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Screening and Time Trend Study of Decabromodiphenylether in Birds

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Executive Summary

In 2001, Swedish scientists reported the presence of decabromodiphenylether (DBDE) in peregrine falcons. Subsequently, we have carried out an extensive study on the presence and time trends of DBDE in predatory birds and water birds in order to provide information on this subject for the European environmental risk assessment of DBDE. The UK Environment Agency and the Bromine Science and Environmental Forum (BSEF) initiated the study. In the first part of the study, 135 samples from various species of predatory birds and water birds (muscle tissue, liver, eggs) were analysed for DBDE. In the second part of the study, sparrow hawk muscle tissue (64 samples) and peregrine falcon eggs (48 samples) sampled at different times between 1973 and 2001 were analysed for DBDE in order to obtain a time trend. The analytical difficulties in the determination of DBDE are substantial, as has been shown by the results from two international interlaboratory studies. Therefore, much attention was paid to the optimisation and validation of the analytical method so as to ensure that blank values were sufficiently low as to allow unequivocal detection of DBDE at very low concentrations. Despite this, a problem with elevated blank values resulted in some delay in one of the laboratories at the start of the second stage (analysis of time trend samples). Comparisons between the two laboratories involved and between one of these laboratories and Stockholm University (who conducted the original Swedish study) showed very good comparability. Otherwise, the study went smoothly and has yielded reliable concentration values for DBDE in the samples studied. The high variability of DBDE concentrations in predatory birds and water birds in combination with the relatively low DBDE concentrations found created some difficulties in detecting and interpreting trends.

This study was carried out by the Netherlands Institute for Fisheries Research, in collaboration with the Centre for Environment, Fisheries and Aquaculture Science (CEFAS). Most samples were obtained from the Centre for Ecology and Hydrology, Monks Wood, Huntingdon (CEH), UK, while some were provided by Stockholm University and the Bureau Waardenburg in The Netherlands. The conclusions and recommendations of this study are given below.

Conclusions

1. The presence of DBDE in peregrine falcon eggs from Sweden has been confirmed.
2. DBDE is also present in peregrine falcon eggs from the UK, as well as in peregrine falcon muscle tissue and liver, but at 3-4 fold lower concentrations than those found in Swedish peregrine falcon eggs.
3. Other UK predatory birds (sparrowhawk, barn owl, kestrel, red kite, Montagu's harrier and merlin) also contain DBDE, although at relatively low concentrations, generally just above the detection limits, while in 65% of all samples (eggs, muscle tissue, liver) of the screening study no DBDE was detected.
4. Some water birds (1 heron, 2 great crested grebes, 1 sea eagle) showed measurable DBDE concentrations, but in the majority of the water birds DBDE was not detected.
5. DBDE was detected neither in golden eagle, marsh harrier, osprey, or gannets (all from the UK), nor in cormorants from the Netherlands.
6. Terrestrial bird samples clearly showed higher DBDE concentrations than in those from water birds. This confirms the hypothesis that water presents a barrier to the transfer of DBDE from suspended particulate matter and sediment to aquatic organisms. Bioaccumulation of DBDE in aquatic food chains, apart from a possible uptake through the gut, is, therefore, highly unlikely.
7. The low DBDE concentrations in terrestrial birds show that DBDE is bioavailable to these birds, but bioaccumulation, if at all present, occurs only to a very limited extent.
8. A statistically significant increase of DBDE concentrations was found in UK peregrine falcon eggs from 1975 to 1995, as well as a statistically significant decrease from 1995 to 2001. More data on DBDE production and consumption in the UK are needed before drawing conclusions on possible correlations.. The sparrow hawk time trend did not show a significant trend (1973-2001), but some samples with higher concentrations were seen in later years.

Recommendations

1. In order to gain a better understanding of the transfer of DBDE through the terrestrial food chain into predatory birds, selected prey items from these food chains, and related soil samples, should be collected and analysed for DBDE.
2. For a prolonged time trend study in predatory birds, peregrine falcon samples (eggs or muscle tissue) are the preferred matrix. The DBDE concentrations in sparrowhawks are too low for this purpose.
3. It would be valuable to determine the DBDE concentrations in predatory birds from a number of other European countries, including birds from both industrialised areas and rural areas. There seems to be no need to analyse water birds from other countries.

