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Polychlorinated biphenyl residues in the golden grey mullet (*Liza aurata*) from Tunis Bay, Mediterranean sea (Tunisia)

Wièm Masmoudi [a,b,c,*], Mohamed Salah Romdhane [b], Souhaila Khériji [a,b,c], M'hamed El Cafsi [a]

[a] *UR Physiologie et Ecophysiologie des Organismes Aquatiques, Département des Sciences Biologiques, Faculté des Sciences de Tunis, Campus Universitaire, 2092 Tunis, Tunisia*

[b] *UR Ecosystèmes et Ressources Aquatiques, Institut National Agronomique de Tunis, 43 av. Charles Nicolle 1082 Tunis, Tunisia*

[c] *Institut Supérieur de Pêche et d'Aquaculture de Bizerte, Rimel 7080, BP 15, Bizerte, Tunisia*

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Abstract

Concentrations of polychlorinated biphenyls (PCBs) were determined in the muscle tissues of mullets (*Liza aurata*) caught from two coastal marine areas in Tunis bay, "Raoued" and "Radès". Twenty congeners were detected in all the fish samples analyzed, among which were 4 congeners recommended by the European Union for assessing pollution and 4 "dioxin-like" congeners. Residue levels of total PCBs ranged from 45 to 194 ng/g wet wt in the mullet from "Radès" and from 43 to 65 ng/g w wt in specimens from "Raoued". Hexachlorobiphenyls were predominant in fishes caught from both "Raoued" and "Radès" areas, accounting for 43% and 40%, respectively. The individual PCB profiles were dominated by congeners 153 in mullet from "Raoued" and PCB 200 in fishes from "Radès". The levels of TEQs of "dioxin-like" congeners were 3.4 pg/g w wt for fishes from "Raoued" and 2.5 pg/g w wt for specimens from "Radès".

Keywords: Polychlorinated biphenyls; Mullet; Tunis bay

1. Introduction

Polychlorinated biphenyls (PCBs) are synthetic chemical compounds that are very widespread in the environment. These persistent organic pollutants (POPs) are commonly used in many industrial activities since they make up the composition of some electrical equipment and ship paint. Also, these compounds could be derived from combustion of some plastic material and from paper mills (Takasuga, Makino, Umetsu, & Senthilkumar, 2003). PCBs are highly lipophilic and are rapidly accumulated in organisms. These very persistant compounds have the potential to affect the physiological functions of widlife. Dallaire et al. (2006) point out that environmental prenatal exposure to PCBs is associated with incidence of acute respiratory infections in children. PCBs were also identified as endocrine disruptors, mainly with estrogenic effects (Munoz-de-Toro et al., 2006). Many authors have revealed that high fish and seafood consumption increases the risk of PCB contamination of the human body (Cuadra, Linderholm, Athanasiadou, & Jakobsson, 2006; Karouna-Renier, Snyder, Allison, Wagner, & Ranga-Rao, 2007).

Tunis Bay is located on the Mediterranean sea of northern Tunisia with an area of 1500 km^2 and 160 km of coasts. This zone is submitted to the impact of many chemical and physical stressors related to human activities: industry, agriculture, tourism and urbanization (Raïs, 1999; Zaïri, Ferchichi, Ismaïl, Jenayeh, & Hammami, 2004). Wastes resulting from these activities are dumped into the sea via

* Corresponding author. Address: UR Physiologie et Ecophysiologie des Organismes Aquatiques, Département des Sciences Biologiques, Faculté des Sciences de Tunis, Campus Universitaire, 2092 Tunis, Tunisia. Tel.: +216 71 882 200; fax: +216 71 885 480.

E-mail address: maswiem@lycos.com (W. Masmoudi).

