ICES CM 2000/S:01

Temporal trends in malformations of pelagic fish embryos from the southern North Sea in relation to anthropogenic xenobiotics

H. von Westernhagen*, V. Dethlefsen and M. Haarich*****

** Alfred-Wegener-Institute for Marine- and Polar Research - Biologische Anstalt Helgoland - Columbusstrasse, 27568 Bremerhaven, Germany*
*** Institut für Fischereiökologie, Bundesforschungsanstalt für Fischerei, Deichstrasse 12, 27427 Cuxhaven, Germany*
**** Institut für Fischereiökologie, Bundesforschungsanstalt für Fischerei, Wüstland 2, 22589 Hamburg, Germany*

The trends of malformation prevalence in embryos of dab, *Limanda limanda*, in the southern North Sea after the year 1990 mirrored the drop in major pollutants in the rivers draining into the German Bight. Despite this general decline we detected a pollution event in the southern North Sea in winter 1995/1996 employing the prevalence of malformations in pelagic dab embryos as an indicator. An abrupt rise in malformation prevalence in the embryos of dab, corresponded to a dramatic increase in DDT levels in parent fish from the same area, indicating a hitherto unnoticed introduction of considerable quantities of DDT into the system.

Keywords: biological effects monitoring; embryo malformations; dab, *Limanda limanda*; German Bight.

Since pollution has been recognized as a major threat to the marine environment, national and international programs have been initiated to monitor its development, with the objective to reduce the input of xenobiotic substances into the marine ecosystem (North Sea Task Force, 1993). With this objective in mind, the 'Monitoring-Programme' of the North Sea commenced in 1974, and started with the determination of xenobiotics in water and sediment. It was soon supplemented by the identification of these substances in biological matrices. With the introduction of the concept of 'biological effects monitoring' (see ICES 1978, MacIntyre and Pearce, 1980) there began a new dimension in thinking on pollution abatement (Stebbing *et al.*, 1992; Stagg, 1998; den Besten, 1998), using a more direct approach by looking for effects on living organisms in the field.

Criteria (endpoints) for assessing the impact of human activities have been selected on the basis of their relevance to the individual, the population or the species. There still is, however, little consensus on appropriate endpoints for assessing impacts on ecosystems or communities. In the absence of *a priori* specification of ecosystem effects, a wide variety of potentially relevant ecological variables (bioindicators) are typically measured in environmental monitoring programs. This approach introduces difficulties from the standpoint of ecosystem protection or management, since routine monitoring of a large suite of ecological state variables can be a formidable task and a considerable strain on available resources. Practical constraints on the expenditure of time, effort, and money often dictate a sampling scheme, which allows routine monitoring of only part of the desired and relevant parameters. Ideally, the desired result is a small subset of easily measured variables, which would provide early, adequate warning of ecosystem and/or population damage. From the point of view of decision-makers, the most adequate number of variables would probably be only one, since this would preclude any contradictory interpretation. On the other hand that single indicator should preferably have an obviously outstanding significance for the ecosystem, that allows it to be accepted without hesitation by all parties concerned as an important member or part of the system.

Embryo malformations in the North Sea

During the life cycle of a fish the embryonic and the larval phase are usually considered the most sensitive (Westernhagen, 1988). During these phases pollutants as well as adverse natural factors may exert the strongest effects on the survival of an individual but less so during the adult phase (Johnson & Landahl, 1994). Therefore the regional occurrence and abundance of malformations (malformation frequency) in pelagic fish embryos were used as a reflection of disturbances in the marine ecosystem of the North Atlantic and the North Sea by Longwell & Hughes (1980) and Cameron et al. (1996) as a relatively easily studied bioindicator (normal or abnormal fish embryos) with a high societal value (fish).

In the southern North Sea investigations on fish embryo health had been initiated in 1984 and conducted at intervals until 2000 on a grid of stations in the coastal waters of Denmark, The Netherlands, Belgium and Germany (Fig. 1). Between 20 000 to 40 000 embryos were examined during each cruise. Although the method has not been included in those adopted for the recommended procedures in biological effects monitoring in the OSPAR convention area (Stagg, 1998) it provided a useful time series for now 17 years. It could be shown that malformation rates were higher in estuaries and in near shore areas off the Danish, Dutch, and German coasts than in offshore areas (Cameron et al., 1996).

