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Accumulation and Long Term Effects of a PCB and DDE Methyl Sulfone Mixture in the Mink (*Mustela vison*)

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Abstract

Fifteen synthesised PCB methyl sulfones ($MeSO_2$-PCBs) and ($MeSO_2$-DDE) were mixed according to their relative concentrations in grey seal (*Halichoerus grypus*) blubber, in a quantity enough to treat female mink with daily doses (0.1 mg sulfone mixture/day and mink) for 12 months. The mink were mated after 9 months. The litter size was significantly increased compared to the control group (7.9 ±2.7 pups per treated mink compared to 5.1 ±2.4 pups in the control group). The mortality rate was significantly higher among the kits from the PCB and DDE methyl sulfone treated group than among the controls. Among several biochemical parameters measured in the adult females after the exposure period, significantly decreased concentrations of the thyroid hormones T_3 and T_4 were observed. All the aryl methyl sulfones were strongly retained in the mink muscle of both maternal mink and offspring, showing an efficient transfer of these compounds from mothers to kits. A highly selective retention of some of the $MeSO_2$-PCBs and of $MeSO_2$-DDE was found in maternal but not in kit livers.

Introduction

Not only anthropogenic substances but also their metabolites may be accumulated in wildlife and humans. This is known to occur for the DDT metabolites DDD and DDE, for aryl methyl sulfones of PCB and DDE [e.g. 1-3], and for hydroxylated PCB metabolites [4]. Even though the presence of PCB and DDE methyl sulfones in wildlife has been known for two decades, [5] it has not been possible to perform congener specific analysis until a few years ago [6]. A prerequisite for these analyses was the synthesis of a number of aryl methyl sulfone standards [7-9]. The availability of larger quantities of several aryl methyl sulfones was also necessary for this study.

Even though the PCB and DDE methyl sulfones have been known for decades, only limited information exists on their toxicological impact. However, $MeSO_2$-DDE has been shown to be strongly toxic to the adrenal cortex (*zona fasiculata*) in mice [10,11]. $MeSO_2$-PCB congeners with the sulfone group in a *meta*-position of the molecule have in several cases been shown to be potent inducers of cytochrome P450 2B1, 2B2, 3A2 and 2C6 in rats, [12-14] but no apparent

toxicity of $MeSO_2$-PCB has so far been reported.

The aim of the present study was to investigate chronic and reproductive toxicity in mink of a mixture of 15 $MeSO_2$-PCBs and $MeSO_2$-DDE known to be present in wildlife, and to determine the accumulation potential of the sulfones, including tissue distribution, and transfer of the sulfones from mother to offspring.

Experimental Methods

A general description of the experiment is given below. A detailed description of all experimental conditions of the present study is given elsewhere [15].

A $MeSO_2$-PCB and $MeSO_2$-DDE mixture (400 mg) of the synthesised compounds [7-9] was dissolved in fish oil (North-Atlantic capelin). The relative concentrations of the 16 aryl methyl sulfones in the mixture was based on the concentrations of the compounds (table 1) in a homogenate of blubber of grey seals from the Baltic [16].

Table 1. Aryl methyl sulfones used in the present study and their abbreviations.

Aryl methyl sulfone	Abbrev.	Aryl methyl sulfone	Abbrev.
3-$MeSO_2$-2,5,2'4'-tetraCB	3-49	4-$MeSO_2$-2,5,2',4',5'-pentaCB	4-101
4-$MeSO_2$-2,5,2',4'-tetraCB	4-49	3-$MeSO_2$-2,5,6,3',4'-pentaCB	3-110
4-$MeSO_2$-2,3,6,4'-tetraCB	4-64	4-$MeSO_2$-2,3,6,3',4'-pentaCB	4-110
3-$MeSO_2$-2,5,3',4'-tetraCB	3-70	3-$MeSO_2$-2,5,6,2',3',4'-hexaCB	3-132
3-$MeSO_2$-2,5,2',3',4'-pentaCB	3-87	4-$MeSO_2$-2,3,6,2',3',4'-hexaCB	4-132
4-$MeSO_2$-2,5,2',3',4'-pentaCB	4-87	3-$MeSO_2$-2,5,6,2',4',5'-hexaCB	3-149
3-$MeSO_2$-2,5,6,2',4'-pentaCB	3-91	4-$MeSO_2$-2,3,6,2',4',5'-hexaCB	4-149
3-$MeSO_2$-2,5,2',4',5'-pentaCB	3-101	3-$MeSO_2$-DDE	$MeSO_2$-DDE

For 12 months, ten female mink obtained 0,1/mink/day of the sulfone mixture via the feed [15]. The mink were one year old when the experiment started. Twenty non-exposed mink were used as controls. The mink were mated after 9 months of exposure to the sulfone mixture. The dams were anaesthetized and killed 6 weeks after delivery. The kits were killed at 5 weeks of age. Blood and samples of muscle, liver, lung, adrenal and kidney were taken for the analyses described below.