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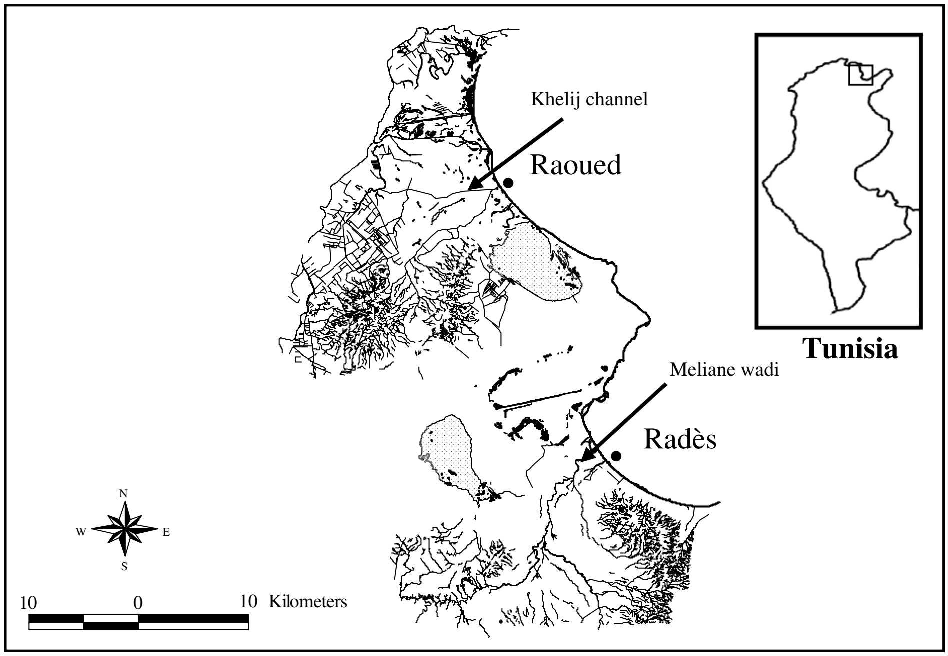

Fig. 1. Location of the sampling sites.

two channels: “Meliane” wadi flowing to “Radès” beach and “Khelij” channel flowing into “Raoued” beach (Fig. 1). Coastal fishing activity is very much developed in these areas. Among the exploited fishes, the golden grey mullet (*Liza aurata*) is very much appreciated by Tunisian consumers. The Mugilidae species are euryhaline fish, inhabiting especially river mouths and lagoons and are known to concentrate pollutants (Ferreira, Antunes, Gil, Vale, & Reis-Henriques, 2004; Masmoudi, Romdhane, Kheriji, & El Cafsi, 2001; Pastor, Boix, Fernandez, & Albaiges, 1996).

A previous study revealed the presence of residual polychlorinated biphenyls in surficial sediment of “Meliane” wadi and in marine stations located offshore (Masmoudi, Romdhane, Kheriji, & El Cafsi, 2005). But, to our knowledge, no data have yet mentioned contamination of marine organisms by the polychlorinated biphenyls from Tunis bay. Therefore, our purpose in the present work is to assess the chemical quality of the golden grey mullet from “Raoued” and “Radès” by revealing the possible presence of PCB residues in the fish flesh in order to evaluate risks to human health.

2. Materials and methods

The present work was carried out in two coastal sites in Tunis Bay: “Raoued”, close to the “Khelij” channel outlet and “Radès” in the proximity of “Méliane” wadi’s mouth (Fig. 1). Specimens of the golden grey mullet (*L. aurata*) (length: 6.4 ± 0.7 cm; weight: 3.7 ± 1.2 g) were sampled using a net, in March 2004. The fish were immediately sacrificed, weighed, measured, dissected and kept frozen (−20 °C) until required for chemical analyses.

The PCB analysis was conducted in composite samples (3 fishes per pool and 3 pools per site) of fish muscles according to a standard procedure (UNEP, 1996). Freeze-dried samples were Soxhlet-extracted with hexane for 8 h. The extracts obtained were dried by rotary evaporator in order to determine (gravimetrically) the fat content. After that, the fat was dissolved in hexane and cleaned-up with concentrated sulphuric acid (H_2SO_4). Then, the treated extract was concentrated (about 1 ml) and was added to an open column packed with florisil (60–100 mesh; activated at 120 °C for 8 h) with a small amount of anhydrous sodium sulphate on top. The column was eluted by hexane (70 ml). The collected eluate was again concentrated and the volume fixed to exactly 1 ml prior to gas chromatographic analysis. The determination of PCBs was carried out using a VARIAN CP 3380 electron capture detector gas chromatograph. The capillary column was a 15 m CP SIL type, with an internal diameter of 0.25 mm. The initial temperature was 50 °C (holding for 1 min), followed by a ramp to 140 °C (holding for 4 min) at a rate of 30 °C/min, then to 250 °C at a rate of 5 °C/min. The injection was in splitless mode at 250 °C and the detector temperature was maintained at 300 °C. Nitrogen was used as the carrier gas and the make up gas. The standards employed were obtained from Supelco (chemical purity 99%) and made up as a mixture of 36 PCB congeners, including the 7 congeners (IUPAC 28, 52, 101, 118, 153, 138 and 180) recommended by the European Union for assessing the pollution by PCBs (Commission of the European Communities, 1999) and also the “dioxin-like” PCBs (four non-ortho PCBs: congeners 77, 81, 126 and 169; and eight mono-ortho PCBs: PCBs 105, 114, 118, 123, 156, 157, 167 and 189) which have been assigned a toxic equivalency factor by the World Health Organisation (WHO-TEF) (Van den Berg et al., 1998).