The state of pollution in the North Sea

After more than a decade of pollution abatement measures in the North Sea, a marine environment that has the 'reputation' of being particularly polluted, the situation slowly changed for the better (de Jong et al., 1999). This is particularly evident with regard to the input of copper, lead and mercury through the river Elbe the North Sea, but also in view of declining levels of some chlorinated hydrocarbons such as PCBs and γHCH. This improvement could likewise be traced in the decreasing concentrations of xenobiotics in organisms (Anders et al., 1996; Dethlefsen et al., 1996; Westernhagen et al., 1997; Broeg et al., 1999). Malformation rates of fish embryos in the southern North Sea also decreased with time. While in 1987 the mean value for malformations in dab (*Limanda limanda*) (dab is representative for all other major fish species) embryos was 28.7% (with maximum values at stations up to 60%), this value decreased continuously and fluctuated around 5% from 1993 to 2000 (Fig. 2), with the exception of 1996. This year showed an exceptionally high value, which was the reason for a closer look into its possible causes. In order to correct for the proven temperature effect on developmental defects (Dethlefsen et al., 1996; , Westernhagen and Dethlefsen, 1997) ($y=31.205*10^{-0.15796x}$; y = malformation rates; x = temperature) total malformation rates were split into temperature induced, a naturally occurring background of 2%, and an unexplained portion which was assumed to be caused by anthropogenic factors. Figure 2 shows, that beginning 1993 the observed malformation prevalence ceased to be related to anthropogenic influence, but could be contributed entirely to the natural background and a temperature effect, i.e. lower than usual temperature; except for the 1996 value. The 1996 value (prevalence 24%) stood out from the hitherto decreasing trend of malformation rates, which up to then was very well in line with the reduced pollution load of the rivers Rhine and Elbe (Friedrich and Schulte-Wülder-Leidig; 1996, Kausch, 1996; de Jong et al., 1999). Although in winter 1995/1996 there was a strong temperature effect (Fig. 2), due to low (as low as -1°C) water surface temperatures in the German Bight in February 1996, the non-temperature effect on the malformation rate was considerable and higher than expected in view of a decreasing trend in contaminant inputs. This trend had been ascertained before for tissue contamination levels of flatfish by various monitoring-type investigations between 1988 and 1996 (Anders et al., 1996; Dethlefsen et al., 1996; Westernhagen and Dethlefsen, 1997, Broeg et al., 1999) in the southern North Sea and the Wadden Sea.

Taking muscle tissue contamination of Elbe flounder as an example, DDT had dropped from 243 µg/kg fat in 1989 to 40 µg/kg fat in September 1995, CB153 from 2346 µg/ kg fat to 411 µg/kg fat, HCB from 1047 µg/kg fat to 221 µg/kg fat. Yet in January 1996 levels of DDT reached tissue concentrations 120 times higher than measured in September 1995 (4748 µg/kg fat in January 1996, compared to 39.8 µg/kg fat measured for September 1995). Similarly, ΣPCB was 17 times higher in January 1996 than in September 1995 (33824 µg/kg fat compared to 1960 µg/kg in September 1995). In the center of the German Bight concentrations of the DDT metabolite DDE in dab liver tissue had increased from 3.8 µg/kg wet weight in December 1994 to 18 µg/kg in 1995 and 27.5 µg/kg in January 1996 (Fig. 3). Interestingly, the tissue concentrations of other chlorinated hydrocarbons remained relatively low, reflecting the low level of contamination measured in fall 1995 in fish from between the Elbe estuary and the island of Helgoland in the German Bight, suggesting that a pollution event restricted only to an input of PCBs and DDT had occurred in the German Bight between autumn 1995 and spring 1996.

In an attempt to localize this input we inferred that it could have been caused by the runoff from the Elbe river. In fact, upriver high levels of organic micropollutants have been reported by Heemken et al. (2000) for August 1995 at a sampling site at Dessau (in the upper Elbe river) and in an Elbe river tributary, associated with suspended particulate matter, thus supporting our view. In agreement with these findings, data from the long term monitoring program of the Elbe river revealed, that concentrations of

DDT and its derivatives in suspended matter from the estuary after increasing in July 1995, reached high levels in September 1995 (Fig. 4, see also de Jong, 1999 for PCBs), which lead eventually to the accumulation of these xenobiotics in fish. The chronology of detecting the pollutant as DDT in flounders in the estuary and as DDE in dab further offshore strongly suggests the existence of a recent input of DDT via the Elbe river.

