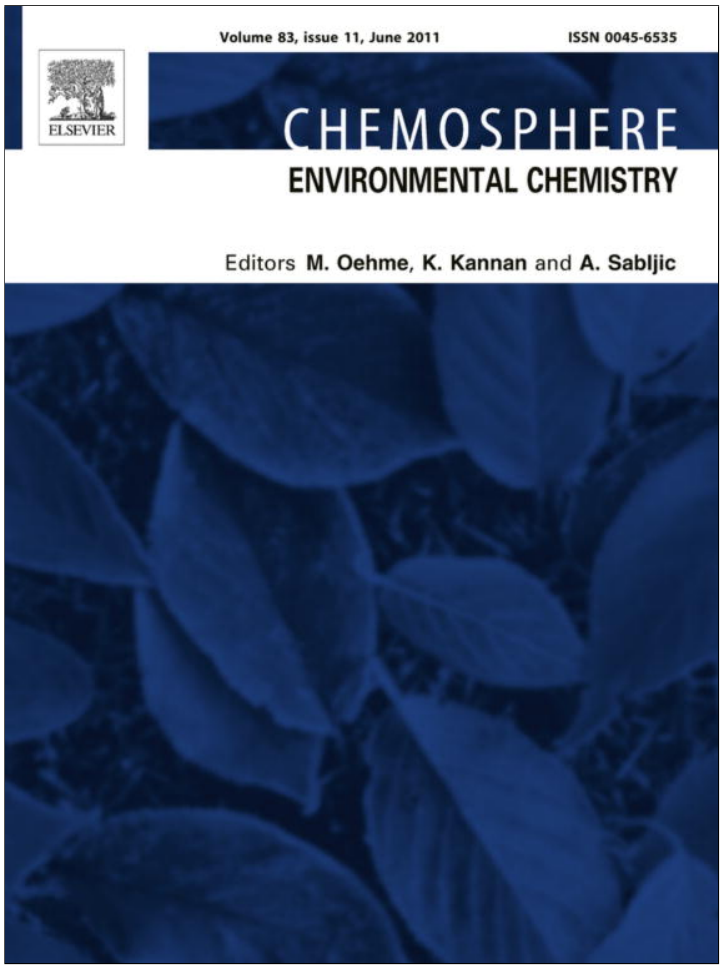

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Polybrominated diphenyl ethers and organochlorine compound levels in brown trout (*Salmo trutta*) from Andean Patagonia, Argentina

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ABSTRACT

Polybrominated diphenyl ethers (PBDEs), polychlorinated biphenyls (PCBs), DDTs and endosulfan residues were analyzed in muscle, liver, gonads, gills and stomach content of brown trout (*Salmo trutta*) from the Andean Patagonia. PBDEs, PCBs and DDTs levels were positively correlated with lipid content, while less hydrophobic endosulfans showed a poor correlation. Endosulfan levels were about 99.9% of total contaminant (highest levels in liver 500×10^3 ng g^{-1} lipid weight), with α-/β-isomers ratio >1 in all organs. These results are in agreement to the current-use of the technical endosulfan and also suggest acute exposure to this insecticide. Conversely, DDT/DDE ratio reflects fish exposure to old DDT sources, showing a DDE predominance. Gills had the highest levels of DDTs, PCBs and PBDEs, indicating they represent the main uptake pathway for such hydrophobic compounds from water column. PCBs showed the lowest levels in all organs and the PBDEs/PCBs ratios >1 agree with worldwide trends. PBDEs levels in gonads, gills, liver and muscle exceeded 80 ng g^{-1} (lipid weight) and were higher than other values reported in the Southern Hemisphere. BDE-47 was the predominant congener, suggesting higher bioaccumulation potential and possible brown trout metabolism of higher congeners. Since there is no point source of PBDEs in the region and residues were dominated by lower brominated congeners, atmospheric transport could be the main source of these compounds. This first report of PBDEs levels in fish from Argentina contributes to the knowledge about environmental trends of these persistent organic pollutants (POPs) in remote areas such as the Andean Patagonia.

