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Environmental specimen banking. Herring gull eggs and breams as bioindicators for monitoring long-term and spatial trends of chlorinated hydrocarbons*

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Abstract: The German Environmental Specimen Bank (GESB) has been an important tool in contaminant monitoring and ecotoxicological research for more than one decade. The annual monitoring program provides a long-term database to determine trends in contaminant burdens in biota samples. Results of ten years of experience in the determination of chlorinated hydrocarbons (CHCs) in herring gull eggs collected in the North Sea and the Baltic Sea are presented. In addition, the study reports CHC concentrations in bream livers from the River Elbe between 1993–1997. Most of the compounds studied show a significant decline over time, especially in samples from East Germany. For specimens taken in West Germany, only small decreases or no changes of CHC levels were observed

INTRODUCTION

In 1985, the decision was made to establish a German Environmental Specimen Bank (GESB) as a permanent institution for the systematic collection, characterization, and storage of representative environmental samples from marine, limnic, and terrestrial ecosystems [1–3]. Specimen banking has made progress, and environmental specimens have been collected and stored continuously for the purpose of spatial, real-time, and retrospective monitoring as well as for ecotoxicological research.

Representative areas within typical ecosystems in Germany have been selected as sampling sites, for instance, North Sea, Baltic Sea, River Elbe, River Rhine, and urban-industrialized regions such as Saarland and the area around Leipzig-Halle-Bitterfeld. The representative matrices are taken annually according to stringent sampling protocols, mainly biological materials such as tree leaves and shoots, earthworms, bird eggs, mussels, fish tissues, and algae. The environmental specimens are deep frozen above liquid nitrogen on site directly after collection, all steps of sample pretreatment are carried out at cyrogenic temperatures, thus minimizing losses of volatile compounds and avoiding sample degradation due to thawing and refreezing. After grinding, homogenizing and dividing in samples of specific size most of the powder subsamples are stored at cryogenic storage conditions ($T = < -150$ °C) to insure sample stability over relatively long periods. Some of the annual homogenates are used for real-time monitoring of known environmental contaminants [metals, metal species, chlorinated hydrocarbons (CHCs), polycyclic aromatic hydrocarbons (PAHs)]. The whole GESB process is standardized and described in standard operating procedures (SOPs) [4]. The GESB monitoring activity is designed

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to quantify the current status and long-term temporal and spatial trends of key contaminant concentration.

Selected recent GESB results about CHC concentrations in the aquatic environment demonstrate that GESB is an effective instrument for the long-term observation of environmental pollution.

MATERIALS AND METHODS

The further discussion is restricted to CHC contamination of herring gull eggs and breams. All samples are taken, prepared, stored, and analyzed according to the SOPs of the GESB [4].

Sampling and sample preparation of herring gull eggs

Fish-eating birds are at the top of the aquatic food chain and can be used to assess chemical contamination in the marine environment. Seabird eggs have earlier been shown to be a very useful monitoring matrix [5–7]. The GESB contains a large number of egg samples which have been collected because they are a relatively non-invasive method of testing the population and are readily accessible in well established colonies. The herring gull *(Larus argentatus)* eggs were collected from the breeding colony on the islands of the North Sea and Baltic Sea (Fig. 1). The islands are located in the estuaries of the River Weser (Island Mellum), River Elbe (Island Trischen), and River Oder (Island Ruegen, sanctuary Heuwiese). With the exception of Heuwiese the egg samples have been taken every May since 1988. To avoid variations in CHC concentration the second egg laid was taken and characterized biometrically (e.g., egg length, diameter, and weight). After transport in the refrigerator at + 4 °C to the GESB facility in Juelich, the egg content was blown out under clean room conditions and homogenized to an annual pooled sample of each sampling locality.