1. Introduction

Regarding the possible occurrence of decabromodiphenylether (DBDE) in birds (Sellström et al., 2001), and the possible implications of that observation for the ongoing EU risk assessment of DBDE (Commission Regulation No. 1488/94 on risk assessment of existing substances), it was considered necessary to determine the concentrations of DBDE in various bird species, as well as to establish any time trends of DBDE concentrations in some of the species studied. The UK Environment Agency and BSEF agreed to organise such a study. The Centre for Ecology and Hydrology, Monks Wood, Huntingdon, UK (CEH), was able to provide bird samples from all over the UK from their established sample bank. These samples were sent to the Netherlands Institute for Fisheries Research (RIVO) and the Centre for Environment, Fisheries, and Aquaculture Sciences (CEFAS) for analysis. This report describes the results of this study.

Given the ongoing input of DBDE in the environment over recent years (de Boer et al., 2001, Boon et al., 2002, Vethaak et al., 2002), the recent report of the presence of DBDE in Swedish peregrine falcon eggs (Sellström et al., 2001), and a possible reduction of the inputs of DBDE due to stewardship programmes of the bromine industry in the future, it was considered desirable to obtain a better understanding of the current levels and any time trends of concentrations of DBDE in European birds. This information would be valuable for use in the final phase of the European risk assessment process of DBDE. Because of the known substantial consumption of DBDE in the UK, and the availability of many bird samples from various species sampled in the UK at CEH, it was proposed to use these samples to study the environmental occurrence of DBDE. Some cormorant samples from the Netherlands were added to the sample set, as well as a number of Swedish peregrine falcon eggs obtained from Stockholm University. Sellström et al. (2001) had reported the presence of DBDE in a number of these samples, and a sub-set of these peregrine falcon samples were re-analysed in this screening study.

In order to establish a time trend, some species from the screening study would be selected of which samples were available from different time periods and in which relatively high or at least measurable DBDE concentrations were found in the first part of the study (screening).

2. Objectives

The objectives of this study are the following:

- To determine DBDE concentrations in a large number of bird samples.
- To establish a time-trend of DBDE in peregrine falcons and sparrow hawks from at least five different time periods.
- To analyse HBCD diastereomers in the samples mentioned under 1.
- To report the results and statistical and ecological interpretation.

This report gives the results of the studies under objectives 1, 2 and 4. The HBCD concentrations will be presented in a separate report.

3. Analytical Requirements and Methods

3.1 Analytical requirements

The analysis of DBDE is difficult and its requirements different from the analyses of most other organohalogen contaminants. It requires good skills and experience, because a number of errors can easily be made which degrade data quality. To date, two international interlaboratory studies (ILS) have been organised (de Boer and Cofino, 2002). Both studies, which were coordinated by RIVO, showed that only a very small number of laboratories could be considered competent to carry out this analysis without making substantial errors. The first ILS showed between-lab standard deviations of 48-78% for sediment samples, whereas for biota these standard deviations were between 82 and >100%. The second exercise, finalised in 2002, showed even higher between-lab standard deviations. The obvious conclusion is that the majority of the laboratories working on DBDE do not have their analytical procedures under control.

A number of possible sources of error can be mentioned. DBDE is a very large molecule, which may not easily bioaccumulate because it may possibly only partly pass the membranes of aquatic organisms, and, due to its extremely low solubility in water (<0.1 ?g/L), it may not be able to migrate from the suspended particulate matter and sediment via the water to the organisms. Therefore, the concentrations in biota will be relatively low. Detection limits of 0.1-0.2 µg/kg wet weight may be needed to assess whether DBDE is present at relevant concentrations. Such sensitivity is difficult to obtain as the DBDE peak elutes relatively late in the chromatogram. GC/ECD analysis is normally not sensitive enough to reach the required detection limits. In addition, GC/ECD is not very specific. GC/MS analysis, using negative chemical ionisation, may provide enough sensitivity. However, a number of precautions must be taken: the GC column should be relatively short (≤ 15m) to reduce the residence time of DBDE as much as possible, as DBDE is sensitive to breakdown at higher temperatures. Also, the injection temperature should not be too high, and a pressure-pulse injection should be used to reduce the exposure to high injector temperatures in order to avoid degradation of DBDE. The analysis and quantification of DBDE analysis should occur separately from the analysis of the other PBDE's, as DBDE concentrations may differ considerably from those of the other congeners. The use of a short GC column for DBDE will also help to focus the peak, resulting in a better response. Incoming UV light in the laboratory must be blocked, as it may have an effect on the DBDE standard solutions and extracts due to photodegradation. Toluene or dichloromethane should be used as solvents, as DBDE may not be sufficiently soluble in other common solvents such as *iso*-octane. Further, DBDE extracts should never be evaporated to dryness, as even with toluene DBDE may not complete redissolve afterwards. This aspect is also relevant to the discussion regarding blanks. If DBDE is adsorbed to the inner walls of glassware and cannot be redissolved in the sample, it may remain to be desorbed into subsequent samples or blanks analysed using the same (though cleaned) glassware. False positive results can easily be obtained in this way if the appropriate precautions are not taken and rigorous protocols followed. This may explain several positive results from the literature. It is known that DBDE is present in dust (Leonards et al., 2001). Consequently, much attention has been paid to the blank analysis and all precautions intending to avoid the problems mentioned above were taken. In addition to these extra analytical efforts, there are of course normal good laboratory practice requirements, such as routine analytical quality control procedures including the use of internal laboratory reference materials, preparation of calibration curves, use of ^{13}C labelled internal standards, and participation in interlaboratory studies and proficiency schemes.

