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Distribution of Aliphatic and Aromatic Hydrocarbons in Red Mullet (*Mullus barbatus*) and Annular Sea Bream (*Diplodus annularis*) from the Izmir Bay (Eastern Aegean)

Enis Darilmaz · Filiz Kucuksezgin

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Abstract Polycyclic aromatic and aliphatic hydrocarbons were analyzed in red mullet and annular sea bream from the Izmir Bay. These fishes were selected because of their multitude, wide distribution and common use in the Turkish diet; they were sampled and analyzed in 2000–2001 and 2004–2005 periods at five locations by trawling. Red mullet showed higher PAHs and aliphatics than annular sea bream. PAH levels for red mullet and annular sea bream ranged from 202 to 556 and 78.7 to 415; aliphatics ranged from 834 to 2,420 and 436 to 1,724 ng/g dw, respectively. Molecular ratios showed pyrolitic inputs for PAHs, biogenic and anthropogenic inputs for aliphatics. The carcinogenic PAH, benzo(a)pyrene, was detected in most fish samples in levels ranged between 22.2 and 64.1 ng/g dw. The average PAH contamination level was within the "moderate" category in fish from Izmir Bay. Results indicate the needing of a systematic monitoring program in order to provide accurately assessment and management of risks for the regional population.

Keywords Polycyclic aromatic hydrocarbons · Aliphatic hydrocarbons · Red mullet · Annular sea bream · Molecular ratios · Izmir Bay (Eastern Aegean)

Petroleum and its derivatives are the major contaminants in marine coastal areas. Petroleum-related hydrocarbons in the marine environment cause great concern due to their persistence in sediment and their toxicity for marine organisms. An oil spill is the release of oil (generally petroleum) into the natural environment. Fuel oils are complex mixtures of aliphatic and aromatic hydrocarbons. These include n-alkanes, branched alkanes, benzene and alkylbenzenes, naphthalenes, and heavier components such as PAHs. PAHs and their metabolites have the highest toxicity, being mutagenic, cytotoxic and potentially carcinogenic (IARC 1989). The United States Environmental Protection Agency (EPA) and World Health Organization (WHO) have identified 16 PAHs as priority pollutants, while some of these, e.g. benzo(a)anthracene, chrysene, benzo(a)pyrene are considered to be potential human carcinogens. PAHs are derived mainly from anthropogenic sources, though some of them may also be of biogenic origin. They enter in the aquatic environment primarily from petroleum sources and the long-range transport on particulates, following incomplete combustion of organic matter.

Marine species are widely used as bioindicators for environmental pollution because contaminants from seawater and their diet can be concentrated in their tissues. This enables the assessment of bio-accumulation, bio-concentration, and bio-magnification through the food chain. The presence of pollutants in fish and shellfish depends on the environmental concentrations of these compounds and on the physiology and ecological characteristics of the species.

Trace metal levels have been investigated extensively in the marine organisms of Izmir Bay (Kucuksezgin et al. 2006). Petroleum hydrocarbons have been studied in sediments from Izmir Bay (Darilmaz and Kucuksezgin 2007). The aim of this study was to evaluate the level, distribution, sources and trend of aliphatic and polycyclic aromatic hydrocarbons in two marine fish species, *Mullus barbatus* and *Diplodus annularis* in different sites within Izmir Bay.

E. Darilmaz · F. Kucuksezgin (✉)
Institute of Marine Sciences and Technology,
Dokuz Eylul University, Inciralti 35340,
Izmir, Turkey
e-mail: filiz.ksezgin@deu.edu.tr

As the species selected are regularly eaten in Turkey, it is important to know as much as possible about pollutants in edible fish with the aim of protecting consumers.

Materials and Methods

Izmir Bay, (Western Turkey), is one of the great natural inlets of the Mediterranean. The main urban conurbation is the Izmir Metropolitan Municipality, covering 88,000 hectares with a population of close to 3 million inhabitants. Izmir is an important industrial, commercial and cultural city. Industrial activities cover a large range of industries including food processing, tanneries, paint, chemicals, textiles and petroleum refining. Izmir Bay has a total surface area of over 500 km^2, water capacity of 11.5 billion m^3, a total length of 64 km. The Gediz River, which flows into the Outer Bay, is the most important input of fresh water.