The reproductive outcome, signs of malformations, survival frequency and body and organ weights were determined. The thyroid and adrenal glands were examined for histopathological changes. Hepatic ethoxyresorufin O-dealkylase (EROD), pentoxyresorufin O-dealkylase (PROD) [17-19], hepatic progesterone catabolism [15], adrenal P450c11 and P450c21 [19] activities, plasma alanine aminotransferase (ALAT), alkaline phosphatase (ALP), cholesterol, fructosamine, bile acids, T_3 and T_4 [20] were analysed. Further, liver, lung and kidney retinol and retinyl palmitate [21] were determined in the mink.

Quantitative chemical analysis of $MeSO_2$-PCBs and $MeSO_2$-DDE in muscle, liver and lung samples of dams and their offspring was performed as previously described [22].

Results and Discussion

Only the major, significant, findings in the present study will be reported in this context. A full report on this study will be published elsewhere [15]. Thus, among all parameters analysed significant differences between the sulfone exposed minks and the control group were observed for litter size, liveborn kits, survival rate at 2 weeks, hepatic PROD and progesterone catabolism, plasma T_3 and T_4 as shown in table 2. Hepatic PROD was also increased in the kits ($p < 0.05$).

Table 2. Effects on mink dosed with a mixture of 15 $MeSO_2$-PCB and 3-$MeSO_2$-DDE

Parameter	Control group	Experimental group
Litter size (number)	5.07 ±2.37	7.86 ±2.73*
Liveborn/litter (number)	4.43 ±2.47	7.00 ±2.31*
Kit survival (%)	73.0 ±14.4	47.1 ±30.6*
Hepatic PROD (pmol product/mg protein/min)	13 ±5 n = 7	143 ±86* n = 6
Hepatic progesterone catabolism (nmol product/mg protein/min	0.29 ±0.07 n = 6	0.48 ±0.06* n = 6
Plasma T_3 (nmol/l)	1.87 ±1.11	0.76 ±0.48**
Plasma T_4 (nmol/l)	55.0 ±44.1	19.3 ±16.1**

* $p < 0.05$; ** $p < 0.01$ Student's t-test

All the sulfones were accumulated to a similar degree in the maternal and kit muscle and lung; 17 - 21 µg/g lipid weight (l.w.). Significantly higher concentration of aryl methyl sulfones was observed in the maternal livers (82.3 ±28.7 µg/g l.w.) as compared to the kit livers (28.2 ±3.22 µg/g l.w.). The relative composition of the PCB methyl sulfones and the DDE methyl sulfone in muscle, lung and liver are compared to the original composition of these sulfones in the mixture used for dosing the mink (Figure 1). It is clear that the general relative abundance of the individual $MeSO_2$-PCBs in the mink muscle and lung is similar to their concentrations in the original sulfone mixture. Minor differences are however observed, e.g. relatively higher concentrations are observed in maternal lung and muscle of 4-49, 4-101, 3-87 and 4-87 (Figure 1, upper diagram). The same PCB methyl sulfones are also magnified in kit muscle and lung tissue (Figure 1, lower diagram). $MeSO_2$-DDE on the other hand, shows a major decrease in the relative concentration in maternal mink and particularly in kit tissues. All the *meta*-substituted PCB methyl sulfones were accumulated in maternal liver tissue, particularly 3-91, 3-101, 3-87, 3-149 and 3-132. No similar strong retention was obtained in the kit livers even though 3-149 and 3-132 were accumulated to some degree also in the kit livers.

The results clearly show that the aryl methyl sulfones are readily bioavailable and accumulate in the mink. Also, the sulfones are transferred to the kits. $MeSO_2$-DDE is not accumulated to the same degree as the $MeSO_2$-PCBs. This is probably due to metabolism of $MeSO_2$-DDE. The strong retention of the 3-$MeSO_2$-PCBs in liver tissue is similar to the observed tissue selective retention of these metabolites in, e.g., seal and otter livers [2] and in polar bear livers [ref].

The significant PROD induction in the mink is in agreement with the observation of strong P450 inductions by 3-$MeSO_2$-CBs in rats [12-14]. The strong accumulation of certain $MeSO_2$-PCBs (cf. above) in the livers raises the possibility that these sulfones are mainly responsible for the induction observed, which is supported by data from Kato and coworkers who have shown that several of them are P450 inducers [12-14]. Also a two-fold enhancement of the progesterone catabolism was determined in the livers of the dams, which may have importance for the embryo development.

Interestingly, the litter size was increased in the sulfone-treated mink. This is not easy to explain, but may indicate a hormonal disturbance of some kind.

