., 2019). Dias et al. in 2013 found levels of PCBs between 1.36 and 127.7 ng g^{-1} ww in shellfish from Sao Paulo.

In relation to wild mussels, ANOVA significant differences between stations were well-defined for ΣPCBs ($p = 0.009$) and for PCBs 52, 118, 153, 105 and 138 ($p = 0.03$, 0.020, 0.019, 0.018, 0.011, respectively). In Fig. 2a, Box–Whiskers plot of ΣPCBs levels (ng g^{-1} dw) in the wild mussel from the studied stations is shown. Samples taken at WM4 (mean levels for period 2010–2019, 58.0 ± 11.0 ng g^{-1} dw) and WM2 (53.4 ± 10.5 ng g^{-1} dw) stations were the most affected by ΣPCBs pollution. It can be because of the effect of the important domestic and industrial activities, mainly in WM4 station, and the effect of port and shipyard (mainly shipyard paints) located in WM2. The WM1 and WM3 stations were the least affected by ΣPCBs pollution with values around 40 ng g^{-1} dw. MW1 is located in the outer area of the Ría and MW3 is further away from industrial and urban sources. In general terms, ΣPCBs concentrations decreased gradually after 2015 (ANOVA, $p = 0.009$). The establishment of measures for environmental improvements has set up important controls of industrial, urban, sanitary and agricultural wastewater and rainwater discharges and has got better the collection and depuration systems of these waters, these actions have been a constant in the last decade in all Galician Rías.

With regard to raft mussels, although samples from RM3 (mean levels for 2010–2019, 50.5 ± 43.9 ng g^{-1} dw), RM4 (41.0 ± 22.5 ng g^{-1} dw) and RM5 (45.2 ± 12.0 ng g^{-1} dw) polygons were more polluted (two times or more) than RM1 (22.1 ± 22.1 ng g^{-1} dw) and RM2 (30.2 ± 9.87 ng/g dw), there was not significant difference between stations for Σ PCBs (ANOVA, $p = 0.112$). However the individual congeners, PCB153, PCB101 and PCB 156, showed differences significant ($p = 0.023$, 0.011 and 0.032, respectively) that are reflected in PCB 153 levels in the Box–Whiskers plot of Fig. 2b. RM1 station is located near the mouth of the Ría and although close to a wastewater treatment plant (WWTP) of a population of 26,000, the mean levels of ΣPCBs contamination were relatively low. The other polygons (RM2, RM3, RM4 and RM5) are placed in the middle or inner part of the estuary. The mean levels of ΣPCBs found were very low, even though RM2 is situated close to WWTP (20,000 inhabitants). The highest values of ΣPCBs were recorded in the first two years of sampling (2010 and 2011). In the following samplings (2012–2019), RM1 and RM2 had a very sharp decrease probably because of measures taken to clean up the estuary. In this case, an ANOVA significant difference between sampling years was observed ($p = 0.003$).

The profile of PCB congeners was similar for both wild and raft mussel (see Figure S1). However for two congeners, PCB 105 and PCB 180, there was a slight difference, PCB 105 contribution was higher in WM than in RM (1.21% for WM and 0.64% for RM) and PCB 180 contribution was higher in RM than in WM (4.76% for WM and 6.24% for RM). The predominating congener was PCB 153 (48.4% for WM and 48.6% for RM), followed by PCB 138 (28.0% for WM and 27.6% for RM). The same trend was recorded in mussels and other marine organisms coming from different aquatic systems such as marine fish from Persian Gulf (Jafarabadi et al., 2019), shellfish from coastal areas of Chinese Xiamen Island (Chen et al., 2002), mussels from Río Ebro in Spain (Campillo et al., 2019), blue mussels from Port Elizabeth Harbour in South Africa (Kampire et al., 2015) and from European coastal waters (Olenycz et al., 2015).