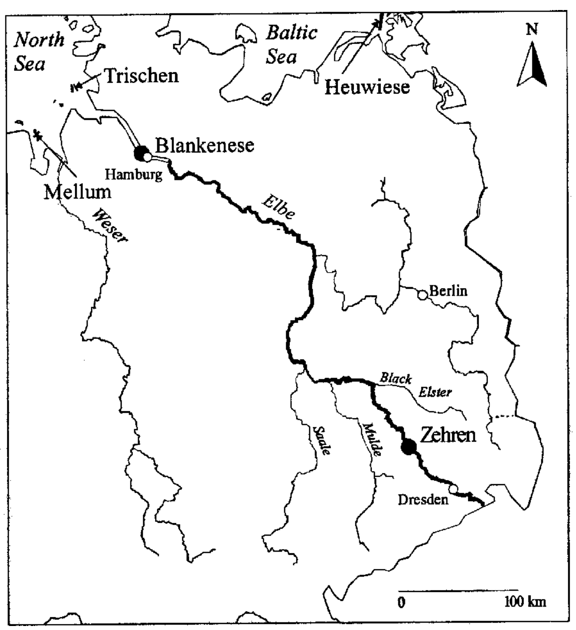

Fig. 1 Sampling locations of herring gull eggs and breams.

Sampling and sample preparation of breams

Breams (*Abramis brama*) are freshwater fish with a small migration radius. They are an ideal organism for monitoring freshwater and sediment contamination because their feeding habits are characterized by permanent direct contact with the sediments [8]. The collection of breams at the sampling sites of the

River Elbe presented here started in 1993 (Fig. 1). During the last two decades the River Elbe has been one of the most polluted rivers in Europe [9]. The river has been strongly influenced by municipal and industrial emissions of the chlorine industry. German reunification in 1990 and political changes in the eastern part of Europe resulted in factory closures and industrial restructuring. Sampling of fish in the River Elbe gives us the opportunity of monitoring the undergoing remediation of the freshwater ecosystem with respect to relevant toxic CHC. The sampling site Blankenese (river km 632,5) is located downstream of the harbor at Hamburg. The input of pollutants from the River Elbe into the North Sea is supposed to be reflected by the contamination of fish caught at Blankenese. Breams from the sampling station Zehren (river km 93) are used to characterize the influence of anthropogenic inputs from the Dresden-Leipzig area.

It is known that concentrations of contaminants in fish show seasonal variations. Therefore, the bream samples were collected annually after the breeding season (July–October). To obtain annual samples from homogenous and comparable bream population, only breams in the range of 8–12 years of age were analyzed. The killed and dead fish were dissected under clean-room conditions at the collection site. Muscle, liver, and kidney were deep-frozen on site in the gaseous phase over liquid nitrogen (–150 °C). Fish specimen were characterized biometrically by age (from scale and otolithes), sex, weight, and length. The homogenized bream livers were analyzed.

Chemical analysis of CHC

For the CHC analysis the powdered subsamples were mixed with sodium sulfate/sea sand and extracted with *n*-hexane/acetone (1:1) in an extraction column. Size-exclusion chromatography (SEC) and silica gel clean-up by high-performance liquid chromatography (HPLC) were used to separate lipids and other biogenic materials from the fraction containing the CHC. The CHC fraction was analyzed by capillary column gas chromatography (GC) equipped with an electron capture detector (ECD). More details about the method are given elsewhere [10].

The following compounds were determined: Pentachlorobenzene (PCBz), Hexachlorobenzene (HCB), α-,β-,γ-Hexachlorocyclohexane (HCH), Heptachlor (HC), Heptachlorepoxide (HE), Aldrin, Dieldrin, o,p'-DDT, p,p'-DDT, p,p'-DDE, p,p'-DDD, Octachlorostyrene (OCS), polychlorinated bi-phenyls (PCB) (B28, B52, B101, B138, B153, B180). Since 1992 the pure PCB congeners have been quantified.

All analytical results are expressed on a fat weight basis. The fat percentage of the matrices is given in Table 1, allowing concentrations to be converted to a fresh basis.

Table 1 Fat content of herring gull eggs and bream liver samples.