In conclusion, one year of exposure of mink to a PCB and DDE methyl sulfone mixture, identical to the mixture of sulfones present in Baltic grey seals, resulted in induced levels of hepatic enzymes, and decreased T_3 and T_4 levels in the plasma. The exposed mink gave birth to a larger number of kits even though their survival rate was lower. The sulfones are bioavailable from the food, bioaccumulate in several tissues, and are transferred to the offspring. Some 3-$MeSO_2$-CBs were strongly retained in the liver of the adult female mink.

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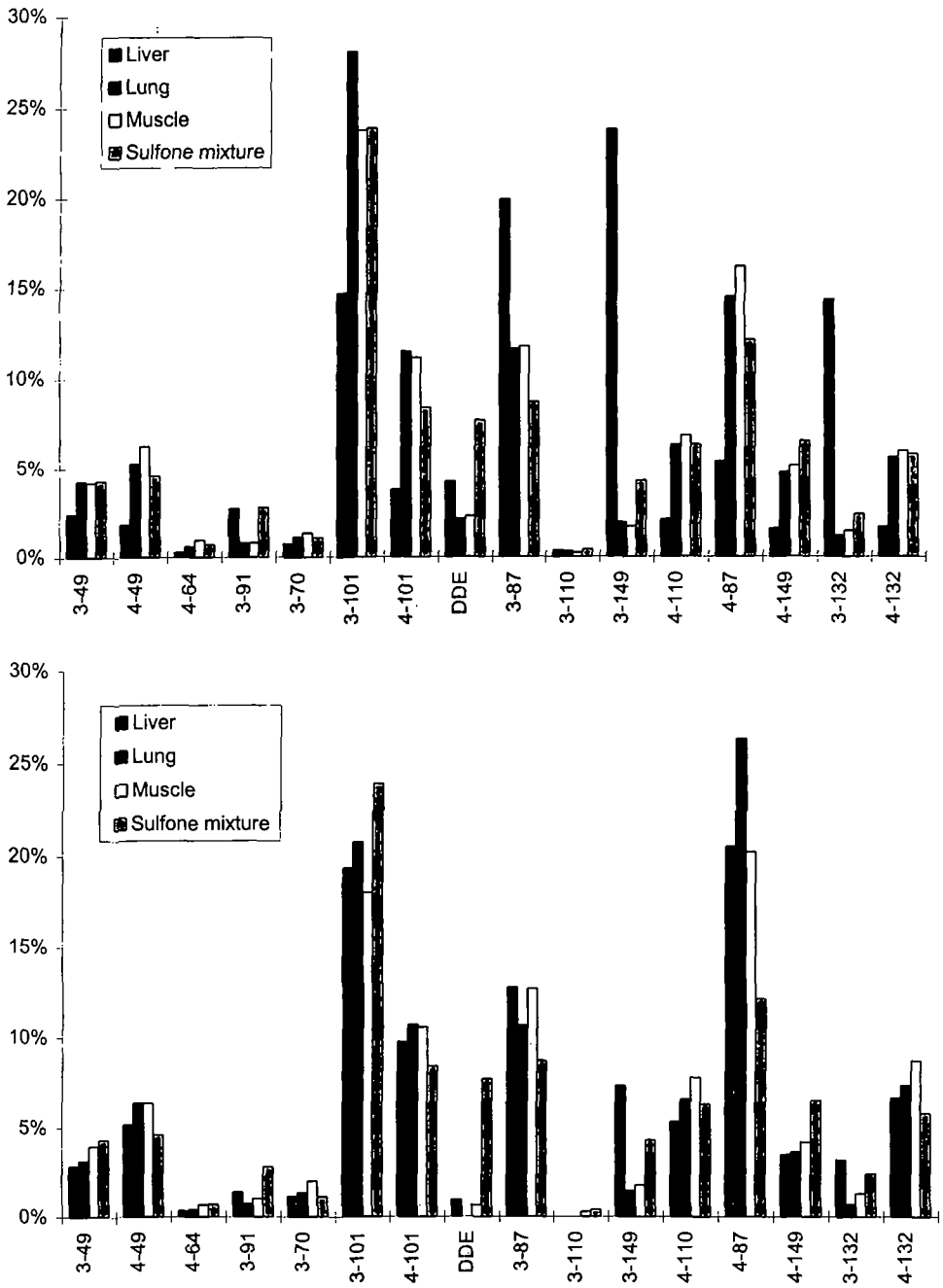
Figure 1. Relative concentrations of the individual $MeSO_2$-PCB congeners and $MeSO_2$-DDE (% of total) in the mixture used for dosing the mink and in liver, lung and muscle of the maternal mink (upper diagram) and the kits (lower diagram). The total concentration of the sulfones is set to 100% in each case.



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