The distribution of congeners of PCBs found in this work and in the most of bibliographic references is consistent with the high bioaccumulation power of the penta and hexachlorinated compounds. The lower chlorinated compounds, such as PCBs 31 and 28, are less bioavailable for mussels because of their high volatility (Lammel and Stemmler, 2012). Other authors relate the greater presence of higher chlorinated compounds with historical applications and the presence of lower ones with recent releases (Breivik et al., 2002). Both PCB 153 and PCB 138 were commonly used in different percentages with other congeners in industrial mixtures (Aroclors 1260 and 1254) purchased by Monsato Corporation until the 1980s.

3.2.2. ΣDDs and HCB levels

The levels of ΣDDs (sum of pp′-DDE and pp′-DDD) and HCB ranged from 0.09 ng g^{-1} dw (RM2 station in 2010) to 28.9 ng g^{-1} dw (WM1 station in 2010) and from 0.01 ng g^{-1} dw (WM2 in 2010, 2015 and 2017 and RM1 and RM4 in 2019) to 1.79 ng g^{-1} dw (WM 4 in 2017), respectively (see Table S2). Levels of DDT metabolites (pp′-DDE and pp′-DDD) were similar or slightly lower to those found in mussels from the River Ebro mouth that ranged from 15.7 to 24.0 ng g^{-1} ww (wet weight) and from Peñiscola with values from 4.8 to 10.0 ng g^{-1} ww (Campillo et al., 2019). Shellfish collected in Sao Pedro and Sao Paulo Archipelago (Brazil) also presented values of the same order of magnitude, 0.87 and 24.4 ng/g ww, respectively (Dias et al., 2013). In this later work, the values of HCB were higher than those found in the present work ranging from 1.36 to 6.33 ng g^{-1} ww. Chen et al. in 2002 also found values of pp′-DDE and pp′-DDD in oyster from Chinese coast that were two to three orders of magnitude higher than those reported in this work (Chen et al., 2002).

The residual HCB levels found in these samples, in the range from 0.07 to 0.27 g^{-1} dw mean levels for the period 2010–2019 (data not shown) are due to the ban on production and use of commercial products. Nevertheless, HCB can also be generated unintentionally during processes of thermal industrial and domestic combustion (Liu et al., 2014; Gong et al., 2017), This is why some peaks of HCB with values above 1 ng g^{-1} dw were found in WM3, WM4 and RM3 stations during 2014 and 2018, 2017 and, 2014, respectively.

Pp′-DDE and pp′-DDD are the majority metabolites of DDT (insecticide banned since the 1980s). The relative concentrations of the parent compounds (op′-DDT and pp′-DDT) and their metabolites (pp′-DDE, result of the aerobic degradation of DDT and without commercial use and pp′-DDD, result of the anaerobic degradation of DDT and used also as pesticide) are very useful to give information about history of input to the environment. In spite of its prohibition, DDT is used in the third word as a control of pests. In Europe, dicofol is a pesticide based on DDT. In 1986 European Commission limited its DDT content to less 0.1%. Finally in 2008 and 2014, European Commission banned the production and use of dicofol, respectively (Regulation (EC) No 850/2004, 0000). In this Galician monitoring programme (2010–2019), pp′-DDT was found in levels below limits of detection so this pesticide was not considered in the present work. Pp′-DDE was the predominant metabolite in all samples (70.8% of ΣDDs).

The most polluted by ΣDDs wild mussel samples came from WM1 (mean level for the period 2010–2019, 6.89 ± 8.96 ng g^{-1} dw) and the least polluted ones from WM3 (mean level 3.80 ± 1.12 ng g^{-1} dw). The wild mussel station with the highest levels of HCB was WM4 (mean levels 0.26 ± 0.54 ng g^{-1} dw). For ΣDDs and HCB, there were no statistical differences between stations (ANOVA, $p = 0.415$ and $p = 0.664$, respectively). Only pp-DDD was statistically significant for sampling years ($p = 0.003$). The first samplings had the highest values of pp′-DDD, mainly from 2010 to 2012. These levels of metabolites are related to an ancient use of DDT.

