www.trjfas.org
ISSN 1303-2712
Turkish Journal of Fisheries and Aquatic Sciences 14: 993-1000 (2014) DOI: 10.4194/1303-2712-v14_4_19

Partitioning and Level of Organochlorine Compounds in the Tissues of the Black Sea Turbot at the South-Western Shelf of Crimea

Ludmila Malakhova[1,*], Vitaly Giragosov[1], Antonina Khanaychenko[1,*], Tetiana Malakhova[1], Viktor Egorov[1], Dmitry Smirnov[1]

[1] Institute of Biology of the Southern Seas NAS Ukraine, 2 Nakhimov, Sevastopol, Ukraine.

* Corresponding Author: Tel.: +380.692 550079; +380.692 550990; Fax: 380.692 557813;
E-mail: malakh2003@list.ru; a.khanaychenko@gmail.com

Received 12 May 2014
Accepted 23 December 2014

Abstract

The organochlorine compounds (OCs) contamination was studied in different tissues of the adult Black Sea turbot (BST) from natural spawning populations of 2008, 2009 and 2013 in the South-Western shelf off Crimea, the Black Sea. Total lipids, six congeners of polychlorinated biphenyl (CB 28, 52, 101, 138, 153, 180) and organochlorine pesticide p,p'- DDT including its metabolites p,p'- DDE and p,p'- DDD were measured in liver, red and white muscles and gonads of males and females of BST.

In BST tissues sum of DDT compounds ranged on average from 2.5 to 223.9 $ng \cdot g^{-1}$ wet weight, and sum of six congeners of PCB dominated by hexa-CB 138 and 153 congeners had an overall range 2.0-234.3 $ng \cdot g^{-1}$ wet weight. The main DDT compound detected in BST were the metabolites p,p'-DDE and p,p'-DDD (83% of ΣDDT) indicative of old DDT residues. OCs concentrations positively correlated with extractable lipid content in BST tissues. Sex-related pattern of OCs accumulation in different tissues of BST was found. On average, OCs concentration (on wet weight basis) in liver was twice higher in male than that in female, while OCs concentration in gonads were lower in male than that in female

Keywords: Fish, Black Sea turbot, organochlorine contaminants, polychlorinated biphenyls, DDT, DDE, DDD.

Introduction

Although production of organochlorine compounds (OCs) such as polychlorbiphenyls (PCB) and persistent organochlorine pesticides (DDT) was banned worldwide already 30 years and their utilization significantly reduced, these toxic compounds still pollute the Black Sea and contaminate its inhabitants. Marine organisms easily uptake persistent organochlorine compounds from marine water (Polikarpov and Egorov, 1986) and accumulate them through food web. Accumulation of OCs in hydrobionts tissues is enabled by their hydrophobic and lipophylic properties and due to a very low degradation rate of DDT and PCB in aquatic environment.

According to earlier data on Baltic flatfish (Courtney and Langston, 1980), the concentration factors for liver and muscle were 10^4 and 10^3, respectively, for uptake of PCB from seawater; therefore, even at low OCs concentration in the marine water, fish can be contaminated significantly. Various adverse effects on reproduction and immunocompetence, increase of malignant tumours and other pathologies were observed in different fishes and other aquatic animals exposed to OCs contaminated environment and food (Popova and Shamrova, 1987; Cogliano, 1998; Khan, 2003).

In late XX century OCs contaminants were still found in all components of the Black Sea ecosystem in the South-Western shelf off Crimea: water, bottom sediments, hydrobionts (Polikarpov and Zherko, 1996), and the most polluted areas were aquatic areas near the industrial centres such as Sevastopol (Malakhova and Voronov, 2008). PCB contamination in pelagic fish exceeded that of bottom fish. The specimens of horse mackerel and mullet contained 454 and 240 $ng \cdot g^{-1}$ w.w., correspondingly, while the Black Sea turbot contamination amounted only to 76-105 $ng \cdot g^{-1}$ w.w. of PCB (Polikarpov and Zherko, 1996).

Besides overfishing, the Black Sea pollution was considered one of the probable causes of significant decrease in number of the Black Sea turbot (further referred as BST), *Psetta* (*Scophthalmus*) *maxima* var. *maeotica*, common name kalkan (Giragosov and Khanaychenko, 2012). High numbers of diseased BST specimens were observed in natural spawning

© Published by Central Fisheries Research Institute (CFRI) Trabzon, Turkey
in cooperation with Japan International Cooperation Agency (JICA), Japan

population (Khanaychenko *et al*., 2012). The health of bottom-dwelling flatfish can be impaired significantly by chronic exposure to sediments contaminated by PCBs (Khan, 2003).

To our knowledge no information is available on the highly persistent PCBs and DDTs in the Black Sea turbot apart from one study based on only two sporadic (1986 and 1989) measurements of PCB (Aroclor 1254) concentration in BST (Polikarpov and Zherko, 1996), and it could not reflect the modern state of its contamination.

Therefore, it was necessary to assess actual OCs contamination of BST and find out the peculiarities of OCs distribution in fish different tissues.

Materials and Methods

Sampling of BST specimens for OCs analysis was conducted during scientific monitoring carried out by the Institute of Biology of the Southern Seas NAS Ukraine. Fish was collected from the gill nets (180-200 mm) captures at the depths 25-95 m during the spawning seasons in 2008, 2009 and 2013 in Sevastopol region (South Western off Crimea, the Black Sea). The study area is presented in Figure 1.

