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RESEARCH ARTICLE

Distribution and human health risk assessment of PAHs in four fish species from a SW Atlantic estuary

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Abstract The aim of this study is to assess—for the first time—the concentration of the 16 polycyclic aromatic hydrocarbons (PAHs) in the muscle tissues of four fish species (*Micropogonias furnieri*, *Cynoscion guatucupa*, *Ramnogaster arcuata*, and *Mustelus schmitti*) from Bahía Blanca estuary, Argentina and to evaluate their sources, distribution, and the human health risks implicated. Considering the four species under study, mean total PAH concentrations showed the following decreasing accumulation trend: *M. schmitti*, *R. arcuata*, *C. guatucupa*, and *M. furnieri.* Low molecular weight PAHs, such as naphthalene and phenanthrene, were generally predominant, displaying properties of PAH mixtures generated from petrogenic pollution. Of the four fish species analyzed, *M. furnieri* was the only one that did not raise any human consumption warning. In the case of the other species, exceeding values were found above the safety human consumption guidelines. Nevertheless, the screening criteria for carcinogenic PAHs proposed by the USEPA indicated a good quality status for these fish species.

Keywords Bahía Blanca estuary · Polycyclic aromatic hydrocarbons · Fish species · Health risk assessment

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Introduction

Polycyclic aromatic hydrocarbons (PAHs) are broadly distributed within marine and coastal environments (Ramalhosa et al. 2012; Storelli et al. 2013). PAHs involve a broad class of compounds consisting of two or more fused benzene rings and emerge as priority pollutants due to their persistence, bioaccumulation ability, and toxicity to both aquatic organisms and human populations (Boitsov et al. 2009). According to the Environmental Protection Agency (USEPA), 16 PAHs have been established as top priority control pollutants. Moreover, according to the International Agency for Research on Cancer (IARC 2010), over half of them are potentially carcinogenic to humans. Due to the ability of the reactive metabolites of some PAHs to bind to cellular proteins and DNA several biological effects of PAHs are known, including tissue and genetic alterations, cancer, effects on growth and development, and effects of immune function, among others (Delistraty 1997; Hoffman 2003). Regarding their origin, PAHs can be originated from petrogenic (i.e., petroleum derivate), biogenic (e.g., perylene, retene), and/or pyrogenic (i.e., burning of organic matter) sources (Wilcke 2000, 2007; Neff et al. 2005). The common routes by which they reach the aquatic environment include atmospheric depositions, oil spills, urban runoff, waste water discharges, and emissions from watercraft and vehicles, among others (Fu and Wu 2005; Vuorinen et al. 2006; Ramalhosa et al. 2012; Abdolahpur Monikh et al. 2014; Bandowe et al. 2014). Once in the aquatic ecosystem, PAHs

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are primarily accumulated in fine grained sediments and suspended particles due to their hydrophobic nature; after that, they could be remobilized into the water column to finally become bioavailable to organisms (Wetzel and Van Vleet 2004). Uptake of PAHs by these aquatic organisms may occur by inhalation, ingestion, or skin surfaces (Neff 1985; Fu and Wu 2005; Oluseyi et al. 2011; Owabor et al. 2010; Zhang et al. 2011; Castro-Gutiérrez et al. 2012).

To assess the environmental condition of coastal zones such as estuaries, the concentration of PAH levels in edible fishes is advisory and is of considerable interest due to the toxic risk effects not only to the fish themselves but also to the top-level organisms that consume these contaminated fish, such as humans (Klumpp et al. 2002; Ashley et al. 2003). Fish communities have been recognized as efficient tools to evaluate possible organic pollution impacts due to their vulnerability to exposure, the visibility of some adverse effects such as tumors and lesions, their centrality in aquatic food webs, and both recreational and commercial importance (Logan 2007).

Bahía Blanca estuary (BBE) is the second largest estuary of Argentina (South America). This environment has shown a significant increment both in the industrial development and in the population growth during the past decades (Marcovecchio et al. 2008). This coastal area supports an intensive anthropogenic activity, including five national harbors and one of the biggest industrial parks in South America that comprise refineries, oil terminals, tanks for storing oil products, and multiple docks. Moreover, several industries are located on the northern coast and directly release hydrocarbons, muds with heavy hydrocarbon fractions, crude oil, and smoke particles into the estuary (Limbozzi and Leitao 2008). In the BBE waters, 30 fish species have been registered, where *Cynoscion guatucupa* (Cuvier, 1829), *Micropogonias furnieri* (Desmarest, 1823), and *Mustelus schmitti* (Springer, 1939) are the most important fishing resource (Lopez Cazorla 2004). *C. guatucupa* and *M. furnieri* are migratory fish species. In BBE, adults perform seasonal migrations, moving into the estuary in April and September (autumn and spring) and since October to March (spring-summer), respectively. Spawning occurs outside estuaries along the Argentinean coast, from spring to mid-autumn. Small juveniles during the first year life (age 0+) are into the estuary (Lopez Cazorla 1996, 2000). *C. guatucupa* feeds on crustaceans on its early stages and its diet shifts to pelagic fish as it develops into adulthood (Lopez Cazorla 1996; Sardiña and Lopez Cazorla 2005a). On the other hand, juveniles of *M. furnieri* feed on polychaetes and adult on crabs (Lopez Cazorla 1987; Sardiña and Lopez Cazorla 2005b). *M. schmitti* is another migratory fish species, in BBE adults moving into the estuary since August (winter) to December (spring), and juveniles stay here during the first year life. It feeds on crabs (Lopez Cazorla 2004). Finally, *Ramnogaster arcuata* (Jenyns, 1842) is a small pelagic, zooplanktivorous fish species. It has often been reported to be an estuarine-resident species that exhibits a wide spatio-temporal distribution and completes its whole life cycle within the BBE (Lopez Cazorla and Sidorkewicj 2009; Lopez Cazorla et al. 2011). *R. arcuata* is not fit for consumption, nevertheless is an important fish species for studying PAH accumulation since, as permanent inhabitant of the BBE, it shows more accurately what happens within the estuarine system.

Even though the occurrence of PAHs in sediments has been intensively addressed in the past (Arias et al. 2010a, b, 2010; Oliva et al. 2015a, b), information regarding the biota contamination with PAHs is scarce (Arias et al. 2009). The lack of available data of PAH accumulation within fish species inhabiting the BBE gives emphasizes to the evaluation of the present condition of this environment. Then, the main aim of this study is to determine the concentration of PAHs in fish muscle tissues, discussing its accumulation through four species with different habitat use under analysis. In addition, this study addresses the safety of fish consumption issue by evaluating PAHs in terms of accumulation trends.

Materials and methods

Sample collection and preparation

Fish samples were trimonthly caught at the BBE (Fig. 1) from August 2013 to June 2014. Fish catches were carried out with shrimp nets at two sampling sites (Galvan Harbor and Embudo Channel) to ensure proper geographical representation. Overall, 536 individuals were collected, corresponding to four fish species: *M. furnieri* ($n = 106$), *C. guatucupa* ($n = 132$), *R. arcuata* ($n = 287$), and *M. schmitti* ($n = 11$). Each of the analyzed individual was classified in size classes following different criteria for each species. In *C. guatucupa* and *M. furnieri*, trophic groups were formed by different sizes throughout the first year of life of each one according to Sardiña and Lopez Cazorla (2005a and b) and adults with sizes larger than 350 mm of total length (TL). In *R. arcuata*, each class corresponds to an age according to Lopez Cazorla and Sidorkewicj (2009). For *M. schmitti*, the classes were juveniles and adults with sizes larger than 450 mm of TL (Lopez Cazorla 1987).

