Accumulation of hexabromocyclododecane (HBCD) diastereoisomers in juvenile rainbow trout (Oncorhynchus mykiss) after single oral exposures

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Introduction

- HBCD is a brominated flame retardant which due to its environmental stability, persistence and high production volume has been found widely distributed over the northern hemisphere
- The highest HBCD concentrations have been found around urban areas and close to production sites (Janak et al. 2005)
- Intake via food is probably the main source of HBCD exposure in wild fish (Kuiper et al. 2007)
- The HBCD diastereoisomers α, β and γ (Fig. 1) behave differently in the environment, and there is a selective biomagnification of the α-isomer in aquatic food webs (Tomy et al. 2004, Law et al. 2006a, Sørmo et al. 2006)
- The elevated level of the α-isomer in biota may be due to isomer selective uptake (Kuiper at al. 2007), or isomer-specific metabolization of β- and γ-HBCD (Law et al. 2006b)
- In the present study, a feeding experiment was performed to assess the role of selective uptake on isomer-specific accumulation of HBCD in fish

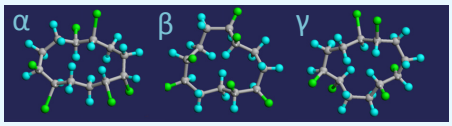

Fig. 1: The three diasereoisomers of HBCD; α, β and γ

Materials and methods

Food preparation

- Homogenate of fish pellets were spiked with technical HBCD (α=1.6, β=8.1, γ=90.3 % contribution) and diluted with distilled water to achieve a concentration of 1 mg/ml. Control food was prepared similarly, but not spiked with HBCD

Animals and treatment

- Juvenile rainbow trout were anesthetized before pellet homogenates was administered orally through a stomach tube (Fig. 2)
- Individual doses of HBCD were 10 mg/kg bodyweight
- A total of ten fish were euthanized at time zero and 3 h, 6 h, 12 h, 24 h, 48 h, 4 d, 8 d and 15 d after exposure

Fig. 2: The pellet homogenate was orally administered

Sample extraction and analysis

- Whole-body fish samples (without GI tract) were analyzed for α-, β- and γ-HBCD using HPLC/MS. Lipid content of the fish was determined gravimetrically to be 7,4 ± 0.1 % (mean ± SE)
- Before analysis, the samples were extracted with ASE, followed by acid treatment

Data analysis

- Inferential statistics were computed in R 2.3.1, wherein the significance level was set at $\alpha \leq 0.05$. ANOVA and Tukey's HSD tests were used to analyze differences in concentration between time groups
- The 3 h data were not included in the statistical analysis due to residues of undigested food in the fish throats, resulting in overestimated accumulation concentrations

Results

- 6 hours after exposure, the accumulated concentration of all three HBCD isomers was significantly higher than in the zero group and control fish

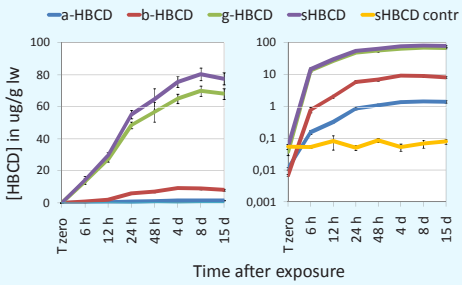

Fig. 3: HBCD accumulation in rainbow trout. The right diagram displays the data on a log-scale. Concentration (ng/g lipid weight) of α- (blue), β- (red), γ- (green) isomers and ΣHBCD (violet) are plotted for the exposed fish. Only ΣHBCD levels (yellow) are plotted for the control fish

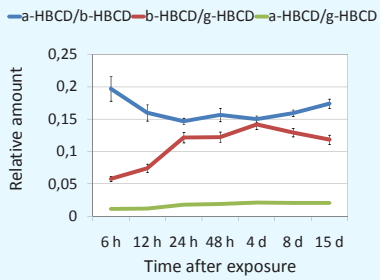

Fig. 4: Relative amounts of α- to β-HBCD (blue), and β- to γ-HBCD (red) and α- to γ-HBCD (green) in exposed rainbow trout

- The concentration of accumulated HBCD reached a maximum level on day eight, after which the level of all isomers decreased, though not significantly (Fig. 3). This decrease is consistent with an earlier study showing that depuration starts approx. one week after exposure (Law et al. 2006b)
- The relative isomer contribution of the administrated HBCD-mixture was reflected well in the analyzed fish after 6 h (α=1.1, β=5.4, γ=93.5%), but changed significantly during the experiment. The relative amount of α and β increased, while γ decreased
- The relative amount of α to β and γ to β decreased significantly from 6 h to 4 d, and then appeared to increase to 15 d (Fig. 4). The ratio α to γ increased significantly from 6 h to 8 d

Conclusions

- HBCD was efficiently accumulated from diet in rainbow trout
- Accumulation of HBCD was evident until day 8, after which the depuration probably dominated
- According to the relative isomer contribution, α and β appeared to be more easily taken up and accumulated than γ in rainbow trout
- The relative amounts of β to α and γ, suggested that β was accumulated and metabolized more efficiently than α and γ in rainbow trout

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