Biological analysis of all caught specimens was carried out; the fish was weighed (g) and measured (cm) before sampling. Sex was determined by examination of the gonads. Age was determined by otoliths analysis. Sampling of fresh BST tissues for OCs contamination was carried out during biological analysis. During the period 2008, 2009 and 2013 in total 139 samples (of the liver, red and white muscles and gonad tissues) from 19 males and 20 females were collected (2008 - 8 males and 9 females, 2009 – 4 males and 7 females, 2013 – 7 males and 4 females). All sampled specimens were adult and either ready for spawning (stage IV in April), or in active phase of spawning (stages IV-V, V, VI-V, April-June). To avoid possible large variations resulted from histological heterogeneity of the same tissue within different localities, samples were taken exactly from the same parts of the tissues. Tissue samples were individually wrapped in aluminum foil and stored at –10°C until analyzed for contaminants. The level of total lipids, six congeners of polychlorinated biphenyl (IUPAC # 28, 52, 101, 138, 153, 180) and organochlorine pesticide dichlorodiphenyl-trichloroethane (p,p'-DDT) including its metabolites (p,p'-DDE and p,p'- DDD) were examined in all samples.

The extractive analytical procedure and the instrumental conditions for identifying the peaks of PCBs and DDTs compounds and for their quantitative analysis have been described in detail elsewhere (Standardized methods for monitoring of background pollution of the environment, 1986). Briefly, after grinding the tissues the homogenates (5 g wet white muscle, 0.5 g wet gonad, red muscle or liver) were dried overnight with sodium sulfate (1:4 gonad or liver tissue:sodium sulfate; 1:3 muscle tissue:sodium sulfate). Pesticide-grade reagents and solvents were used for the analysis. The tissue–sodium sulfate mixture was extracted with acetone:hexane (1:3 v/v). The extracts were then concentrated and subsamples were taken in order to determine the tissue fat content by gravimetry. An aliquot of the remaining extract was dissolved in hexane (5 ml) and mixed with concentrated H_2SO_4 for the clean-up, following the procedure described by Murphy (1972). After centrifugation, the hexane solution was concentrated (about 1 ml) and transferred to a glass column (i.d. 5 mm) filled with 1 g of florisil (activated at 120°C for 16 h) for the separation of OCs from other organic compounds. The OCs fraction eluted with n-hexane.

To determine levels of PCBs and DDTs, samples were analyzed using a gas chromatograph (Model Varian 3800) equipped with an electron-capture

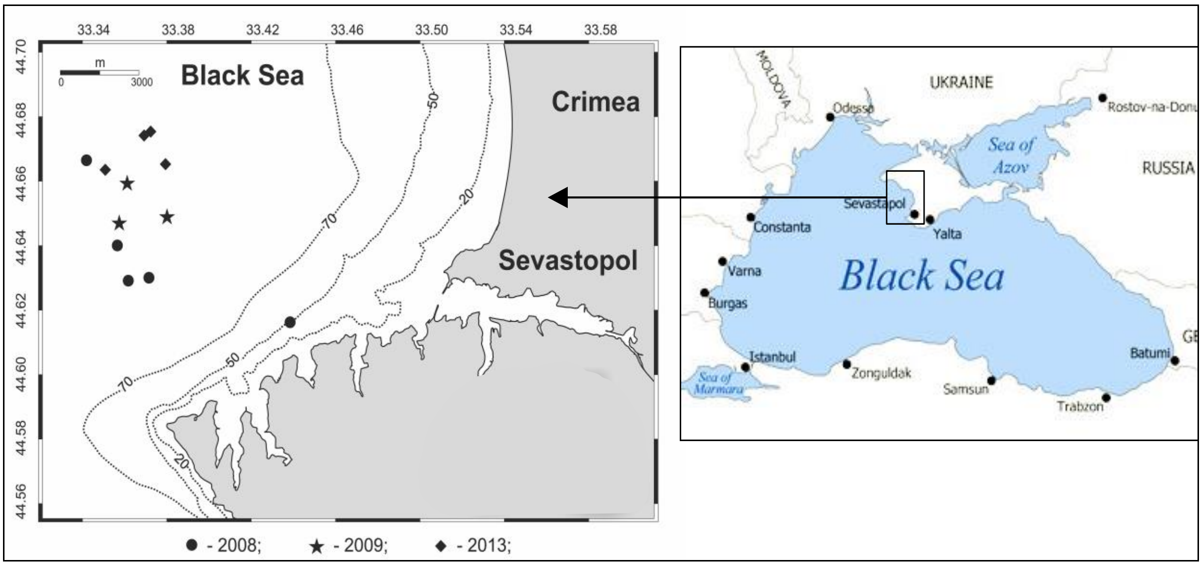

Figure 1. Schematic map showing the Black Sea turbot sampling area and points.

detector (ECD). A fused silica capillary column (25 m x 0.25 mm i.d.) coated with CP Sil-8CB having a film thickness of 0.45 gm was used for the separation of the organochlorine compounds (OCs). Oven operated under the following conditions: 90°C, 1 min hold, ramp to 120 at a rate of 25°C/min, 0.5 min hold, ramp to 290°C at a rate of 5°C/min, 9 min hold with a final hold of 10 min. Injection of 1 µl into a split/splitless injector was used in splitless mode, with 1 min of injection time. The injector and detector temperatures were kept at 280°C and 300°C. Nitrogen was used as the carrier and the make-up gas.