1. Introduction

Organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs), and polybrominated diphenyl ethers (PBDEs) are organic, lipophilic and persistent compounds that accumulate in sediments and biota, including human tissues. Some of them are known to undertake long-range atmospheric transport leading to a widespread occurrence in the environment (Wania and MacKay, 1996; Sabljic, 2001; Borghesi et al., 2008). Because of these, PCBs and most of OCPs are on the list of persistent organic pollutants (POPs) (UNEP). Although the use of PCBs and the organochlorine insecticide DDT was banned in Argentina since 2005 and 1998, respectively, their residues can still be found in freshwater environments (Gonzalez et al., 2010; Ondarza et al., 2010), Special attention should be given to other organochlorine insecticide, endosulfan, which is highly toxic to the aquatic biota (Nowak et al., 1995) but is still being used on a wide variety or crops. As a result, endosulfan residues have been found in both biotic and abiotic compartments from different regions of Argentina (Jergentz et al., 2005; Lanfranchi et al., 2006; Gonzalez et al., 2010).

PBDEs are a class of flame retardants that are widely used in plastics, textiles and electronic appliances (de Wit, 2002). The three commercial mixtures (penta, octa and deca-BDEs) are now banned in Europe. Penta- and octa-mixtures were forbidden in the United States and Canadian markets at the end of 2004 and they has been recently included into the POPs list which regulates their elimination (UNEP, 2010. http://chm.pops.int). Moreover, an increase trend in environmental PBDEs levels have been reported (Hites, 2004).

Aquatic environments are under the pressure of direct and indirect discharges from urban, industrial or agricultural activities and fishes are often used as biomonitors of aquatic pollution. In addition to indicate the likelihood of exposure, the analysis of contaminants in specific tissues (i.e., gills, stomach content, liver, muscle and gonads) allows the identification of uptake pathways. The importance of each pathway will depend mainly on the feeding preference, trophic level and contaminant characteristics (Hellou et al., 1995). Although brown trout (*Salmo trutta*) is an introduced

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species in all rivers from Argentinean Patagonia, it can be considered a good biomonitor of aquatic environment. It is on top of the trophic food web (feeds on invertebrates, crustaceous and fishes), has non-migratory behavior and is widely distributed (Macchi et al., 1999). El Bolson city constitutes a particular and representative area of a region known as the Andean shire of parallel 42 from the Andean Patagonia, where the main economic activities are conformed by small-case agricultural practices and tourism.

The aim of this study was to evaluate levels, and compositional and tissue distribution of endosulfans, DDTs, PCBs and PBDEs in brown trout (*S. trutta*) from Quemquentreu river (Andean Patagonia). Furthermore, it focus specifically on endosulfan levels in order to check if the concentrations found could be linked with acute exposure by point discharges in the environment.

2. Material and methods

2.1. Study area

The sampling site is located in the Quemquentreu river (41°56′S–72°29′W), which is nearby El Bolson city (Andean Patagonia, Argentina). The main activities in the area are small-scale agriculture and tourism. The climate is temperate-cold with means rain of 800 mm y^{-1}, Most of the rain is concentrated during autumn and winter seasons, while supplementary irrigation is needed when temperature rises during spring time. The favorable microclimate and fertile soils allows traditional farming devoted mostly to the production of berries, hop and plums, representing the main production area of the country for these products. Moreover, manufactured products such as ice-cream, chocolate, beer, wool textiles and a great variety of crafts led to industrial settlements.

2.2. Sampling and preparation

Brown trout individuals ($n = 9$) were sampled in November of 2006 following standard fishing procedures with multifilament gillnet. At the laboratory, fishes were measured, weighted and dissected. Muscle, gills, liver, gonads and stomach content were wrapped in an aluminum foil and kept frozen (−20 °C) until analysis. Condition index (KI = total weight × 100/total length^3), hepatic index (HI = liver weight × 100/total weight) and gonadosomatic index (GI = gonad weight × 100/total weight) were calculated.

2.3. Age determination

The age of each fish was estimated using scales and otoliths measurements. Four scales (not regenerated) were extracted from the right and left side, upper the lateral line and just after the opercula. The total ratio of each scale was measured and the annuli were counted under microfiche readers (27 times). The otoliths were observed under optical microscope (10 times) and the annullis were counted in order to confirm the age found in the scales.