Sampling took place in November 2000, in January, April and in August 2001, in November 2004 and in January, February, April, August and September 2005 at five locations shown in Fig. 1. *Mullus barbatus* and *Diplodus annularis* were collected by trawling from Izmir Bay with vectors based at Foca, Gediz, Uzunada, Gulbahce and Guzelbahce. Muscle tissue of the samples was removed in the field and preserved at −20°C until analyzed. Prior to extraction, about 2.5–3 g of freeze-dried fish samples placed into the microwave extraction tubes and internal, non-naturally occurring aliphatic and PAH standards (nC19 d40, nC32 d66, Hexamethylbenzene, Cadalene, Naphthalene-d8) are added for the quantitative calculations. All samples were extracted using a microwave extraction system for 30 min, with methanol. After the extraction is completed, 2 M KOH were added to the flask to saponify the lipids. Extractable organic matter (HEOM) was determined by evaporating a small measured volume of the extract.

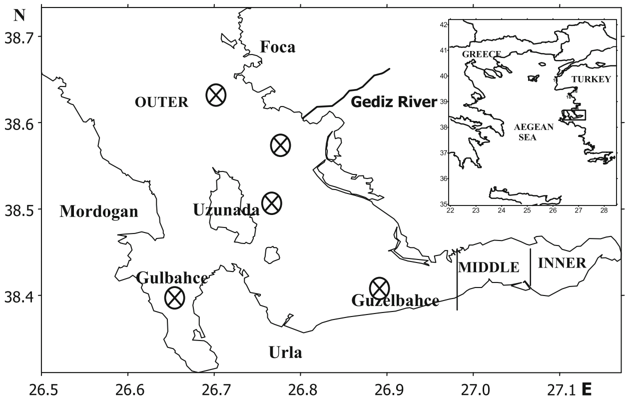

Fig. 1 Sampling areas (*circle* with *cross*) in the Izmir Bay

Extracts are then separated into two fractions, using a column chromatography on deactivated silica, deactivated alumina and anhydrous Na_2SO_4. A first fraction was eluted with hexane. The second saturated fraction was eluted with a solution of hexane:dichloromethane at a ratio of 90:10. The first fraction has saturated aliphatic hydrocarbons, the second fraction has unsaturated and aromatic hydrocarbons. Detection limits were 2.01–9.1 ng g^{-1} dw for aliphatics and 3.97–9.79 ng g^{-1} dw for PAHs. Fractions were then analyzed by high-resolution gas chromatography with a split/splitless capillary injection system and flame ionization detector. The samples were analyzed in the splitless mode using a fused silica capillary column (DB-5MS Column 30 m × 0.25 mm, film thickness 0.25 μm). Nitrogen was used as a carrier gas, at flow rates of 1.6 mL min^{-1}. The injector and detector temperatures were 270°C and 300°C, respectively. The GC temperature programmed was from 60°C (3 min) to 300°C at 3°C min^{-1}, and held at the final temperature for 15 min.

Hydrocarbon data are mean of two replicate analyses. For every set of 10 samples, a procedural blank and spike sample are run to check for interference and cross contamination. The hydrocarbon recoveries are determined relative to the ratio of direct injection of the extract and the working standards prepared in hexane. Also, the quality of the analytical data is assured using the reference materials of an IAEA-435 tuna fish sample (from the IAEA, Monaco). The recoveries for fish fell within a fairly narrow range, for internal standards between 77.0 and 87.1%.

Statistical analyses were performed using STATISTICA 6.0 (Stat. Soft. Inc., 1995). Pearson's Product-Moment Correlation test was used to check for significant relationships between hydrocarbons and HEOM in muscle. In all case, the level of significance was set at $p < 0.05$. One-way analysis of variance (ANOVA) was used to detect temporal and spatial differences in hydrocarbon concentrations. In this study, principal component analysis (PCA) was applied to the fish samples. The standardization of the data is reasonable when variables are in different units or the variance of the different columns of raw data is substantial. The data were standardized to eliminate scaling before PCA processing.