	Herring gull eggs			Bream livers	
Year	Mellum Fat %	Trischen Fat %	Heuwiese Fat %	Zehren Fat %	Blankenese Fat %
1988	8.2	9.3			
1989	10.7	8.9			
1990	9.4	9.3			
1991	9.6	10.0	9.4		
1992	9.2	9.2			
1993	9.3	9.4	8.9	5.5	5.1
1994	10.3	10.1		4.6	5.1
1995	9.6	9.2		6.1	5.4
1996	9.3	9.1	8.4	6.6	4.1
1997	9.5	9.4		6.0	4.9
1998	12.7	9.1	11.6		

RESULTS AND DISCUSSION

The different economic and technical development of the former two German states [East Germany (GDR) and West Germany (FRG)] led to a partly different pattern of environmental pollution. Due to economical reasons the production and application of low volatile CHC played a more important role in the chemical pest control in agriculture and forestry of GDR than in the FRG [11]. This report describes regional variation of the CHC contamination in GDR and FRG and attempts to recognize temporal trends in the environmental pollution of both German parts. The discussion is predominately focused on the major organochlorine contaminants present in the samples analyzed: HCB, OCS, p,p'-DDE, p,p'-DDD and the PCB congeners (B52, B138, B153). More information can be found in the annual reports of the GESB [12]. HCB and OCS are mainly generated as by-products of chlorine production and combustion processes. p,p'-DDE is the major metabolite of DDT, whereas p,p'-DDD is a degradation product of DDT by anaerobic conversion as well as a by-product of the DDT synthesis. PCB comprise a class of 209 individual congener compounds. They have been used, e.g., as electric fluids in transformers, hydraulic lubricants, and flame retardants. Relatively large amounts were released due to inappropriate disposal practices and leakages from industrial facilities.

Herring gull eggs

Comparisons of the DDE, HCB, and B138 concentrations in herring gull eggs from typical marine ecosystems in former GDR and FRG over the last decade are presented in Figs. 2–4. The DDE levels in specimens from the Baltic Sea clearly demonstrate the importance of DDT in the former GDR (Fig. 2). DDE values in eggs sampled in Heuwiese in 1993 were ten times higher compared to specimens from the North Sea. Recent data (1996–1998) show a significant decline of the DDE content in eggs from Heuwiese by a factor of 3.8, but the levels in samples from the Baltic Sea are still two times higher than the average concentration found in species from Trischen. Due to the influence of the River Elbe, concentration of DDE in Trischen is higher during the entire study period than those reported for Mellum. With the exception of 1989, when higher concentrations have been measured, the DDE levels in Trischen remained practically constant. A similar trend has been found for Mellum. The DDE data from this location show a decreasing trend over the last two years. Additional analyses of egg specimens are

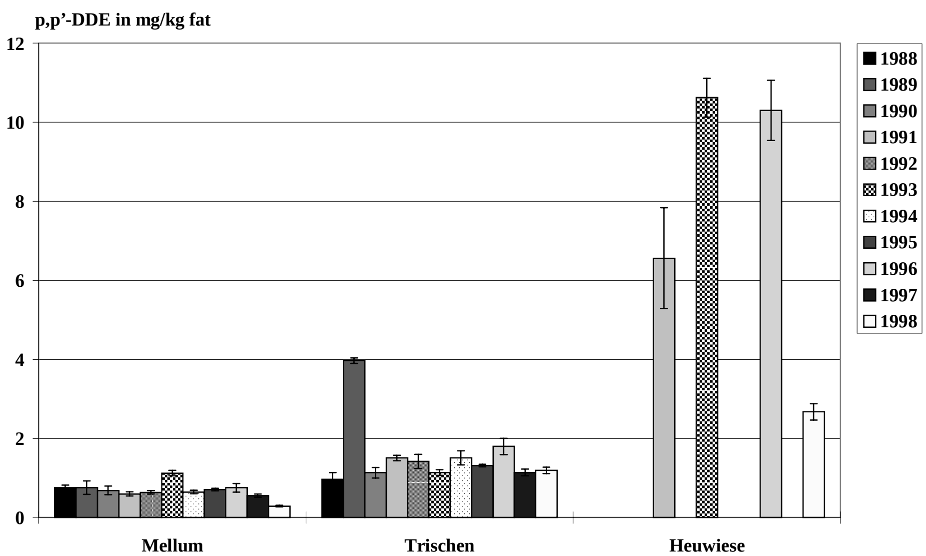

Fig. 2 Mean concentrations of p,p'-DDE in homogenates of herring gull eggs from the North and Baltic Seas.