Pure reference standard solutions (Supelco Pesticides Mix - 2000 µg/ml and Sigma-Aldrich Laborchemikalien GmbH PCB Mix 7-10 µg/ml) were used for instrument calibration and quantification of compounds. Quantification was performed using internal standards calibration.

The recoveries for the PCB congeners and DDT compounds, varying between 70% and 110%, were determined by adding known amounts of PCBs and DDTs standards to empty samples before extraction (method of additions). Reproducibility was calculated on replicate analysis giving an overall variability of 15–20%. The detection limits for individual PCBs and for DDT ranged between 0.1 and 0.5 ng/g.

Total PCB content was based on the summed concentrations of 6 congeners (CB 28, 52, 101, 138, 153, 180, further reported as ΣPCB_6) – ecological indicators recommended by European Commission to assess marine pollution (Commission Regulation (EU) No 1259/2011), and total DDT – on summed concentrations of residues p,p'-DDT, p,p'-DDE and p,p'-DDD (further reported as ΣDDT). Concentrations of organochlorine compounds, means of duplicate measurements, are presented as ng/g on the wet weight and lipids basis.

Standard statistics (mean, standard deviation, coefficients of variations (C_v) and significance of differences between sex- and tissue–related values, determined by Student's test at 1 and 5% significance level was carried out using Excel.

Results

The average values, standard deviations, and ranges for total lipids content and concentrations of ΣPCB_6 and ΣDDT metabolites on a wet weight (w.w.) and lipid weight (l.w.) basis in four different tissues of BST males and females collected in different years (2008, 2009 and 2013) are presented in Table 1.

Lipid content (on the basis of wet weight) increased in the following order of BST tissues: white muscles, gonads, red muscles, liver with statistically significant differences ($P \leq 0.01$) amongst the four tissues regardless of sex and year of sampling (excluding differences in lipid level in white and red muscles). Liver presented the highest lipid content, with average mean for all years, significantly different ($P \leq 0.01$) amongst the sexes, with 17.5% in males and 6.9% in females. White muscles presented the lowest lipid content amongst the tissues, with non-significant difference between males (mean 0.66%) and females (0.5%).

The level of OCs contamination (on w.w. basis) varied significantly among the tissues of BST (Figure 2) and was characterized by high values of coefficients of variations (C_v): up to 86 % in females and 122 % in males and differed non-significantly between years of sampling.

The liver contamination on wet weight basis was generally significantly ($P \leq 0.01$) higher relatively to gonads and muscle, and red muscle *versus* white muscle in both genders. The lowest and similar contamination was determined for both genders in white muscles: ΣDDT (0.6 $ng \cdot g^{-1}$ w.w.) and ΣPCB_6 (1.2 $ng \cdot g^{-1}$ w.w.). Maximum levels of ΣDDT and ΣPCB_6 (564 and 443 $ng \cdot g^{-1}$ w.w., respectively) were found in liver. Significant differences ($P \leq 0.05$) amongst the level of contamination by different OCs contaminants was found in the BST liver tissues where ΣDDT value was permanently about 1.5 higher than that of ΣPCB_6. Females livers contained significantly ($P \leq 0.01$) lower concentration of ΣPCB_6 and ΣDDT (mean 95 and 66 $ng \cdot g^{-1}$ w.w., respectively) than the males livers (163 and 166 $ng \cdot g^{-1}$ w.w., respectively). Female gonads *vice versa* permanently contained higher OCs concentration than the male gonads. Red muscles contamination by OCs was by an order of magnitude higher than that of white muscles.

When OCs concentration was expressed on the basis of total lipid weight basis the C_v decreased to 21 % in females and to 48% in males indicating rather uniform distribution of OCs within lipid content of tissues. Positive linear regression was found between both log-transformed ΣPCB_6 and ΣDDT concentrations (R=0.75 and R=0.72, correspondingly) and extractable lipid content in all BST tissues (Figure 3). Therefore, the revealed uneven distribution of OCs between tissues found on the basis of wet weight was related to lipid content in different tissues. Pooled for tissues lipid content could be arranged in order of decreasing as following: liver (mean 13.7% lipid), red muscles (4.6%), gonads (mean 2.3%), white muscles (0.6%). Significantly higher ($P \leq 0.05$) contamination by ΣDDT per lipid weight was determined in gonads of females in comparison with males.

Since the OCs concentrations in white muscle of both genders was the less variable parameter, these values were used to analyze the age-related level of fish contamination, and both ΣPCB_6 and ΣDDT concentrations in white muscles tended to decrease with age, yet, the differences were found not significant (Figure 4).

Proportion of PCB congeners varied amongst BST tissues (Figure 5). The higher hexachlorinated congeners PCB 138 and PCB 153 comprised 28-41% and 21-41%, respectively, from sum of PCBs (% from

Table 1. Mean ± standard deviation and range (min.–max.) of concentrations of total lipids (TL, % wet weight) and organochlorine compounds (ΣPCB_6 and ΣDDT) on the basis of wet (ng·g^{-1} w.w.) and lipid weight (ng·g^{-1} l.w.) in the tissues of Black Sea turbot from Sevastopol region of the Black Sea in 2008, 2009 and 2013

