EROD-inducing potencies of extracts from sediments collected in a PCBcontaminated Swedish lake

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## INTRODUCTION

Biota and sediments from Lake Järnsjön (26 ha, maximum depth 2.5 m, average depth 0.7 m) in the water system of the Emån River located in southern Sweden contain high levels of polychlorinated biphenyls (PCBs)<sup>1,2</sup>. The main source of the PCBs is a paper recycling plant located just upstream of Lake Järnsjön. The paper plant has been using in part PCB-contaminated self-copying paper as a raw material. The sediments of Lake Järnsjön also contain high levels of polychlorinated naphthalenes<sup>1</sup>. Lake Järnsjön is situated about 60 km upstream of the river mouth and the transport of PCBs into the Baltic from the Emån River has been estimated at about 5 kg per year. The contaminated sediments will be dredged in 1993. As part of a comprehensive examination programme before dredging starts, sediments have been sampled in Lake Järnsjön, Lake Flögen, located upstream of the paper recycling plant, and Lake Grönskogssjön, located about 50 km downstream from Lake Järnsjön. The ethoxyresorufin-O-deethylase (EROD)-inducing potencies of the compounds extracted with toluene from the sediments are presented in this paper.

## MATERIALS AND METHODS

Lake Järnsjön was divided into twelve areas and surface sediment samples (0-5 cm) were taken in each area. Sediment samples were also collected in various parts of Lake Flögen and Lake Grönskogssjön. The sediment samples were Soxhlet-extracted for 24 h with toluene. The toluene was then evaporated and the residues redissolved in hexane, followed by the clean-up of the hexane solution on a Si-column. Aliquots of the hexane solution were evaporated and the residues redissolved in DMSO.

The livers of 8-day-old chick embryos (White leghorn, Shaver) were transferred to glass vials containing RPMI 1640 supplemented with penicillin, streptomycin, L-glutamine, HEPES, and 10% heat-inactivated fetal calf serum (0.5 ml medium per liver, 6 livers per vial). DMSO solutions of the extracts as well as TCDD were added to the cultures (3  $\mu$ l DMSO/ml medium). After having been cultured for 48 hours, the EROD activity of each liver was determined individually as previously described<sup>3</sup>. The concentration-response relationships were determined and compared with that for 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in the liver culture system. EC<sub>50</sub> values, i.e. the concentrations of the

extracts that caused half the maximal induction responses, were estimated from the concentration-response curves.

# **RESULTS AND DISCUSSION**

All extracts tested caused a concentration-dependent increase in EROD activity. The concentration-response curve for EROD induction with TCDD, used here as a reference substance, is shown in Fig. 1. The dose-response curve for induction with an extract from a pooled sediment sample from the twelve areas in Lake Järnsjön is shown in Fig. 2. In contrast to the concentration-response relationship for TCDD, there was a decrease in activity at high concentrations of the pooled sample extract. This decline indicates that the extract contained compounds that inhibited EROD activity. The EC<sub>50</sub> value, presented in Table 1, was calculated from the curve adapted to the data points showing a concentration-dependent increase in activity. Due to the possible presence of inhibiting substances in the extract, the potency of the inducing compounds present in the extract might have been underestimated. Extracts from sediments collected from the Emån River inlet to Lake Järnsjön were about twice as potent as the extract from the pooled sample (Table 1).

Extracts from sediments collected in Lake Flögen and Lake Grönskogssjön caused similar concentration-response curves as the extracts from sediment samples obtained from Lake Järnsjön. The  $EC_{50}$  value for the extract collected in Lake Flögen (upstream of the paper recycling plant) was approximately 50 times higher than the value for the pooled sample from Lake Järnsjön (Fig. 3, Table 1). The extract from sediment obtained from Lake Grönskogssjön (50 km downstream from Lake Järnsjön) was intermediate in EROD-inducing potency compared with the other two locations. Thus, the toxic potency of compounds associated with the sediments downstream from the paper recycling plant in the Emån River was considerably higher than the potency of compounds in sediments located upstream of the plant.

By comparing the  $EC_{50}$  values obtained for the sediment extracts with the  $EC_{50}$  value for TCDD, the concentrations of TCDD equivalents (TEQs) in the sediments could be estimated (Table 1). However, the different shapes of the concentration-response curves for TCDD and the sediment extracts makes a stringent determination of TEQs difficult.

Previous studies have shown high levels of coplanar PCBs in the sediments of Lake Järnsjön<sup>1</sup> and it is most likely that these compounds accounted for the majority of the EROD inducing potency of the samples collected downstream from the paper recycling plant. The *in vitro* EROD induction test, here applied on extracts from sediments, seems to be a useful tool for estimating the concentrations and potencies of dioxin-like compounds in environmental samples.

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Fig. 2. Concentration-response curve for EROD induction with an extract from sediments collected in Lake Järnsjön. Quantity is given as mg (dry weight) of sediment from which the added compounds were extracted. The  $EC_{50}$  value was calculated from the curve adapted to the data points showing a concentration-dependent increase in EROD activity. Each point represents the mean activity of six livers and variation is given as S.D.

Table 1.  $EC_{50}$  values for extracts from sediments collected in the water system of the Emån River and estimated concentrations of TCDD equivalents (TEQs) in the sediments.

Extract	EC <sub>50</sub> <sup>a</sup> Quantity <sup>b</sup> /ml medium	TEQs <sup>c</sup> pg/g dw
Lake Järnsjön, pooled sample	6	300
Lake Järnsjön, the inlet	3	600
Lake Flögen, pooled sample	300	6
Lake Grönskogssjön, pooled sample	50	40

<sup>a</sup> The  $EC_{so}$  values were determined from the concentration-response curves adapted by computer-aided nonlinear curve-fitting.

<sup>b</sup> Quantities are given as mg (dry weight) of sediment from which the added compounds were extracted,

<sup>c</sup> Calculated by comparing the  $EC_{50}$  value for the extract with the  $EC_{50}$  value for TCDD.



Fig. 3. Inverted  $EC_{50}$  values for extracts from sediments collected in various parts of the Emån River water system. The  $EC_{50}$  values were determined from the concentration-response curves adapted by computer-aided nonlinear curve-fitting.

### REFERENCES

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