After being caught, for each fish, TL was measured to the nearest millimeter and the samples were transported to the laboratory with ice. Feeding activity for each species and month was analyzed by the vacuity index (VI). VI indicates the percentage of individuals in the population who have been feeding, and it was calculated as follows: (number of empty stomachs/total number of stomachs examined) × 100 (Molinero and Flos 1992). Dissection was performed with a

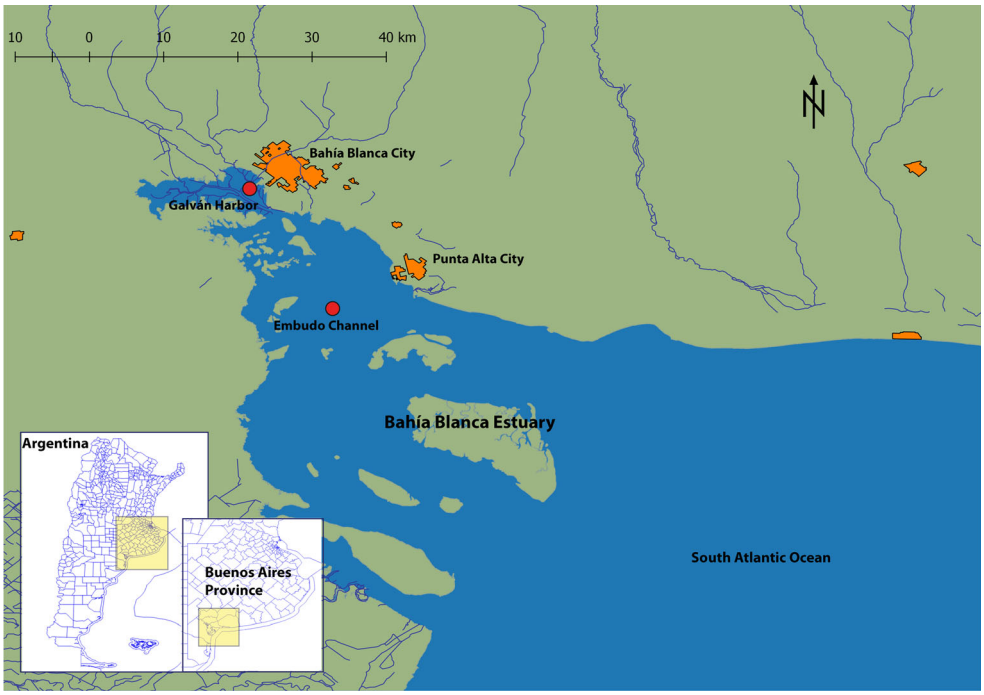

Fig. 1 Map of the Bahía Blanca Estuary, indicating with *circles* the two sampling zones

stainless steel knife in order to obtain tissue subsamples from dorsal muscle. After that, with pooling criteria adjusted to discriminate among size classes, samples were weighed, homogenized, and pooled if necessary. A total of 23 composite samples were analyzed. The samples were then lyophilized during 48 h, smashed in a mortar, and stored in desiccators prior to analyses.

Analytical procedure

The analytical procedure for PAH analyses involved an extraction according to the method of UNEP/IAEA/FAO/IOC (1993). Before extraction, 100 μL of the mixture of four predeuterated PAHs (napthalene-d_8, acepnapthene-d_{10}, phenanthene-d_{10}, crysene-d_{12}) was added as subrogate standards. Muscle tissue (5 g) was digested under reflux with methanol for 8 h, and then potassium hydroxide (0.7 M) and tridistilled water were added and left to reflux for two more hours. The non-saponifiable fraction was extracted with *n*-hexane; the organic phase was dried with anhydrous sodium sulfate and concentrated close to 5 mL in a rotary evaporator with a low-temperature thermostatic bath. Furthermore, the concentrate was reduced to 1.5 mL under a gentle high purity nitrogen flow. The extract was seeded in an alumina-silica (2:1) gel column to carry out the sample clean- up. PAHs were eluted with 70 mL of hexane-dichloromethane (9:1), and the volume of eluates was then reduced to 5 mL by rotary evaporator and further to 1.5 mL under nitrogen flow. Finally, just before the GC/MS injection, 100 μL of deuterated internal standard (benzo-[a]-anthracene-d_{12}) was added to the extract vials for recovery asses.

PAHs were quantified using a gas chromatograph (Agilent 7890 B, Santa Clara, USA) coupled with a mass spectrometer (Agilent 5977A, Santa Clara, USA), equipped with a fused silica column (HP-5MS; 30 m; 0.25 mm i.d.; 0.25 μm film thickness). Helium was used as a carrier gas. The mass spectrometer was operated in selected ion monitoring mode (SIM) and electron impact mode (70 eV). The samples were injected in the splitless mode at 250 °C, and the temperature program used was as follows: initial temperature 70 °C for 2 min; heated to 150 °C at 30 °C/min and then to 310 °C at 4 °C/min; and held for 10 min. The 16 priority PAHs proposed by USEPA were analyzed: naphthalene (NA), acenaphthylene (ACY), acenaphthene (ACE), fluorene (FL), phenanthrene (PHE), anthracene (AN), fluoranthene (FLU), pyrene (PY), benzo[a]anthracene (BaA), chrysene (CHR), benzo[b]-fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), indeno[1,2,3-cd]pyrene (IP), dibenzo[a,h]anthracene (DBA), and benzo[ghi]perylene (BPE). Each individual PAH compound was confirmed by the retention time and the abundance of quantification/confirmation ions with respect to authentic PAH standards. Quantification of individual compounds was based on the ratios of analyte peak areas/surrogate standards areas (naphthalene-d_{12}, acenaphthene-d_{10}, phenanthrene-d_{10}, chrysene-d_{12}, internal standard method) using the corresponding calibration curves.

Quality control and assurance

To ensure quality control, procedural blanks were regularly performed during the extraction process. Blanks were prepared following the same procedure but without adding the

fish tissue. Quality controls for the PAH analyses were carried out by monitoring the recovery of the internal standard (benzo-[a]-anthracene-d_{12}) spiked just before GC injection; recoveries ranged from 76 to 107%.

Sample concentrations were expressed as nanograms per gram wet weight (ng/g ww). For that, results were normalized to a water content of 70% (Soclo et al. 2008).The laboratory detection limits of the method (DLs) for individual PAH ranged from 0.5 to 1.3 ng/g dry weight. DL was set at five times the detected amount of the procedural blank. A PAH standard mixture of 16 PAHs, deuterated internal standard solutions, and benzoanthacene-d_{12} were obtained from Supelco (Bellefonte, PA, USA). All solvents used for sample processing and analyses (hexane, methanol, and dichloromethane) were of analytical and chromatographic grade from Merck (Darmstadt, Germany). Merck silica gel 60 (70–230 mesh ASTM) and aluminum oxide activated at 450 °C were heated at 120 °C for 12 h prior to use. Glassware was washed with non-ionic detergent, rinsed with ultrapure water and acetone/hexane, and dried at 120 °C prior to use.

Health risk assessment

The carcinogenic potential of PAHs was evaluated using the toxic equivalent of benzo[a]pyrene (TEQ BaP) and was calculated as follows

$$\text{TEQ BaP} = \sum ci \times \text{TEF} \quad (1)$$

in which *ci* is the concentration of the individual PAH (ng/g) and TEF is the toxic factor of PAHs relative to BaP (USEPA 1993; Nisbet and Lagoy 1992).

The daily dietary intake (DDI) value via fish consumption was calculated based on Eq. 2

$$\text{DDI} = C \times \text{IR} \quad (2)$$

where *C* is the concentration of single or sumatory PAHs in fish muscle (ng/g) and IR is fish ingestion rate (13 g/day per person; FAO 2016)

The excess cancer risk (ECR) resulting from a lifetime fish consumption was also calculated by the following equation (Bandowe et al. 2014)

$$\text{ECR} = \frac{\sum Q \times \text{TEQ BaP} \times \text{IR} \times \text{ED}}{\text{BW} \times \text{AT}} \quad (3)$$

where *Q* is the potential cancer factor of BaP (7.3 mg/kg/day), ED is the life expectancy (70 years for adults), BW is the average adult body weight (70 kg), and AT is the average life span for carcinogens (25,500 days).