Sex	Tissue	TL, % w.w.	ΣDDT, ng·g^{-1}w.w.	ΣDDT, ng·g^{-1}l.w.	ΣPCB_6, ng·g^{-1}w.w.	ΣPCB_6, ng·g^{-1}l.w.
2008						
Male	Liver	14.5±6.6 (8-25)	224±147 (105-564)	1622±868 (856-3565)	137±94 (69-324)	976±531 (481-2048)
	Gonad	1.1±0.4 (0.7-1.8)	6.7±4.0 (1.1-12.4)	617±416 (90-817)	7.9±4.0 (2.2 -13.4)	803±611 (257-2005)
	Red muscle	4.0±1.8 (2.5-6.3)	50.8±34.8 (8.8-82.7)	1310±919 (244-2684)	49.5±27.4 (28 - 95)	1366±720 (817- 633)
	White muscle	0.6±0.4 (0.3-1.3)	6.2±3.8 (1.4-11.9)	1217±764 (468-2702)	4.2±2.8 (1.7-10.3)	1007±802 (305-2450)
Female	Liver	5.5±2.5 (2.2-10.3)	104±81 (15-252)	1851±1018 (363-3581)	54±26 (14-97)	1138±624 (341-2047)
	Gonad	2.5±1.0 (1.2-4.2)	50±45.3 (13-134)	1609±1075 (565-3731)	26.9±16.3 (6.2-54.5)	1124±831 (353-3048)
	Red muscle	3.9±1.8 (2.1-6.7)	72±54.2 (24-178)	2298±1525 (523-2646)	31.1±14.1 (9.6-50.6)	878±478 (415-1650)
	White muscle	0.5±0.4 (0.2-1.4)	8.1±4.9 (1.6-15.3)	1828±769 (298-5106)	5.9±5.0 (1.2-17.5)	1432±1007 (595-3494)
2009						
Male	Liver	13.1±1.8 (11-15)	162±39 (131-201)	1266±380 (823-1685)	128±29 (94-159)	1006±286 (619-1279)
	Gonad	1.5±1.2 (0.7-2.9)	7.8±9.3 (2.2-18.3)	418±176 (300-621)	11.9±10.6 (3.7 - 24)	726±157 (544-819)
	Red muscle	4.8±1.0 (3.8-5.9)	97±70 (45-172)	1765±999 (1183-2919)	61±44 (22-109)	1244±712 (459-1850)
	White muscle	0.4±0.2 (0.3-0.7)	2.5±3.3 (0.6-6.3)	477±449 (195-975)	2.1±1.3 (1.2-3.6)	455±449 (195-975)
Female	Liver	7.6±2.4 (5.3-11.2)	96±53 (29-175)	1226±469 (496-1689)	65±26 (25-100)	872±337 (436-1536)
	Gonad	1.4±1.4 (0.7-2.4)	16.4±10.9 (6.5-33.2)	922±394 (515-1510)	12.8±7.6 (4.9-25.3)	1068±825 (654-2746)
	Red muscle	4.1±1.8 (2.8-7.1)	48 ±16 (32-65)	1306±464 (814-2069)	29±7.1 (27-54)	971±310 (566-1038)
	White muscle	0.3±0.1 (0.1-0.4)	3.5 ±3.2 (1.0-9.4)	492±189 (334-736)	2.0±2.1 (1.3-6.2)	655±351 (451-1181)
2013						
Male	Liver	25.3±14.6 (12.6-38.2)	111.6±57.4 (77.5-187.6)	467±96.8 (399-613)	234.3±149.6 (44.8-443.2)	1013±430 (452-1258)
	Gonad	2.3±1.9 (0.4-5.2)	9.1±7.7 (4.2-23.8)	648±630 (163-1715)	26.2±19.8 (9.5-56.2)	1870±1537 (253-4045)
	Red muscle	4.0±1.9 (2.5-7.2)	40.2±19.5 (17.8-69.7)	1276±894 (246-2488)	42.7±31.5 (15.2-88.5)	1495±1381 (210-3480)
	White muscle	1.0±1.4 (0.15-3.47)	6.8±2.3 (3.9-10.2)	1960±1556 (213-3999)	17.2±12.9 (7.8-39.7)	3702±3006 (1145-8884)
Female	Liver	8.1±5.3 (1.9-13.5)	85.4±57.2 (20.3-148.6)	1182±648 (513-2071)	78.3±40.9 (32.7-121.8)	1233±642 (497-1849)
	Gonad	4.7±4.3 (1.2-10.8)	37.5±19.9 (13.8-61.3)	1210±924 (395-2507)	31.7±14.7 (11.5-45.7)	894±351 (421-1262)
	Red muscle	6.7±3.9 (2.2-11.4)	81.3±55.6 (22.2-136.7)	1345±816 (406-2272)	89.3±88.2 (15.5-216.2)	1318 ±1020 (432-2706)
	White muscle	1.0±0.8 (0.4-2.1)	5.4±3.6 (1.9-10.6)	653±291 (471-1087)	15.1±14.8 (3.9-36.3)	1445±334 (976-1694)

ΣPCB_6) found in BST tissues with maximum percent of these congeners in the liver. Congeners of polychlorinated biphenyl (101, 138, 153 и 180) and metabolites of DDT (p,p'-DDE and p,p'- DDD) were detected in all samples of BST. The original organochlorine pesticide p,p'-DDT was present only in 85% liver samples, and less chlorinated PCB congeners 28 and 52 were found in 67 % of liver samples. The concentration of DDT metabolites increased in all samples within the following range: p,p'-DDT < p,p'-DDD <p,p'-DDE. Metabolites p,p′-DDE and p,p′-DDD comprised 83% of ΣDDT in BST tissues, especially in BST liver where the level of p,p'-DDE contamination was 8 times higher than parent p,p'-DDT compound level. Rather high quota of p,p'- DDT were found in BST gonads (up to 19%)