Results and Discussion

Aliphatic and aromatic hydrocarbon concentrations are reported in Tables 1, 2 for *M. barbatus* and *D. annularis* in the years 2000, 2001, 2004 and 2005. The total mean concentrations ranged from 0.83 to 2.42 μg g^{-1} and 0.44 to 1.72 μg g^{-1} dw in *M. barbatus* and *D. annularis*, respectively. The concentrations of total aliphatics were generally higher than 1 μg g^{-1} dw in Izmir Bay. Higher concentration

Table 1 Concentrations of aliphatics and aromatic hydrocarbons (ng/g dw), extractable organic matter (mg/g), molecular ratios and fork length (FL) in *Mullus barbatus*

	Gulbahce				Uzunada			Foca			Guzelbahce		Gediz	
	2000	2001	2004	2005	2000	2001	2005	2000	2001	2004	2004	2005	2004	2005
n (sample)	17	23	20	69	10	15	75	19	23	10	10	10	11	43
FL (mm)	111–169	124–184	146–177	92–158	130–183	140–198	110–195	130–202	127–184	86–103	145–159	111–120	140–149	122–170
Aliphatics														
nC17	74.0	61.0	168	103	51.0	56.0	98.7	49.6	61.7	46.0	121	165	190	62.0
Pri	138	152	171	185	52.5	83.1	112	212	156	47.4	128	170	203	64.5
nC18	88.3	83.0	92.3	48.0	25.2	43.8	63.8	73.3	28.0	37.1	73.0	34.0	97.3	56.2
Phy	96.3	94.8	114	153	47.6	51.9	72.5	127	120	45.9	76.2	65.0	102	61.3
Pri/Phy	1.88	1.49	1.44	1.41	1.10	1.60	1.71	1.66	1.29	1.03	1.68	2.62	2.00	1.06
nC17/Pri	0.61	0.60	0.98	0.73	0.97	0.67	0.87	0.24	0.45	0.97	0.95	0.97	0.94	0.94
nC18/Phy	0.88	0.87	0.79	0.60	0.53	0.84	0.92	0.58	0.23	0.81	0.96	0.52	0.96	0.91
Σ(C10–C34)	1,387	1,425	1,371	1,167	711	1,149	1,320	1,409	877	720	1,394	826	2,107	1,669
ΣAliphatics	1,666	1,731	1,889	1,563	841	1,293	1,525	1,793	1,169	834	1,710	1,075	2420	1,843
Aromatics														
Nap	304	282	245	197	239	323	218	321	288	109	205	350	266	234
1-m-Nap	15.3	18.5	21.1	14.2	16.4	22.3	10.9	13.5	16.8	8.52	27.9	31.7	21.3	17.4
1 e-Nap	8.89	nd	13.8	2.06	nd	nd	4.64	16.4	9.62	nd	nd	nd	nd	nd
Phe	18.0	18.6	nd	4.59	nd	15.0	14.7	11.3	nd	nd	nd	nd	8.06	nd
Ant	43.0	50.9	nd	nd	nd	78.8	nd	30.6	nd	nd	nd	nd	43.4	nd
2-m-Phe	14.0	nd	nd	6.16	nd	nd	nd	7.72	nd	nd	nd	nd	10.3	nd
1-m-Phe	7.30	nd	nd	nd	nd	nd	nd	21.1	nd	nd	18.0	nd	nd	nd
Flt	4.34	5.73	nd	nd	nd	11.7	2.36	8.07	nd	nd	nd	nd	9.42	23.6
Pyr	3.79	4.15	nd	nd	nd	10.5	1.84	4.10	nd	nd	nd	nd	7.80	13.5
BbF	3.86	nd	nd	nd	nd	7.78	1.97	3.47	nd	nd	7.44	nd	nd	nd
BkF	nd	nd	nd	nd	nd	7.83	1.62	4.60	nd	nd	8.69	nd	nd	nd
BeP	3.29	4.12	nd	6.97	12.4	nd	6.58	5.04	nd	36.9	nd	nd	7.30	nd
BaP	42.5	54.4	39.4	44.1	59.0	56.0	45.4	57.4	64.1	47.9	45.7	42.2	40.4	nd
Per	21.6	21.6	7.76	6.12	32.0	23.6	31.1	20.9	54.3	nd	nd	24.5	25.1	20.6
ΣPAHs	489	460	327	281	359	556	339	525	433	202	313	448	439	309
BeP/BaP	0.08	0.08	–	0.16	0.21		0.14	0.09	–	0.77	–	–	0.18	–
Phe/Ant	0.42	0.37	–	–	–	0.19	–	0.37	–	–	–	–	0.19	–
Flt/Pyr	1.14	1.38	–	–	–	1.11	1.28	1.97	–	–	–	–	1.21	1.76
HEOM	97.1	132	118	53.6	52.0	167	94.8	174	149	95.0	114	56.0	103	45.2

nd not detected

Table 2 Concentrations of aliphatics and aromatic hydrocarbons (ng/g dw), extractable organic matter (mg/g), molecular ratios and fork length (FL) in *Diplodus annularis*