Statistical analyses

All statistical analyses were carried out using STATISTICA 7.0 (StatSoft, Inc.), following Zar (1996). One-way analysis of variance (ANOVA) and Scheffé contrast were performed to assess differences in PAH concentrations between fish species and fish size classes. If necessary, data was previously transformed to meet the required assumptions of homogeneity and normality for the parametric tests. When the data did not meet the assumptions, a non-parametric test was used (Kruskal-Wallis). The acceptable level of statistical significance used throughout the study was $p < 0.05$. PAH concentrations reported as below the laboratory detection limit (DLs) were substituted by half of the DL for statistical analyses.

Results and discussion

Concentration of PAHs

PAH concentrations in muscle tissues of *M. furnieri*, *C. guatucupa*, *R. arcuata* and *M. schmitti* are listed in Table 1. The levels of total PAHs found in the different fish species (sum of 16 PAHs analyzed) ranged from 8.42 to 661.15 ng/g ww. Mean total PAHs accumulated in each of the four fish species, including both juveniles and adults, showed the following decreasing concentrations: *M. schmitti* (308.96 ng/g) > *R. arcuata* (182.35 ng/g) > *C. guatucupa* (98.25 ng/g) > *M. furnieri* (34.87 ng/g). Beyond this, statistical comparisons indicated that there were no significant differences in total PAH concentrations among the different species under analysis (ANOVA, $p < 0.05$). In regard to the fish size classes, there were no significant differences in PAH concentration between them; nevertheless, *C. guatucupa* achieved the maximum PAH levels in adult tissues, while the lowest values were found in the smallest individuals (class II). As bioaccumulation is generally non-demonstrable for PAHs in fish (Varanasi et al. 1989), this suggested a possible quite recent PAH uptake for this species. contrast, for the other analyzed species, no clear trends were observed. Species-specific differences in biochemical and physiological parameters, such as basal levels of xenobiotic-metabolizing enzymes and lipid content of tissues, appear to have significant effects on the disposition of PAHs and their metabolites (Varanasi and Stein 1991), and could be responsible for the variations in PAH accumulation. The differential trends in PAH accumulation have already been shown in other studies, since PAH levels can be naturally higher in different fish species because of diet or habitat use (Escartín and Porte 1999), also indicating that organisms show different selectivities towards contaminants.

According to the NOAA (National Oceanic and Atmospheric Administration), PAH concentration in muscle

Table 1 Concentration of PAHs in fishes from Bahía Blanca Estuary (ng/g wet weight)

	M. furnieri		*C. guatucupa*			*R. arcuata*			*M. schmitti*	
	Juveniles	Adults (n = 1)	Juveniles		Adults (n = 3)	Juveniles	Adults		Juveniles	Adults (n = 1)
	Class III (n = 3)		Class II (n = 2)	Class III (n = 2)		Class II (n = 3)	Class III (n = 3)	Class IV (n = 2)	Class III (n = 3)	
NA	23.53 ± 23.45	<DL	33.87 ± 47.68	51.01 ± 60.09	63.46 ± 50.73	147.63 ± 175.26	200.15 ± 308.53	7.11 ± 9.91	193.36 ± 326.44	366.16
ACY	0.12 ± 0.03	<DL	0.15 ± 0.03	12.75 ± 17.89	8.75 ± 14.98	0.38 ± 0.48	0.75 ± 1.13	0.14 ± 0.06	9.39 ± 14.52	1.2
ACE	0.46 ± 0.62	1.23	0.77 ± 1.09	2.11 ± 2.85	10.28 ± 16.34	<DL	4.23 ± 6.9	<DL	4.77 ± 8.18	<DL
FL	0.89 ± 1.06	1.22	1.38 ± 1.43	3.44 ± 4.72	3.76 ± 3.71	10.51 ± 5.78	6.69 ± 10.61	10.70 ± 14.26	7.66 ± 13.1	13.72
PHE	3.23 ± 2.72	4.81	3.17 ± 0.83	7.32 ± 10.21	6.72 ± 7.79	9.77 ± 12.23	14.57 ± 23.65	1.11 ± 1.42	19.01 ± 22.12	27.06
AN	7.68 ± 11.13	<DL	1.10 ± 1.13	11.97 ± 16.93	9.03 ± 15.18	9.56 ± 12.79	9.34 ± 15.55	19.83 ± 24.58	10.85 ± 12.97	1.10
FLU	0.35 ± 0.43	0.88	1.49 ± 1.97	8.48 ± 9.11	2.07 ± 2.25	7.33 ± 11.13	15.63 ± 17.56	12.49 ± 17.66	1.52 ± 2.54	6.08
PY	7.41 ± 12.07	1.28	3.16 ± 3.54	6.12 ± 1.57	3.33 ± 4.51	3.56 ± 4.58	5.86 ± 9.38	8.63 ± 3.84	0.03 ± 0.06	5.88
BaA	<DL	<DL	<DL	1.41 ± 1.85	0.65 ± 1.04	<DL	<DL	<DL	6.76 ± 11.70	<DL
CHR	<DL	<DL	<DL	2.34 ± 3.16	1.02 ± 1.69	<DL	<DL	<DL	<DL	<DL
BbF	<DL	<DL	<DL	<DL	<DL	0.14 ± 0.09	<DL	<DL	<DL	<DL
BkF	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL
BaP	<DL	<DL	<DL	<DL	8.67 ± 13.74	0.13 ± 0.11	0.34 ± 0.5	<DL	7.95 ± 13.77	<DL
IP	<DL	<DL	<DL	<DL	<DL	<DL	0.12 ± 0.13	<DL	<DL	<DL
DBA	<DL	<DL	<DL	<DL	10.82 ± 18.27	<DL	<DL	<DL	10.44 ± 18.07	<DL
BPE	<DL	<DL	<DL	<DL	<DL	<DL	0.11 ± 0.1	<DL	<DL	<DL
Total PAHs	43.35 ± 30.36	9.42	45.10 ± 44.45	106.64 ± 37.66	128.08 ± 48.93	188.80 ± 178.72	257.54 ± 351.03	59.86 ± 68.70	271.64 ± 342.26	421.21
∑LMW	35.89 ± 24.35	7.25	40.44 ± 49.94	88.60 ± 43.05	102.00 ± 52.9	177.85 ± 181.28	235.74 ± 343.04	38.89 ± 47.27	245.04 ± 359.71	409.25
∑HMW	7.76 ± 11.87	2.16	4.65 ± 5.42	18.34 ± 5.67	26.66 ± 27.29	11.15 ± 8.38	22.12 ± 18.09	21.12 ± 21.50	26.69 ± 42. 44	11.96
cPAHs	23.53 ± 23.45	<DL	33.87 ± 47. 77	54.76 ± 65.11	84.73 ± 27.55	147.90 ± 175.32	200.67 ± 309.16	7.11 ± 9.91	218.50 ± 307.54	366.16

Results are expressed as average ± SDs

n number of pools; <*DL* lower detection limit; ∑*LMW* sum of NA, ACY, ACE, FL, PHE, and AN; ∑*HMW* sum of FLU, PY, BaA, CHR, BbF, BkF, BaP, IP, DBA, and BPE; *cPAHs* sum of carcinogenic PAHs: NA, BaA, CHR, BbF, BkF, BaP, IP, and DBA

