FIRST WORLDWIDE INTERLABORATORY STUDY ON POLYBROMINATED DIPHENYL ETHERS (PBDEs)

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Introduction

Polybrominated diphenyl ethers (PBDEs) are being produced as flame reatrdants since the early 1970s. They are being found in the aquatic environment since the late 1970s[1,2]. However, since they were found to be present in sperm whales from deeper Atlantic waters[3] and since Norén and Meyronité[4] indicated an exponential increase of tetra and penta BDE concentrations in Swedish human milk, many laboratories have started to work on PBDE analysis in the environment. PBDEs are very similar in structure to polychlorinated biphenyls (PCBs)[5]. The two congeners which are most frequently reported in environmental samples are BDE 47 (2,4,2',4'-tetra BDE) and BDE 99 (2,4,5,2',4'-penta BDE). These congeners most likely originate from the (mainly) historical use of Penta mix, a PBDE mixture of mainly pentabrominated congeners, which is not used in electronic enclosures. However, the production of PBDEs has been gradually shifted towards BDE 209 (deca BDE)[6]. Until now, there are only few laboratories that have reported BDE 209 values in the environment, mainly because their methods did not include the analysis of deca BDE. Several laboratories have recently started to develop deca BDE methods.
The performance of the laboratories for PBDEs had not been tested until now. Therefore, an interlaboratory study on PBDEs was initiated. This study included most laboratories in the world which are carrying out PBDE analysis in the aquatic environment.

Methods

Seven samples were prepared: one sterilised canned fish sample: eel (*Anguilla anguilla*) (coded 1), one sterilised canned shellfish sample: mussels (*Mytilus edulis*) (coded 2), one bird sample: freeze-dried cormorant liver (*Phalacrocorax carbo*) (coded 3), two marine mammal samples: freeze-dried harbour porpoise (*Phocoena phocoena*) liver (coded 4) and ampouled harbour propoise oil (melted blubber) (coded 5), and two freeze-dried sediment samples (coded 6 and 7). In addition two BDE solutions, one containing the BDEs 47, 85 and 99 in iso-octane (coded 8), and one containing BDE 209 in toluene (coded 9) in undisclosed concentrations were prepared and ampouled. All biological samples were tested for homogeneity. The eel, porpoise blubber and porpoise liver were completely homogeneous. There was a small contribution of inhomogeneity of 3.7-5.9% in the mussels and of 0.7-2.6% in the cormorant liver. However, this contribution was considered negligible in comparison with the expected analytical error. The sediment samples had been used in previous interlaboratory studies on organic contaminants and trace metals[7] and had shown to be suitable for that purpose.
The BDEs 47, 99 and 85 in solution 8 were a gift of mrs. Dr. U. Sellstrøm of the Stockholm University (ITM). There were no significant impurities in this solution. The purity of the BDE 209 in solution 9 was 97%. The target concentration was corrected for the percentage of purity. The laboratories were asked to determine the concentrations of the BDEs 47 (2,4,2',4'-tetra BDE), 99 (2,4,5,2',4'-penta BDE) and 209 (2,3,4,5,6,2',3',4',5',6'-deca BDE) in all samples. This was the mandatory part of the exercise. In addition, a secondary, voluntary set consisting of the BDEs 28 (2,4,4'-tri BDE), 66 (2,4,3',4'-tetra BDE), 71 (2,6,3',4'-tetra BDE), 75 (2,4,6,4'-

tetra BDE), 77 (3,4,3',4'-tetra BDE), 85 (2,3,4,2'4'-penta BDE), 100 (2,4,6,2',4'-penta BDE), 119 (2,4,6,3',4'-penta BDE), 138 (2,3,4,2',4',5'-hexa BDE), 153 (2,4,5,2',4',5'-hexa BDE), 154 (2,4,5,2',4',6'-hexa BDE), and 190 (2,3,4,5,6',3',4'-hepta BDE) could be determined. The emphasis of the study was on the between-laboratory agreement. So, only one result per determinand per sample was required. The participants were asked to use their own method. Some advice was given on how to avoid specific errors during the determination. It was recommended to use at least two GC columns of different polarity to check for co-elution. The best result to the judgement of the participant should be reported. The reported results should be corrected for recovery. The participants were requested to send chromatograms (one of the eel sample, one of the sediment sample and one of each standard solution from all GC columns used) and their method description to the co-ordinator. A period of 4.5 months was given to the participants to complete this work.