	Gulbahce			Uzunada			Foca		Guzelbahce	Gediz	
	2000	2001	2005	2000	2001	2005	2000	2001	2005	2004	2005
n (sample)	13	27	28	10	20	56	10	14	25	15	24
FL (mm)	116–138	111–150	105–122	116–130	121–150	120–168	120–125	105–150	110–129	110–116	100–120
Aliphatics											
nC17	65.8	44.3	54.7	62.1	39.5	51.5	51.5	56.7	78.0	60.3	38.8
Pri	73.3	54.0	57.9	64.2	47.7	55.7	55.1	65.4	80.9	64.7	41.7
nC18	55.4	18.3	43.3	38.8	31.7	40.0	50.7	44.1	50.6	57.2	29.8
Phy	71.4	41.2	53.9	43.3	34.3	50.3	52.3	55.4	60.4	61.6	32.4
Pri/Phy	1.03	1.31	1.07	1.48	1.39	1.11	1.05	1.18	1.34	1.05	1.29
nC17/Pri	0.90	0.82	0.95	0.97	0.83	0.92	0.93	0.87	0.96	0.93	0.93
nC18/Phy	0.78	0.44	0.80	0.90	0.92	0.80	0.97	0.79	0.84	0.93	0.92
Σ(C10–C34)	941	1,097	368	1,078	441	950	884	1562	587	953	362
ΣAliphatics	1,085	1,196	489	1,204	540	1,059	1,006	1724	753	1,112	436
Aromatics											
Nap	288	120	208	254	269	160	nd	243	127	172	18.1
1-m-Nap	18.7	3.96	8.12	8.89	26.6	14.3	nd	17.5	9.37	17.5	2.45
1 e-Nap	nd	nd	nd	nd	20.5	12.4	nd	10.5	nd	nd	4.71
Phe	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Ant	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
2-m-Phe	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
1-m-Phe	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Flt	12.6	4.76	9.08	nd	nd	nd	17.8	22.0	nd	nd	10.7
Pyr	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
BbF	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
BkF	nd	nd	14.0	nd	nd	nd	15.3	6.15	nd	nd	nd
BeP	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
BaP	58.0	43.1	50.5	45.4	38.0	40.7	25.0	22.2	35.8	42.2	34.8
Per	38.0	27.8	41.0	44.0	23.6	8.39	31.0	35.3	13.1	24.1	8.00
ΣPAHs	415	200	331	352	377	236	89.1	357	185	256	78.7
Flt/Pyr	3.61	1.36	2.59	–	–	–	5.08	6.30	–	–	3.06
HEOM	74.0	51.2	26.8	42.0	60.0	32.4	31.0	60.1	32.3	76.0	13.7

nd not detected

for red mullet occurred at the Gediz site. The recorded concentrations were higher than those recorded for annular sea bream. The values of nC17 and nC18 ranged between 46–190 and 25–97 ng/g for red mullet; 39–78 and 18–57 ng/g for annular sea bream, respectively. High concentrations at the Gediz sites in different sampling periods (during 2004 and 2005), are probably related to fisheries and to anthropogenic activities. The origin of these two isoprenoid hydrocarbons, C17 and C18 are plankton (Lacaze 1980), algae (Blumer et al. 1971) and bacteria (Albaigés et al. 1984).

However, the majority of the resolved alkanes had odd-carbon chain lengths. This suggested that the presents of aliphatics due not only to anthropogenic activity but also result of marine phytoplankton and terrestrial vegetation. This is indicated by the presence of phytane and pristane at all stations. In general, pristane has a biochemical origin while phytane comes from geochemical processes. Pristane and phytane are often considered as good indicators of petroleum contamination. The ratio of pristane to phytane varies between oils, reflecting the depositional environment of the original source. As a rule, a high ratio of pristane to phytane or the predominance of a single isoprenoid (such as pristane) indicates a biogenic source (UNEP/IOC/IAEA 1992). At all sampling stations Pri/Phy ratios were >1 indicating the biogenic source (Table 1). The highest values were found in Gulbahce region during 2004.