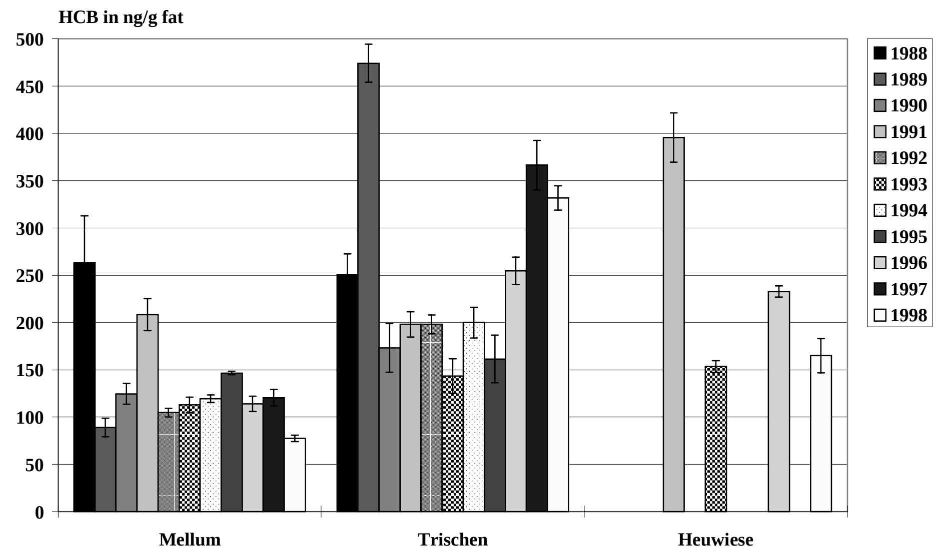

Fig. 3 Mean concentrations of HCB in homogenates of herring gull eggs from the North and Baltic Seas.

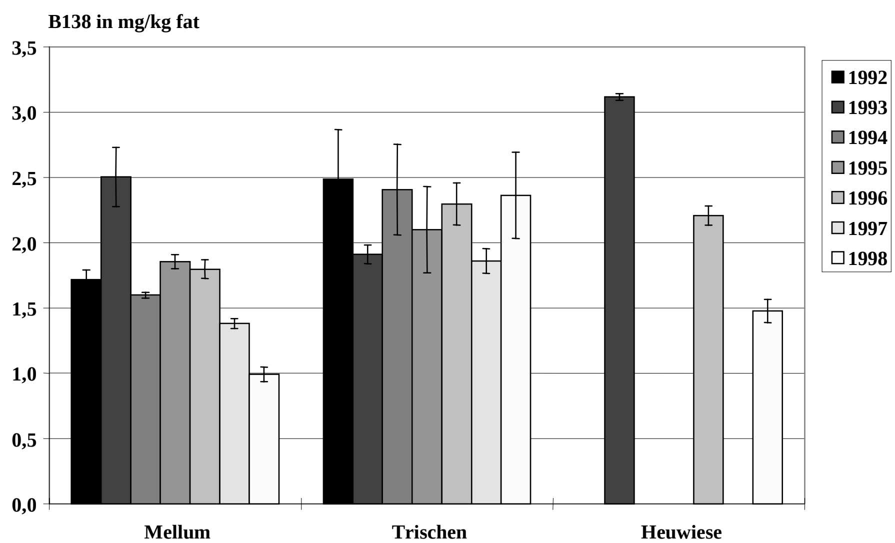

Fig. 4 Mean concentrations of B138 in homogenates of herring gull eggs from the North and Baltic Seas.

necessary to determine if this observation indicates the onset of a further decline.

The highest HCB concentrations in herring gull eggs were found in Trischen, which is consistent with relatively high concentrations of HCB being found in the River Elbe (Fig. 5). In contrast to the other sampling sites an increase of HCB has been measured since 1996. B138 data decline significantly in Mellum and Heuwiese over the study period, except in eggs from Trischen, where the PCB contamination remained practically constant (Fig. 4).