Results and discussion

Results of twenty laboratories from ten different countries were received. For several reasons five laboratories were not able to carry out the task. The preliminary results (16 sets) of this study are given in Table 1. Only those BDEs are given which were found in relevant concentrations in the samples. The BDEs 28, 71, 75, 77, 119, 138 and 190 were generally below the detection limits of the laboratories in all samples. The BDEs 66 and 119 were only found by some laboratories in the harbour porpoise samples.

BDE 47

The results for BDE 47 are satisfactory with a range of relative standard deviations (Rsd) of 10-42%. Actually, for some of the samples the result is extremely good. In eel an Rsd value of 10% was found. Such a low Rsd value is rarely found in PCB interlaboratory studies[8]. Most participating laboratories have experience in analyzing this BDE, but this was the first interlaboratory study ever held for this compound. Apparently, methods are well under control in most laboratories. The purity of the standards used by the laboratories is also acceptable, although, given the very good result for the eel and other samples, a better Rsd value than the now obtained 20% would have been expected for the unknown solution. Two laboratories reported strongly deviating values for the unknown solution. For the biological and sediment samples the number of outliers was limited to one or two.

BDE 99

The results for BDE 99 are not as good as for BDE 47. This is partly due to the generally lower concentrations of BDE 99 in the samples compared to BDE 47. For example, in eel the BDE 99 concentration is more than 10-fold lower than the BDE 47 concentration. In the two sediments in which the level of BDE 99 is higher than that of BDE 47, the Rsd values approach those of the BDE 47, but are still somewhat higher. This points to a chromatographic problem. The result of the analysis of the standard solution (Rsd 28%) is not much different from that of BDE 47, which means that a significant problem with the purity of the standards is not expected for most of the laboratories.

BDE 209

Given the shift in production of PBDEs towards deca BDE, a good quality of the BDE 209 analysis is important. Until now laboratories were not very experienced in this analysis. This is clearly reflected in the results. The comparability of the participating laboratories for BDE 209

Table 1. Summary of preliminary results of BSEF PBDE interlaboratory study

Sample	47			99			209		
	m	Rsd	n	m	Rsd	n	m	Rsd	n
1 eel	11.6	10	13	0.92	77	15		>100	11
2 muss.	0.53	42	13	0.29	59	13		>100	11
3 corm.	49.7	18	16	18.0	32	15		>100	12
4 por. l.	133	20	15	20.4	25	15		>100	13
5 por.b.	643	25	14	108	43	14		>100	11
6 sed.	2.5	22	13	3.2	35	11	2.9	48	10
7 sed.	12.5	24	12	9.2	36	11	64.5	78	5
8 sol.	819	20	15	1094	28	15			
Target	910			900					
9 sol.							296	46	12
Target							298		

Sample	100			153			154		
	m	Rsd	n	m	Rsd	n	m	Rsd%	n
1 eel	3.4	26	11	0.54	38	11	0.61	16	9
2 muss.	0.15	35	9	0.04	55	5	0.04	46	5
3 corm.	33.5	20	11	12.7	31	14	11.4	26	12
4 por. l.	29.9	26	10	7.3	37	13	14.2	31	12
5 por.b.	105	39	10	29.1	34	12	51.6	25	10
6 sed.	0.46	33	10	0.47	37	11	0.29	43	9
7 sed.	3.1	21	9	3.2	32	9	1.4	35	9

m: mean in ng/g, Rsd: relative standard deviation in %, n: number of observations.