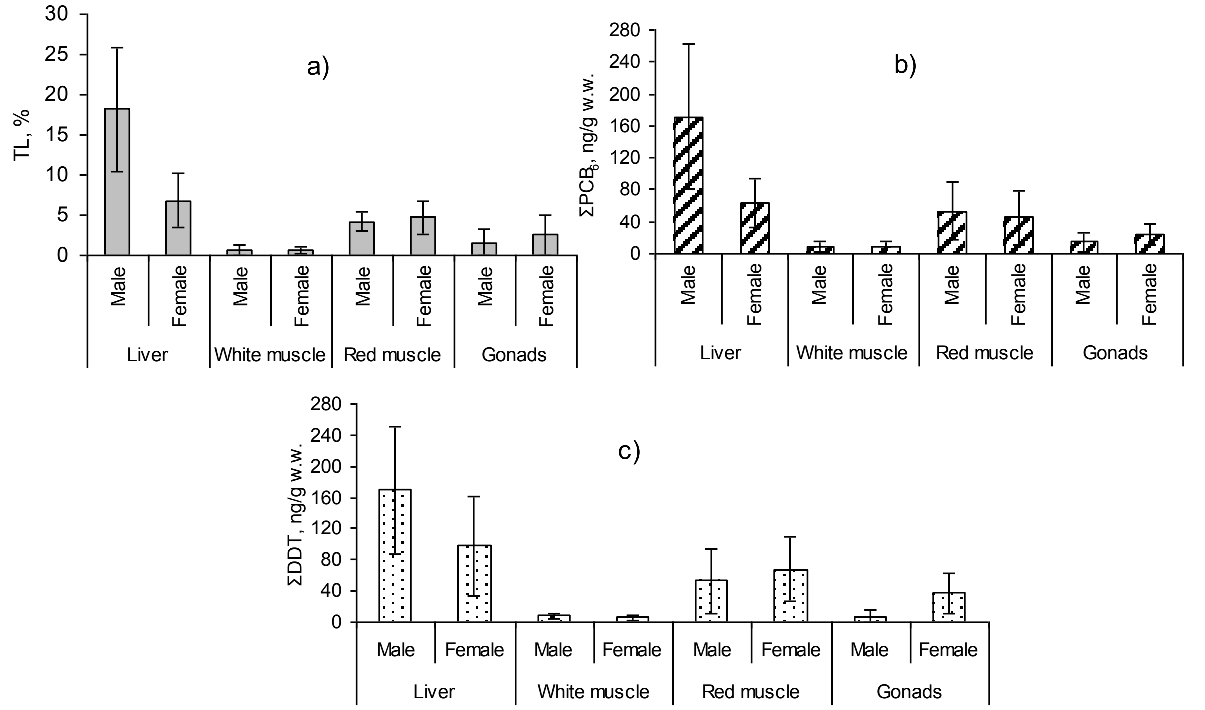

Figure 2. Comparison of average total lipids (TL, % w.w.) (a) and ΣPCB_6 (b) and ΣDDT (c) levels, $ng{\cdot}g^{-1}$w.w. (mean±SD) between different genders and tissues of the Black Sea turbot.

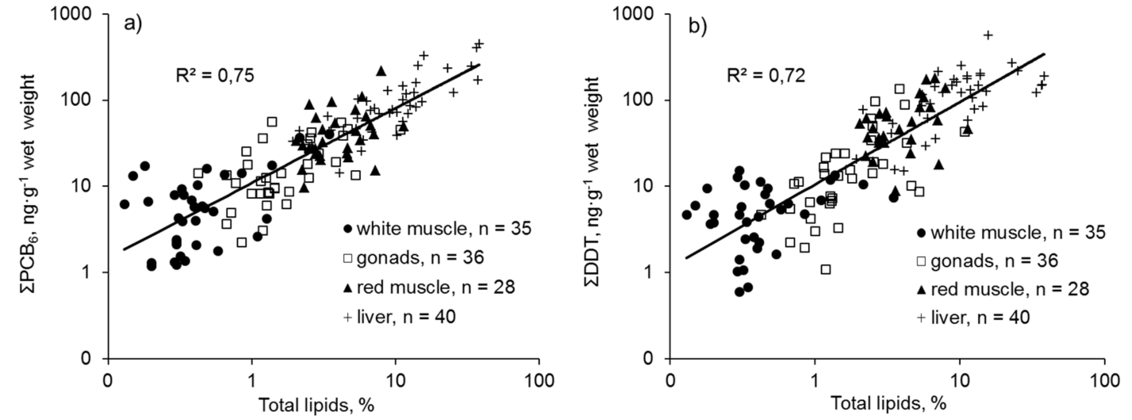

Figure 3. Total ΣPCB_6 (a) and ΣDDT (b) ($ng{\cdot}g^{-1}$ w.w.) concentrations *vs* total lipids (%) content in the Black Sea turbot in Sevastopol area.