Over the past five years, RIVO and CEFAS have built up considerable experience in the analysis of DBDE. These laboratories have undertaken a large study for BSEF concerning PBDEs in aquatic biota (de Boer et al., 2001)), on studies for UK agencies on PBDEs in marine mammals (CEFAS), and in several studies for Dutch governmental organisations, such as the LOES project (National Study on Endocrine Disruptors) (RIVO) (de Boer et al., 2003). RIVO and CEFAS obtained good results in the two interlaboratory studies on PBDEs. RIVO is accredited for the analysis of DBDE, other PBDE congeners and HBCD, and is also certified under ISO 9001.

So as to demonstrate the comparability of the data produced by the two collaborating laboratories, a number of replicate samples were analysed in both laboratories. In this way a reliable impression of the analytical performance of the two laboratories was obtained.

3.2 Analytical methods

Method RIVO

Samples were weighed and extracted with a Soxhlet apparatus for six hours with hexane:acetone (3:1, v/v). ^{13}C labelled DBDE and BDE116 were added as internal standard to the Soxhlet extract, which was evaporated to 10 mL using a rotary evaporator. The extract was dried with sodium sulphate. For the lipid determination, an aliquot of 1 mL of the extract was taken, evaporated to dryness and weighed. The other 9 mL were evaporated to 1 mL and 1 mL dichloromethane (DCM) was added. Gel permeation chromatography (GPC) using a 600 mm x 25 mm PLgel column and DCM as elution solvent was used for the removal of lipids. The GPC fraction from 17 to 24 min. was collected and evaporated to 1 mL. The extract was cleaned with 1 mL of sulphuric acid. The organic layer was separated and evaporated to 1 mL. An additional clean-up step was carried out with silica gel using 12 mL isooctane and 25 mL isooctane:diethylether (85:15, v/v).

The final extract was evaporated to 200 µl, and analysed using coupled gas chromatography (GC) / mass spectrometry (MS) in the negative chemical ionisation mode (NCI). A 15 m (0.25 mm ID, 0.1 ?m film thickness) DB-5 column was used. A pulsed splitless injection (1 ?L) was used. All analyses were carried out under the specific conditions for the DBDE analysis as described above. Two blank samples, a procedural recovery sample, and one internal reference material (IRM) were analysed within each series of twelve samples. The quantification of DBDE was based on detection of the fragment ions at m/z 485 and 487, and concentrations were corrected for the recovery of ^{13}C labelled DBDE. The limit of quantification (LOQ) was set at the level of the lowest calibration standard. A correction for the procedural blank was made if the concentration in the sample was higher than two times the blank value. More details of the method can be found in de Boer et al. (2001) and Covaci et al. (2003).

Method CEFAS

Samples were weighed and extracted with a Soxhlet apparatus for six hours with hexane:acetone (1:1, v/v). ^{13}C labelled DBDE were added as internal standard to the Soxhlet extract, which was evaporated to 10 mL using a rotary evaporator. The extract was dried with sodium sulphate. For the lipid determination an aliquot of 1 mL of the extract was taken and evaporated to dryness and weighed. The other 9 mL were concentrated to 2 ml and transferred to the top of an alumina column (i.d. 6mm, 3g deactivated alumina (Merck, 70-230 mesh, 90 active neutral, no. 1077, topped with 1 cm anhydrous sodium sulphate). Two fractions were collected: 1) 2 mL *n*-hexane, 2) 12 mL *n*-hexane. The first fraction was concentrated to 1 ml and subjected to further fractionation on a 3 g 3% deactivated silica column (Merck, 70-230 mesh, Kieselgel 60, no. 7734). Two *n*-hexane fractions were collected, 1) 7mL, 2) 16 mL. The second fraction was combined with the second alumina fraction and the whole volume was reduced to 1 mL. The hexane in this fraction and the first silica fraction was replaced by *iso*-octane and both fractions were reduced in volume to 200 ?L. The first fraction contains DBDE, the second fractions the other PBDE congeners.