In relation to raft mussel, samples from RM3 and RM5 stations had the highest levels of ΣDDs (4.61 ± 7.01 ng g^{-1} dw and 3.82 ± 1.73 ng g^{-1} dw, respectively), similarly no statistically differences between stations were observed ($p = 0.664$). RM3 station was affected by nearby discharges. RM5 is very close to a very populous city (Vigo town, 400 000 inhabitants) with a strong human impact and industrial presence. No difference between years was found ($p = 0.268$) for ΣDDs in raft mussel. Levels remained stable throughout the years, except for RM3 in 2011 where there was a peak of ΣDDs (24.3 ng g^{-1} dw).

3.2.3. PAHs levels and profile

The concentration of ΣPAHs ranged from 3.56 ng g^{-1} dw (RM1 station in 2018) to 57.6 ng g^{-1} dw (RM5 station in 2010) (ANOVA, $p = 0.01$). All congeners of PAHs, except DBahA ($p = 0.424$), presented differences between stations ($p < 0.05$). PAHs concentrations were the same order of magnitude as levels found in mussel from several sites in European coastal waters, the Bay of Faxaflói (Iceland), the Baltic Sea, the North Sea, the English Channel, the Bay of Biscay, the Mediterranean Sea, the Black Sea (Olenycz et al., 2015). In mussels (*Mytilus galloprovincialis*) from Adriatic sea (Bajt et al., 2019), total PAHs levels ranged from 5.3 to 61.8 ng g^{-1} dw with a similar range. However in Bizerte lagoon (Tunisia) (Barhoumi et al., 2016), PAHs levels in mussel (*Mytilus galloprovincialis*) ranged from 107.4 to 430.7 ng/g dw, being one order of magnitude higher than levels found in the present work. The main PAHs in decreasing order of abundance were CHR (32.8% of total PAHs), BbF (21.2% of total), BaA (11.4) and IcdP (10.4% of total). This pattern is similar to that found by Leon et al. in bivalves from a Mediterranean coastal lagoon (Leon et al., 2013) and by Rodil et al. in bivalve mollusc coming from Galician Rías (Rodil et al., 2019). All congeners except, BaP ($p = 0.061$), BghiP ($p = 0.146$) and IcdP ($p = 0.133$), presented significant differences between years.

In relation to wild mussels, samples coming from WM3 station had the lowest levels of ΣPAHs (mean level for the period 2010–2019, 16.7 ± 3.1 ng g^{-1} dw) and the other stations (WM1, WM2 and WM4) presented similar values (around 25 ng g^{-1} dw). The main congener in wild mussel is also CHR (33.7% of total PAHs) followed by BbF (20.6% of total PAHs), BaA (11.9% of total PAHs) and IcdP (9.55% of total). There was statistical significance between stations ($p = 0.033$). Neither the individual congeners nor ΣPAHs showed differences between sampling years ($p > 0.05$). Table 1 shows that unlike organochlorine compounds, PAHs did not present a clear downward trend over the years. The input sources of PAHs into the marine system are different to the OCs ones. Since they were banned, the sources of PCBs and OCPs are most likely from old pollutants discharges, whereas the sources of contamination of PAHs are most likely to be diffuse.

In relation to raft mussels, RM3, RM4 and RM5 stations presented the high levels of PAHs (mean value of all years, 24.7 ± 9.85 ng g^{-1} dw, 25.2 ± 11.4 ng g^{-1} dw and 22.1 ± 13.6 ng g^{-1} dw, respectively). RM1 station had the lowest levels of ΣPAHs, mean value 7.55 ± 4.67 ng g^{-1} dw, because it is the closest to the mouth of the Ría with a higher rate of water renewal. So, there were significant differences between stations for ΣPAHs ($p = 0.005$), BbF ($p = 0.014$), BkF ($p = 0.009$), Bap ($p = 0.003$), IcdP ($p = 0.001$) and BghiP ($p = 0.000$). In the case of raft mussels, the main congener is CHR (31.8% of total PAHs) followed by BbF (22.0% of total PAHs), IcdP (11.4% of total) and BaA (10.7% of total PAHs).

