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Organochlorine Compounds in Pine Needles from Croatia

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Abstract Levels of seven organochlorine pesticides and 17 congeners of polychlorinated biphenyls (PCBs) were analysed in pine needles collected at eight sites along eastern Adriatic coast (Croatia). The concentration of organochlorine pesticides and PCBs ranged between 0 (below determination limit) and 7.44 ng g^{-1} in 1-year-old needles and between 0 (below determination limit) and 15.57 ng g^{-1} dry weight in 2-year-old needles. Organochlorine levels were compared with those found in urban and mountain areas.

Keywords PCBs · OCPs · POPs · Air pollution

Polychlorinated biphenyls (PCB) and organochlorine pesticides (OCP) are well known persistent organic compounds according to UN-ECE POPs protocol (Lerche et al. 2002). Despite of their ban or restriction of usage, they still were found in all parts of environment (air, water, soil, animals, humans etc.).

Accumulation of these pollutants into epicuticular wax of foliage come from atmosphere because root uptake is not significant for compounds with octanol–water partitioning coefficients larger than 3. Because of this reason, epicuticular wax of pine needles has been used for monitoring local and regional ambient air pollutions (Wyrzykowska et al. 2006).

In previous studies we investigated evels and distribution of PCBs and OCPs in pine needles from Croatian mountains (Herceg Romanić and Krauthacker 2008) and urban and semi-urban cities (Herceg Romanić and Krauthacker 2004, 2006; Kožul and Herceg Romanić 2008). In this study, we collected 1- and 2-years old pine needles along Adriatic coast in Croatia in order to determine their load with PCBs and OCPs.

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Materials and Methods

Pinus strobus and *Pinus nigra* needle samples were collected in March 2005 at eight sites along eastern Adriatic coast (Croatia) (Fig. 1 and Table 1). Branches were collected at approximately 1.5 m above the ground level and stored in plastic bags. One- and two-year-old needles were manually separated and dried at room temperature to constant weight.

The following compounds were analysed: PCB congeners PCB-28, PCB-52, PCB-101, PCB-138, PCB-153, PCB-180, PCB-105, PCB-114, PCB-118, PCB-123, PCB-156, PCB-157, PCB-167, PCB-170, PCB-189, PCB-60, and PCB-74 (altogether 17, numbered according to IUPAC)] and OCPs hexachlorobenzene (HCB), α-HCH, β-HCH, γ-HCH (α-, β-, γ-hexachlorocyclohexanes), 1,1-dichloro-2,2-di(4-chlorophenyl)ethylene (DDE), 1,1-dichloro-2,2-di(4-chlorophenyl) ethane (DDD), and 1,1,1-trichloro-2,2-di(4-chlorophenyl) ethane (DDT).

The analytical procedure has been described by Kožul and Herceg Romanić (2008). Briefly, dried ground needles (5 g) were mixed with dichloromethane (20 mL) in a Teflon PFA extraction vessel and extracted using the Microwave Accelerated Reaction System for Extraction MarsX (CEM, USA) at 1,200 W and 40°C for 15 min. The extract was cleaned by alkaline and acidic hydrolysis and by adsorption chromatography on a multilayer silica column. After adsorption chromatography, the eluate was evaporated to residues under a

gentle stream of nitrogen. Before gas chromatography, the residues were dissolved in *n*-hexane. High resolution gas chromatography with electron capture detector(s) (HRGC/ECD) was done using at "ATI UNICAM" 610 SERIES chromatograph with two ^{63}Ni detectors. The compounds were analyzed simultaneously on two capillary columns ("Supelco", SAD): (1) 60 m × 0.25 mm, SPB-5 film thickness 0.25 lm, temperature programme 100°C, then 4°C min^{-1} to 240°C, 50 min isothermally; and (2) 30 m × 0.25 mm, SPB-1701 film thickness 0.25 lm, temperature programme 110°C, then 4°C min^{-1} to 240°C, 50 min isothermally. Carrier gas was nitrogen. The injector and detector temperature were 250°C and 270°C respectively, and the volume of injected sample was 5 µL. Qualitative and quantitative analyses were done by comparison with external standards. Each sample was analyzed on both columns and only compounds identified on both columns were evaluated. The detection limit was 0.4 ng mL^{-1} for each compound.

Results and Discussion

Table 2 shows the concentration ranges and medians of organochlorine compounds. HCB, α-HCH, β-HCH, γ-HCH, DDE and indicator PCBs (PCB-28, PCB-52, PCB-101, PCB-138, PCB-153 and PCB-180), PCB-60 and PCB-74 were found in all 1- and 2-year-old needle samples. The ranges of compounds are wider in the 2-year-old needle samples than in 1-year-old needles, and also, ranges overlap. The concentration of organochlorine pesticides ranged between 0 (below determination limit) and 7.44 ng g^{-1} in 1-year-old needles and between 0 (below determination limit) and 15.57 ng g^{-1} dry weight in 2-year-old needles.