Concluding remarks

From the above it can be concluded that elevated tissue contamination levels in fish from the Elbe estuary and further offshore in fall 1995/1996 were a direct consequence of an unusual high DDT and PCB input through the river Elbe. The ensuing contamination of the gonads of parent fish in the German Bight and the southern North Sea was responsible for elevated malformation rates in pelagic fish embryos detected in this area spring 1996. This relationship, which has been experimentally proven for marine fish at several occasions (Hansen et al., 1985; Westernhagen et al., 1981; Westernhagen et al., 1989) represents a reliable biomarker which can be used successfully in biological effects monitoring.

Although no plea is made to deviate from the concept of applying a suite of techniques in biological effects monitoring, it can be taken from the above findings that the *in situ* quantification of developmental defects of pelagic fish embryos provides a simple, straightforward and easily applicable method to detect pollution events. This is among other due to the sensitivity of the embryo towards external influences and the fact that a morphological deviation from normal development as an integrated signal stands for a variety of biochemical malfunctions.

References

Anders, K., Landwüst, C. (1996) Fischkrankheiten in der Nordsee. *UBA Texte, Berlin* **57/96**, 1-557.

Broeg, K., Zander, S., Diamant, A., Körting, W., Krüner, G., Paperna, I. and Westernhagen, H. von (1999) The use of fish metabolic, pathological and parasitological indices in pollution monitoring. I. North Sea. *Helgoland Mar. Res.* **53**, 171-194.

Cameron, P., Berg, J. and Westernhagen, H. von (1996) Biological effects monitoring of the North Sea employing fish embryological data. *Envir. Monit. Assess*. **40**, 107-124.

De Jong, F., Bakker, J. F., Van Berkel, C. J. M., Dankers, N. M. J. A., Dahl, K., Gätje, C., Marencic, H. and Potel, P. (1999) *Wadden Sea Quality Status Report*. Wadden Sea Ecosystem No. 9. Common Wadden Sea Secretariat, Trilateral Monitoring and Assessment Group, Quality Status Report Group, Wilhelmshaven, Germany.

Den Besten, P. J. (1998) Concepts for the implementation of biomarkers in environmental monitoring. *Mar. Environ. Res*. **46**, 253-256.

Dethlefsen, V., Westernhagen, H. von and Cameron, P. (1996) Malformations in North Sea pelagic fish embryos during the period 1984-1995. *ICES J. mar. Sci.* **53**, 1024-1035.

Friedrich, G. and Schulte-Wülder-Leidig, A. (1996) In *Warnsignale aus Flüssen und Ästuaren*, eds. J. Lozan and H. Kausch pp. 65-75. Parey, Berlin.

Hansen, P.-D., Westernhagen, H. von and Rosenthal, H. (1985) Chlorinated haydrocarbons and hatching success in Baltic spring spawning herring. *Mar. Environ. Res.* **15**, 59-76.

Heemken, O. P., Stachel, B., Theobald, N. and Wenclawiak, B. W. (2000). Temporal variability of organic micropollutants in suspended particulate matter of the river Elbe at

Hamburg and the river Mulde at Dessau, Germany. *Arch. Environ. Cont. Toxicol.* **38**, 11-31.

ICES (1978) On the feasibility of effects monitoring . ICES Coop. Res. Rep. **75**, 1-42.

Johnson, L. L. and Landahl, J. T. (1994) Chemical contaminants, liver disease, and mortality rates in English sole (*Pleuronectes vetulus*). *Ecol. Appl.* **4**, 59-68.

Kausch, H. (1996) In *Warnsignale aus Flüssen und Ästuaren* eds. J. Lozan and H. Kausch pp. 43-52. Parey, Berlin.

Longwell, A. C. and J. B. Hughes, J. B. (1980) Cytologic, cytogenetic and developmental state of Atlantic mackerel eggs from sea surface water of the New York Bight, and prospects for biological effects monitoring with ichthyoplankton*. Rapp. P.-v. Réun. Cons. perm. int. Explor. Mer* **179**, 275-291.

MacIntyre, A. D. and J. B. Pearce (eds.) (1980) Biological effects of marine pollution and the problems of monitoring. Rapp. P.-v. Réun. Cons. int. Explor. Mer 179, 1-346.