In general, the first samplings (2010–2011) presented the highest load of PAHs in raft mussel. This fact can be due to change in wastewater outfall position or improvements in water treatment of the Ría de Vigo related to mussel culture. In this case unlike wild mussel, the levels of PAHs can be related to a not diffuse pollution caused by the raft and culture activity. Except IcdP, all congeners and ΣPAHs had ANOVA year term ($p < 0.05$).

3.2.4. Correlations between environmental pollutants levels

There was a high Pearson correlation between PCBs congeners ($p < 0.05$) which indicated that they came from a common source, possibly the same Aroclor. For example, the higher chlorinated congeners and majority in industrial mixtures (PCBs 153 and 138) had a very high correlation with the other congeners (PCBs 31, 28, 118, 101, $p = 0.000$). Lower chlorinated congeners (PCBs 31, 28 and 52) and higher chlorinated ones (PCBs 118,

156) were correlated with OCPs (HCB, pp′-DDE and pp′-DDD) suggesting the same origin derived possibly from human activity, PCBs 28 and 52 were highly correlated with HCB and pp′-DDE, with p-values of 0.000. The same for PCBs 52 and 118 and pp′-DDD. The possible uses of PCBs in pesticide formulations may be the response. In all this area there is multitude of rivers and streams that drain adjacent urban and agricultural zones, later these rivers discharge sanitary and municipal wastes into the Ría.

The higher chlorinated PCBs were also correlated with all congeners of PAHs ($p < 0.05$), except with DBahA. It is well known the multivariate source of PAHs in comparison to the industrial source of PCBs, so this correlation can be related to the similar distribution of industrial sources for PCBs and PAHs.

Pp′-DDD and pp′-DDE were correlated with all congeners of PAHs ($p = 0.002–0.022$ and $p = 0.000–0.036$, respectively), except with the higher molecular weight congeners DBahA, BghiP and IcdP. The DBahA origin is related to oil spilt environments. It is the least volatility and more recalcitrant between the PAHs with the highest log Kow. These properties make DBahA different from the rest of hydrocarbons.

3.2.5. Relationship between biological parameters and priority pollutants levels

Our work revealed statistically significant relationships ($p < 0.05$) and negatively Pearson correlated between shell length and some contaminant levels (ΣPCBs, PCB 31, PCB 28, PCB 52, PCB 101, PCB 118, PCB 138, PCB 156, PCB 180, pp′-DDE, ΣPAHs, IcdP and BghiP). These findings can be observed for PCBs content in mussels from Galician Rías (Carro et al., 2015) and in other species as clams (*Ruditapes decussatus)* in Ría de Formosa (Ferreira and Vale, 1998) where PCB residues were slightly higher in the smaller than in the larger individuals.

As is well known, condition index is an important data in Watch Mussel Monitoring because it provides information about mussel biological status and the possible effects of contaminants. In this work, there was a significant relationship between the condition index and many pollutants (ΣPCBs, PCB 31, PCB 28, PCB 52, PCB 118, PCB 153, PCB 138, PCB 156, PCB 180, pp′-DDE, pp′-DDD, ΣPAHs, BaA, BbF, BkF, BaP, BghiP and CHR, $p < 0.05$). It is found that in general CI was positively correlated with the concentration level of chemical contaminants, it is logical because as mussel grows, it accumulates contaminants present in seawater.