2.4. Chemical analysis

2.4.1. Extraction procedure

Endosulfans, DDTs, PCBs and PBDEs were extracted according to Metcalfe and Metcalfe (1997) with modifications of Miglioranza et al. (2003). Briefly, subsamples of muscle (5 g), liver, gonads, gills and stomach content (3 g) were homogenized with anhydrous sodium sulfate, fortified with PCB #103 as surrogate standard. Total lipids and organic compounds were Soxhlet extracted with *n*-hexane: dichlorometane (55:45) for 6 h and then concentrated. Lipid content was determined gravimetrically after removed from the extract by gel permeation chromatography using Bio-Beads S-X3 (200–400 mesh). The fraction containing Endosulfans, DDTs, PCBs and PBDEs was further purified with activated silica gel chromatography. Extracts were concentrated to 1 mL and kept in vials at −20 °C prior to gas chromatography analyses.

2.4.2. Analytical procedure

Endosulfans, DDTs and PCBs were identified and quantified using a gas chromatograph Shimadzu 17-A equipped with an autosampler and a ^{63}Ni electron capture detector (GC-ECD). The GC capillary column used was a SPB-5 (30 m × 0.25 mm i.d. × 0.25 μm film thickness). One microliter was injected on a splitless mode (275 °C) and detector was kept at 290 °C. The oven temperature was held at 100 °C for 1 min, followed by an increase of 5 °C min^{-1} up to 150 °C (held for 1 min), 1.5 °C min^{-1} up to 240 °C and, then, 10 °C min^{-1} up to 300 °C (help for 10 min). Helium was used as carrier (1.5 mL min^{-1}) and nitrogen (1.5 mL min^{-1}) as make-up gas. Identification and quantification of compounds were performed by injection of OCPs and PCBs standard solutions (Absolute Standards, USA) and PCB #103 as internal standard (Ultra Scientific, USA).

PBDEs were identified and quantified using a Perkin Elmer Clarus 500 gas chromatograph equipped with a mass spectrometer (GC/MS) fitted with an ELITE 5MS capillary column (30 m × 0.25 mm i.d. × 0.25 μm film thickness). The data acquisition was done in SIFI mode (Selected Ion and Full Ion Scanning). Two microliter was injected on a splitless mode. The injector temperature was kept at 130 °C for 0.01 min and, then, increased to 295 °C at 100 °C min^{-1}. Helium was used as carrier gas, with an initial flow of 1.3 mL min^{-1} and after 30 min increased to 3.0 mL min^{-1}. The oven temperature was kept at 130 °C for 1 min, followed by an increase of 15 °C min^{-1} up to 180 °C and, then, 4 °C min^{-1} up to 295 °C (held for 3 min). Source and interface temperature were kept at 280 °C. The mass spectrometer operated in the electron impact mode (EI) at 70 eV and multiplier at 450 V. Each PBDE (IUPAC #28, 49, 47, 66, 100, 99, 154, 153 and 183) was identified and confirmed by their relative retention time towards PCB #103 and three main fragmentation ions (one for quantification and two of confirmation) considering a ±10% deviation of standard proportion. The quantification was undertaken against a standard mixture of PBDEs (BDEs–LMS, Bromodiphenyl Ethers-Lake Michigan Study; AccuStandard INC., USA).

2.4.3. Quality control and assurance

Procedural and instrumental blanks were analyzed throughout the procedure to check for interference and laboratory contamination. PCBs and PBDEs values in the blanks were below the detection limit, whereas α- and β-endosulfan ranged between 1 and 2 ng mL^{-1} (<0.01% of levels found in fish samples). Surrogate recovery (PCB #103) was greater than 90%. Detection limits calculated according to Keith et al. (1983) ranged between 0.08 and 33 ng mL^{-1} for DDTs (*p,p′*-DDE, *p,p′*-DDD and *p,p′*-DDT), endosulfans (α-, β- and sulphate) and PCBs (IUPAC #8, 18, 28, 52, 44, 66, 101, 87, 110, 149, 118, 153, 138, 126, 187, 128, 167, 156, 157, -180, 169, 189, 195, 206 and 209), whilst for PBDEs (IUPAC #28, 49, 47, 66, 100, 99, 154, 153 and 183) ranged from 0.2 to 1.1 ng mL^{-1}.