is still not at the desired level. The Rsd value obtained for sediment sample 6 (48%) is, however, almost acceptable given this was the first exercise ever for this compound with relatively unexperienced laboratories, and given also the degree of difficulty of the analysis. This result could not be repeated by the participating laboratories for sediment 7, although the BDE concentrations in that sample were higher. Apparently, the within-laboratory variance is still not completely under control. The result for the unknown solution is less than expected. Apart from the relatively large Rsd value (46%), a few laboratories reported values which differed an order of magnitude from the mean and target value. For the biota samples the result was rather poor. The presence of BDE 209 in the fish and shellfish samples in measurable amounts is unlikely, so these samples are not representative for a good laboratory comparison. However, it is important for the laboratories to be able to determine whether or not deca BDE is present in marine mammals and birds. However, for example in the harbour porpoise liver, some laboratories reported a BDE 209 value of < 0.1 ng/g, whereas other laboratories found 6.3 or 25 ng/g. Obviously, the BDE 209 analysis includes a number of analytical difficulties. The compound is not stable at higher temperatures in the injector and at the GC column. The compound is

sensitive for degradation by UV light. The behaviour in the MS source is different from that of chlorinated and lower brominated compounds. These issues should be studied in detail in order to make further progress with this group of laboratories.

BDE 100
The results of BDE 100 are surprisingly good. Even in the mussels at the low level of 0.16 ng/g an Rsd value of 20% is found. Six laboratories reported a value below their detection limits. The chromatographic separation of BDE is apparently less disturbed by interferences than that of BDE 99.
BDEs 153and 154
The Rsd values of the BDEs 153 and 154 are in the range of 25-55%. The concentrations are generally low, which explains the difficulties which the laboratories experience in determining these BDEs.

Four laboratories used high resolution (HR) MS. The results reported by these laboratories for the BDEs 47 and 99 were not different from the other results which were obtained by low resolution (LR) MS or GC/ECD. In some cases the HRMS laboratories reported lower BDE 47 and /or 99 values, in some cases these values were higher. Also for BDE 209 the differences between the two groups of laboratories were small. HRMS laboratories may be able to obtain somewhat lower detection limits. Two HRMS laboratories reported extremely low BDE 47 and 99 concentrations in the solutions 8 and 9.

Conclusions
- The first interlaboratory study on PBDEs shows that there is a good agreement between the laboratories for BDE 47 with Rsd values of 10-25% for seven of the eight samples. Also the results of BDE 100 show a good agreement.
- The results of the BDEs 99, 153 and 154 show that further improvement is required. Particularly for BDE 99, a better resolution is required to separate this BDE from interferences or other BDEs.
- The analysis of BDE 209 is not under control yet in most laboratories. For one of the sediments an Rsd value of 48% was obtained, but this was not repeated for the second sediment sample. The results of BDE 209 in biota were very variable.
- By improving the calibration the performance of the laboratories can be improved substantially.

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References
1. Wolf M. and Rimkus, G. (1985) Dtsch. Tierärtzl Wschr. 92, 174.
2. de Boer J. (1989) Chemosphere, 18, 2131.
3. de Boer J., Wester P.G., Klamer H.J.C., Lewis W.E and Boon JP (1998) Nature, 35, 1187
4. Norén K.and Meyronité D. (1998) Organohalogen Compounds, 38, 1
5. Ballschmiter K., Bacher R., Mennel A., Fischer R., Riehle U. and Swerev M. (1992) J. High Resolut. Chromatogr., 15, 260.
6. de Boer J., de Boer K. and Boon J.P. (2000) in: The Handbook of Environmental Chemistry (Paasivirta J., Ed): Vol 3 Part K , 61.
7. Houba, V.J.G., Uittenbogaard, J., Cofino, W., Pellen, P.J., van Dijk, D. and Brader, A. (1999) SETOC report 1998, Wageningen University, Wageningen, the Netherlands.
8. de Boer J. and Wells, D.E. (1997) Mar. Pollut. Bull. 35, 52.