No statistical difference between lipid content and organic contaminant levels in mussel tissues was observed ($p > 0.05$), except for PCB 180 ($p = 0.003$). There is some author that did not observe any relationship between lipid content and concentration of pollutants (PAHs, PCBs or DDTs), for example Thompson et al. (1999) who studied several bivalve species collected in the Arcachon Bay (France). However many other authors have found this relationship in aquatic biota. Carro et al. (2004) reported Pearson product–moment correlation coefficients that suggested the existence of significant relationships ($p < 0.05$) between levels of some organochlorine compounds and the lipid content in mussel from Galician Rías. Deribe et al. (2011) found relationships between log transformed DDTs and PCBs and the relative lipid levels in four fish species from Lake Koka, Ethiopia. Jafarabadi et al. (2019) also identified possible similarity of patterns and correlations between total PCBs and PCB congeners with lipid content of fish samples coming from Persian Gulf. There are no conclusive results in this respect since there is no homogeneity between laboratories for the determination of the lipid content in biota samples.

3.2.6. Influence of mussel culture

Only condition index and shell length parameters had significant differences between wild (intertidal) and raft mussel ($p = 0.000$). This can be explained by the different physiological behaviour observed between cultivated and wild mussels. The food conditions of raft mussel are very different from those of intertidal mollusc. In this last case, there is variability of the particulate matter suspended (mussel food) in the water because wild mussel is subjected to large periods of air exposure. These factors can have a strong impact on mussel growth (Labarta et al., 1997), leading to cultivated mussels have better physiological conditions than intertidal mussels (see Fig. 3a).

There is a clear ANOVA difference between wild and raft mussel for the higher chlorinated PCBs (IUPAC number 105, 118, 153, 105 and 138, $p = 0.010$, 0.042, 0.040, 0.010 and 0.042, respectively) and for pp′-DDE and BaA ($p = 0.041$). Discharges of organochlorine compounds usually occur from land to sea in contrast to oil spills that also occur from sea to sea. This is the reason why PCBs concentration was higher in intertidal mussels and PAHs level similar to both, cultivated and wild mussels (see Fig. 3b).

3.2.7. Position in the Ría

Since the Ría de Vigo is not very wide (it has widths that vary from 11 km near the mouth to 0.7 km at the head), no statistical difference of biological parameters and chemical levels in studied mussels has been observed between the sampling points located in the north and south margins of the estuarine bay. However important differences were found between biological parameters (condition index and length, $p = 0.004$ and $p = 0.000$, respectively) and chemical levels (ΣPCBs, PCBs 101, 153, 138 and 156, ΣPAHs, BaA, BaP, CHR, BbF, BkF, IcdP and BghiP, $p < 0.05$) of mussels coming from the inner, outer and middle part of the Ría de Vigo. Samples from the outermost zones had higher length and lower content in contaminants.

3.2.8. Relation with sampling timepoints

Only three stations were sampled twice a year, WM1, WM3 and RM5. The hydrological regime of the rivers and streams can influence the transport, entry and release of pollutants into the Ría de Vigo so in the WM1 and WM3 stations, samples collected in the first sampling (February–March) presented the highest levels of ΣPCBs (mean values 53.1 and 53.4 ng g^{-1}, respectively) against the levels belonging to the second sampling (September–October) (mean values 34.0 and 32.3 ng g^{-1}, respectively) (data not shown). The same was happening in WM1 and WM3 stations for OCPs and PAHs (mean values for ΣPAHs, 33.6 and 21.8 ng g^{-1} respectively in the first sampling, and 16.9 and 12.7 ng g^{-1} in the second one) (data not shown). In relation to raft mussel (RM5) occurred the opposite, the highest values of priority pollutants were present in the second sampling against the first one, for ΣPCBs 58.0 against 32.5 ng g^{-1}dw, for pp′-DDE 2.9 against 1.6 ng g^{-1}dw and for ΣPAHs 28.1 against 21.3 ng g^{-1}dw (data not shown). February and March are the wettest months in Galicia when the rivers flow really strong, in addition wild mussels live on the rocks of the coast and the loading of pollutants do not suffer the dilution effect such as it happens offshore.