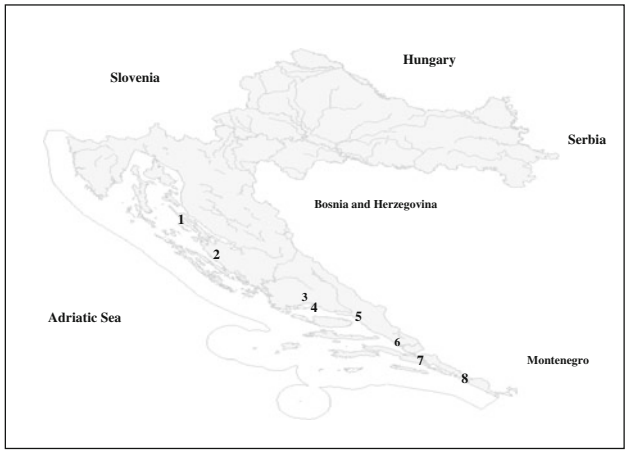

Fig. 1 Map of Croatia with sampling sites of pine needles

The concentration of indicator PCBs in 1-year-old needles ranged between 0.17 and 7.05 ng g^{-1} and between 0.26 and 12.14 ng g^{-1} dry weight in 2-year-old needles. The remaining PCB congeners ranged between 0 (below determination limit) and 1.45 ng g^{-1} in 1-year-old needles and between 0 and 2.84 ng g^{-1} dry weight in 2-year-old needles. Medians of all compounds are higher in 2-year-old needles and in all needles medians follow the same order: organochlorine pesticides - β-HCH > γ-HCH > HCB, α-HCH > DDE; PCBs - PCB-28, PCB-101 > PCB-138, PCB-52, PCB-153 > PCB-60, PCB-74 > PCB-180, PCB-118, PCB-123. α-HCH/γ-HCH ratio is often used to indicate a fresh input of γ-HCH in environment. Low ratios, particularly below one, indicate recent input (Ballschmiter and Wittlinger 1991). In 1-year-old needles α-HCH/γ-HCH is lower than in 2-year-old needles. α-HCH/γ-HCH ratio range between 0.16 and 0.82 and between 0.38 and 1.71 in 1-year-old needles and 2-year-old needles, respectively. α-HCH/γ-HCH ratio median in 1-year-old needles is 0.47 and 0.8 in 2-year-old needles. This implies a recent lindane release into the environment.

No particularly high or particularly low levels of all analysed compounds were found at any single site. However, the levels of certain compounds (in both, 1-year and 2-year-old needles) do stick out such as for β-HCH (3.39 and 15.5 ng g^{-1}), PCB-101 (7.05 and 12.14 ng g^{-1}) at site 8. In addition, site 8 was the only location where DDT (7.44 and 6.13 ng g^{-1}) and DDD (2.01 and 4.19 ng g^{-1}) were found. At site 5, levels of α-HCH (2.32 and 4.04 ng g^{-1}) and γ-HCH (4.47 and 8.63 ng g^{-1}) stick out.

Table 1 Details of pine needles sampling sites

Sampling site	Species	Latitude	Longitude
1 island Pag	*Pinus nigra*	N 44° 26′ 56.1″	E 15° 03′ 36.9″
2 Zadar	*Pinus nigra*	N 44° 08′ 05.5″	E 15° 15′ 07.3″
3 Kaštela	*Pinus nigra*	N 43° 33′ 06.1″	E 16° 21′ 13.3″
4 Split	*Pinus strobus*	N 43° 33′ 05.3″	E 16° 25′ 13.7″
5 Makarska	*Pinus strobus*	N 43° 17′ 52.5″	E 17° 01′ 45.6″
6 Opuzen	*Pinus nigra*	N 43° 02′ 16.4″	E 17° 32′ 50.5″
7 Doli	*Pinus strobus*	N 42° 48′ 50.4″	E 17° 47′ 14.1″
8 Dubrovnik	*Pinus strobus*	N 42° 41′ 48.5″	E 18° 03′ 03.1″

Table 2 Concentrations of organochlorine compounds (ng g^{-1} dry weight) in pine needles collected along Adriatic coast