2.5. Statistical analysis

Statistical analyses were performed using STATISTICA 6.0. Pearson correlation coefficient was performed between lipid content and contaminant levels (ng g^{-1} wet weight) in each tissue. Differences in contaminant levels among tissues were tested using a Friedman ANOVA analysis for multiple dependent samples and

pairwise comparisons were made using a Wilcoxon matched pair test ($\alpha = 0.05$).

3. Results and discussion

3.1. Biological parameters and physiological conditions of brown trout

The analysis of the scales and otoliths showed that fish ages ranged from +1 to +5 and age increase was related to length and weight enhancement. Based on that, fishes were divided in four groups (Table 1), since the uptake rate for hydrophobic compounds is mainly a function of size of the organism, whereas the elimination rate is mainly a function of size, lipid content, biotransformation process and compounds lipophilicity (Landrum and Fisher, 1998).

KI, HI and GI indexes assess the physiological state of the body, the food reserves available for metabolism and the degree of gonad development, respectively (Fechhelm et al., 1995). Table 1 shows that all fishes were in good condition, with low energetic reserves in liver and at low gonadal development, which reflects organisms either in juvenile or post-spawning stages. Since the reproductive period of brown trout occurs from May to July (winter), the indexes values found reflected the post-spawning and resting periods when they were captured (spring time).

3.2. Lipids distribution and total contaminants accumulation in brown trout tissues

Since no clear relationship was observed between contaminants bioaccumulation and size or age of the individuals (data not shown), and to obtain conclusive results about the dynamics of these contaminants into *S. trutta*, the data was pooled together and presented as an average of all fishes caught (Tables 2 and 3).

The highest lipid content was recorded in gills (9.1%; $p < 0.05$), while for liver, stomach content and gonads, lipid levels ranged between 3.7% and 4.8%. Muscle lipid content designates the brown trout as a lean fish (1.2%) (Table 2). DDTs, PBDEs and PCBs levels correlate well with lipid content, which is in agreement with the high hydrophobicity of these compounds (Table 2). Therefore, contaminant levels were normalized on lipid content to reduce tissues variability (Table 3). Conversely, for endosulfans with a low K_{ow} (3.83, Sabljic et al., 1995), lipids may not be the unique biochemical parameter involved in its accumulation. Bertelsen et al. (1998) suggest that in relatively lean tissues, such as those of brown trout, proteins and other non-lipid components can contribute substantially to chemical accumulation.

Endosulfans represented about 99.9% of total contaminants in all fish tissues. Liver showed the highest levels ($592 \pm 97 \times 10^3$, $p < 0.05$), while the lowest concentrations were found in stomach content (Table 3). Leaner fish exhibits faster hydrophobic contaminants elimination rates than fatty organisms (Landrum and Fisher, 1998). Brown trout fits into this hypothesis even though the results demonstrated that these fishes were exposed to high loads of endosulfans.

Acute or chronic exposure was assessed through the $c_{liver}/(c_{liver} + c_{muscle})$ ratio, where c represents concentration expressed on lipid basis. Values lower than 0.5 suggest a chronic exposure (Erdrogul et al., 2005). DDTs and PBDEs showed a chronic exposure (0.5), while endosulfans and PCBs ratios suggest an acute exposure (0.8).

3.3. Endosulfans: α-, β- isomers and endosulfan sulfate

Technical endosulfan is recognized as primarily responsible for events of fish kills worldwide, e.g. Germany and Sudan (Schulz, 2004). Levels of endosulfans in gills of fishes dead after acute exposure experiments and in dead fishes collected from cotton-growing areas were higher than 0.5×10^3 ng g^{-1} wet weight (Herzberg 1986; Nowak et al., 1995).

Technical endosulfan (a mixture of α- and β- isomers in a ratio 7:3) is used on blueberries, strawberries, hop and tomato crops in the study area. Runoff represents an effective process of input to aquatic environments and it is particularly important for endosulfan during storm events, since both α- and β-isomers and the endosulfan sulfate metabolite persist sorbed to soil organic matter (Jergentz et al., 2005).