3. Results and discussion

Among the 36 investigated PCBs, only 20 congeners were omnipresent in all fish samples of both sites. Residue levels of total PCBs ranged from 45 to 194 ng/g w wt with a median of 96 ng/g w wt in the mullet from "Radès". The concentrations of PCB residues in fishes from the "Raoued" site were between 43 and 65 ng/g w wt with a median of 52 ng/g w wt. (Table 1). Profiles of polychlorinated congeners, according to the degree of chlorination, revealed that hexachlorobiphenyls were predominant in fishes caught from both "Raoued" and "Rades" areas, accounting for 43% and 40%, respectively, followed by octachlorobiphenyls with contributions of 20% and 32% in fishes from Raoued and Radès, respectively. Hepta- and pentachlorobiphenyls exceed 10% in fishes from both Raoued and Rades while tetra- (8% in Raoued fishes and 1% in Radès fishes) and nonachlorobiphenyls (2 % in both Raoued and Radès fishes) made up the smallest percentages of total PCB residues (Fig. 2).

This kind of distribution of PCB congeners was also registered in other studies on fishes from the Mediterranean sea (Garcia, Porte, & Albaiges, 2000; Garritano et al., 2006; Porte, Escartin, Garcia, Sole, & Albaiges, 2000; Storelli, Giacominelli-Stufer, Storelli, & Marcotrigiano, 2003; Storelli & Marcotrigiano, 2006; Storelli, Barone, Garofalo, & Marcotrigiano, 2007). These reports noted that hexachlorobiphenyls were the most abondant congeners. Studying the bioaccumulation patterns of PCBs in mesopelagic fishes, Garcia et al. (2000) demonstrated that the less chlorinated PCB congeners were the most eliminated by liver metabolic activities. In fact, the biodegradability of PCBs in the environment depends essentially on the number of chlorines. PCBs with more chlorine atoms are more lipophilic and show a strong adsorption capacity (Barakat, Kim, Qian, & Wade, 2002; Nie, Lan, Wei, & Yang, 2005; Tolosa et al., 1997).

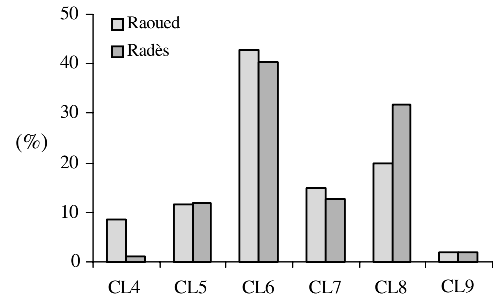

Fig. 2. Percentage of PCBs represented as chlorination levels in the mullet muscle from Raoued and Radès.

Table 1
Minimum, maximum and mean concentrations (ng/g w wt) of individual PCB congeners in the mullet muscle from Raoued and Radès