The final extract was analysed using coupled gas chromatography (GC) / mass spectrometry (MS) in the negative chemical ionisation mode (NCI). A 15 m (0.25 mm ID, 0.1 ?m film thickness) HP-1 column and a pulsed splitless injection (1 ?L) was used. All analyses were carried out under the specific conditions for the DBDE analysis as described above. Two blank samples, a procedural recovery sample, and one internal reference material (IRM) was analysed within each series of twelve samples. The quantification of DBDE was based on detection of the fragment ions at m/z 485 and 487, and concentrations were corrected for the recovery of ^{13}C labelled DBDE. The limit of quantification (LOQ) was set at the level of the lowest calibration standard. A correction for the procedural blank was made if the concentration in the sample was higher than two times the blank value. More details can be found in de Boer et al. (2001).

4. Samples

Samples were obtained from the specimen tissue bank of CEH after cataloguing. A selection of samples was carried out in cooperation with a bird specialist from Bureau Waardenburg, Culemborg, The Netherlands, Mr. Theo Boudewijn, who evaluated the suggestions of CEH on the suitability of the suggested bird species in relation to their feeding behaviour. Bureau Waardenburg is well known for their expertise in bird studies and environmental research. Bureau Waardenburg also provided a number of cormorant samples from the Netherlands for inclusion in the screening study. The final selection mainly consisted of predatory birds and water birds. The driving force for this selection was the report on the presence of DBDE in Swedish peregrine falcons (Sellström et al., 2001). It was considered to be interesting and of importance to see whether other terrestrial predatory birds would contain similar DBDE concentrations. A number of peregrine falcon samples were included in this study. Also, ten peregrine falcon eggs from the Swedish study were obtained for re-analysis for confirmatory purposes, as well as ten new Swedish peregrine falcon egg samples. In addition, it was thought that it would be interesting to study the possible bioaccumulation of DBDE in water birds, as bioaccumulation of DBDE in fish has not been confirmed as yet. Some of the bird species selected have a fish diet (osprey, cormorant, great crested grebe, heron, gannet) and others have a mammal/bird/reptile diet (red kite, barn owl, golden eagle, kestrel, peregrine falcon, sparrowhawk, Montagu's harrier, marsh harrier). It was decided to include three different types of tissues in the study: liver, muscle and egg (white plus yolk), as these were the most likely compartments in which DBDE could be expected to be found. Table 1 shows the samples that were made available for the screening study. All samples are from the period 2000-2002. Details are given in Annex 1.

Table 1. Overview of samples analysed in screening study

Species	Tissue	Country	Number
Peregrine Falcon	Egg	Sweden	20
Peregrine Falcon	Egg	UK	11
	Muscle tissue	UK	5
	liver	UK	5
Sparrow hawk	Muscle tissue	UK	5
	Liver	UK	4
	Egg	UK	5
Kestrel	Muscle tissue	UK	4
	Liver	UK	3
Great crested grebe	Muscle tissue	UK	3
	Liver	UK	4
Barn owl	Muscle tissue	UK	4
	Liver	UK	5
	Egg	UK	4
Montagu's harrier	Egg	UK	4
Marsh harrier	Egg	UK	2
Gannett	Egg	UK	12
Sea eagle	Egg	UK	1
Merlin	Egg	UK	2
Red kite	Egg	UK	2
Heron	Muscle tissue	UK	5
	Liver	UK	4
Cormorant	Muscle tissue	The Netherlands	2
	Liver	The Netherlands	3
	Egg	The Netherlands	5
TOTAL			124

Based on the results of the screening study (see chapter 5) peregrine falcon eggs and sparrowhawk muscle tissue samples were selected for the time trend study. Table 2 gives an overview of the samples selected for the time trend study. Details are given in Annex 2.