3.3. Spatial environmental pollutants distribution: Relationship with human pressures

Principal Components Analysis (PCA) was used to compare the priority pollutants profiles of investigated mussel and the potential sources of contamination. PC analyses helped to identify the variables (pollutants concentrations) that contributed powerfully to the total variance of data set. The PC analyses were based on

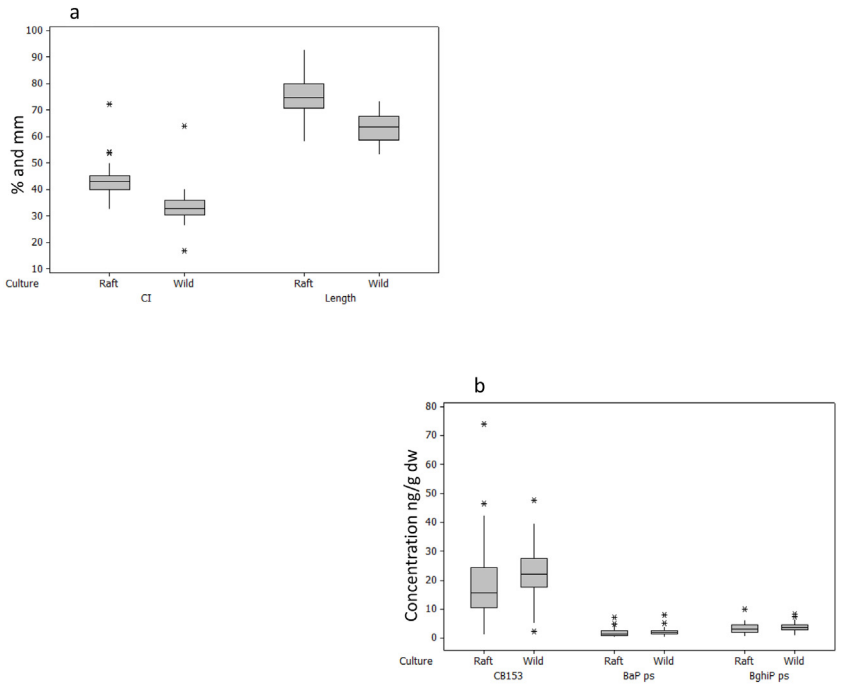

Fig. 3. a: Average levels of condition index and shell length in wild and raft mussel coming from Ría de Vigo. b: Average levels of PCB153, BaP and BghiP levels in wild and raft mussel coming from Ría de Vigo.

all significant pollutants levels (log concentration values) of the wild mussel and raft mussel samples dataset by separately.

For wild mussel, the first two principal components accounted for 77.6% of the total variance. PC1 was positively correlated with PAHs (BaA, CHR, BbF and BaP) and PC2 negatively correlated with PCBs 52, 105, 118 and 138. Figure S2a shows the plane of the two first components, the most of samples contaminated by PAHs and PCBs came from station WM4 and were distributed in the positive PC1 and the negative PC2 plane. Near this station in an earlier period, 2006–2014, there were discharges of both industrial and domestic activities with presence of pollutants, it was confirmed by high concentrations of PCBs mainly, which are the main indicators of anthropogenic pollution. The station WM4 is very close to WWTP and small urban centres. The most of samples from WM1 and WM2 stations were distributed in the plane of the positive axis of PC1 and PC2, with high levels of hydrocarbons and low levels of PCBs and collected in the first sampling (February or March). However some samples from WM1 and WM2 stations were distributed along the negative axis of PC2 with high levels of PCBs, in this case, samples from WM1 were collected in the second sampling (September or October) and samples from WM2 in the first one (February or March). Both stations are located in the northern bank of the Ría. WM1 station is situated on a closed inlet where two rivers flow and during the summer months (June-September) the population increases considerably. In this station, it has also been found that in 2014, 2015 and later years, there have been discharges of both industrial and domestic with presence of pollutants. WM2 station is located in a small fishing harbour where there are several rivers with high flow rate in the winter months (February–March). Therefore this station is very close to a WWTP, and in 2015 and previous years there have been important domestic spillways and discharges, reflected in the high PCBs levels found before 2015 (Table 1). However dealing with fishing harbour with intense maritime activity and an intense traffic related to a nearby motorway (AP 9), the PAHs concentrations have not shown any downward trend during the sampled years, they have been found in samples from WM2 in very high and disperse values (the most of samples from WM2 station are along the positive PC1). The most of samples from WM3 station were distributed along the negative axis of PC1, indicating low levels of PAHs, of those one part (associated to the positive axis of PC2) contained low levels of PCBs and mainly were collected in the first sampling (February–March), the another one (associated to the negative axis of PC2) with high levels of PCBs and collected in the second samplings (September–October). Although WM3 station was near neighbourhood sewage treatment plant, it was the least affected station by persistent compounds pollution.