North Sea Task Force (1993) North Sea Quality Status Report 1993, Oslo and Paris Commissions, 1-132. International Council for the Exploration of the Sea, London.

Stagg, R. M (1998) The development of an international programme for monitoring the biological effects of contaminants in the OSPAR convention area. *Mar. Environ. Res.* **46**, 307-313.

Stebbing, A. R. D., V. Dethlefsen and M. Carr (Eds.) (1992) Biological Effects of Contaminants in the North Sea. *Mar. Ecol. Progr. Ser. Special* **91**, 1-361.

Westernhagen, H. von, Rosenthal, H., Dethlefsen, V., Ernst, W., Harms, U. and Hansen, P.-D. (1981) Bioaccumulating substances and reproductive success in Baltic flounder *Platichthys flesus*. *Aquatic Toxicol.* **1**, 85-99.

Westernhagen, H. von (1988) Sublethal effects of pollutants on fish eggs and larvae. In *Fish Physiology*, eds. W. S. Hoar and D. J. Randall, pp. 253-346. Academic Press, London.

Westernhagen, H. von, Cameron, P., Dethlefsen, V. and Janssen, D. (1989) Chlorinated haydrocarbons in North Sea whiting (*Merlangius* merlangus L.), and effects on reproduction. I. Tissue burden and hatching success. *Helgoländer Meeresunters.* **43**, 45-60.

Westernhagen, H. von and Dethlefsen, V. (1997) The use of malformations in pelagic fish embryos for pollution assessment. *Hydrobiologia* **352**, 241-250.