Endosulfans levels were the highest found so far for fishes in Argentina, even considering other studies from areas particularly associated to the use of technical endosulfan (Ondarza et al., 2010). High endosulfans concentrations are likely to result from events such as accidental overspray or storm events. Endosulfans concentrations higher than 7 mg L^{-1}, that could persist for at least 48 h were recorded during storm events (Preece and Whalley, 1993). However, fishes were sampled in November, which corresponds to the dry season (rains lower than 25 mm) in the area. Thus, the unusual levels found in brown trout are more likely related to an acute exposure to technical endosulfan (i.e. an accidental direct discharge) than runoff processes.

Total endosulfans exceed 500×10^3 ng g^{-1} lipid weight in liver, while gills and gonads showed levels around 130×10^3 ng g^{-1} lipid weight (Table 3). The relative low K_{ow} and the comparative gills levels indicate that endosulfans are available in both particulate and dissolved phase of water column. According to Georgakopoulos-Gregoriades et al. (1991), gills are primarily responsible for the uptake of chemicals from the aquatic environment, which is determined by passive diffusion and is well correlated with their K_{ow}. In addition, gills accumulate more endosulfans during short-time exposures to lethal concentrations than over an extended period to low concentrations (Nowak et al., 1995). Our results agree with this statement since they show the relevance of gills over all uptake pathways. They also showed the low contribution of diet on endosulfans accumulation (Table 3), which was also reported for the Atlantic salmon (*Salmon salar*) (Petri et al., 2006). Gills from brown trout contained 12.1×10^3 ng g^{-1} wet weight of endosulfans, supporting the idea of an acute exposure.

Though the acute endosulfans levels could not be lethal, will lead to long term accumulation and subsequently damage in all organs, Particularly, OCPs residues up to 10 μg g^{-1} (wet weight) in gonads had been linked to diminishing levels of testosterone and

Table 1
Biological parameters, condition, hepatic and gonadosomatic indexes of brown trout (*Salmo truta*) individuals.

Group	N	Age	Sex/RE	TL (cm)	SL (cm)	TW (g)	KI (%)	HI (%)	GI (%)
1	4	1+	nd	9.7–13.0 (11.3)	8.4–11.4 (9.9)	9.4–22.3 (15.8)	1.0–1.3 (1.1)	0.6–1.1 (0.8)	nd
2	3	2+	F/1	18.1–20.5 (19.3)	15.8–18 (16.9)	70.1–104.5 (87.3)	1.2	0.8–1.5 (1.1)	0.5
3	1	3+	F/1	26.0	23.3	239.3	1.4	0.7	0.3
4	1	5+	F/2	35.2	30.9	665.1	1.5	0.9	0.5

RE: Reproductive stage; F: Female; TL: Total length; ST: Standard length; TW: Total weight; KI: Condition index; HI: Hepatic index; GI: Gonadosomatic index; nd: No determinated; length and weight data are expressed as range and arithmetic mean is showed between brackets.

Table 2
Lipid content (%), levels of endosulfans, DDTs, PCBs and PBDEs (ng g^{-1} wet weight, mean ± standard deviation) and correlation coefficient (r^2) in brown trout tissues.

	Muscle	Liver	Gonads	Gills	Stomach content	r^2
Total lipids	1.2 ± 0.1	4.7 ± 4.9	3.7 ± 2.1	9.1 ± 2.4	4.8 ± 3.8	–
Σ endosulfans	1.4 ± 0.3 × 10^3	12.0 ± 2.2 × 10^3	3.6 ± 0.6 × 10^3	12.1 ± 3.1 × 10^3	2.6 ± 0.4 × 10^3	0.7
Σ DDTs	1.7 ± 0.9	7.4 ± 3.4	6.6 ± 3.6	83.8 ± 38.2	8.4 ± 3.6	0.9
Σ PCBs	0.1 ± 0.01	0.3 ± 0.02	2.9 ± 0.3	5.8 ± 0.4	0.4 ± 0.1	0.8
Σ PBDES	1.1 ± 0.2	1.6 ± 0.2	15.3 ± 4.5	10.1 ± 3.4	1.3 ± 0.2	0.9

Σ endosulfans = Σ (α-, β-, endosulfan sulfate); Σ DDTs = Σ (*p,p′*-DDE, *p,p′*-DDD, *p,p′*-DDT); Σ PCBs = Σ (#44, 87, 110, 149, 118, 153, 138, 187, 180); Σ PBDEs = Σ (#47, 99 100).

