# TOXICOLOGY AND ENVIRONMENTAL LEVELS OF HALOGENATED PHENOLIC COMPOUNDS

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## Introduction

Interest has increased in a relatively new class of compounds called halogenated phenolic compounds (HPCs). HPCs are unusual because they are predominantly, but not exclusively, metabolites of persistent organic pollutants (POPs); some HPCs have been produced and used as chemical agents. They are generally aromatic and contain both halogen groups and a minimum of one hydroxyl group.

Environmental measurement of POPs has concentrated on detecting parent compounds because of their persistence in biota but recent research has focused on biotransformation products and their biological effects, especially the degradation products of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers.

PCBs were discovered by Soren Jensen as he investigated DDT and its metabolites in Baltic Sea fish and eagle samples in 1964 [1]. Metabolism of PCBs was well studied in the early 1970s, but all biotransformation products of PCBs formed in *in vivo* experiments were believed to be rapidly excreted from the body, and environmentally persistent PCBs were not degraded resulting in their persistence. In the early 1980s, PCB biotransformation products became better known with evidence that certain hydroxylated metabolites of PCBs (HO-PCBs) were at measurable concentrations in blood and could interact with vitamin A and thyroid hormone homeostasis in exposed rats and mice [2;3]. This led to the first quantification and characterization of HO-PCBs in the environment and further exploration of HPCs found circulating in the blood of wildlife and humans [4]. This summary will discuss the formation and known biological effects of HPCs, different types of HPCs in the environment, and current levels of these contaminants in wildlife and humans.

## Discussion

*Formation and Relevance*

HPCs are formed mostly as biotransformation products of POPs by the enzyme cytochrome P450 (CYP P450) system. These structurally diverse heme-containing enzymes can hydroxylate many types of POPs thought to be completely impervious to metabolic degradation. The four-step process can be summarized as the substrate (PCB) binding to the enzyme, metabolic activation of the heme functionality of the CYP P450, insertion of the oxygen, and release of the compound so the cycle can repeat again. Upon release, the metabolite can either be further metabolized by the plethora of metabolic enzymes found in the cell or expelled from the cell. Once expelled from the cell, the slightly polar metabolite (because of its new functionality) can be either excreted or retained.

Retention of HPCs is thought to involve the thyroid hormone transport system, in particular, transthyretin (TTR) [5]. TTR is one of three known proteins responsible for transporting

thyroid hormones to the organs and cells that require thyroid hormones for their cellular processes. One of the main functions of thyroid hormones is brain development, cell generation, and differentiation in the brain. Certain biotransformation products of PCBs and organohalogen compounds structurally resemble the thyroid prohormone, tetraiodothyronine (T4) and can bind with high affinity to TTR [6-8]. This results in the interference of thyroid hormone transport possibly resulting in inadequate hormone supply to the tissues. Retinol binding protein (RBP) is also closely associated to TTR, in that it binds to TTR to form a 1:1 protein dimer. HO-PCBs bound to TTR cause a structural distortion of the protein, which inhibits dimer formation and can result in a decrease of circulating retinol levels [3]. These endpoints have been well documented in *in vivo* dosing experiments.

HPCs have other biological effects not involving retinol or thyroid hormone transport. HPCs at the cellular level also affect thyroid hormone and estrogen metabolism by inhibiting the sulfation metabolism mechanism [9;10]. This can result in elevated cellular levels of estrogen and thyroid hormones because the cells will be deficient in the sulfation detoxification reaction.

*Types of HPCs and Environmental Levels*

Investigation of HPCs in blood of humans and wildlife began with the measurement of HO-PCBs in blood components [4]. A recent publication indicated that over 50 HO-PCBs could be determined in human plasma; 38 positively identified with authentic standards [11]. Availability of authentic standards has been the limiting factor in HO-PCBs becoming a part of routine analysis. Two main metabolites of PCBs found in humans and wildlife are listed in Table 1 along with their concentrations and the sum HO-PCBs for that specie. Some are species, such as albatrosses from Midway Atoll or Baltic white-tailed eagle nestlings had very few detectable PCB metabolites, [12;13] while others, such as the polar bear, had over 30 [14]. Polar bears have the highest known concentration of HO-PCBs of all species analyzed, with concentrations equal to or higher than the PCB concentrations. The high concentrations of HO-PCBs probably result from the polar bears' metabolic capability and the high concentrations of PCBs in their diet.