Congeners	Raoued	Radès
% Fat	(1.2–5) 3.2	(1.1–8.5) 3.7
PCB 77	(4.1–5.4) 4.5	(0.9–1.1) 1
PCB 99	(0.1–1.1) 0.5	(0.2–0.4) 0.3
PCB 100	(0.6–2.1) 1.2	(1.8–7.1) 3.8
PCB 101	(0.5–1) 0.8	(0.9–6.8) 3.2
PCB 110	(0.4–1.8) 1.3	(ND-2.1) 1
PCB 118	(2.3–2.5) 2.4	(2.1–4.9) 3.2
PCB 138	(3.9–4.3) 4.1	(2.7–14.6) 6.8
PCB 141	(ND-0.4) 0.2	(0.1–1.7) 0.6
PCB 149	(5.7–6.9) 6.2	(5–26.2) 12.4
PCB 153	(ND-20.4) 11.6	(8.1–36.2) 18.8
PCB 156	(0.1–0.7) 0.4	(ND-0.4) 0.2
PCB 169	(0.1–0.5) 0.3	(0.2–0.3) 0.2
PCB 170	(1.1–3.1) 2.1	(0.6–9.9) 3.9
PCB 176	(ND-4.9) 2.7	(ND-3.6) 2.4
PCB 187	(2.6–3.1) 2.8	(2.1–10.9) 6
PCB 194	(ND-4.9) 2.7	(0.1–0.6) 0.4
PCB 198	(0.2–0.6) 0.4	(ND-0.5) 0.2
PCB 200	(ND-11.7) 7.7	(6.8–64.3) 26.4
PCB 203	(0.9–2.9) 1.9	(1.6–7.8) 3.8
PCB 206	(0.1–2.3) 1	(1.4–2.5) 1.9
$\sum$ PCBs	(43–65) 52	(45–194) 96
Target PCBs[a]	(224–2225) 961	(185–5566) 2350

[a] Sum of the PCB 101, PCB 118, PCB 138 and PCB 153 (ng/g fat wt).

PCB 153 was the predominant congener in the mullet flesh from "Raoued" (11.6 ng/g w wt). Similar results have been reported for PCB 153 as the predominant congener in fishes from the Mediterranean sea (Ferreira et al., 2004; Pastor et al., 1996; Porte & Albaiges, 1993; Stefanelli et al., 2004; Storelli, Storelli, & Marcotrigiano, 2005; Storelli et al., 2007) but also in fishes from other seas (Coelhan, Strohmeier, & Barlas, 2006; Yang, Matsuda, Kawano, & Wakimoto, 2006). PCB 153 have Chlorines at 2–4 or 5 positions in one or both ring. This feature seems to be responsible of the persistency and bioaccumulative properties (Bright, Grundy, & Reimer, 1995). The highest level of PCB congener residues in mullet from Radès was for PCB 200 (26.4 ng/g w wt), followed by PCB 153 (18.8 ng/g w wt) and PCB 149 (12.4 ng/g w wt).

Among the seven "target" congeners, only PCBs 101, 118, 138 and 153 were detected in all samples from both sites. PCBs 28, 52 and 180 were below the detection limits. The sum of these congeners are expressed also on a fat weight basis in order to compare our results with data cited in the literature. Concentrations of the target PCB congeners in mullet from "Raoued" were 224–2225 ng/g fat wt, with a median of 961 ng/g fat wt, while, in fishes caught from "Radès", PCB concentrations were 185–5566 ng/g fat wt with a median of 2350 ng/g fat wt. These levels (found here) are higher than those reported in mullet species from the Ebro delta in the western Mediterranean (109–611 ng/g fat wet) (Pastor et al., 1996), from the Marmara sea in Turkey (508 ng/g fat wt) (Coelhan et al., 2006) and from the Tyrranean sea in Italy (9 ng/g w wt) (Garritano et al., 2006). Comparable levels were reported in other fish species from the Mediterranean sea, such as in a benthic species *Lophius budegassa* (3791 ng/g fat wt) (Storelli,

Storelli, Barone, & Marcotrigiano, 2004) and in mackerel (652–1094 ng/g fat wt) (Stefanelli et al., 2004). It seems that habit, physiological factors, lipid content, geographical origin and feeding behaviour of fishes are important aspects that explain pollutant storage and elimination (Perrugini et al., 2004). As already deduced by Ferreira et al. (2004), higher levels of PCBs in the mullet tissues can be attributed to the feeding strategies or life style, which lead to a high uptake of contaminants.

