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In vitro TOXICITY OF DECABROMODIPHENYL ETHANE (DBDPE)

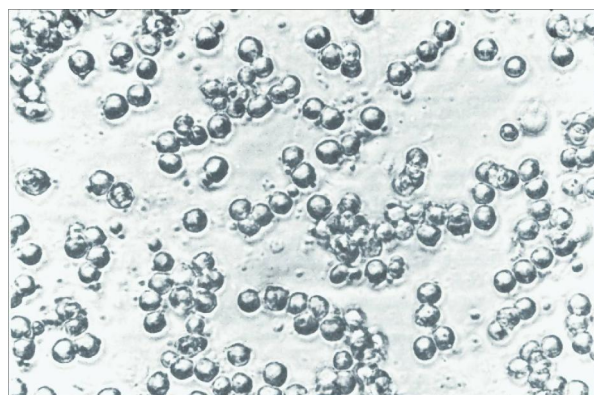
Introduction

Decabromodiphenyl ethane (DBDPE) came into the market in the early 1990s as an alternative to decabromodiphenyl ether (BDE-209), which was assumed not to be bioavailable due to its high molecular weight of 971 g/mol. However, the BDE-209 has been detected in humans as well in the environment. DBDPE has been used as an additive flame retardant in different types of plastics and textiles, and it is predicted to become one of the major flame retardants used by the thermoplastics industry. There is evidence of its existence in aquatic food web but amount of data concerning the toxicity of DBDPE is limited.

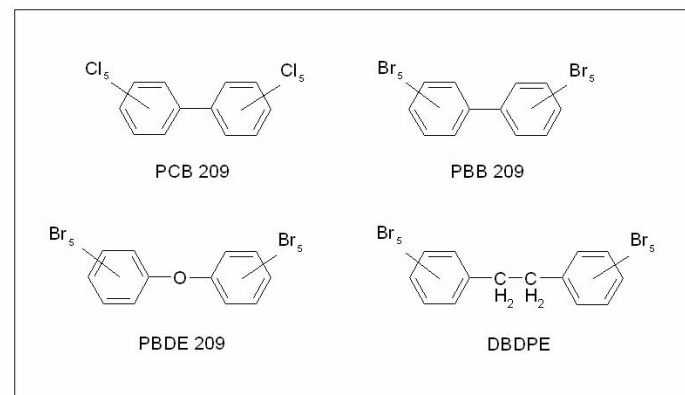
Materials and Methods

Freshly isolated hepatocytes from hatchery-reared brown trout (*Salmo trutta*) males were used to test the bioaccumulation, detoxification metabolism and estrogenic activity of DBDPE. Ethoxyresorufin-O-deethylase (EROD) and uridinediphosphoglucuronyltransferase (UDPGT) activities were used to assess the detoxification activity. Cell vitellogenin induction was the variable for the estrogenic activity.

The hepatocyte cell culture samples were extracted with dichloromethane (DCM). The solvent was changed to isooctane and the extract was treated with concentrated sulphuric acid. The quantitation was done with ¹³C-labelled DBDPE as an internal standard. The analyses were conducted on Varian CP-3800 GC coupled to Varian 1200L triple quadrupole mass spectrometer. Short analytical column (10m x 0.53mm) was operated at diminished pressure (Rapid-MS™). Mass spectrometer was operated at EI/SIM mode. The most abundant fragment and the molecular ion was monitored (m/z 971 $[M+10]^+$ and 485 $[C_6Br_5CH_2]^+$).



Pic. 1. Trout hepatocytes after 2h in culture.**



Pic. 2. Molecular structures of PCB-209, PBB-209, PBDE-209 and DBDPE

Results

DBDPE was detected from the hepatocytes at concentrations corresponding accumulation of 42% (26 ng/18milj cells) at test concentration of 6.25 µg/l with decrease to 17% (84ng/18 milj. cells) at test concentration of 50 µg/l. DBDPE was not detected from the cell culture blank or from the procedural blank.

DBDPE caused a significant vitellogenin induction in the separated hepatocytes already at the concentration of 6.3 µg/l (Fig.1.). Inhibition was observed at higher test concentrations. Hepatocyte EROD activity (Fig.2.) followed similar courses with that of the vitellogenin. The activity increased significantly up to the concentration of 12.5 µg/l. After that DBDPE started to inhibit the enzyme activity so that at the concentration of 25 µg/l the cell EROD activity was decreased to zero. What comes to the cell UDPGT activity (Fig.3.) the trend was increasing till the highest test concentration, 50 µg/l.