Bream livers

Figures 5–7 compare the data from 1993–1997 obtained from the analysis of bream livers from the River Elbe sampled in Zehren and Blankenese. The CHC concentrations in breams from Blankenese were substantially lower than concentrations found in fish collected in Zehren. Differences in contamination levels may be explained by the considerable pollution of the River Elbe by industrialized areas and leaching dumps of the former CSFR [13] as well as industrial activities in the area of Dresden, Coswig, and Meissen.

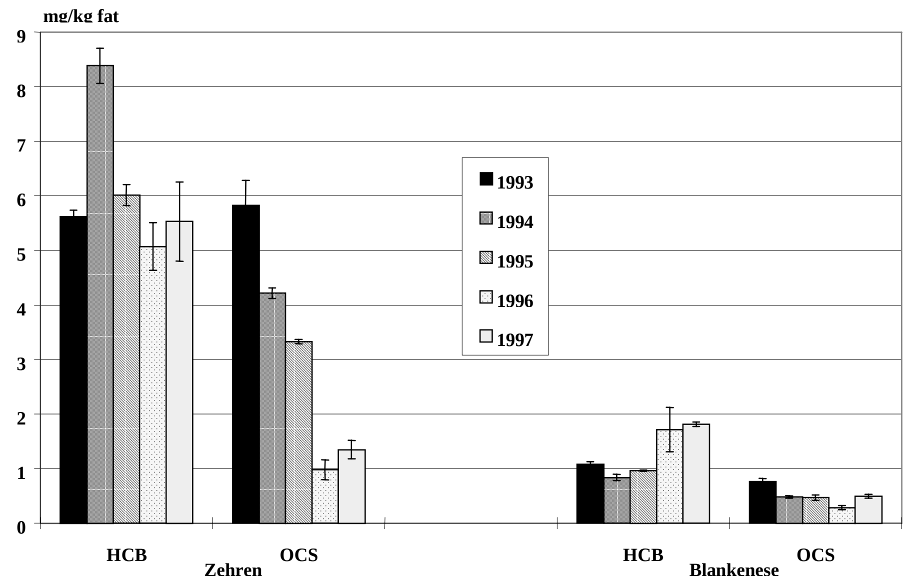

Fig. 5 Mean concentrations of HCB and OCS in homogenates of bream livers from the River Elbe.

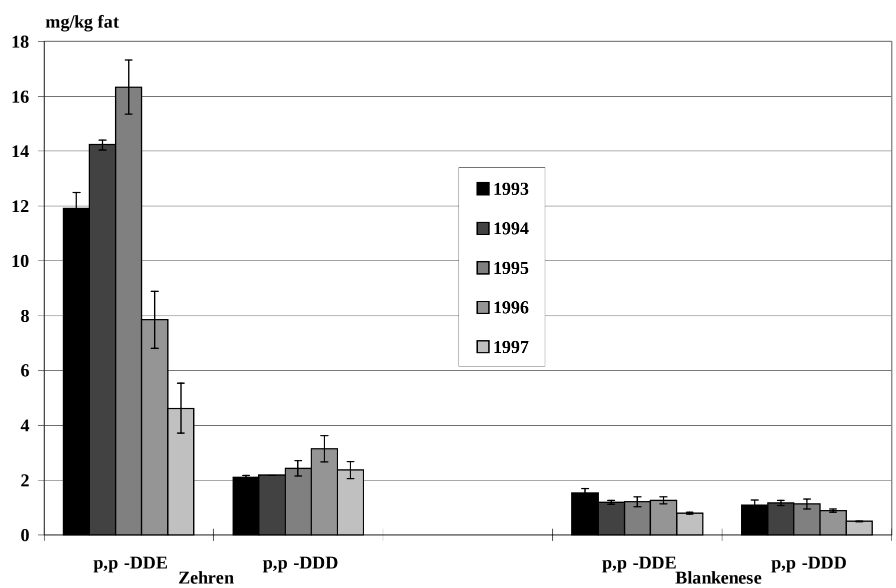

Fig. 6 Mean concentrations of p,p’-DDE and p,p’-DDD in homogenates of bream livers from the River Elbe.