The limitations of using n-alkanes to isoprenoid ratios, such as nC18/phytane and nC17/pristane, for monitoring biodegradation were demonstrated in this study. The ratio of nC17/pristane and nC18/phytane are often used as qualitative measures of biodegradation (Ezra et al. 2000), because pristane and phytane are relatively persistent biomarkers. As the fuel contaminant is biodegraded, bacteria preferentially consume the nC17 and nC18 compounds, resulting in a relative enrichment of pristane and phytane in the residue. The relative nC18/Phy and nC17/Pri ratios at all stations ranged from 0.23–0.96 to 0.24–0.98 for red mullet, 0.44–0.97 and 0.82–0.97 for annular sea bream, respectively.

Aliphatic hydrocarbon concentrations in fish from Izmir Bay were lower than those in North Western Mediterranean coast (Villeneuve et al. 1999), Gulf of Naples (Amodio-Cocchieri and Cirillo 2003), Gulf of Oman (Tolosa et al. 2005), and higher than those in Egyptian Mediterranean Sea (El-Deeb et al. 2007).

The total mean PAH concentrations ranged from 202 to 556 and 78.7 to 415 ng g^{-1} dw in *M. barbatus* and *D. annularis*, respectively. The concentrations of total aromatics were generally lower than 1 µg g^{-1} dw in Izmir Bay. Higher and lower concentrations for red mullet occurred at the Uzunada and Gediz sites. The recorded concentrations were higher than those recorded for annular sea bream. The values of naphthalene ranged between 109 and 350 ng.g^{-1} for red mullet; nd-288 ng g^{-1} for annular sea bream in the study area. Naphthalene is the dominant PAHs for both fish species. Thomann and Komlos (1999) reported that, among PAHs with Log K_{ow} (octanol/water partition coefficient) less than 5, no metabolism was documented for naphthalene, phenanthrene, and anthracene in fish. Therefore, direct exchange with the water through the gills served as higher levels of naphthalene in red mullet and annular sea bream.

The specific PAH Ace, Acy, Flu, 3,6-dm-Phe, 1-m-Pyr, BaA, Chr, IP, dBA and BPer were under the detection limits in most of the sites or at very low levels in both species. In addition, Phe, Ant, Pyr and BbF were also measured below detection limit in annular sea bream. The highest values were found for Nap, 1-m-Nap and BkF in Guzelbahce, for 1-e-Nap, 1-m-Phe, BeP, BaP and Per in Foca, for Phe and 2-m-Phe in Gulbahce, for Ant and BbF in Uzunada, for Flt and Pyr in Gediz for red mullet; for Nap and BaP in Gulbahce, for 1-m-Nap, 1-e-Nap and Per in Uzunada, for Flt and BkF in Foca for annular sea bream.

The molecular ratios were calculated to monitor the pyrolytic or petrogenic processes generating PAH (Tolosa et al. 2004). In general, a ratio of Phe/Ant of <10, Flt/Pyr of >1 and Flt/(Flt + Pyr) >0.5 suggests that PAH contamination arises from pyrolytic origins; all samples were characterized as Phe/Ant <10 and Flt/Pyr >1 in red mullet. In annular sea bream, when values for Pyr and BbF were not detected, a value of half the detection limit was used for the calculations of ratios (Table 2). This is probably due to shipping activities, fisheries and residential heating by coal. El-Deeb et al. (2007) reported a method to indicate the sources of PAHs by using the ratios BbF/BaP and BkF/BaP (pyrolitic <0.5; petrogenic >0.5). All the BbF/BaP and BkF/BaP ratios lower than 0.5 suggest a pyrolitic origin of PAHs in both species.

Polycyclic aromatic hydrocarbon concentrations in fish from Izmir Bay were lower than those in Abu Qir Bay (El-Deeb et al. 2007) and higher than those in Gulf of Oman (Tolosa et al. 2005), Adriatic Sea (Perugini et al. 2007).

Baumard, in a study targeting sedimentation, had concluded that “low” could be defined as 0–100 ng/g dw, “moderate” as >100–1,000 ng/g dw, “high” as >1,000–5,000 ng/g dw and “very high” as >5,000 ng/g dw. If the classification developed by Baumard et al. (1998) is applied to the present study it is seen that the average PAH contamination levels fall within the “moderate” category.

Total hexane extractable organic matter (HEOM) ranged from 45 to 174 (mg g^{-1}) in red mullet, from 14 to 76 (mg g^{-1}) in annular sea bream (Tables 1, 2). Total aliphatics and total aromatic hydrocarbon concentrations in fish showed no relation to HEOM.

