Polychlorinated biphenyls and naphthalenes: long-lasting induction of oxidative stress in the rat

Mäntylä, E.[A], Ahotupa, M.[B]
[A]Orion Corporation Farmos, Toxicological Laboratory, SF-20101 Turku, Finland
[B]Department of Physiology, University of Turku, SF-20520 Turku, Finland

The toxic effects of polychlorinated biphenyls (PCBs) are numerous: wasting syndrome, immunotoxicity, hepatotoxicity, epidermal toxicity, reproductive toxicity etc. The biochemical changes involved include e.g. hormonal changes, vitamin A depletion, changes in lipid metabolism, porphyria, drug metabolizing enzyme induction.[1,2,3] Of the 209 possible PCB congeners the most toxic are the ones that are approximate stereoisomers of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) (non*ortho* coplanar PCBs). Further, quite toxic are also the mono*ortho* coplanar PCBs, and even the di*ortho* coplanar PCBs, too, and the toxicologic significance of these latter two groups is increased by the fact that some of the congeners occur in abundance in our environment.[4,5] Polychlorinated naphthalenens (PCNs) have been used as replacing chemicals for PCBs. Those PCN congeners that are approximate stereoisomers of TCDD have the same kind of toxic properties as TCDD.[6] Several studies have indicated that TCDD and PCBs could cause toxic responses by increasing lipid peroxidation.[8] In a recent study of Dogra *et al.*[8] the long-term effects (up to 30 days) of a PCB mixture (Aroclor 1254) on lipid peroxidation were studied in rats. An 2-fold increase in endogenous ethane production, and in hepatic malondialdehyde concentration was found, but only at the 30 days time point. It was also suggested that both coplanar and noncoplanar PCB congeners can induce lipid peroxidation. The aim of the present work was to study further the long-term effects of PCBs and PCNs on hepatic lipid peroxidation and to elucidate the concomitant changes in antioxidant defence system functioning and cytochrome P450 activities.

Male Sprague-Dawley rats were used. PCBs (Clophen A 50, CloA50) or PCNs (Halowax 1014, H1014) were given ip to rats (100 or 20 mg/kg, respectively). A group of animals was sacrificed at 1, 3, 7 or 14 days or at 3 months time points. The MC-type induction (3-methylcholanthrene-type; P448 induction) or PB-type (phenobarbital-type) induction of P450 activity was evaluated by measuring microsomal ethoxyresorufin O-deethylase (EROD) or pentoxyresorufin O-deethylase (PROD) activities, respectively. The extent of lipid peroxidation was studied by quantitating tissue conjugated dienes and thiobarbituric acid (TBA) reactive material. The functioning of the hepatic antioxidant defence system was evaluated (vitamin A and E and reduced glutathione (GSH) contents; superoxide dismutase, catalase, GSH-peroxidase, glucose-6-phosphate

dehydrogenase and GSH S-transferase activities) using different subcellular fractions.

ClоA50 (100 mg/kg) induced EROD activity maximally in 7 days (50-fold), after which time the activity decreased slowly, but was still almost 10-fold after 3 months. The induction time profile after treatment with H1014 (20 mg/kg) was despite of the lower dose almost identical with the curve obtained with PCBs, and the activities were only somewhat lower. PROD activity in PCB-treated rat liver rose somewhat faster than EROD activity: induction was 18-fold at 1 day and reached maximum (24-fold) at 7 days, being back at the control level at 3 months. In the H1014 group the PROD activity was 4-fold at 7 days and below 2-fold at 14 days. Hepatic lipid peroxidation, as evaluated by conjugated diene content, was elevated from 1 to 14 days, up to 2-fold. Changes in the amount of TBA reactive material were minor. In hepatic GSH content there was a slight increasing trend between 7 and 14 days. Hepatic GSH S-transferase activity increased slowly after PCB-treatment and reached maximum (2-fold) at 14 days. PCNs caused 2 activity peaks. Hepatic GSH-peroxidase activity did not change. PCNs did not alter hepatic total vitamin A content, but PCBs decreased it slowly (40% decrease at 14 days). Hepatic vitamin E content was changed only at 14 days (a 30% decrease with PCBs). Hepatic SOD activity showed a decreasing trend, catalase activity decreased by 20-30% in 3 days after treatment and was recovered slowly. Further hepatic glucose-6-phosphate dehydrogenase activity was increased 2-fold up to 14 days.