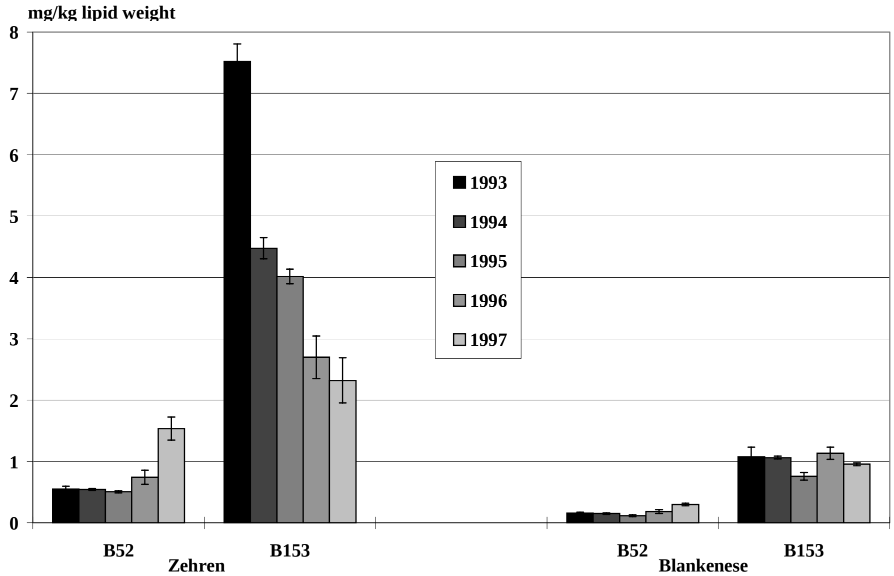

Fig. 7 Mean concentrations of B52 and B153 in homogenates of bream livers from the River Elbe.

DDD constituted higher percentages of DDT-related compounds in fish than in herring gull eggs. This confirms findings by Ruus *et al*. [14], who reported on an increasing proportion of DDE, together with decreasing proportions of DDD from fish to marine mammals in food chain studies. At the beginning of time series, the DDE/DDD ratio in Zehren was significantly higher (1994:6.5) than in Blankenese (1994:1.0) (Fig. 6). Recent data show a considerable decrease of the DDE/DDD ratio in Zehren (1997:2.0), which can be attributed to the rapid decrease of DDE at this station, whereas DDD remained constant over the study period. B153 and OCS data in breams from Zehren followed a similar fast decrease indicating that the CHC input from the Czech Republic into the River Elbe has been reduced significantly during the last years. In contrast to these observations the DDT-metabolites, OCS, and B153 levels in livers from Blankenese exhibited only small decreases or were within the same range as at the beginning of the study. For HCB a different temporal trend was observed at both sampling sites (Fig. 5). With the exception of 1994, when higher concentrations have been measured, the HCB levels in Zehren remained practically constant. Recent HCB results in fish from Blankenese showed an increasing trend, which is consistent with higher concentrations of HCB being found in herring gull eggs from Trischen (Fig. 3). An increase was also measured for B52 at both stations. Further research is needed to determine whether this trend will continue.

CONCLUSION

Apart from sample archivation, the GESB has proved to be an effective instrument for providing a long-term database to determine temporal and spatial trends in contaminant burden in the environment. Results based on the annual chemical analysis of aquatic samples have shown that concentrations of DDE, PCB (B138, B153), and OCS have declined significantly in specimens from former GDR (Heuwiese, Zehren) during the study period. The results demonstrate that measures to protect the environment have had a positive and measurable effect. However, concentrations of DDT-metabolites are still the dominant pollutants in former GDR. In samples collected in FRG the declining tendency is less obvious showing only small decreases or similar levels when compared to corresponding data found at the beginning of the study.

Recent data of HCB and B52 show an increase in species sampled in the River Elbe or Trischen located in the esturary of this river. Further investigations must be done to evaluate this time trend. The presented results clearly demonstrate the necessity of having environmental samples that are properly collected and stored. As the database continues to grow, it will provide a valuable resource for the assessment of the environmental pollution in Germany.

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