Table 3
Levels of endosulfans, DDTs, PCBs and PBDEs (ng g^{-1} lipid, mean ± standard deviation).

Compounds	Muscle	Liver	Gonads	Gills	Stomach content
Total	**113 ± 56 × 10^3**	**592 ± 296 × 10^3**	**133 ± 66 × 10^3**	**134 ± 66 × 10^3**	**64 ± 31 × 10^3**
α-endosulfan	66 ± 11 × 10^3	283 ± 303 × 10^3	68 ± 46 × 10^3	80 ± 34 × 10^3	34 ± 10 × 10^3
β-endosulfan	36 ± 6 × 10^3	203 ± 209 × 10^3	48 ± 42 × 10^3	3916 × 10^3	19 ± 7 × 10^3
Endosulfan sulfate	10 ± 1 × 10^3	105 ± 109 × 10^3	15 ± 12 × 10^3	13 ± 4 × 10^3	10 ± 5 × 10^3
Σ endosulfans	**113 ± 4 × 10^3**	**592 ± 97 × 10^3**	**132 ± 18 × 10^3**	**133 ± 15 × 10^3**	**64 ± 2 × 10^3**
p,p′-DDE	125.6 ± 143.7	99.5 ± 69.9	136.2 ± 111.8	831.9 ± 255.9	193.8 ± 88.3
p,p′-DDD	2.0 ± 1.6	0.7 ± 0.7	0.5 ± 0.3	8.7 ± 2.7	5.6 ± 3.7
p,p′-DDT	3.4 ± 0.6	38.7 ± 73.1	7.2 ± 9.1	117.9 ± 48.0	31.8 ± 21.2
Σ DDTs	**131.0 ± 82.3**	**139.0 ± 40.9**	**143.9 ± 62.0**	**958.5 ± 135.0**	**231.2 ± 44.6**
44	0.5 ± 0.6	5.6 ± 8.5	3.0 ± 2.1	6.3 ± 3.8	<ld
87	0.9 ± 0.4	2.5 ± 3.6	1.1 ± 1.2	<ld	<ld
110	0.8 ± 0.6	4.8 ± 7.5	2.8 ± 3.5	7.2 ± 5.4	1.8 ± 2.2
149	0.2 ± 0.2	3.4 ± 4.5	1.1 ± 1.1	3.6 ± 0.3	0.5 ± 0.5
118	0.4 ± 0.4	1.8 ± 3.0	0.6 ± 0.7	4.1 ± 1.8	1.7 ± 3.7
153	0.4 ± 0.4	4.0 ± 5.4	2.2 ± 1.9	12.5 ± 5.2	5.1 ± 6.7
138	0.9 ± 1.2	3.5 ± 5.5	3.3 ± 4.7	14.3 ± 7.6	4.7 ± 6.2
187	0.2 ± 0.1	0.4 ± 0.5	0.2[a]	3.3 ± 1.5	0.05[a]
180	0.5 ± 0.3	1.0 ± 1.4	0.9 ± 1.0	4.1 ± 2.7	0.3 ± 0.3
Σ PCBs	**4.7 ± 0.3**	**26.9 ± 1.7**	**15.2 ± 1.1**	**55.4 ± 4.7**	**14.1 ± 2.1**
47	25.7 ± 10.9	55.0 ± 56.7	52.3 ± 61.7	48.5 ± 29.5	21.1 ± 18.3
100	13.4 ± 14.6	21.9 ± 41.5	6.8 ± 7.2	<ld	16.4 ± 14.6
99	41.5 ± 34.3	4.5 ± 2.3	32.3 ± 35.8	30.7 ± 29.1	10.1 ± 3.0
Σ PBDEs	**80.7 ± 14.1**	**81.3 ± 25.7**	**91.4 ± 27.3**	**79.2 ± 22.8**	**47.6 ± 5.5**

Totals = Σ (Σ endosulfans, Σ DDTs, Σ PCBs, Σ PBDEs); Σ endosulfans = Σ (α-, β-, endosulfan sulfate); Σ DDTs = Σ (*p,p′*-DDE, *p,p′*-DDD, *p,p′*-DDT); Σ PCBs = Σ (#44, 87, 110, 149, 118, 153, 138, 187, 180), Σ PBDEs = Σ (#47, 99, 100); <ld: below the detection limit.
[a] Since only one data was available no standard error could be calculated.

estradiol in fish, indicating a vulnerability to reproductive dysfunction (Singh and Singh, 2008). In the present work endosulfans level itself in gonads accounts for about 36% of that value (Table 2), suggesting that the development of brown trout in the study area could be at risk.