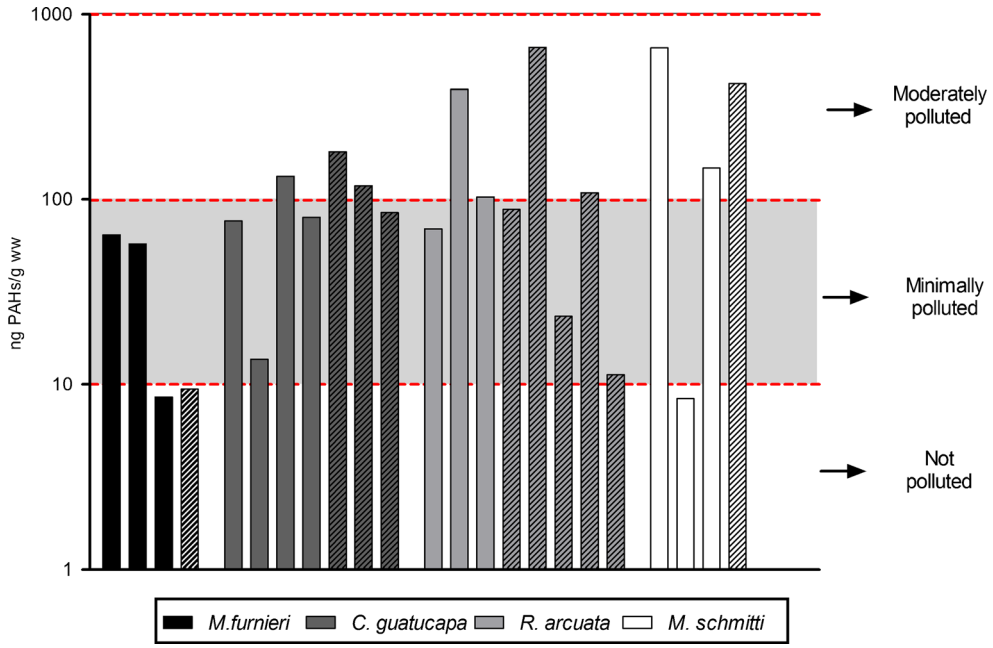

Fig. 2 Classification of PAH levels in fishes from the BBE according to NOAA criteria. *Striped bars* and *full bars* represent adults and juveniles samples, respectively

tissues can be grouped into four categories: not polluted (<10 ng/g), minimally polluted (10 to 99 ng/g), moderately polluted (100 to 1000 ng/g), and highly polluted (>1000 ng/g) (Varanasi et al. 1993; Soares-Gomes et al. 2010). The application of the NOAA criteria in the Bahía Blanca estuarine samples (Fig. 2) revealed that 43.5% of the samples analyzed could be categorized as minimally polluted (11.29 to 88.10 ng/g), whereas 43.5% could be classify as moderately polluted (102.86 to 661.15 ng/g). In fact, at least two samples of each species could be categorized as moderately polluted. Finally, only three samples (13.0%) showed no polluted levels at all (8.35 to 9.42 ng/g) (Fig. 2).

Comparison of these data with other studies should be applied with caution due to differences in fish species, number and type of PAH compound analyzed, and methodology employed, among others. However, the range of PAH levels found in this study for *M. furnieri* showed to be slightly higher than records from the same fish species inhabiting the estuarine environment of Guanabara Bay, Brazil (Da Silva et al. 2007; Meniconi et al. 2001), but lower than those previously reported for another fish species (*Odontesthes* sp.) from the BBE (Arias et al. 2009).

Regarding the time of the samples catches, the lowest mean values of PAHs were recorded for all the fish species during June 2014 (late autumn). Considering the ingested food as a source of PAHs to fishes, in this study, the vacuity indexes for both *C. guatucupa* and *R. arcuata* were evaluated. These fish species were selected upon their better representative. According to the results (Fig. 3), differences were found between the indexes of vacuity (VI), with June 2014 as the time of the year with the higher proportion of empty stomachs concordantly with the lowest PAH values. Thereafter, an increment in the vacuity levels could be, at least in part, responsible for

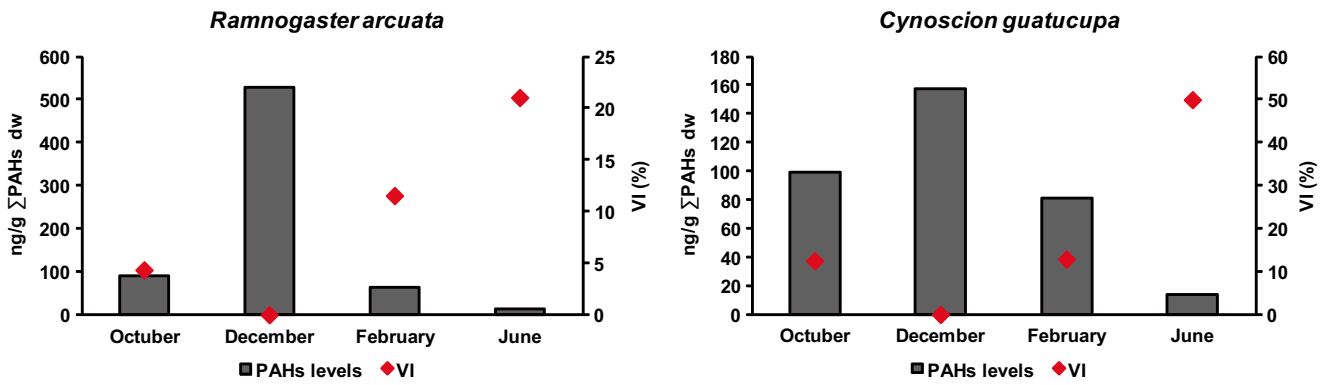

Fig. 3 Relation within indexes of vacuity (%) and PAH values from *R. arcuata* and *C. guatucupa*

the observed lower PAH values found for the colder sampling time. In agreement with this, the maximum levels of PAHs and the lower VI were detected in December 2013 (late spring), a period whit higher feeding activity for these species. This allowed to positively correlating the feeding cycle/behavior with the PAH muscle content at the area of study. Previous studies with juveniles of *C. guatucupa* from the BBE (Sardiña and Lopez Cazorla 2005a) had already found during the time of year with cold temperatures the highest vacuity levels and shorter quantity of ingested food items.

It is well known that the lipid content is an important factor for determining the distribution of lipophilic compounds, such as PAHs (Bruner et al. 1994, Hellou et al. 2003; Barhoumi et al. 2016), and it has been suggested that the rate of uptake of hydrophobic chemicals in fish increase with a higher lipid content of the biological membranes (Spacie and Hamelink 1982; Van der oost et al. 2003). As the lipid content is related to the reproductive stage period (Sprung 1993; Bruner et al. 1994), it is important to note the time when the spawning season takes place. In the case of the fish species from the BBE, the spawning season takes place throughout the spring, the summer, and the beginning of autumn. Thus, the low PAH concentrations found in late autumn could be at least in part attributed to a natural decrement in the lipid concentrations after the spawning season.

Composition of PAHs

PAHs in marine environments commonly originate from pyrogenic or petrogenic sources. Pyrogenic PAHs are dominated by compounds with high molecular mass (HMW-PAHs) with four to six rings, whereas petrogenic PAHs are dominated by PAHs with lower molecular weight (LMW-PAHs) with two to three rings (Sanders et al. 2002; Dahle et al. 2003). In this work, similar trends in the distribution and composition pattern of PAH congeners can be advised for all fish species (Fig. 4). The preferential accumulation of PAHs is determined by their solubility and bioavailability, related to the octanol-water partition coefficient (K_{ow}), molecular weight, exposure route, and ingestion of PAHs (Conell and Miller 1984). In this study, LMW-PAHs prevailed in all the samples, achieving together between 37.5 and 100% of total PAHs in each species. In contrast, PAHs with more than five rings were usually lower than the DL. The dominance of LMW-PAHs in fish muscles has also been reported in several other studies (Ramalhosa et al. 2009, 2012; Xu et al. 2011; Storelli et al. 2013; Barhoumi et al. 2014, 2016). PAHs composed of two to three fused rings have higher water solubility, bioavailability, and uptake rates than PAHs with four to six rings and can be assimilated by these species by ingestion, direct absorption from water, or via passive diffusion through the gills and skin. As a consequence, organisms from the marine environment