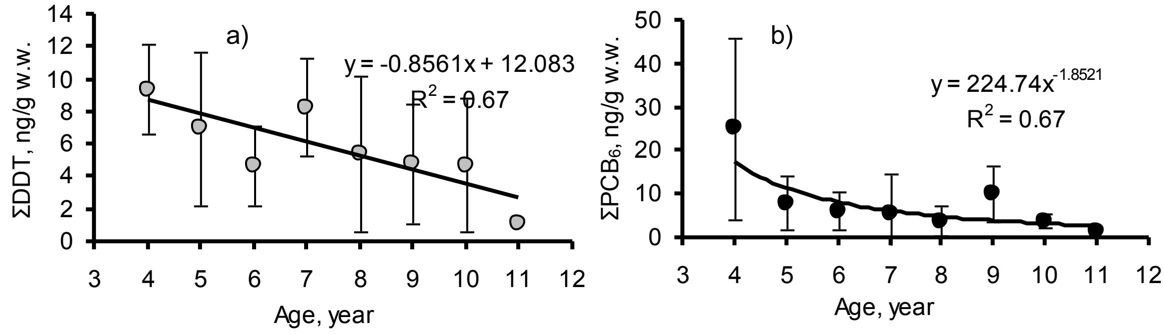

Figure 4. ΣDDT (a) and ΣPCB (b) concentration ($ng{\cdot}g^{-1}$ w.w.) (±SD) in the Black Sea turbot white muscles *vs*. age.

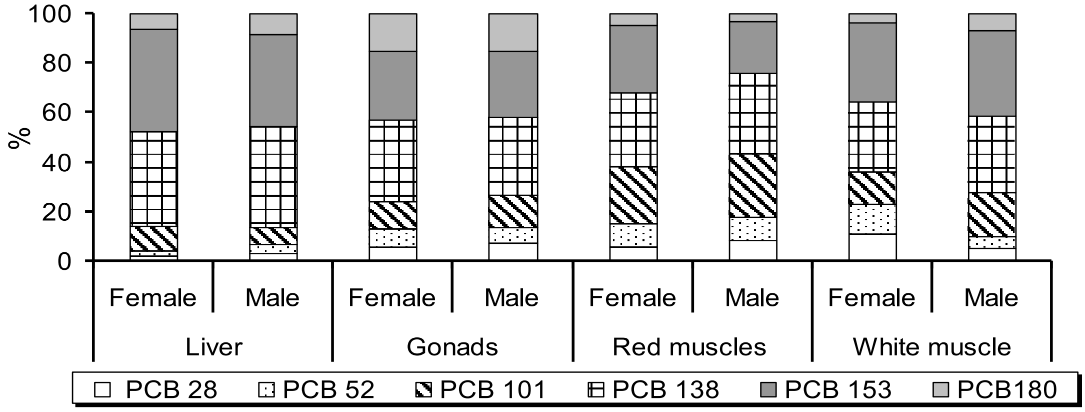

Figure 5. Average proportion of PCB congeners (% from the sum of six congeners ΣPCB_6) in the Black Sea turbot tissues.

and white muscle (up to 39.6%).

Discussion

Our results show that all tissues of the Black Sea turbot accumulated OCs contaminants, yet to different degree, and their accumulation was found related to tissue and sex. Mean OCs concentration in male liver was twice higher than that in female liver, while it was lower in male gonads than in female gonads. Despite the overall prohibition of utilization of DDT these compounds still dominated among organochlorine contaminants in majority of BST samples. The lowest contamination by ΣPCB_6 and ΣDDT was observed in white muscles in both genders.

OCs concentration in the BST tissues increased with the total lipids content of the tissue which increased in the following order: white muscles, gonads, red muscles, liver. High OCs enrichment in liver tissue is likely related to lipophilic OCs uptake in the liver mainly via food sources, and selective accumulation of lipid residues from food into the liver. Lower contamination by ΣPCB_6 and ΣDDT of females livers than males livers was supposed due to the active redistribution of OCs linked to specific lipids accumulation from the liver to female gonads during gametogenesis.

Predominance of the metabolites p,p′-DDE and p,p′-DDD in BST tissues was indicative that DDT did not penetrate into BST organisms recently and the intensive destruction of DDT in BST tissues with higher rate of their metabolism in the liver and the red muscles. The residues of DDT still remain in BST tissues and reveal that the accumulated earlier amount of organochlorine pesticide was not still totally metabolized as the time of DDT decomposition can vary from 240 days to 10 years (Isidorov, 1999).

The BST liver tissue was significantly enriched by hexachlorinated biphenyls 138 and 153. Predominance of hexachlorinated congeners PCB 138 and PCB 153 among other PCB congeners in the BST livers was also found in food objects of BST and in coastal waters according to our own (Malakhova and Voronov, 2008; Çakıroğulları *et al.*, 2010) studies. Apparently, such selective accumulation of certain PCB congeners is related to gradual fermentative metabolism of low chlorinated trichlorobiphenyl-28 and tetrachlorobiphenyl-52, and pentachlorobiphenyl-101 to hydrophilic compounds in the liver followed by their wash-out off organism while the high chlorinated congeners are degraded metabolically slowly (Isidorov, 1999).

Assessment of ecological state of environment suggests the comparison of contamination level of its components with the maximum permissible concentration (MPC) of pollutants and recommended maximum levels (ML) of PCB in marine fish. MPC for OCs in the fish and marine were standardized as 200 $ng{\cdot}g^{-1}$ w.w. of ΣDDT and 2000 $ng{\cdot}g^{-1}$ w.w. of ΣPCB; MPC of ΣPCB in the liver and fish oil – 5000 $ng{\cdot}g^{-1}$ w.w. (Belyaev *et al.*, 1993). In European Community, maximum level of six congeners of PCB – 75 $ng{\cdot}g^{-1}$ w.w. is permissible for muscle meat of fish while ML of PCBs in fish liver is permissible up to 200 $ng{\cdot}g^{-1}$w.w. (Commission Regulation (EU) No 1259/2011). Maximum content of DDT (sum of DDT-, DDD- and DDE- isomers, expressed as DDT) in $mg{\cdot}kg^{-1}$ (ppm) relative to feed materials with a moisture content of 12 % – 50 $ng{\cdot}g^{-1}$w.w. and for fats and oils – 500 $ng{\cdot}g^{-1}$ lipids (Commission regulation (EU) No 574/2011).