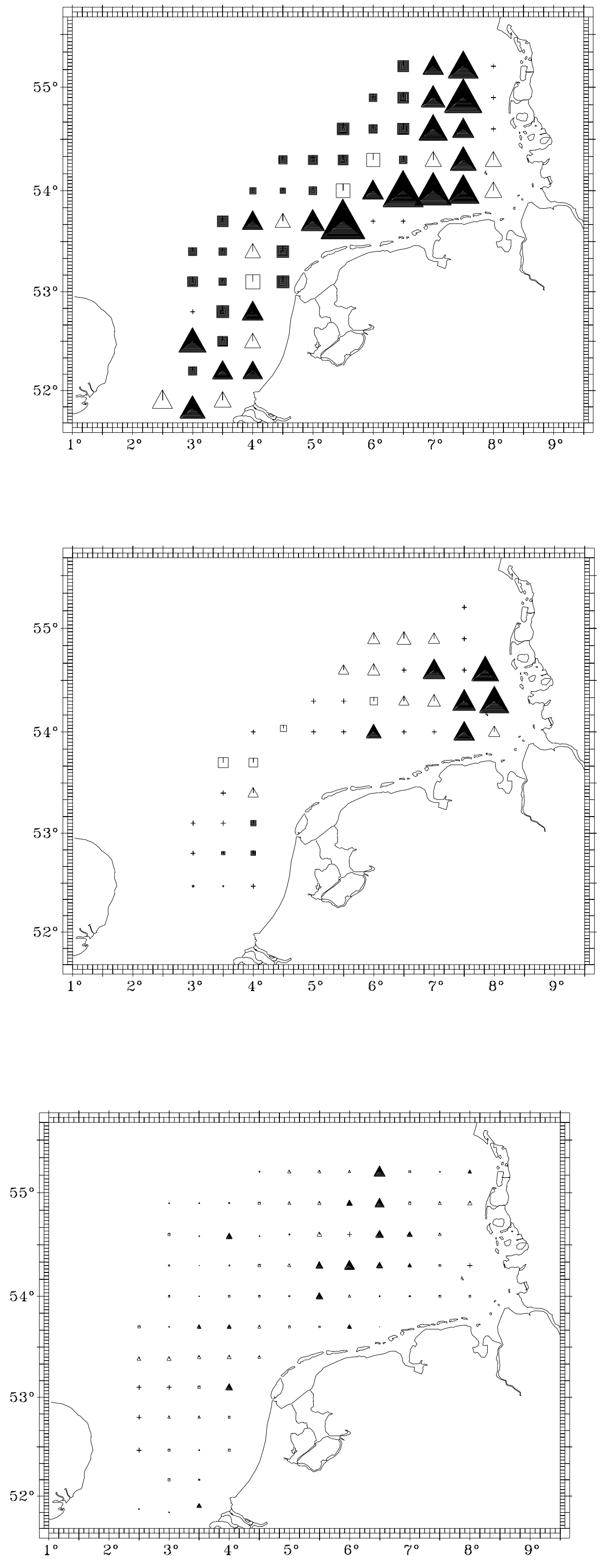


Figure 1. Malformation rates in embryos of dab, *Limanda limanda*, in the southern North Sea during the years 1987, 1996 and 1999 for all developmental stages until hatching. Note highest malformation rates in near coastal areas (1987) or in the Elbe river plume (1996). Maximum: 1987 - 61.8%, 1996 - 41.4%, 1999 - 16.1%. +: not enough eggs for evaluation (threshold n>50); χ^2 evaluation: black triangles - significantly above the mean; black squares - significantly below the mean; open triangles and squares - not significantly above or below the mean.

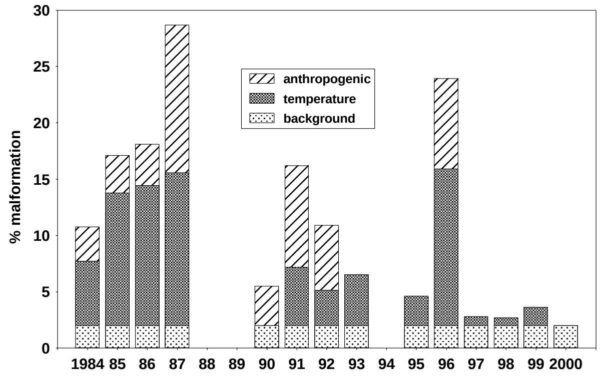


Figure 2. Mean malformation rates (%) in embryos of dab (*Limanda limanda*) between 1984 and 2000 from the southern North Sea (for area see Fig. 1). Temperature effects calculated on the basis of actual *in situ* measurements. Natural background value for unpolluted areas was arbitrarily set at 2%.

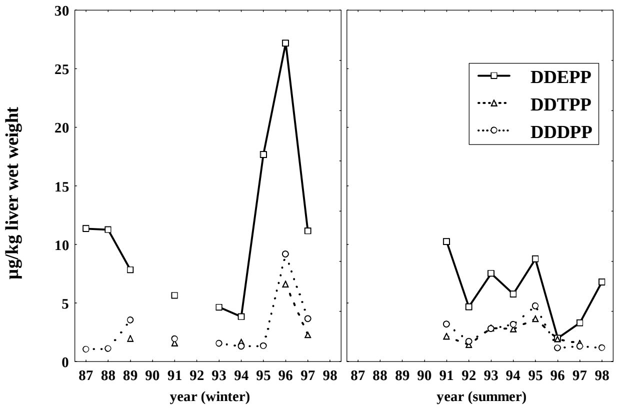
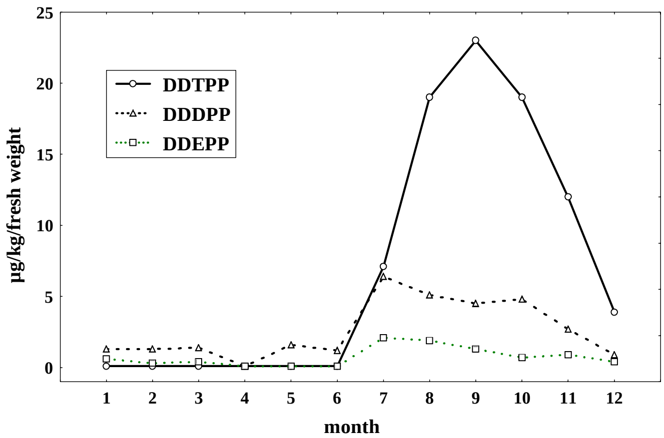


Figure 3. Concentrations of DDT and derivatives in livers (wet weight) of dab (*Limanda limanda*) in the German Bight, 1987-1998, winter and summer



.

Figure 4. Concentrations of DDT and its derivatives in suspended matter in the Elbe estuary (monthly pools in sediment traps, wet weight) in 1995. Data from the Arbeitsgemeinschaft für die Reinhaltung der Elbe, ARGE, Hamburg).