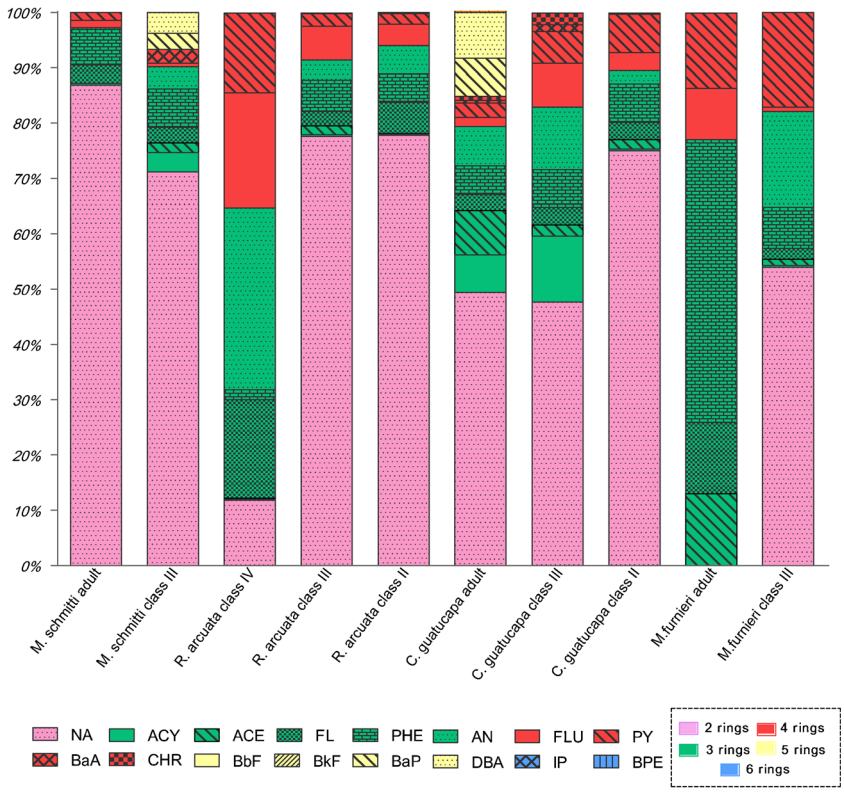

Fig. 4 Average percentual composition of PAH congeners in fish tissues from each species and size classes from the BBE

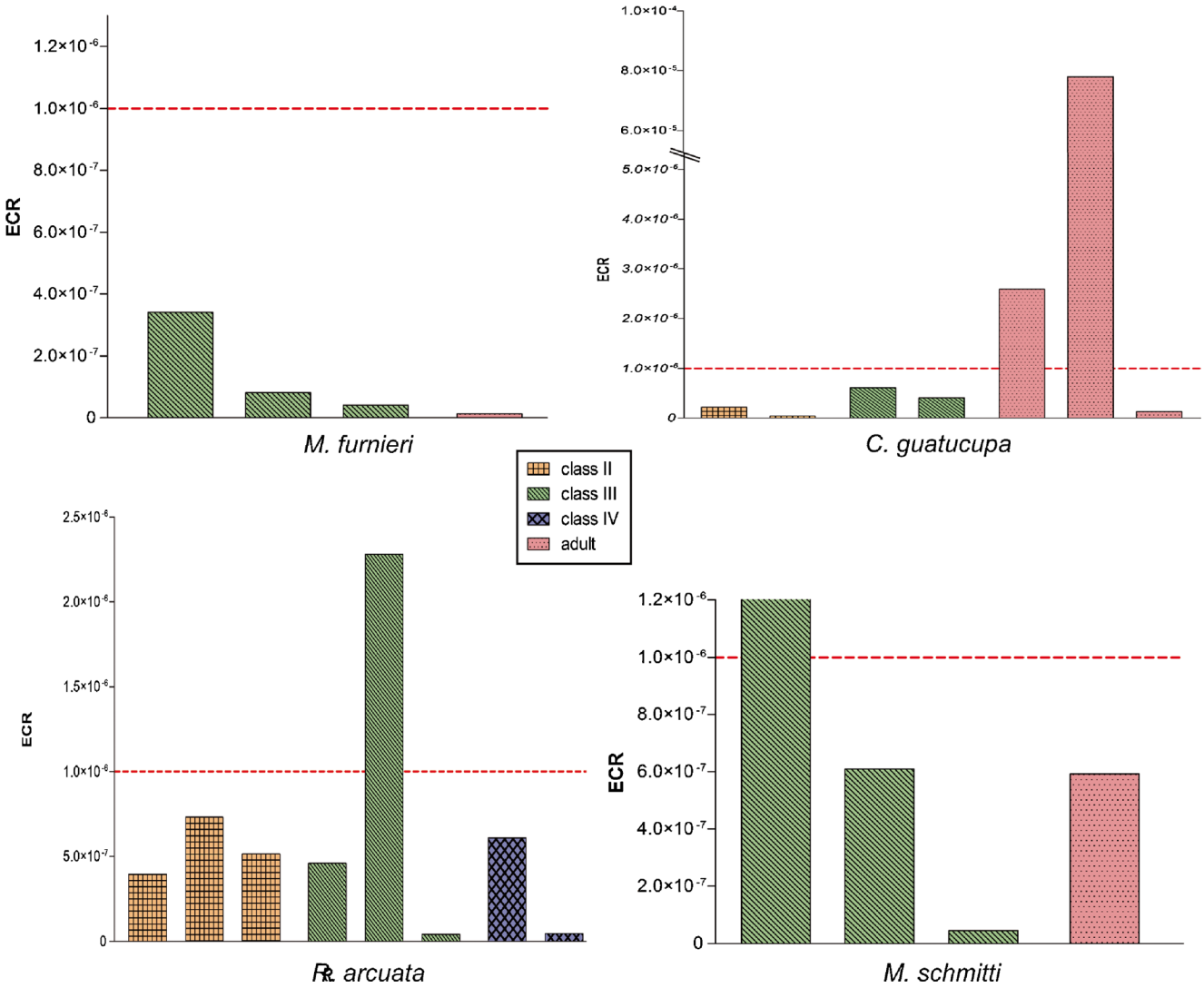

Fig. 5 Excess cancer risk (ECR) values in each size class from the four fish species. *Dashed lines* represent the acceptable risk level proposed by USEPA

are enriched in LMW compounds, whereas HMW-PAHs tend to be absorbed onto or associated with organic particles present in the water column or sediments, making them less bioavailable to fishes (Porte and Albaiges 1993; Baumard et al. 1998; Thorsen et al. 2004; Tolosa et al. 2005).

Finally, the selective accumulation for LMW-PAHs could also be an artifact attributed to the metabolic transformations of heavier PAHs occurring in the fish liver through the Cytochrome P450 System (Meador et al. 1995), since fish can rapidly convert up to 99% of the PAHs to metabolites within 24 h of uptake, changing the pattern and concentrations of PAHs in their tissues (Varanasi et al. 1989; Barhoumi et al. 2016). Half-life of parental PAHs is generally very short, ranging from 1 day for ACE to 9 days for PHE (Meador et al. 1995), and showing that the presence of these PAHs in fish muscle is an indicator of recent episodes of pollution exposure in the surrounding environment (Zhao et al. 2014; Barhoumi et al. 2016). Nevertheless, it is important to highlight that *C. guatucupa and R. arcuata* showed an increasing trend in the abundance of HMW-PAHs as the size classes of these fish species increased (Fig. 4). Particularly for *R. arcuata*, it has been reported to be an estuarine-resident species that completes its whole life cycle within the area of study. Then, this characteristic could have lead to a differential exposure pattern in comparison with the rest of the fish species, which could have resulting in a higher HMW PAH intake.

In terms of individual compounds, NA was the most abundant PAH in almost all the samples, accounting for 47.8 to 86.9% of total PAHs (Fig. 4). PHE was the second PAH compound in terms of abundance. Such patterns are properties of PAH mixtures generated from petrogenic pollution (Sauer et al. 1993; Cheung et al. 2007). Petrogenic inputs could be associated to the intense shipping activity in the study area, like fishing fleet activities, tanker traffic, petroleum buoys, spills of fuel, and lubricating oils.