Table 2. Samples analysed in the time trend study

Species	Periods	Number of individuals
Peregine falcon eggs	1973-2002	48
Sparrowhawk muscle	1975-2001	64
Exchange of samples RIVO/CEFAS		6 (2 of each species)
TOTAL		**118**

5. Results and discussion

5.1 Screening study

All the results of the screening study are given in Annex 3, and a summary is given in Table 3. A number of initial observations can be made on the results. Firstly, the data analysis was hampered by the fact that in 65% of the samples DBDE was non-detectable with multiple detection limits (0.08-20 ?g/kg wet weight) scattered across the entire measured concentration range of 0.08-32??g/kg wet weight.

Secondly, the Swedish results have been confirmed. Table 4 shows the results of the re-analysis by RIVO of ten Swedish peregrine falcon egg samples. The correspondence with the original Swedish data is excellent. This result shows not only that the Swedish peregrine egg samples do indeed contain DBDE, but also that the analyses undertaken by RIVO and Stockholm University are comparable. Only one sample from a sample taken in 1999 shows a strong disagreement: 2.4 ?g/kg ww as analysed by the Stockholm University and 32 ?g/kg, as analysed by RIVO. The latter value is most likely to be in error as the result of sample contamination. All the other results are in agreement to within ca. 30% or less from each other. At this relatively low DBDE level, and given the high degree of difficulty of the DBDE analysis, this may be considered a very good analytical result.

In addition to the re-analysed Swedish peregrine egg samples, ten other peregrine falcon egg samples from Sweden were obtained (Annex 3a). These eggs had not been analysed previously. The results are in the same range as those of the re-analysed samples: between non-detectable and 16 ?g/kg ww for the new samples and between non-detectable and 19 ?g/kg for the re-analysed samples. This is another confirmation of the presence of DBDE in Swedish peregrine falcon eggs, although in this second sample set 6 out of 10 samples resulted in non-detects (3 non-detects in the set of re-analysed samples).

Furthermore, it has been confirmed that the peregrine falcons from the UK contain DBDE, although at lower concentrations than those found in the Swedish egg samples. The UK peregrine falcons show the presence of DBDE not only in their eggs, but also in their liver and muscle tissue. Liver and muscle tissue of the Swedish peregrine falcons were not available for analysis. The eggs actually showed more non-detects than either the muscle tissue or the liver samples. The DBDE level in the Swedish peregrine falcon eggs seem to be ca. 3-4-fold higher than in the peregrine falcon eggs from the UK. However, the sample number is low as there are only two positive samples from the UK and the natural variation is thought to be high.

All sparrowhawk muscle tissue samples from the UK contained DBDE. Sparrowhawks are mammal eating birds, as are peregrine falcons. The mean concentrations on a lipid weight basis (101 ?g/kg) were comparable to those in peregrine falcon muscle tissue also from the UK (137 ?g/kg), but on a wet weight basis the DBDE concentrations were 4-fold lower in the sparrowhawks (1.1 – 4.0 ?g/kg). In sparrowhawk liver no DBDE was found, and two out of five egg samples showed non-detectable levels of DBDE.

The DBDE concentrations found in most of the other bird species were relatively low, in many cases just above the detection limits. A large number of non-detects was also found. One kestrel liver sample contained 5.5 ?g/kg DBDE on a wet weight basis, but 4 out of 5 muscle tissue samples and 3 out of 5 liver samples resulted in non-detects (Annex 3c). Great crested grebe samples showed non-detects, apart for one muscle tissue and one liver sample (Annex 3d). Barn owls contain low concentrations of DBDE (liver, muscle tissue and eggs), but also a substantial number of non-detects was found (2 out of 5 liver samples, 4 out of 5 muscle tissue samples and 1 out of 4 egg samples (Annex 3e).

All heron samples (liver and muscle tissue) analysed were negative apart from one muscle tissue that contained 4.5 ?g/kg on a wet weight basis (Annex 3f). All gannet samples were negative (12 samples), as well as all cormorant samples from The Netherlands (Annex 3g and 3h). Golden eagle egg, osprey egg and marsh harrier eggs were all negative, but in 3 out of 4

Table 3. DBDE concentrations in predatory birds from the UK.