Other HPCs have been determined in many of the studies listed in Table 1. The most abundant HPC in humans was pentachlorophenol (PCP) [15-17]. PCP is also found in lake trout [18] and polar bears [14] but at much lower concentrations. Other compounds, such as the hydroxylated metabolite of octachlorostyene [19-21] and halogenated phenols [22] recently have been discovered and probably will become candidates for routine HPC monitoring in the future.

*Future Research*

Hydroxylated metabolites were thought to be excreted and thus were no longer a problem to the environment. With the recent discovery that Great Lakes fish have appreciable levels of HPCs in their blood, new research into the potential bioaccumulation and bioavailability of these compounds in the environment is probably underway [20;21]. Fish do not possess the enzyme system necessary to metabolize persistent PCBs, yet they have circulating levels of these metabolites in their blood, leading to questions about the origin of these HPCs in fish blood.

Many studies are now investigating the interaction of HPCs and endocrine disruption endpoints such as thyroid and retinol homeostasis. Studies aimed at linking HPC exposure and these endpoints will attempt to explain some of the effects witnessed in species environmentally exposed to large concentrations of anthropogenic compounds.

Further research into the discovery of unknown compounds and the synthesis of authentic standards also will help with the determination of HPCs in blood. Due to the biased detection

Table 1 - Concentrations of pentachlorophenol (PCP) and hydroxylated metabolites of PCBs in wildlife and human plasma. Concentration units are given for each species as well as the number of samples analyzed. The number of compounds included in the sum HO-PCB calculations and the concentrations are study/specie dependent and are listed for each study separately. All concentrations are based on wet weight.