The ratio between endosulfan isomers has been used to indicate the time elapsed since technical endosulfan applications (Leonard et al., 2001). Since α-isomer degrades faster than β-isomer, which remains mainly adsorbed onto soil organic matter, result in deviations from the α-/β-ratio of the technical mixture (7:3). Thus, the α-/β-isomer ratios >1 in all tissues indicate an exposure to recent applications of technical endosulfan (Table 3). Furthermore, α- and β-endosulfan have a half life of days in natural waters, since this pesticide could be hydrolyzed, photodegraded and metabolized. However, their metabolite (endosulfan sulfate) is more stable and has a similar toxicity in comparison to both α- and β-isomers. Therefore, the occurrence of this metabolite suggests that brown trout is capable of biotransforming endosulfan isomers in all tissues. However, more studies are necessary to elucidate how important the fish metabolism and the accumulation from the environment (either by ingestion and gills) are to endosulfan sulfate levels.

Another aspect that must be taken in consideration is the human exposure through fish consumption, which is an important source of contamination. In this particular case, an ingestion of only 300 g of brown trout muscle represents an input of endosulfans similar to the maximum levels allowed for human consumption (0.006 mg kg^{-1} body weight) (Codex Alimentarius Comission, 2001).

3.4. DDTs

DDTs were the second most important group. Levels were significantly higher in gills ($p < 0.05$) followed by stomach content, gonads, liver and muscle (Table 3). DDTs have a high hydrophobicity ($\log K_{ow} > 6$) resulting in a large adsorption onto dissolved and particulate organic matter (Robinson et al., 2008). Thus, suspended particulate matter represents the main source for DDTs to brown trout. Muscle levels were below the guideline set by US-EPA (2000) of 14.4 ng g^{-1} (wet weight).

p,p′-DDE metabolite represented about 72% and 96% of total DDTs in liver and muscle of brown trout, respectively. It is known that *p,p′*-DDT can be metabolized into *p,p′*-DDE and *p,p′*-DDD under aerobic and anaerobic environmental condition, respectively. Moreover, fishes are also capable of transforming of *p,p′*-DDT into *p,p′*-DDE (Vives et al., 2005). As a consequence, predominance of DDE over DDT indicates an exposure to old or legacy DDT sources (Borrell and Aguilar, 1987). Although DDT use has been restricted in Argentina since 1998, natural and agricultural soils of Patagonia are highly contaminated by *p,p′*-DDE residues due to its historical and extensive use on fruit cultures (Gonzalez et al., 2010). Thus, the predominance of *p,p′*-DDE in brown trout tissues could be the result of both processes: (1) transformation of DDT by fish

metabolism, and (2) exposure to legacy DDTs sources that are enriched in *p*,*p*′-DDE residues.

3.5. PCBs

Differences among tissues were found for PCBs accumulation (Table 3), where high levels in gills indicate their preferential uptake from the water column. Muscle showed lower PCBs levels than those reported for both slightly and highly contaminated industrial areas (Falandysz et al., 2004; Erdrogrul et al., 2005), which was expected considering the scarce industrial development of the region. PCBs congeners profile was similar in all tissues, with the predominance of penta (#110, 118) and hexachlorobiphenyls (#153, 138) (Table 3). This pattern agrees with Arochlor 1254 and 1260 compositions, which were historically used in Argentina. This pattern could be a result of both, the low mobility of higher chlorinated congeners and the higher transport of lighter congeners. Nevertheless, the same profile was reported for fishes from other areas of Argentina (Colombo et al., 2007). However, (DDTs + Endosulfans)/PCBs ratios higher than 1 demonstrated that agriculture is the main contamination source for brown trout in the studied area.