For raft mussel samples, the two PCs, PC1 and PC2 explained 64.9% and 11.4%, respectively of the total between-sample variability. As it happened with wild mussels, positive PC1 was mainly constructed by PAHs levels, except DBahA, this congener was strongly correlated to positive axis of PC2. Negative PC2 axis was correlated with PCBs. Projecting the data onto PC1 and PC2 plane a distinct group can be observed (see Figure S2b), it is the group of samples coming from RM1 station collected during the studied period, with the lowest PC2 scores (the lowest loading of PCBs). This group was also associated to the negative part of PC1, this means that samples also had a low concentration of PAHs. It is worth remembering that RM1 station is close to the mouth of the Ría where there is an easier oceanic water exchange. However, one of the samples had high values of PAHs and PCBs (highly correlated with positive PC1 and negative PC2), it was collected in 2011. It is known the existence of spillways and points of discharges of natural, domestic and industrial type in 2011. Industrial and commercial activities in this zone were related to wholesale of fish, shellfish and other food products, in addition to shellfish and fish companies. Samples from RM2 were distributed in the three planes, positives PC1 and PC2 (high levels of PAHs and low ones of PCBs), negative PC1 and positive PC2 (low levels of PAHs and PCBs, except of DBahA levels) and negatives PC1 and PC2 (high levels of PCBs). RM2 station is located in a pristine zone, levels of pollutants are relatively low. However during the period 2010–2011 mussels had relatively high values, the reason is the existence of points of domestic and industrial discharges during the years prior 2012. Samples of RM3 station were distributed in

three planes too, in the positive PC1 and PC2 one, in the negative PC1 and positive PC2 one and in the positive PC1 and negative PC2 one, this later with high levels of pollutants. Samples from this station were very contaminated by priority pollutants, RM3 station is located from the Rande Strait to the interior part of the estuary. Furthermore RM3 was situated in front of the emissary of the Redondela village and near the fish hatchery and cannery. As it is the case of the other samples, the first samplings 2010–2011 were the most polluted and were placed in the positive PC1 and negative PC2 plane. Samples from RM4 and RM5 stations were distributed in the four planes forming PC1 and PC2. In the event of RM4, six samples were placed in the positive part of PC1 with high load of hydrocarbons, and four in the negative part with low levels of them. Some samples collected in the first samplings and placed along the negative part of PC2 had high levels of PCBs. RM4 station was the most contaminated by PAHs and in 2011 there were several domestic and industrial spills that could justify the high levels of PCBs. Both RM3 and RM4 are within Community Interest Zone SCI (Site of Community Importance). RM5 station is mainly situated along the positive part of PC1 with high levels of PAHs, some of them are also distributed along the negative part of PC2 with high levels of PCBs, this later samples were collected in 2012, 2013 and 2018. RM5 station is situated in Vigo town, with a dense population, an important fishing harbour and industrial activity. RM5 station is also near a WWTP and, in 2011 there were urban-domestic inputs in the zone.