Actually in Tunisia, a maximum residue limit (MRL) for PCBs in fish is not yet established. However, the European Union has recommended a tolerance limit of 200 ng/g fat wet, calculated as the sum of the seven target PCBs in terrestrial edible animals but not in fish (Commission of the European Communities, 1999). Our values exceeded the MRL of 200 ng/g fat wt but we cannot take into account this limit for the evaluation of the contamination in our fish samples. Nevertheless, a population that ingests high amounts of fish could encounter health risks. To evaluate these risks, we are also interested to the coplanar PCBs, also called dioxin-like PCBs, which are considered as the most toxic of all 209 congeners. Mean concentrations of mono (PCB 118 and PCB 156) and non-ortho coplanar PCBs (PCB 77 and PCB 169) present in all samples examined are listed in Table 2. In fishes from Raoued, coplanar PCBs represent a fraction of 14% of the total PCBs while, in mullet from Radès, these congeners made up a lower fraction of the total PCBs (5%). In Raoued samples, PCB 77 accounted for most of the PCB coplanar content with a percentage of 60.3%, followed by PCB 118 (31.6%), PCB156 (4.8%) and PCB 169 (3.3%). In Radès fishes, PCB 118 contributed with the highest percentage (68.9%), followed by PCB 77 (22.2%), PCB 156 (4.5%) and PCB 169 (4.3%) (Fig. 3). Moreover, the non-ortho PCB concentration patterns suggest a possible detoxifying activity in wildlife when a depletion of PCB 77 is registered (Tanabe, Kannan, Subramanian, Watanabe, & Tatsukawa, 1987). In our case, PCB 77 > PCB 169, and this kind of pattern of non-ortho coplanar PCBs shows little or non-existent detoxifying metabolic activity in fishes (Corsolini, Aurigi, & Focardi, 2000).

Table 2
Mean concentrations of non- and mono-ortho coplanar PCBs (ng/g w wt) and their TEQs (pg/g w wt) in the mullet muscle from Raoued and Radès

Coplanar PCBs	TEF	Concentrations		TEQs	
		Raoued	Radès	Raoued	Radès
Non-ortho					
PCB 77	0.0001	4.5	1.0	0.45	0.1
PCB 169	0.01	0.3	0.2	3.00	2.0
Total		4.8	1.2	3.45	2.1
Mono-ortho					
PCB 118	0.0001	2.4	3.2	0.24	0.32
PCB 156	0.0005	0.4	0.2	0.20	0.10
Total		2.8	3.4	0.44	0.42
Total		7.6	4.6	3.9	2.5

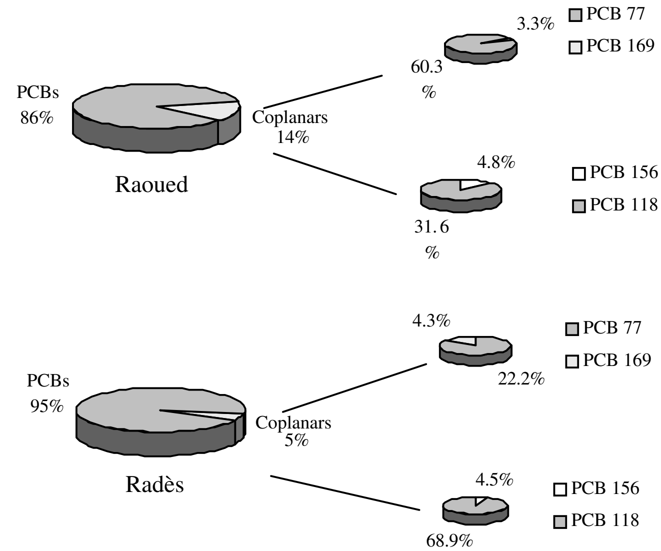

Fig. 3. Total PCB/Coplanar ratio and their percentages in the mullet muscle from Raoued and Radès.

Coplanar PCBs are also used in order to estimate the toxicity potential (TEQs) of PCB exposure, using the Toxic equivalency factors (TEFs) for fish developed by Van den Berg et al. (1998). TEQs of the 4 congeners was 3.9 pg/g w wt for fishes from Raoued and 2.5 pg/g w wt for specimens from Radès. The comparaison of the TEQ values in mullet with those in the literature showed high levels when compared with the levels of TEQs in sardine from Spanish Atlantic southwest coast (0.75pg/g w wt) (Bordajandi, Martin, Abad, Rivera, & Gonzalez, 2006). Our TEQ values are lower than those in salmon from the Baltic sea (12.6 pg/g w wt) (Isosaari et al., 2006).

In conclusion, we show here the first data about polychlorinated biphenyl contamination of the golden grey mullet from Tunis bay. PCB profiles showed similarities between the two investigated sites. On the other hand, PCB levels registered in the muscle of all fishes examined are not insignificant. Moreover, it is important to set limits for PCBs in fishes, in order to better estimate the risk to human health.

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