Human health risk assessment

Dietary intake has been recognized as the main route to uptake persistent organic pollutants within humans (Barhoumi et al. 2016). Some PAHs and their metabolic products are considered priority pollutants of environmental concern, representing a threat to human health due to their toxic, carcinogenic and mutagenic characteristics, reproductive impairments, and endocrine disruption in lower and higher trophic levels (USEPA 2002, IARC 2010). Among PAHs, the IARC has classified the priority PAH compound BaP as carcinogenic to humans (group 1), DBA as a possible human carcinogenic (group 2A), and NA, BaA, CHR, BbF, BkF, and IP as probable human carcinogenics (Group 2B) (IARC 2010). In this study, the sum of the PAHs within groups 1, 2A, and 2B (i.e., ∑cPAHs) in fishes from the BBE varied from <DL to 570.27 ng/g ww and accounted for up to 88.5% of total PAHs. NA was the largest contributor to the cPAH levels while BaP, one of the most toxic and well-investigated carcinogenic PAHs, was detected in seven of the analyzed fish muscle samples. Of these samples, 43% corresponded to *C. guatucupa* (two adults and one juvenile class II), 43% corresponded to *R. arcuata* (two juveniles class II and one adult), and 14% corresponded to *M. schmitti* (juvenile). On the other hand, all *M. furnieri* samples presented values of BaP lower than DL.

BaP is the only PAH for which toxicological data are sufficient for derivation of a carcinogenic potency factor among all known potentially carcinogenic PAHs (Peters et al. 1999). Thus, the carcinogenic potential of PAHs was evaluated using the toxic equivalent of BaP (TEQ BaP). Total TEQ BaP calculated for samples varied from 0.009 to 57.492 ng TEQ BaP/g ww, and the median of total TEQ BaP in the four fish species achieved the following concentrations in decreasing order: *M. schmitti* > (0.569 ng/g) > *R. arcuata* (0.354 ng/g) > *C. guatucupa* (0.295 ng/g) > *M. furnieri* (0.044 ng/g). In this study, only five samples showed levels above the screening values for the TEQ BaP (0.67 ng/g ww) suggested by USEPA (2000) for human fish consumption. From those five samples, four of them corresponded to fish species that are fishing resource in the area (adults of *C. guatucupa* and juveniles *of M. schmitti*).

The Human Health Risks of fish consumption, due to differences in food consumption rates, were assessed and compared using the concept of daily dietary intake (DDI) (Shi et al. 2016) (Eq. 2). Then, the mean DDI value via fish consumption and for total PAHs was calculated in the present study, reaching 2118.6 ng/day ww. It is important to highlight that the DDI was calculated only for the fishes species normally consumed (adults of *M. schmitti*, *C. guatucupa*, and *M. furnieri*). This value was lower than those reported from fishes collected in Haimen bay, China (Shi et al. 2016), Gulf of Guinea, Ghana (Bandowe et al. 2014), and from fish and shellfish found in the coastal system of Catalonia, Spain (Martorell et al. 2010).

In addition, the ECR values resulting from the consumption of fishes during a lifetime were calculated according to Eq. 3 and are presented in Fig. 5. The ECR obtained from fishes from the BBE ranged from 1.29×10^{-08} to 7.90×10^{-05}. Compared to the screening criteria for cancerigens proposed by the USEPA, four samples (17.4% of the samples) exceeded the acceptable risk level (1×10^{-06}) above which consequences are expected to occur. The samples that exceeded the risk levels belong to one of *M. schmitti* (*juvenile of class III*), *two* adults of C. *guatucupa*, and one adult of *R. arcuata* (Fig. 5). On the other hand, all the samples of *M. furnieri* presented ECR values lower than the acceptable risk level. Finally, the *serious risk level* (1×10^{-04}) was not achieved by any sample caught in the BBE. At first sight, these results suggested a low carcinogenic risk posed by fish consumption; however, some considerations should be made since PAHs can be metabolized and biotransformed with the develop of metabolites that could be more toxic and carcinogenic than their parent PAHs (Johnson-Restrepo et al. 2008).

Conclusion

For the first time, the concentration of PAHs in muscle tissues of four fish species (*M. furnieri*, *C. guatucupa*, *R. arcuata*, and *M. schmitti*) from the BBE was evaluated. In general, while a dominance of PAH petrogenic sources was strongly suggested by the experimental evidence, feeding cycle/behavior studies allowed to point to the relation predator/prey as a possible route by which PAHs accessed these organisms. Considering the four species under study, exceedances of safety human consumption thresholds and carcinogenic risk warnings were low to null, indicating a good quality status for these fish species. In general, when compared with other worldwide locations, warning values were located in the low range of impact. Authors remark the need and usefulness of long-term monitoring and further studies to deepen the present analysis and confirm/discard trends.

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References

Arias AH, Spetter CV, Freije RH, Marcovecchio JE (2009) Polycyclic aromatic hydrocarbons in water, mussels (Brachidontes sp., Tagelus sp.) and fish (Odontesthes sp.) from Bahía Blanca estuary, Argentina. Estuar Coast Shelf Sci 85(1):67–81

Arias AH, Vazquez-Botello A, Tombesi N, Ponce-Velez G, Freije H, Marcovecchio J (2010a) Presence, distribution, and origins of polycyclic aromatic hydrocarbons (PAHs) in sediments from Bahía Blanca estuary. Argentina Environ Monit Assess 160(1):301–314

Arias AH, Marcovecchio JE, Freije RH, Ponce-Velez G, Vázquez BA (2010b) Análisis de fuentes y toxicidad equivalente de sedimentos contaminados con PAHs en el estuario de Bahía Blanca, Argentina. Hidrobiológica 20(1):41–56

Abdolahpur Monikh F, Hosseini M, Kazemzadeh Khoei J, Ghasemi AF (2014) Polycyclic aromatic hydrocarbons levels in sediment, benthic, benthopelagic and pelagic fish species from the Persian Gulf. Int J Environ Res 8(3):839–848

Ashley JT, Horwitz R, Steinbacher JC, Ruppel B (2003) A comparison of congeneric PCB patterns in American eels and striped bass from the Hudson and Delaware River estuaries. Mar Pollut Bull 46(10): 1294–1308

Bandowe BAM, Bigalke M, Boamah L, Nyarko E, Saalia FK, Wilcke W (2014) Polycyclic aromatic compounds (PAHs and oxygenated PAHs) and trace metals in fish species from Ghana (West Africa): bioaccumulation and health risk assessment. Environ Int 65:135–146

Barhoumi B, Clerandeau C, Gourves PY, Le Menach K, El Megdiche Y, Peluhet L, Budzinski H, Baudrimont M, Driss MR, Cachot J (2014) Pollution biomonitoring in the Bizerte lagoon (Tunisia), using combined chemical and biomarker analyses in grass goby, *Zosterisessor ophiocephalus* (Teleostei, Gobiidae). Mar Environ Res 101:184–195

Barhoumi B, El Megdiche Y, Clérandeau C, Ameur WB, Mekni S, Bouabdallah S, Driss MR (2016) Occurrence of polycyclic aromatic hydrocarbons (PAHs) in mussel (*Mytilus galloprovincialis*) and eel (*Anguilla anguilla*) from Bizerte lagoon, Tunisia, and associated human health risk assessment. Cont Shelf Res 124:104–116

Baumard P, Budzinski H, Garrigues P, Sorbe JC, Burgeot T, Bellocq J (1998) Concentrations of PAHs (polycyclic aromatic hydrocarbons) in various marine organisms in relation to those in sediments and to trophic level. Mar Pollut Bull 36(12):951–960

Boitsov S, Jensen HKB, Klungsøyr J (2009) Natural background and anthropogenic inputs of polycyclic aromatic hydrocarbons (PAH) in sediments of south-western Barents Sea. Mar Environ Res 68(5):236–245