Species, Country	Tissue type	No. of positive samples/ Total no. of samples	Concentration range	
			?g/kg wet weight	?g/kg lipid
Peregrine falcon UK	Liver	4/6	<0.17-6.7	<5.7-181
	Muscle	5/5	1.8-9.5 (mean 4.0)	53-344 (mean 137)
	Egg	2/6	<0.08-7.5	<1.8-108
Per. falcon Sweden	Egg	11/20	<0.3-21	<4-412
Sparrowhawk UK	Liver	0/4	<3.2-<9.8	<82-<200
	Muscle	5/5	0.26-2.2 (mean 1.1)	13-275 (mean 101)
	Egg	3/5	<0.16-1.5	<2.1-38
Kestrel UK	Liver	2/5	<0.26-5.5	<5.8-120
	Muscle	1/5	<0.11-0.29	<4.2-10
Great crested grebe	Liver	1/4	<0.11-0.52	<1.5-9.1
UK	Muscle	1/3	<0.4-1.2	<8.1-31
Barn owl UK	Liver	3/5	<0.13-2.5	<2.6-37
	Muscle	1/5	<0.5-1.2	<6.3-14
	Egg	3/4	<2-1.7	<20-30
Heron UK	Liver	0/4	<0.08-<0.25	<2.3-<5.7
	Muscle	1/5	<0.32-4.5	<6.3-563
Gannett UK	Egg	0/12	<0.2-<2.2	<4-<57
Cormorant	Liver	0/4	<0.2-<1.2	<7-<36
(The Netherlands)	Muscle	0/2	<0.4	<24-<25
	Egg	0/5	<0.2-<2.2	<4-<31
Red Kite UK	Egg	1/4	<0.09-2.3	<2.1-29.1
Montagu's harrier UK	Egg	3/4	<0.12-1.3	<2.1-28
Marsh harrier UK	Egg	0/2	<0.09	<2-<2.4
Sea eagle UK	Egg	1/1	0.48	6.2
Osprey UK	Egg	0/3	<0.2-<1.5	<4-<27
Golden eagle UK	Egg	0/5	<0.2	<4.1
Merlin UK	Egg	1/2	<3.8-0.3	<43-4.3

Table 4. Re-analysis of peregrine falcon egg samples from Sweden.

Sampling year	Original concentration (Sellström et al., 2001)		Concentration determined on re-analysis (RIVO, 2003)	
	?g/kg wet weight	?g/kg lipid	?g/kg wet weight	?g/kg lipid
1988	<0.7	<8	<0.3	<4
1990	14	210	19	229
1995	1.7	28	1.2	18
1996	20	430	21	412
1998	0.46	8.6	<0.4	<9
1999	5.0	83	4.5	79
1999	11	370	9	155
1999	1.3	28	<1	<19
1999	2.4	46	*32*	*485*
1999	9.7	170	13	197

Montagu's harrier eggs DBDE was found at measurable concentrations (0.15–1.3 ?g/kg). One out of 4 red kite eggs contained DBDE (2.3 ?g/kg wet weight, as well as one merlin egg out of two (0.3 ?g/kg wet w.) and one sea eagle egg (0.48 ?g/kg wet w.). The DBDE concentrations found should be regarded as trace concentrations. It is not clear whether the low concentrations observed in the birds studied is because uptake is limited by the physical properties of DBDE or by the ability of the test species to eliminate DBDE. A more detailed study of relevant food chains should give additional evidence, which will allow these possible explanations to be assessed.

It is remarkable that the positive samples, apart from an occasional exception, occurred mainly in terrestrial predatory birds. Most water bird samples showed no detectable DBDE concentrations. This confirms the hypothesis that DBDE does not bioaccumulate significantly in aquatic organisms, presumably mainly because the water acts as a barrier in the transport of DBDE from suspended particulate matter and sediments via the gills to as fish. DBDE uptake in aquatic organisms through their food may still be an alternative uptake route, but seems to contribute only marginally to concentrations in predators. Other PBDEs, such as tetra and pentaBDE congeners, which have higher water solubility, have been found in fish, sometimes at high concentrations (de Boer et al., 2001). It is, though, apparent that DBDE can enter terrestrial organisms and rises up the food chains to predatory birds, although at low levels. It is likely that DBDE in dust and other fine particles are taken up by small terrestrial animals, which are preys of small birds, which are subsequently prey of the predatory birds studied.

Due to the combination of low DBDE concentrations and a relatively high natural variation of contaminant levels in predatory birds it is difficult to make a differentiation according to the geographical origin of the samples. There is a slight tendency towards higher DBDE concentrations in birds from more densely populated and industrialised areas, such as North of London, Kent, Cardiff/Bristol area and Mersey area (Annexes 3a and 3b). More samples would be needed to draw more firm conclusions.