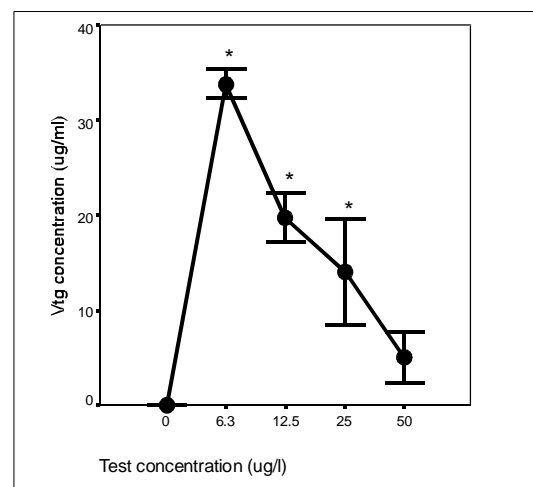


Fig. 1. Vitellogenin response (mean ± SD) of hepatocytes to DBDPE. Asterisks denote the significant difference from the control ($p < 0.05$).

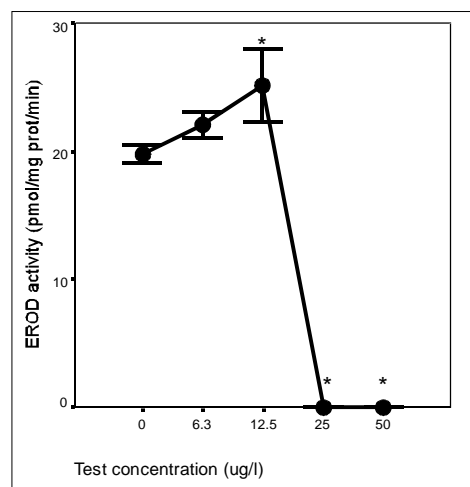


Fig. 2. Hepatocyte EROD activity. Asterisks denote the significant difference from the control ($p < 0.05$).

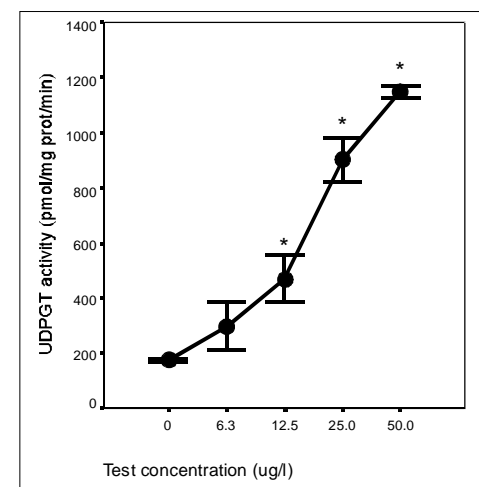


Fig. 3. Hepatocyte UDPGT activity. Asterisks denote the significant difference from the control ($p < 0.05$).

Conclusions

Even though the assumption has been that DBDPE is not bioavailable it was observed that DBDPE does accumulated into the hepatocytes. According to the observed changes in the hepatocyte detoxification activities it can be stated that there is metabolism of DBDPE in the brown trout liver cells. The toxic effects of DBDPE and the effective concentration levels has to be investigated. Also the occurrence of DBDPE in the environment needs to be further studied.

References

- American Chemical Council: Brominated flame retardant industry panel. (2001) Document Number Arias, P.A. 2001.
 Castren, M. and Oikari, A. 1983. Comp. Biochem. Physiol. 76C, 365-369.
 Firemaster 2100 Technical Information. Great Lakes Polymer Additives. www.polyadd.com/products/firemaster:210.pdf
 Hodson, P.V. et al., 1991. Can. Tech. Rep. Fish. Aquat. Sci. 1829, Quebec, Canada.
 Kierkegaard, A., Björklund, J., 2003. Organohalogen compounds. 61, 183-186
 Kierkegaard, A., Björklund, J., Friden, U., 2004. Environ. Sci. Tech. 38, 3247-3253
 Klotz, A.V. et al., 1984. Anal. Biochem. 107, 150-155.
 Konstantinov, A. et al., 2006. Chemosphere 64, 245-249.
 Law, R.J. et al., 2006. Chemosphere 64 (2), 187-208.
 Moon, TW et al., 1985. Can. J. Fish. Aquat. Sci.
 Nielsen, B. et al., 1998. Mar. Environ. Res. 46, 153-157
 The second international workshop on brominated flame retardants, Stockholm Sweden, May 14-16.
 ** picture from Pesonen, M., Andersson T. Xenobiotica. 1991, Vol. 21 461-471