Bruner KA, Fisher SW, Landrum PF (1994) The role of the zebra mussel, *Dreissena polymorpha*, in contaminant cycling: I. The effect of body size and lipid content on the bioconcentration of PCBs and PAHs. J Gt Lakes Res 20(4):725–734

Castro-Gutiérrez VM, Rodríguez-Rodríguez CE, Vargas-Azofeifa I (2012) Hydrocarbon degrading microflora in a tropical fuel-contaminated aquifer: assessing the feasibility of PAH bioremediation. Int J Environ Res 6(1):345–352

Cheung KC, Leung HM, Kong KY, Wong MH (2007) Residual levels of DDTs and PAHs in freshwater and marine fish from Hong Kong markets and their health risk assessment. Chemosphere 66(3):460–468

Connell DW, Miller GJ (1984) Chemistry and ecotoxicology of pollution (vol. 65). John Wiley & Sons

Da Silva TF, de Almeida AD, de Aquino Neto FR (2007) Polycyclic aromatic hydrocarbons in fishes and sediments from the Guanabara Bay, Brazil. Environ Forensics 8(3):257–264

Dahle S, Savinov VM, Matishov GG, Evenset A, Næs K (2003) Polycyclic aromatic hydrocarbons (PAHs) in bottom sediments of the Kara Sea shelf, Gulf of Ob and Yenisei Bay. Sci Total Environ 306(1):57–71

Delistraty D (1997). A critical review of the application of toxic equivalency factors to carcinogenic effects of polycyclic aromatic hydrocarbons in mammals. In: Neilson, A.H. (Ed.), The handbook of environmental chemistry Vol 3, Anthropogenic compounds, Par J: PAHs and related compounds: biology. Springer Science & Business Media, pp. 311–359

Escartin E, Porte C (1999) Assessment of PAH pollution in coastal areas from the NW Mediterranean through the analysis of fish bile. Mar Pollut Bull 38(12):1200–1206

FAO (2016) Food and Agriculture Organization of the United Nations. El estado mundial de la pesca y la acuicultura 2016. Contribución a la seguridad alimentaria y la nutrición para todos. Roma. 224 pp

Fu CT, Wu SC (2005) Bioaccumulation of polychlorinated biphenyls in mullet fish in a former ship dismantling harbour, a contaminated estuary, and nearby coastal fish farms. Mar Pollut Bull 51(8):932–939

Hellou J, Yeats P, Steller S, Gagné F (2003) Chemical contaminants and biological indicators of mussel health during gametogenesis. Environ Toxicol Chem 22:2080–2087

Hoffman DJ (2003) Handbook of ecotoxicology. CRC Press, Boca Raton, Florida

IARC (2010). International Agency for Research on Cancer. Some non-heterocyclic polycyclic aromatic hydrocarbons and some related exposures, vol 92. http://monographs.iarc.fr/ENG/Monographs/vol92/mono92.pdf.

Johnson-Restrepo B, Olivero-Verbel J, Lu S, Guette-Fernández J, Baldiris-Avila R, O'Byrne-Hoyos I, Aldous MK, Addink R, Kannan K (2008) Polycyclic aromatic hydrocarbons and their hydroxylated metabolites in fish bile and sediments from coastal waters of Colombia. Environ Pollut 151(3):452–459

Klumpp DW, Huasheng H, Humphrey C, Xinhong W, Codi S (2002) Toxic contaminants and their biological effects in coastal waters of Xiamen, China.: I. Organic pollutants in mussel and fish tissues. Mar. Pollut. Bull 44(8):752–760

Limbozzi F, Leitào TE (2008). Characterization of Bahía Blanca main existing pressures and their effects on the state indicators for surface and groundwater quality. En: Neves, R., Baretta, J., Mateus, M. (Eds.), Perspectives on integrated coastal zone management in South America, Lisboa, 315–331.

Logan DT (2007) Perspective on ecotoxicology of PAHs to fish. Hum Ecol Risk Assess 13(2):302–316

Lopez Cazorla, A. (1987). Contribución al conocimiento de la ictiofauna marina del área de Bahía Blanca. PhD thesis, Universidad Nacional de La Plata, Argentina, 247 pp.

Lopez Cazorla AL (1996) The food of *Cynoscion striatus* (Cuvier) (Pisces: Sciaenidae) in the Bahía Blanca area, Argentina. Fish Res 28(4):371–379

Lopez Cazorla A (2000) Age structure of the population of weakfish *Cynoscion guatucupa* (Cuvier) in the Bahía Blanca waters, Argentina. Fish Res 46(1–3):279–286

Lopez Cazorla A (2004) Peces. In: Piccolo CM, Hoffmeyer MS (Eds.). Ecosistema del estuario de Bahía Blanca, Instituto Argentino de Oceanografía, Bahía Blanca, 191–201

Lopez Cazorla A, Pettigrosso RE, Tejera L, Camina R (2011) Diet and food selection by *Ramnogaster arcuata* (Osteichthyes, Clupeidae). J Fish Biol 78(7):2052–2066

Lopez Cazorla AL, Sidorkewicj N (2009) Some biological parameters of Jenyns' sprat *Ramnogaster arcuata* (*Pisces: Clupeidae*) in south-western Atlantic waters. Marine Biodiversity Records 2:e127

Marcovecchio JE, Botté S, Delucchi F, Arias A, Fernández Severini M, De Marco S, Tombesi N, Andrade S, Ferrer L, Freije RH (2008) Pollution processes in Bahía Blanca Estuarine environment. In: Neves R, Baretta J, Mateus M (Eds) Perspectives on integrated coastal zone management in South America, Lisbon, pp 303–316

Martorell I, Perelló G, Martí-Cid R, Castell V, Llobet JM, Domingo JL (2010) Polycyclic aromatic hydrocarbons (PAH) in foods and estimated PAH intake by the population of Catalonia, Spain: temporal trend. Environ Int 36(5):424–432

Meador JP, Casillas E, Sloan CA, Varanasi U (1995) Comparative bioaccumulation of polycyclic aromatic hydrocarbons from sediment by two infaunal invertebrates. Mar Ecol Prog Ser 123:107–124

Meniconi MFG, Santos AF, Moreira IMN, Scofield ADL, Salmito TM, Romão CM, Machado GAWC. (2001) Fisheries safety monitoring in the Guanabara Bay, Brazil following a marine fuel oil spill. In