Also no differentiation with respect to age (adult/juvenile) could be made. This was the conclusion of Vangheluwe and Verdonck (2003) who carried out a statistical analysis of the data derived from this study. No significant differences ($p < 0.05$) in DBDE concentration were observed between adults and juveniles (based on a two sample Kolmogorov Smirnov test).

The influence of the sex (male or female) was also explored. There were no data available for eggs. Again, no significant differences ($p < 0.05$) in DecaBDE concentration were observed between males and females (based on a two sample Kolmogorov Smirnov test) (Vangheluwe and Verdonck, 2003).

The highest DBDE concentrations were found in peregrine falcon egg, liver and muscle tissue samples, followed by sparrow hawk muscle tissue samples. Because a relatively large number of peregrine falcon egg samples and sparrow hawk muscle tissue was available from different time periods, these two matrices have been selected for the time trend study.

5.2. Time trend study

All the results of the time trend study are given in Annex 4. Figures 1 and 2 show the graphical plot of the data for sparrowhawk muscle tissue and peregrine falcon eggs, respectively. Again a large number of non-detects was found. According to Vangheluwe and Verdonck (2003) and Hoogerbrugge (2000) values corresponding to 50% of the quantification limits were assigned to non-detects in order to obtain these trend graphs.
The quality of the samples was not always very good. Several of the older samples seemed to be dehydrated and several egg samples were actually almost full-grown chickens instead of yolks. The possible dehydration of some samples may have caused higher DBDE values. The effect may be somewhat limited, as most of the older samples did not contain DBDE at measurable levels.

Both figures show a large number of non-detects in the period before 1990, although a limited number of positive samples were found for peregrine falcon eggs sampled during earlier periods. Figure 1 shows no significant trend for DBDE in sparrowhawks ($p<0.05$). However, it can be seen that a larger number of positive samples were found in later years, particularly in the period 1995-2001. Figure 2 (peregrine falcon eggs) is somewhat different. In these samples also most of the positive samples were found in the period 1995-2001, but the results from 1995 are significantly higher than those from other years. It may be that there is an association between this peak value and DBDE consumption and/or production in the UK but more data on production and consumption are needed to draw conclusions.

The different patterns in the two figures also make it difficult to predict future trends. However, based on the higher number of positive samples, it seems fruitful to follow the time trend for a longer period, especially in peregrine falcons, as these birds appear to show the highest concentrations and so yield the more robust statistical treatment.