ClоA50 is a mixed-type P450 activity inducer and it has been previously shown to have long-lasting effects (up to 4 weeks) on rat hepatic drug metabolizing enzyme activities[9]. In the present study EROD activity was significantly elevated as late as 3 months after a sigle dose. H1014 is concidered to be a MC-type P450 activity inducer and the results of the present work supported this: H1014 induced hepatic EROD activity 37-fold, but only 4-fold increase in PROD activity was seen. In the present study conjugated diene content was shown to be elevated (up to 2-fold) from 1 to 14 days after treatment with ClоA50 or H1014. This increase in lipid peroxidation was more closely associated with MC-type P450 induction. This agrees well with the observation that the most potent MC-type inducer, TCDD, has also been shown to be a very potent inducer of lipid peroxidation. The TCDD congeners not inducing P448 activity (nontoxic congeners) do not increase lipid peroxidation.[7] On the other hand, the data presented by Dogra *et al.*[8] suggest that both inducer types can equally be involved. The changes in the antioxidant defence system noticed in this work were mostly favouring or reflecting the increased oxidative stress. **In conclusion,** a single dose of PCBs or PCNs was shown to cause an long-lasting increase in P448 activity and lipid peroxidation in rat liver. Concomitantly some noxious changes in the components of the antioxidant defence system were seen. The observed increase in oxidative stress could give explanation to some of the long-term toxic properties of PCBs and related compounds, e.g. involvement in tumour promotion or in immunosuppression process.[4,10,11]

1 *Health effects of halogenated aromatic hydrocarbons*, Nicholson, WJ, Moore, JA, eds. *Ann NY Acad Sci* 1979;320:1-730.

2. Parkinson A, Safe S. Aryl hydrocarbon hydroxylase induction and its relationship to the toxicity of halogenated aryl hydrocarbons. *Toxicol Env Chemistry Rev* 1981;4:1-45.

3. Poland A, Knutson, JC. 2,3,7,8-Tetrachlorodibenzo-p-dioxin and related halogenated aromatic hydrocarbons: examination of the mechanism of toxicity. *Ann Rev Pharmacol Toxicol* 1982;22:517-54.

4 Safe S. Polychlorinated biphenyls (PCBs) and polybrominated biphenyls (PBBs): biochemistry, toxicology, and mechanism of action. *CRC Crit Rev Toxicol* 1984;13:319-95.

5. McFarland VA, Clarke JU. Environmental occurence, abundance, and potential toxicity of polychlorinated biphenyl congeners: considerations for a congener-specific analysis. *Env Health Persp* 1989;81:225-39.

6. Goldstein JA. Structure-activity relationships for the biochemical effects and the relationship to toxicity. In: Kimbrough, RD, ed. *Halogenated biphenyls, terphenyls, naphthalenes, dibenzodioxins and related compounds.* Amsterdam: Elsevier, 1980:151-90.

7. Stohs, SJ. Oxidative stress induced by 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). *Free Rad Biol Med* 1990;9:79-90.

8. Dogra S, Filser JG, Cojocel C, Greim H, Regel U, Oesch F, Robertson LW. Long-term effects of commercial and congeneric polychlorinated biphenyls on ethane production and malonialdehyde levels, indicators of in vivo lipid peroxidation. *Arch Toxicol* 1988;62:369-74.

9. Parkki MG, Marniemi J, Vainio H. Long-term effects of single and combined doses of DDT and PCB on drug-metabolizing enzymes in rat liver. *J Toxicol Env Health* 1977;3:903-11.

10. Oesterle D, Deml E. Promoting effect of polychlorinated biphenyls on development of enzyme-altered islands in livers of weanling and adult rats. *J Cancer Res Clin Oncol* 1983;105:141-47.

11. Silkworth JB, Grabstein EM. Polychlorinated biphenyl immunotoxicity: dependence on isomer planarity and the Ah gene complex. *Toxicol Appl Pharmacol* 1982;65:109-115.