International Oil Spill Conference (March 2001, Vol. 2001, No. 2, pp. 951–957). American Petroleum Institute
Molinero A, Flos R (1992) Influence of season on the feeding habits of the common sole *Solea solea*. Mar Biol 113:499–507
Neff JM (1985) Polycyclic aromatic hydrocarbons. In: Rand GM, Petrocelli SR (eds) Fundamentals of aquatic toxicology, methods and applications. Hemisphere Publishing Corporation (McGraw-Hill International Book Company), Washington, New York, London, pp 416–454
Neff JM, Stout SA, Gunster DG (2005) Ecological risk assessment of polycyclic aromatic hydrocarbons in sediments: identifying sources and ecological hazard. Integr Environ Assess Manag 1(1):22–33
Nisbet IC, LaGoy PK (1992) Toxic equivalency factors (TEFs) for polycyclic aromatic hydrocarbons (PAHs). Regul Toxicol Pharmacol 16(3):290–300
Oliva AL, Quintas PY, La Colla NS, Arias AH, Marcovecchio JE (2015a) Distribution, sources, and potential ecotoxicological risk of polycyclic aromatic hydrocarbons in surface sediments from Bahía Blanca estuary, Argentina. Arch Environ Contam Toxicol 69(2):163–172
Oliva AL, Ovaert J, Arias AH, Souissi S, Marcovecchio JE (2015b) Mussels as Bioindicators of PAHs pollution within Argentinean coastal environments, South America. Int J Environ Res 9(4): 1293–1304
Oluseyi T, Olayinka K, Alo B, Smith RM (2011) Improved analytical extraction and clean-up techniques for the determination of PAHs in contaminated soil samples. Int. J. Environ. Res. 5(3):681–690
Owabor CN, Ogbeide SE, Susu AA (2010) Model simulation of biodegradation of polycyclic aromatic hydrocarbon in a microcosm. Int. J. Environ. Res. 4(4):807–816
Peters CA, Knightes CD, Brown DG (1999) Long-term composition dynamics of PAH-containing NAPLs and implications for risk assessment. Environ Sci Technol 33:4499–4507
Porte C, Albaiges J (1993) Bioaccumulation patterns of hydrocarbons and polychlorinated biphenyls in bivalves, crustaceans, and fishes. Arch Environ Contam Toxicol 26(3):273–281
Ramalhosa MJ, Paíga P, Morais S, Delerue-Matos C, Prior Pinto Oliveira MB (2009) Analysis of polycyclic aromatic hydrocarbons in fish: Evaluation of a quick, easy, cheap, effective, rugged, and safe extraction method. J Sep Sci 32(20):3529–3538
Ramalhosa MJ, Paíga P, Morais S, Ramos S, Delerue-Matos C, Oliveira MBPP (2012) Polycyclic aromatic hydrocarbon levels in three pelagic fish species from Atlantic Ocean: inter-specific and inter-season comparisons and assessment of potential public health risks. Food Chem Toxicol 50(2):162–167
Sanders M, Sivertsen S, Scott G (2002) Origin and distribution of polycyclic aromatic hydrocarbons in surficial sediments from the Savannah River. Arch Environ Contam Toxicol 43:438–448
Sardiña P, Lopez Cazorla AL (2005a) Feeding habits of the juvenile striped weakfish, *Cynoscion guatucupa* Cuvier 1830, in Bahía Blanca estuary (Argentina): seasonal and ontogenetic changes. Hydrobiologia 532(1–3):23–38
Sardiña P, Lopez Cazorla AL (2005b) Trophic ecology of the whitemouth croaker, *Micropogonias furnieri* (*Pisces: Sciaenidae*), in southwestern Atlantic waters. J Mar Biol Assoc UK 85(02):405–413
Sauer TC, Brown JS, Boehm PD, Aurand DV, Michel J, Hayes MO (1993). Hydrocarbon characterization of intertidal and subtidal sediment of Saudi Arabia from the Gulf War oil spill, vol. II. Marine Spill Response Corporation, Washington DC, MSRC Technical Report Series
Shi J, Zheng GJS, Wong MH, Liang H, Li Y, Wu Y, Li P, Liu W (2016) Health risks of polycyclic aromatic hydrocarbons via fish consumption in Haimen bay (China), downstream of an e-waste recycling site (Guiyu). Environ Res 147:233–240
Soares-Gomes A, Neves RL, Aucélio R, Van Der Ven PH, Pitombo FB, Mendes CL, Ziolli RL (2010) Changes and variations of polycyclic aromatic hydrocarbon concentrations in fish, barnacles and crabs following an oil spill in a mangrove of Guanabara Bay, Southeast Brazil. Mar Pollut Bull 60(8):1359–1363
Soclo HH, Budzinski H, Garrigues P, Matsuzawa S (2008) Biota accumulation of polycyclic aromatic hydrocarbons in Benin coastal waters. Polycycl Aromat Compd 28(2):112–127
Spacie A, Hamelink JL (1982) Alternative models for describing the bioconcentration of organics in fish. Environ Toxicol Chem 1: 309–320
Sprung M (1993) The other life: an account of present knowledge of the larval phase of *Dreissena polymorpha*. In: Nalepa TF, Schloesser DW (eds) Zebra mussels: biology, impacts and control. Lewis Publishers, USA, pp 39–54
Storelli MM, Barone G, Perrone VG, Storelli A (2013) Risk characterization for polycyclic aromatic hydrocarbons and toxic metals associated with fish consumption. J Food Compos Anal 31(1):115–119
Thorsen WA, Cope WG, Shea D (2004) Bioavailability of PAHs: effects of soot carbon and PAH source. Environ Sci Technol 38(7):2029–2037
Tolosa I, De Mora SJ, Fowler SW, Villeneuve JP, Bartocci J, Cattini C (2005) Aliphatic and aromatic hydrocarbons in marine biota and coastal sediments from the Gulf and the Gulf of Oman. Mar Pollut Bull 50(12):1619–1633
UNEP (1993) United Nations Environment Programme. UNEP/FAO/IOC/IAEA. Guidelines for monitoring chemical contaminants in the sea using marine organisms. Reference Methods For Marine Pollution Studies No. 6, pp 28
USEPA (1993) Schoeny, R. and K. Poirier. Provisional Guidance for Quantitative Risk Assessment of Polycyclic Aromatic Hydrocarbons. U.S. Environmental Protection Agency, Office of Research and Development, Office of Health and Environmental Assessment, Washington, DC, EPA/600/R-93/089 (NTIS PB94116571).
USEPA (2000) Guidance for assessing chemical contaminant data for use in fish advisories. Risk assessment and fish consumption limits, 3th edn. Office of Water, Washington DC
USEPA (2002). Polycyclic organic matter. US Environmental Protection Agency. Available from: https://www.epa.gov/sites/production/files/2016-09/documents/polycyclic-organic-matter.pdf
Van der Oost R, Beyer J, Vermeulen NP (2003) Fish bioaccumulation and biomarkers in environmental risk assessment: a review. Environ Toxicol Pharmacol 13(2):57–149
Varanasi U, Stein JE, Nishimoto M (1989) Biotransformation and disposition of PAH in fish. In: Varanasi U (ed) Metabolism of polycyclic aromatic hydrocarbons in the aquatic environment. CRC Press, Boca Raton, FL, pp 1–40
Varanasi U, Stein JE (1991) Disposition of xenobiotic chemicals and metabolites in marine organisms. Environ Health Perspect 90:93
Varanasi U, Brown DW, Hom T, Burrows DG, Sloan CA, Field LJ, Stein JE, Tilbury KL, McCain BB, Chan S (1993). Survey of Alaskan subsistence fish, marine mammal, and invertebrate samples collected 1989–91 for exposure to oil spilled from the Exxon Valdez, vol. 1. NOAA Technical Memorandum NMFS-NWFSC-12.
Vuorinen PJ, Keinanen M, Vuontisjarvi H, Barsien J, Broeg K, Forlin L, Gercken J, Kopecka J, Kohler A, Parkkonen J, Pempkowiak J, Schiedek D (2006) Use of biliary PAH metabolites as a biomarker of pollution in fish from the Baltic Sea. Mar Pollut Bull 53(8):479–487
Wetzel DL, Van Vleet ES (2004) Accumulation and distribution of petroleum hydrocarbons found in mussels (*Mytilus galloprovincialis*) in the canals of Venice. Italy Mar Pollut Bull 48(9):927–936
Wilcke W (2000) Synopsis polycyclic aromatic hydrocarbons (PAHs) in soil—a review. J Plant Nutr Soil Sci 163(3):229–248
Wilcke W (2007) Global patterns of polycyclic aromatic hydrocarbons (PAHs) in soil. Geoderma 141(3):157–166
Xu FL, Wu WJ, Wang JJ, Qin N, Wang Y, He QS, He W, Tao S (2011) Residual levels and health risk of polycyclic aromatic hydrocarbons

in freshwater fishes from Lake Small Bai-Yang-Dian, Northern China. Ecol Model 222(2):275–286

Zar JH (1996) Biostatistical analysis, 3rd edn. Practice Hall, New Jersey b25

Zhang Y, Li F, Wang J (2011) Determination of retardation effect of SOM on aqueous leaching of polycyclic aromatic hydrocarbons using confocal laser scanning microscope. Int J Environ Res 5(4):999–1008

Zhao Z, Zhang L, Cai Y, Chen Y (2014) Distribution of polycyclic aromatic hydrocarbon (PAH) residues in several tissues of edible fishes from the largest freshwater lake in China, Poyang Lake, and associated human health risk assessment. Ecotox Environ Safe 104:323–331