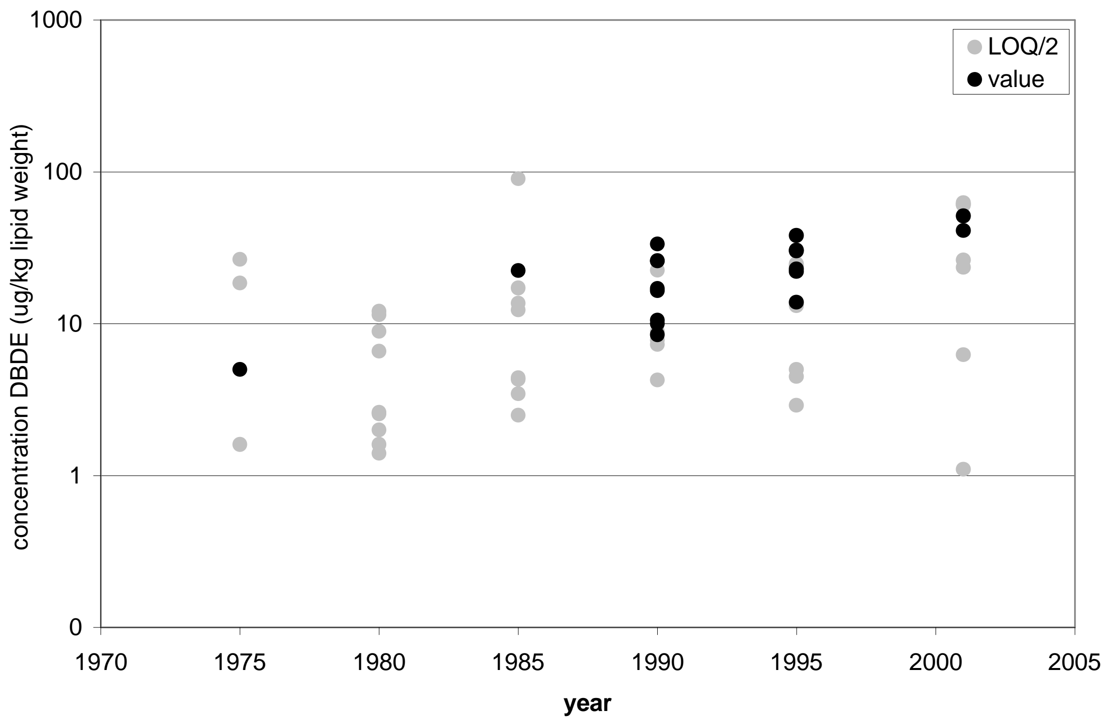

Figure 1. Concentration of DBDE on lipid weight basis in sparrow hawk muscle from the UK for the period of 1975 to 2001. Grey points are limit of quantification (LOQ) divided by two, and black points are values above LOQ.

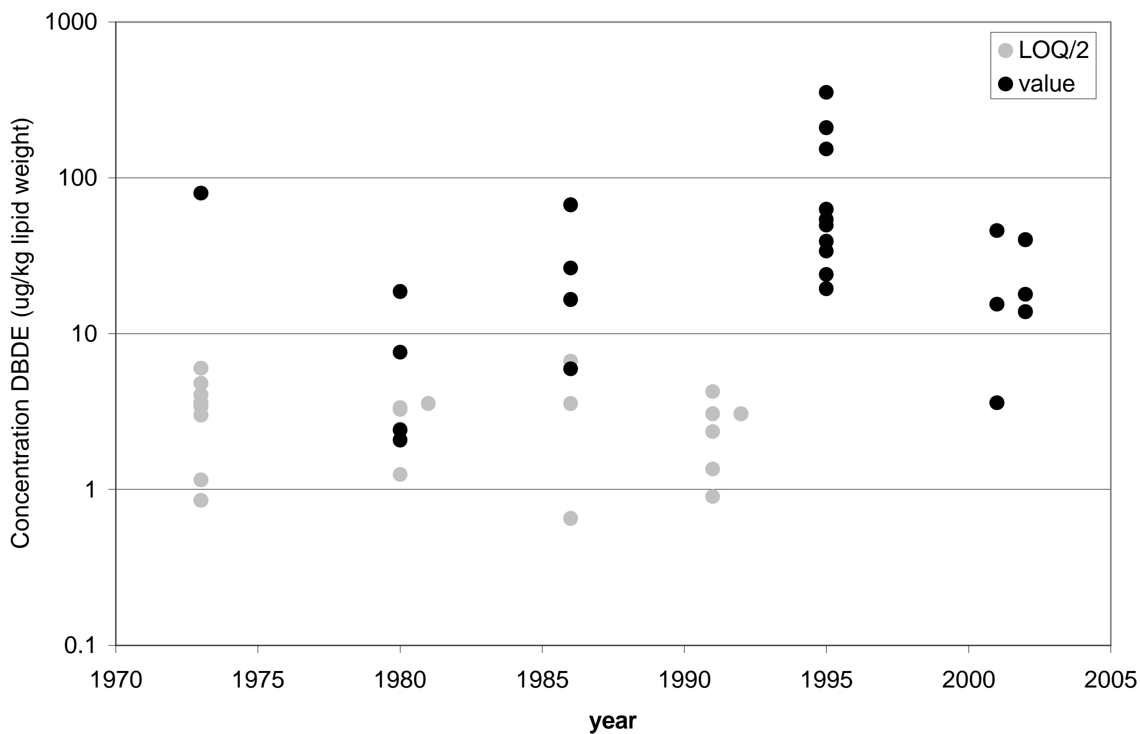

Figure 2. Concentration of DBDE on lipid weight basis in peregrine falcon eggs from the UK for the period 1973 to 2002. Grey points are limit of quantification (LOQ) divided by two, and black points are values above LOQ.

Annex 5 shows the results of the comparison between RIVO and CEFAS. In fact only one deviating result has been found: annex 5b, CEH LSN 11797 – sparrowhawk muscle. The exceptional high value found by one of the two laboratories can most likely be explained by interference. This outlying result has not been used in figure 1. The remaining results of this comparison, as well as the results of the comparison between RIVO and Stockholm University (Table 4) show that the results reported are very comparable. Also, the lipid contents are in good agreement.

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