Compound	1-year old needles		2-year old needles	
	Range	Median	Range	Median
HCB	0.60–0.97	0.76	0.72–1.61	1.19
α-HCH	0.58–2.32	0.86	0.73–4.04	1.11
β-HCH	0.71–3.39	2.10	1.7–15.57	3.92
γ-HCH	0.34–4.47	1.09	0.89–8.63	3.67
DDE	0.47–1.35	0.66	0.46–2.56	0.88
DDD	0–2.01	0	0–4.19	0
DDT	0–7.44	0	0–6.13	0
PCB-28	1.37–3.39	1.73	2.14–8.47	4.57
PCB-52	0.36–2.25	2.21	1.17–2.85	1.96
PCB-101	1.11–7.05	1.78	2.03–12.14	4.51
PCB-138	0.34–1.97	1.21	0.45–2.04	1.58
PCB-153	0.70–1.72	1.11	1.07–2.28	1.79
PCB-180	0.17–0.72	0.32	0.26–1.04	0.60
PCB-60	0.34–1.39	0.63	0.61–4.06	1.36
PCB-74	0.38–1.45	0.58	0.46–2.84	1.19
PCB-105	0	0	0–2.78	0.54
PCB-114	0–0.8	0	0–0.94	0
PCB-118	0–0.56	0.30	0.17–1.03	0.60
PCB-123	0–0.42	0.26	0–0.75	0.30
PCB-156	0	0	0–0.45	0
PCB-157	0	0	0–0.62	0
PCB-167	0	0	0–0.33	0
PCB-170	0–0.79	0	0–0.78	0
PCB-189	0	0	0–0.44	0

Comparison with PCBs and OCPs levels in urban and mountain samples (Croatia): In 2002 we collected pine needle samples from mountains across Croatia, and investigated the distribution of OCPs and PCBs in 1- and 2-years-old needles (Herceg Romanić and Krauthacker 2008). They ranged between 0.02 ng g^{-1} for α-HCH and PCB-180 and 8.63 ng g^{-1} for β-HCH and 8.98 ng g^{-1} dry weight for PCB-28 in 1-year-old and 2-year-old needles, respectively. In both generations of needles, medians follow the same order: OCPs: β-HCH > γ-HCH, DDT > DDE, DDD > α-HCH, HCB; PCBs: PCB-28 > PCB-101, PCB-138, PCB-52 > PCB-153, PCB-123, PCB-118 > PCB-180, PCB-74 > PCB-167. In samples from urban sites of Croatia collected in 1998, the concentration of organochlorine pesticides ranged between 0 (below determination limit) and 4 ng g^{-1} in 1-year-old needles and between 0.05 and 10.4 ng g^{-1} dry weight in 2-year-old needles. The concentration of PCBs in 1-year-old needles ranged between 0 (below determination limit) and 5.83 ng g^{-1} and between 0.15 and 9.91 ng g^{-1} dry weight in 2-year-old needles. In both, 1- and 2-year-old needles from urban sites, medians of OCPs and PCBs followed the same order: organochlorine pesticides - γ-HCH > DDE > HCB > β-HCH > α-HCH > DDT > DDD; indicator PCBs - PCB-28 > PCB-101 > PCB-138 > PCB-52 > PCB-153 > PCB-180; other PCBs congeners – PCB-118, PCB-123 > PCB-60 > PCB-74 > PCB-105 > PCB-170 (Herceg Romanić and Krauthacker 2006).

It could be noted that OCP profiles differ between urban, mountain and sea coast areas. In mountains and sea coasts pine needles β-HCH is dominant compound in contrast to urban pine needles. Common for all three regions is presence of γ-HCH at high level and the same PCB profile. Also, at all three regions concentration levels of compounds overlapped. In respect with different sampling year and fact that OCPs and PCBs use has been banned for long (also in Croatia), it could be expected some decreasing trend of organochlorine levels, but that was not observed. This is in agreement with results obtained in two previous studies (Kožul and Herceg Romanić 2008; Herceg Romanić and Krauthacker 2004) when levels of organochlorine compounds are not decreasing over the 8 years period, on the contrary, the levels of some organochlorine compounds have increased.

Global comparison: Pine needles are useful for determinating average profile of ambient air pollution and recent inputs of organochlorine compounds into the air, as they reflect on their levels in pine needles. Hellström et al.

(2004) propose that pine needles can be used to map the distribution of lipophilic airborne pollutants and clearly show local hot spots but suggest that we may never be able to calculate absolute air concentrations because of difficulties caused by biological factors affecting the uptake. Wyrzykowska et al. (2006) in their study confirmed the suitability of pine needles as passive samplers for PCBs in air, reflecting current pollution sources and also historical production, storage and use of PCBs. Pine needles are useful for relative comparison of the sampling sites (Klanova et al. 2009; Herceg Romanić and Krauthacker 2007), and comparison of organochlorine levels in pine needles from Croatia (this study; Herceg Romanić and Krauthacker 2004, 2006, 2008; Kožul and Herceg Romanić 2008) and another parts of world (Xu et al. 2004; Yang et al. 2008; Grimalt and van Drooge 2006; Loganathan et al. 2008; Nizzeto et al. 2006) show that levels in Croatia are characteristic for urban areas.

As mentioned above, levels from urban, mountain and sea coast areas are overlapped, probably due to geographic reasons. Croatia is small country with no presence of very high mountains and isolated areas. Results imply that the levels found in pine needles originate from the past use and/or from long range transport via air because usage of OCP and PCB is banned in Croatia. So, due to their persistence, PCB and OCPs will be present in the environment for many years.

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