| Species | Pools | n | PCP | 4-HO-CB146 | 4-HO-CB187 | Sum HO-PCBs | units | # Cpds in Sum | Mean | Reference |
|---|---|---|---|---|---|---|---|---|---|---|
| **Aves** | | | | | | | | | | |
| White Tailed Eagle nestlings - Baltic Sea | - | 15 | n/d | 0.89 | 2.8 | n/a | ng/g | n/a | G | (13) |
| Black Footed Albatross - Midway Island | - | 5 | n/d | n/a | n/a | 27 | ng/g | 6-7 | A | (12) |
| Laysan Albatross - Midway Island | - | 5 | n/d | n/a | n/a | 11.5 | ng/g | 6-7 | A | (12) |
| Herons - St. Lawrence River | 1 | 8 | 4.6 | 0.16 | 2.3 | 9.3 | ng/g | 27 | A | (unpublished) |
| Herring Gull - Lake Superior | 1 | 3 | 0.52 | 0.16 | 3.5 | 7.9 | ng/g | 27 | A | (unpublished) |
| Herring Gull - Lake Erie | 1 | 4 | 0.55 | 0.45 | 12 | 26 | ng/g | 27 | A | (unpublished) |
| Herring Gull - Lake Ontario | 1 | 2 | 0.42 | 0.41 | 5.8 | 16 | ng/g | 27 | A | (unpublished) |
| Bald Eagles - Strait of Georgia | - | 10 | 0.45 | 0.24 | 0.76 | 1.4 | ng/g | 11 | A | (23) |
| Bald Eagles - Clayoquot Sound | - | 7 | 0.21 | 0.13 | 0.42 | 0.76 | ng/g | 11 | A | (23) |
| **Fish** | | | | | | | | | | |
| Trout - Lake Superior | - | 5 | 240 | 0.9 | 61 | 260 | **pg/g** | 17 | A | (20) |
| Trout - Lake Ontario | - | 5 | 360 | 4.1 | 110 | 330 | **pg/g** | 17 | A | (20) |
| Largemouth Bass - Detroit River | 3 | 3 | 1.6 | n/a | 5.6 | 11 | ng/g | 14 | A | (21) |
| Northern Pike - Detroit River | 1 | 4 | 1.1 | n/a | 0.20 | 2.0 | ng/g | 14 | A | (21) |
| White Bass - Detroit River | 1 | 3 | 0.37 | n/a | 9.4 | 30 | ng/g | 14 | A | (21) |
| Bowfin - Detroit River | 2 | 3 | 0.23 | n/a | 6.2 | 79 | ng/g | 14 | A | (21) |
| Longnose Gar - Detroit River | 2 | 3 | 0.21 | n/a | 14 | 130 | ng/g | 14 | A | (21) |
| Black Crappie - Detroit River | 1 | 2 | 1 | n/a | 37 | 90 | ng/g | 14 | A | (21) |
| White Sucker - Detroit River | 1 | 4 | 3.4 | n/a | 0.12 | 2.8 | ng/g | 14 | A | (21) |
| Common Carp - Detroit River | 5 | 3 | 0.36 | n/a | 1.2 | 9.2 | ng/g | 14 | A | (21) |
| Bigmouth Buffalo - Detroit River | 1 | 3 | 0.64 | n/a | 0.80 | 42 | ng/g | 14 | A | (21) |
| Freshwater Drum - Detroit River | 1 | 3 | 2.6 | n/a | 5.5 | 24 | ng/g | 14 | A | (21) |
| Brown Bullhead - Detroit River | 2 | 4 | 0.09 | n/a | 0.02 | 0.57 | ng/g | 14 | A | (21) |
| **Reptiles** | | | | | | | | | | |
| Watersnake - Lake Erie | - | 8 | 2.2 | 0.17 | 0.14 | 4.2 | ng/g | 27 | A | (unpublished) |
| Crocodiles - Florida | - | 16 | 79 | 0.68 | 36 | 59 | **pg/g** | 23 | A | (unpublished) |
| **Arctic - Mammals** | | | | | | | | | | |
| Polar Bear - males - Svalbard, Norway | - | 18 | 0.14 | 6.4 | 20 | 110 | ng/g | 25 | A | (14) |
| Polar Bears - females - Svalbard, Norway | - | 15 | 0.096 | 16 | 92 | 220 | ng/g | 25 | A | (14) |
| Polar Bears - males - Barrow Strait, Canada | - | 12 | 0.14 | 2.3 | 19 | 58 | ng/g | 25 | A | (14) |
| Polar Bears - females - Barrow Strait, Canada | - | 13 | 0.23 | 7.5 | 61 | 110 | ng/g | 25 | A | (14) |
| Walrus - Nunavik, Canada | - | 8 | 46 | 8.9 | 32 | 110 | **pg/g** | 27 | A | (unpublished) |
| Ringed Seal - Nunavik, Canada | - | 5 | 170 | 1.3 | 17 | 87 | **pg/g** | 27 | A | (unpublished) |
| Beluga Whale - Nunavik, Canada | - | 6 | 25 | 0.19 | 8.6 | 25 | **pg/g** | 27 | A | (unpublished) |
| Humans | | | | | | | | | | |
| Men - low fish consumer - Latvia | | 19 | 3.7 | 0.19 | 0.20 | 1.2 | ng/g | 5 | M | (16) |
| Men - low fish consumer - Sweden | | 20 | 9.6 | 0.23 | 0.44 | 1.1 | ng/g | 5 | M | (16) |
| Men - high fish consumer - Latvia | | 26 | 2.0 | 0.96 | 0.72 | 4.5 | ng/g | 5 | M | (16) |
| Men - high fish consumer - Sweden | | 12 | 6.6 | 0.40 | 0.41 | 1.4 | ng/g | 5 | M | (16) |
| Men - Canadian Inuit | | 13 | 5.5 | 0.44 | 0.59 | 3.5 | ng/g | 11 | G | (15) |
| Women - Canadian Inuit | | 17 | 3.2 | 0.22 | 0.30 | 2.0 | ng/g | 11 | G | (15) |
| Umbilical Cord - Canadian Inuit | | 10 | 1.9 | 0.037 | 0.047 | 0.29 | ng/g | 14 | G | (17) |
| Umbilical Cord - Quebec City, Canada | | 10 | 1.7 | 0.012 | 0.028 | 0.23 | ng/g | 14 | G | (17) |
| Women - low whale consumers - Faroe Islands | | 21 | n/a | 0.15 | 0.27 | 0.75 | ng/g | 5 | M | (23) |
| Women - high whale consumers - Faroe Islands | | 15 | n/a | 1.1 | 1.6 | 5.0 | ng/g | 5 | M | (23) |

**Abbreviations - G - geometric mean, A - arithmetic mean, M - median, n/d - not determined, n/a - not available**

**Note - Canadian Inuit Data converted from whole blood to plasma equivalents assuming whole blood is 50% plasma, Fish consumer data converted from lipid weight to wet weight assuming 0.6% lipid by weight.**

systems currently employed for HPC analysis, species (including humans) have not been adequately evaluated for exposure to lower halogen containing compounds. As more commercial standards become available, HPC analysis probably will become part of routine biomonitoring programs because of the toxicology associated with this class of compounds.