The ANOVA test showed no significant temporal differences for total aliphatics ($p < 0.05$) in *M. barbatus* ($p = 0.7923$, $F = 0.3461$, $df = 3$) and in *D. annularis* ($p = 0.1715$, $F = 1.9279$, $df = 3$). Non-significant spatial differences were found in species. The ANOVA test indicated significant temporal differences for total PAHs in *M. barbatus* ($p = 0.029$, $F = 3.6313$, $df = 3$). However, no significant differences were detected in *D. annularis* ($p = 0.6051$, $F = 0.6343$, $df = 3$).

In this study, principal component analysis (PCA) was deployed in order to analyze the data. The first step in the multivariate statistical analysis was application of PCA with the aim of grouping the individual aliphatic. The first three principal components explained 92.4% of the total variability in *D. annularis*. These three principal components were highly weighted due to the contributions of aliphatic hydrocarbons. After auto scaling, the two significant components were identified giving account for 55.4% and 26.6% of the total variation, respectively. The third component takes into account only 10.5% of the total variation.

After PCA calculations had been made, three principal aliphatic compounds were seen to define almost 90.2% of the total variability. These principal components account for 65.6%, 15.3% and 9.3%, respectively of the total variation of aliphatics found in *M. barbatus*.

The score plot of first two components allows for the characterization of the aromatic compounds in terms of the first and second component. In *D. annularis*, three principal components account for 99.8% of the total variation, with values for PC1, PC2 and PC3 of 83.3%, 12.6% and 3.9%, respectively. Component 1 was represented by compounds 1-m-Nap, 1-e-Nap, BkF, Flt and Per in the positive part. BkF is a component of fossil fuels and a portion of BkF is associated with its combustion. PC2 represented Nap in the positive part. Nap dominated every sample and was clearly separated from the other compounds by its ubiquity. BaP is well differentiated by having the higher scores on the PC3 in the positive part. BaP is associated with traffic emission/fosil fuel combustion component. In general the heavier PAHs are the result of the combustion and/or pyrolitic processes and are absent in crude oil or refined products.

In *M. barbatus*, the first three principal components explained 99.3% of the total variation with the values for PC1, PC2 and PC3 being 97.2%, 1.5% and 0.6%, respectively. The first component was mainly positively related to all measured aromatics except BaP and Nap, whereas PC2 was positively related to BaP. PC3 represented Nap in the positive part. Consequently PC1 was defined as a general traffic emission/fossil fuel combustion component. Flt and Pyr are typical markers for pyrolysis or in complete combustion. On the other hand, focusing at fuel combustion emission profile predominantly contains Phe, Flt and Pyr. 2-m-Nap, Phe and Ant belong to the low molecular weight PAHs with 2–3 rings or alkyl-substituted PAHs, which are abundant in petrogenic sources mainly caused by petroleum spills.

Since February 2005 the European Regulation 208/2005/EC fixed at 5 ng g^{-1} wet weight (ww) the maximum admissible concentrations for BaP in crustaceans and cephalopods, at 2 and 10 ng g^{-1} ww in fish muscle and in shellfish, respectively (Commission Regulation 2005). The highest values were observed in Gulbahce for annular sea bream and in Foca for red mullet. In present study average value of dry weight to wet weight is about 0.20. This is because all samples analyzed showed values of BaP for red mullet and annular sea bream from Izmir Bay as being well below the values established by the law.

The present work represented the first detailed study of the distribution and origin of petroleum hydrocarbons in fish from the Izmir Bay, an area economically important for the western part of Turkey due to maritime, industrial and tourism activities. This investigation showed petroleum hydrocarbons in the bay consist of a mixture of compounds from natural and anthropogenic sources. Considering concentration ratios between the different PAHs, it was possible to indicate pyrolysis processes as the most probable source of contamination. The ratios of pristane and phytane were higher than 1 in two species, indicating a high contribution from biogenic hydrocarbons. Levels of total PAHs in fish at various sampling locations of the Izmir Bay were found moderate concentrations, registering a wide spectrum of molecular weights. The levels of BaP all the fish samples were below the guideline of 10 ng g^{-1} ww reported by European Commission. The levels of petroleum hydrocarbons in two species were comparable to the concentrations observed in relatively unpolluted areas elsewhere in the world. Regular monitoring of the area and more specific analyses are recommended in Izmir Bay. Further studies should be necessary to improve the state of knowledge on the relative potency of individual PAHs and estimate their carcinogenic toxicity.

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