3.4. Temporal trends

Figure S3a shows the temporal trend of ΣPCBs and pp′-DDD for the last ten years (2010–2019) in wild mussel samples and Figure S3b exhibits the temporal trends of BaA and DBahA (2010–2019) in raft mussels from Ría de Vigo. These compounds showed a clear trend to the reduction except DBahA, the most recalcitrant compound that showed a tendency to increase. In the case of organochlorine compounds is mainly due to the actions taken to ban their production and use. In the European Union, both PCBs and DDT production was severely restricted in 1985 and 1981, respectively. The emissions of PCBs fell around 74% during the period 1990–2016 (EEA, 2018). For PAHs, several administrative measures have also been taken but the bilge cleaning of vessels, the increase of combustion processes involved in many domestic activities and industrial waste incineration plants can be the reason for the rise. Besides, DBahA is found in gasoline and diesel exhaust gases of boats. The declining trend observed in organochlorine compounds was consistent with the general decline on some POPs reported by other authors. Carro et al. (2010) showed a decrease of PCB levels in wild mussel from Galician Rías along the period 1998–2008. Bellas et al. (2011) reported PCBs decreasing trends in mussels from North-Atlantic Spanish coast from 1991 to 1999. Sturludottir et al. (2013) observed that concentrations of the persistent organic pollutants, *p,p′*-DDE, HCB and PCB153, had decreased in the most of the sampling locations from Iceland during the period 1990–2010. Hung et al. in 2016 under the Arctic Monitoring and Assessment Programme (AMAP) in four Arctic stations found the slow declining of PCBs and DDTs in air. However, Deudero et al. (2007) had not observed significant temporal trend of PCBs in mussels from the Balearic Islands. Campillo et al. (2017) only found significant declines on DDTs levels in the most of the areas studied in Spanish Mediterranean and no trends or weak downtrends for PCBs in the areas where inputs may be primarily from the atmosphere. In Alborán Sea upward trends probably related to PCBs inputs from the Strait of Gibraltar were observed.

4. Conclusions

A report of the levels and trends (from 2010 to 2019) of PCBs, OCPs and PAHs in wild and raft mussel collected in several sites located in the Ría de Vigo and influenced by human pressures has been presented. All levels found in these samples for the compounds regulated in seafood products (i.e. PCBs and PAHs) were below the allowable limits for human consumption. The overall pollution levels were low or similar compared to those in other countries. The results showed that the levels of priorityl pollutants in mussel were related to water discharges from rivers, urban (WWTP) or industrial emissaries. In the case of PAHs, despite the administrative measures the bilge cleaning of vessels and the increase of combustion processes involved in many domestic activities, industrial waste incineration plants or transport were the main reason of occasional rises. Mussels from the outer stations were the least contaminated. However wild mussels were more contaminated by priority pollutants than culture mussels. Raft mussels are permanently submerged in seawater where contaminants are subject to a dilution effect. Unlike organochlorine compounds, PAHs did not show such a strong downward trend.

CRediT authorship contribution statement

N. Carro: Methodology, Resources, Writing - review & editing, Supervision, Conceptualization, Funding acquisition. **J. Cobas:** Methodology, Resources, Writing - review & editing, Funding acquisition. **I. García:** Investigation, Formal analysis, Visualization, Writing - original draft. **M. Ignacio:** Investigation, Formal analysis, Visualization, Writing - original draft. **A. Mouteira:** Investigation, Formal analysis, Visualization, Writing - original draft. **M. Miranda:** Investigation, Formal analysis, Visualization, Writing - original draft. **L. Picado:** Investigation, Formal analysis, Visualization, Writing - original draft.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary material related to this article can be found online at https://doi.org/10.1016/j.rsma.2021.101742.

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