**References**


1. Jensen, S. (1966) New Sci. 32, 612
2. Brouwer, A. (1989) Arch Toxicol Suppl. 13, 440
3. Brouwer, A., van den Berg, K. J. (1986) Toxicol Appl Pharmacol. 85, 301
4. Bergman, Å., Klasson-Wehler, E., and Kuroki, H. (1994) Environ Health Perspect. 102, 464
5. Lans, M. C., Klasson-Wehler, E., Willemsen, M., Meussen, E., Safe, S., and Brouwer, A. (1993) Chem -Biol Interactions. 88, 7
6. Meerts, I. A. T. M., van Zanden, J. J., Luijks, E. A. C., van Leeuwen-Bol, I., Marsh, G., Jakobsson, E., Bergman, Å., and Brouwer, A. (2000) Toxicol Sci. 56, 95
7. Chauhan, K. R., Kodavanti, P. R. S., and McKinney, J. D. (2000) Toxicol Appl Pharmacol. 162, 10
8. Lans, M. C., Klasson-Wehler, E., and Brouwer, A. (1994) Organohalogen Compounds. 20, 481
9. Schuur, A. G., Bergman, Å., Brouwer, A., and Visser, T. J. (1999) Toxicol in vitro. 13, 417
10. Kester, M. H. A., Bulduk, S., Tibboel, D., Meinl, W., Glatt, H., Falany, C. N., Coughtrie, M. W. H., Bergman, Å., Safe, S. H., Kuiper, G. G. J. M., Schuur, A. G., Brouwer, A., and Visser, T. J. (2000) Endocrinology. 141, 1897
11. Hovander, L., Malmberg, T., Athanasiadou, M., Athanassiadis, I., Rahm, S., Bergman, A., and Wehler, E. K. (2002) Arch Environ Contam Toxicol. 42, 105
12. Klasson-Wehler, E., Bergman, Å., Athanasiadou, M., Ludwig, J. P., Auman, H. J., Kannan, K., van den Berg, M., Murk, A. J., Feyk, L. A., and Giesy, J. P. (1998) Environ Toxicol Chem. 17, 1620
13. Olsson, A., Ceder, K., Bergman, Å., and Helander, B. (2000) Environ Sci Technol. 34, 2733
14. Sandau, C. D., Newson, S. C., Duffe, J., Ramsay, M. A., Derocher, A. E., Wiig, Ø., and Norstrom, R. J. (in prep) Environ. Toxicol. Chem.
15. Sandau, C. D., Ayotte, P., Dewailly, E., Duffe, J., and Norstrom, R. J. (2000) Environ Health Perspect. 108, 611
16. Sjödin, A., Hagmar, L., Klasson-Wehler, E., Björk, J., and Bergman, Å. (2000) Environ Health Perspect. 108, 1035
17. Sandau, C. D., Ayotte, P., Dewailly, E., Duffe, J., and Norstrom, R. J. (2002) Environ Health Perspect. 110, 411
18. Fisk, A. T.; Newson, S. C.; Sandau, C. D.; Backus, S.; Brooks, S.; Norstrom, R. J.; Whittle, M. 2001 .
19. Sandau, C. D., Meerts, I. A. T. M., Letcher, R. J., McLees, A., Chittim, B., Brouwer, A., and Norstrom, R. J. (2000) Environ Sci Technol. 34, 3871
20. Campbell, L. M., Muir, D. C. G., Whittle, D. M., Backus, S., Norstrom, R. J., and Fisk, A. T. (2003) Environ Sci Technol. 37, 1720
21. Li, H. X., Drouillard, K. G., Bennett, E., Haffner, G. D., and Letcher, R. J. (2003) Environ Sci Technol. 37, 832
22. Hovander, L., Malmberg, T., Athanasiadou, M., Athanassiadis, I., Rahm, S., Bergman, A., and Wehler, E. K. (2002) Arch Environ Contam Toxicol. 42, 105
23. Newson, S. C., Sandau, C. D., Brown, S. B., Elliott, J. E., Gill, C., and Norstrom, R. J. (in prep) Environ.Toxicol.Chem.
24. Fangstrom, B., Athanasiadou, M., Grandjean, P., Weihe, P., and Bergman, A. (2002) Environ Health Perspect. 110, 895