The amount of ΣPCB_6 in analyzed BST samples from Sevastopol area never exceeded 6% of MPC level (Belyaev *et al.*, 1993), and in majority of BST of samples the ΣPCB_6 level was less than 2% MPC. The maximum allowed PCB level in fish meat by EC (Commission Regulation (EU) No 1259/2011) was also never reached while permissible EC maximum level of ΣPCB_6 in fish liver was exceeded in the samples of the BST males liver in 2013.

Contamination by ΣDDT in all BST liver and red muscle samples, in 25% of gonad samples and in 3 % of white muscle samples exceeded 14 $ng{\cdot}g^{-1}$ w.w. - the reference value of ecological norm recommended for protection of aquatic life in Canada (of wildlife consumers of aquatic biota) (Canadian environmental quality guidelines, 1999). Content of ΣDDT in some BST tissues (liver, red muscles, gonads) exceeded EU maximum content relative to feed materials (Commission regulation (EU) No 574/2011).

It is obvious that the main sources of DDT for fish are water and food (Rhead and Perkins, 1984). Uptake via food sources are shown to be the main way for PCB contamination in flatfish, turbots (Zeng and Tran, 2002), besides seawater and sediments (Courtney and Langston, 1980). The Black Sea turbot is a typical predator fed mainly on abundant pelagic fishes - sprat, and near-bottom fishes, whiting, red mullet and gobies (Popova, 1969) most of them known to be significantly contaminated by OCs (Table 2). The ΣDDT contamination level (on lipid weight basis) in BST from Sevastopol area during 2008, 2009 and 2013 was of the same order of magnitude as that found for sprat (*Sprattus sprattus*), horse mackerel (*Trachurus mediterraneus*), bluefish (*Pomatomus saltatrix*) and shad (*Alosa pontica*) (Stoichev *et al.*, 2007; Stancheva *et al.*, 2010) from Bulgarian region of Black Sea in 2003-2006 (Table 2). Compared with the level of contamination of different fishes from the Turkish coastal waters in 1993 (Tanabe *et al.*, 1997), the level of both ΣPCB_6 and ΣDDT in the tissues of BST from Ukrainian coast was significantly lower. Variations in OCs contamination obviously depend on different fish species, ecological niches of fish, areas, years, and seasons.

In conclusion, marine ecosystem in Sevastopol coastal area is undoubtedly contaminated by various technogenic pollutants from earlier and recent anthropogenic activities. Pollution of the seawater by such indicators of anthropogenic activities as PCB and DDT, resulted in OCs contamination of all components of coastal ecosystem. Yet, according our data, the OC contamination of the Black Sea turbot in Sevastopol area is lower than in some species of pelagic fish, and does not exceed the maximum permitted level of OC contaminants in marine fish.

References

Belyaev, M.P., Gneushev, M.I., Glotov, Y.K. and Shamov, O.I. 1993. Reference Book of Maximum Permissible Concentrations of Harmful Substances in Food and Habitat. Nauka, Moscow, 142 pp. (In Russian).

Canadian environmental quality guidelines, 1999. Canadian tissue residue guidelines for the protection of wildlife consumers of aquatic biota: DDT (total). Canadian Council of Ministers of the Environment, Winnipeg Canadian Council of Ministers of the Environment 1999. Publication No. 1299, Ottawa.

Courtney, W. and Langston, W. 1980. Accumulation of polychlorinated biphenyls in turbot (*Scophthalmus maximus*) from seawater sediments and food. Helgolander Meeresunters, 33: 333-339. doi: 10.1007/ BF02414759.

Çakıroğullari, G.Ç., Yunus, U., Oymael, B., Bozkurt, E.N. and Kılıç, D. 2010. PCDD/F, dl-PCB and indicator PCBs in whiting, horse mackerel and anchovy in

Table 2. Variations of average values and ranges of concentration of ΣDDT and ΣPCB on the basis of wet and lipid weight in the Black Sea fishes

Location and survey year	Fish (species)	ΣDDT, ng·g^{-1} wet weight	ΣDDT, ng·g^{-1} lipid weight	PCB[1], ng·g^{-1} wet weight	PCB[1], ng·g^{-1} lipid weight	Reference
Turkey coastal area from Yakakent and Sinop, 1993	*Engraulis encrasicolus,* whole body	170	2700	50	920	Tanabe *et al.*, 1997
	Merlangius merlangus euxinus, whole body	280	7000	140	3500	-«-
	Mullus surmuletus, whole body	250	1700	20	130	-«-
	Alosa sp., whole body	370	12000	95	3200	-«-
	Trachurus mediterraneus, whole body	490	3800	210	2000	-«-
Bulgarian Black Sea coast, Spring 2004	*Sprattus sprattus,* whole body	-[2]	1374±1707 (362-3345)	n.a.	n.a.	Stoichev *et al.*, 2007
	Trachurus mediterraneus ponticus, whole body	-	833 (159-1509)	n.a.	n.a.	-«-
	Alosa pontica, whole body	-	952±634 (542-1682)	n.a.	n.a.	-«-
Bulgarian Black Sea coast (near Varna), 2003-2006	*Pomatomus saltatrix,* muscle	n.a.	595.0 (367.1-879.5)	n.a.	- (1.2 – 384.9)[3]	Stancheva *et al.*, 2010

[1] – PCB represent the Aroclor 1254; [2] – ΣDDT is not shown in the article; [3] – sum of 14 congeners of PCB: 28, 31, 52, 77, 101, 105, 118, 126, 128, 138. 153, 156, 169, 180; n.a. – not available.