3.6. PBDEs

PBDEs were detected in all tissues of brown trout with similar lipid weight levels in gonads, liver, gills and muscle, whereas stomach content showed the lowest concentration (Table 3). Gills and stomach content showed the highest and the lowest PBDEs concentrations, respectively; which indicate the importance of pollutants uptake from water column, as was observed for DDTs and PCBs. PBDEs concentrations in muscle of brown trout were of in the same order of magnitude as those reported for rainbow trout (*Oncorhynchys mykiss*) from a nearby watershed (138.3 ng g^{-1} lipid weight, Paola Ondarza – unpublished data). Both results are comparable to those in fishes from China (120 ng g^{-1} lipid weight, Xian et al., 2008) and Europe (119 ng g^{-1} lipid weight, Hites, 2004), but lower than fishes from North America (340 ng g^{-1} lipid weight, Dodder et al., 2002). However, levels here reported are quite higher when comparing with muscle of Chilean farmed salmon (*S. salar*) and Chinook salmon (*Oncorhyncus tshawytscha*) (Montory and Barra, 2006; Montory et al., 2010).

PBDEs/PCBs ratios >1 are coherent with the worldwide trend of an increasing levels of PBDEs and decreasing/stable levels of PCBs (Borghesi et al., 2008). Industrial activities, recycling and final disposal sites and waste incinerators constitute *in situ* PBDEs sources. Furthermore, the long-range transport led to the occurrence of PBDEs (mainly lower brominated) in remote regions (Borghesi et al., 2008).

The pattern BDE-47 > BDE-99 and BDE-100 was found in all tissues with the exception of muscle where a BDE-47/BDE-99 ratio was equal to 0.6 (Table 3). Lower brominated compounds, BDE-47 and BDE-99, are more volatile, water soluble and bioaccumulative than higher brominated congeners. Consequently, BDE-47 and BDE-99 are expected to be more mobile and bioavailable, and show an environmental behavior similar to PCBs with four chlorines (Watanabe and Sakai, 2003). In addition, several studies in aquatic organisms reported that BDE-47 and BDE-99 show higher uptake than BDE-100, and the metabolism of BDE-99 is faster than 47 and 100, leading to a higher BDE-47 bioaccumulation (Gustafsson et al., 1999). This relative abundance of BDE-47 was consistent with the general pattern found in fishes from other regions of the world (Vives et al., 2004), suggesting a possible elevated uptake rate from the environment or intestinal or tissue debromination of BDE-99, as was previously described in teleost fish (Stapleton et al., 2004). The commercial Bromkal-70 is a mixture of tetra and pentabrominated congeners (BDE-47:BDE-99 1:1; de Wit, 2002). Our results denote the technical pentabrominated mixture as the main PBDE source although they do not exactly reflect the Bromkal-70 mixture.

4. Conclusions

Concentration and tissue distribution of endosulfans, DDTs, PCBs and PBDEs in brown trout from the Andean Patagonia demonstrate differential exposure scenes to these compounds. Thus, the predominance of endosulfan over all contaminants as well as its high levels reflects an acute exposure to technical mixture. Besides, isomers proportion in all tissues, environmental parameters such as precipitation and temperature, and sampling time support the hypothesis that an acute spillage of this insecticide in the river have occurred. Moreover, consume off fish with such levels represents a risk for human being. Therefore, an improvement in the control of technical endosulfan use and fish consumption in the Andean Patagonia is recommended.

On the other hand, DDTs and PCBs both legacy compounds showed a chronic exposure. The highest levels of DDTs with the predominance of the metabolite *p*,*p*′-DDE show that DDTs still represent a potential risk for aquatic biota in the region although they do not surpass the limits for environmental quality. Regarding to PCBs, the congeners pattern agree with the past use of Arochlor 1254–1260 and values are not relevant.

The occurrence of PBDEs dominated by BDE-47 and BDE-99 and at higher levels than PCBs and similar to highly polluted areas indicate that following the worldwide trend of PBDEs increases, they would represent a risk to the aquatic environment. Thus, future studies should focus on PBDEs sources, bioaccumulation, metabolism and effects in fish to generate tools for environmental risk assessment. Considering their recent inclusion in the POPs list, this first report on PBDEs levels will be useful for evaluating temporal trends for future regulation actions.

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