Black Sea in Turkey. Turkish Journal of Fisheries and Aquatic Sciences, 10: 357-362. doi: 10.4194/trjfas. 2010.0308.

Cogliano, J.V. 1998. Assessing cancer risk from environmental PCBs. Environmental Health Perspectives, 106 (6): 317-323.

Commission regulation (EU) No 574/2011 of 16 June 2011// Official Journal of the European Union. L, 159: 7-24. Brussel.

Commission Regulation (EU) No 1259/2011 of 2 December 2011 amending regulation (EC) no. 1881/2006 as regards maximum levels for dioxins, dioxin-like PCBs and non-dioxin like PCBs in foodstuffs. Official Journal of the European Union, L 320: 18-23. Brussel.

Giragosov, V. and Khanaychenko, A. 2012. The state-of-art of the Black Sea Turbot spawning population off Crimea. Turkish Journal of Fisheries and Aquatic Sciences, 12: 377-383. doi: 10.4194/1303-2712-v12_ 2_25.

Isidorov, V.A. 1999. Introduction to chemical ecotoxicology. Himizdat, St. Petersburg: 90-97. (In Russian).

Khan, R.A. 2003. Health of flatfish from localities in Placentia Bay, Newfoundland, contaminated with petroleum and PCBs. Archives of Environmental Contamination and Toxicology, 44(4): 0485-0492. doi: 10.1007/s00244-002-2063-9.

Khanaychenko, A.N., Giragosov, V.E. and Gaevskaya, A.V. 2012. Epizootological state of the wild Black Sea turbot (kalkan). Grossly visible pathology: preliminary data. Marine Ecological Journal, 11(4): 86-93.

Malakhova, L.V. and Voronov, V.K. 2008. Organochlorine pollution of mariculture objects of the Crimea coastal area (Black Sea). In: B. Faye, Yu. Sinyavsky (Ed.). Impact of Pollution on Animal Products, Zurich: Springer Science+Business Media B.V.: 169-176. doi: 10.1007/978-1-4020-8359-4_18.

Murphy, P.G. 1972. Sulfuric acid for the cleanup of animal tissues for analysis of acid-stable chlorinated hydrocarbon residues. Journal of Association of Official Analytical Chemists, 55: 1360-1362.

Polikarpov, G.G. and Egorov, V.N. 1986. Marine dynamic radiochemoecology. Moscow, Energoatomizdat: 150-151. (In Russian).

Polikarpov, G.G. and Zherko, N.V. 1996. Environmental aspects of the study of Black Sea pollution organochlorine xenobiotics. Ecology Sea, 45: 92-100. (In Russian).

Popova, G.V. and Shamrova, L.D. 1987. Accumulation of pesticides in the reproductive system of fish and their gonadotoxic impact. Experimental Water Toxicology, 12: 191-201. (In Russian)

Popova, V.P. 1969. Some features of the dynamics of turbot fat of the Black and Azov Seas. Proc. AzCherNIRO, 26: 69-79. (In Russian).

Rhead, M.M. and Perkins, J.M. 1984. An evaluation of the relative importance of food and water as sources of p,p’-DDT to the goldfish *Carassius auratus* (L.). Water Research, 6: 719-725. doi:10.1016/0043-1354 (84)90167-2.

Stancheva, M., Rizov, T., Makedonski, L. and Georgieva, S. 2010. Organochlorine pollutants in bluefish (*Pomatomus saltatrix*) from Bulgarian Black Sea coast. Plovdiv University “Paisii Hilendarski”. Scientific Papers. Chemistry, 37(5): 125-130.

Standardized Methods for Monitoring of Background Pollution of the Environment. 1986. Gidrometeoizdat, Moskow, 182 pp. (In Russian)

Stoichev, T., Makedonski, L., Trifonova, T., Stancheva, M. and Ribarova, F. 2007. DDT in fish from the Bulgarian region of the Black Sea. Chemistry and Ecology, 23(3): 191-200. doi:10.1080/02757540701 339851.

Tanabe, S., Madhusree, B., Öztürk, A., Tatsukawa, R., Miyazaki, N., Özdamar, E., Aral, O., Samsun, O. and Öztürk, B. 1997. Persistent organochlorine residues in harbour porpoise (*Phocoena phocoena*) from the Black Sea. Marine Pollution Bulletin, 34(5): 338-347. doi:10.1016/S0025-326X(96)00081-1.

Zeng, E.Y. and Tran, K. 2002. Distribution of chlorinated hydrocarbons in overlying water, sediment, polychaete, and hornyhead turbot (*Pleuronichthys verticalis*) in the coastal ocean, Southern California, USA. Environmental Toxicology and Chemistry, 21(8): 1600-1608. doi: 10.1002/etc.5620210810.