

Is PCB responsible for embryo toxicity in white-tailed sea eagle (*Haliaeetus albicilla*) from the Swedish Baltic coast?

Anders Olsson¹, Björn Helander^{2,3}, Anders Bignert³, Kerstin Litzén⁴,
Lillemor Asplund⁴, Åke Bergman¹

¹Department of Environmental Chemistry, Stockholm University, 106 91 Stockholm, Sweden,

²Swedish Society for Nature conservation, 116 91 Stockholm, Sweden,

³Contaminant Research Group, Swedish Museum of Natural History, 104 05 Stockholm, Sweden

⁴Institute of Applied Environmental Research, Stockholm University,
106 91 Stockholm, Sweden

Introduction

White-tailed sea eagle (*Haliaeetus albicilla*) (WTSE) in Europe and bald eagle (*Haliaeetus leucocephalus*) in north America decreased dramatically during the 50ties and 60ties following the introduction of chlorinated substances to the environment (e.g. DDT) (1, 2). The strong correlation between high concentrations of environmental contaminants and low reproduction have put those substances in focus for explaining the population decreases. The close co-variation between PCB and DDT in eagle eggs have, however, made it difficult to separate the effects of these chemicals and distinguish one of them as the major reason for observed biological effects. Nevertheless, in several studies *p,p'*-DDE, the major metabolite of *p,p'*-DDT, have been most closely correlated to reproduction failure, even though concentrations of PCB have often been significantly correlated too (1-4). In recent years some studies have suggested that PCB, due to the TCDD like effects of some PCB congeners, is the contaminant that today affects the eagle productivity (5, 6). Nevertheless, even in those studies the correlation between productivity and contaminant burden has been stronger for DDE than PCB (6).

Historically the major effect of DDE in avian species have been associated with eggshell thinning (7) whereas PCB has been reported to cause embryo mortality or a disease complex, including oedema and developmental abnormalities (8-10). Eaglets with developmental abnormalities have been reported for both bald and WTSE (11, 12).

The aim of this study was to investigate if concentrations of PCB and/or DDE can be linked to embryo mortality in WTSE eggs from the Swedish Baltic coast.

Materials and methods

The materials studied in this paper are WTSE eggs collected between 1988 and 1997. The eggs were collected on the Swedish Baltic coast in connection with annual surveys in order to monitor the reproductive success of this highly contaminated population. All eggs were dead, collected

4-7 weeks past normal hatching time. According to Helander et al 1998 (13) a problem for the WTSE at the Baltic coast has been an abnormal dehydration of the eggs. In a try to separate this type of symptom from embryo mortality only eggs with a desiccation index (D_i)(13) higher than 0.57 were selected. A total of 26 WTSE eggs with $D_i > 0.57$ was collected between 1988-97 at the Swedish Baltic coast. The eggs were divided into two groups, with or without embryo development. Seven out of the 9 embryonated eggs and 13 out of the 17 undeveloped eggs were found in nests that also contained a nestling.

The eggs were extracted and purified for contaminant analysis according to a previously described method (14). Eggs collected between 1988-90 were analysed by packed column, whereas the eggs from 1991-92 were analysed by capillary column giving individual PCB congener concentrations. CB-138 was selected as a measure of the total load of PCB. CB-138 is strongly correlated to total PCB ($r^2=0.99$) and to the planar PCB congener 3, 3', 4, 4', 5'-pentachlorobiphenyl (CB-126) ($r^2=0.95$) in WTSE eggs from the Swedish Baltic coast (15). In packed column GC analysis peak number 10 (#10) is mainly composed of 2, 2', 3, 4, 4', 5'-hexachlorobiphenyl (CB-138). The relative proportion of CB-138 to #10 was determined in 13 eggs collected in 1991 to 1992 from different parts of Sweden and analysed in parallel on both packed and capillary columns. The ratio of #10 / CB-138 was 1.326±0.044. Thus, in eggs analysed before 1991 the concentration of CB-138 was estimated from peak #10.

Results and discussion

Infertile and/or undeveloped eggs occurs naturally and are found also in much less polluted areas like northern Sweden (1). The undeveloped eggs in this study have therefore been assumed to represent the range of concentrations within the Baltic coast population.

The mean concentrations in eggs uncorrected for embryo length (see below) of both CB-138 and DDE were significantly higher in the eggs with embryos (Table 1). This is probably partly due to the metabolism of lipids by the growing embryo, which thereby increases the concentrations of xenobiotica in the eggs on a lipid weight basis (1). The p value for CB-138 is however much lower than for DDE (Table 1). According to (1) the concentration increase on a lipid weight basis due to lipid metabolism is negligible in embryos smaller than 75 mm, whereas considerable concentration increase occurs in larger embryos. Assuming an approximate linear relationship between the increase in residue concentrations and embryo growth after 75 mm (2), a rough

Table 1. Residue concentrations and desiccation index (D_i) of white-tailed sea eagle eggs from the Swedish Baltic coast collected between 1988 and 1997. Arithmetic mean and 95 % C.I.().

	n	D_i	Residues $\mu\text{g/g l.w.}$	
			<i>p,p'</i> -DDE	CB-138
A. Infertile or undeveloped eggs	17	0.82 (0.026)	163 (34)	59 (11)
B1. Eggs with embryo	9	0.68 (0.069)	252 (64)	105 (25)
B2. Estimate of original conc. in embryo eggs	9		208 (49)	87 (18)
Students T-test (A versus B1)			p=0.014	p=0.0009
Students T-test (A versus B2)			p=0.14	p=0.011

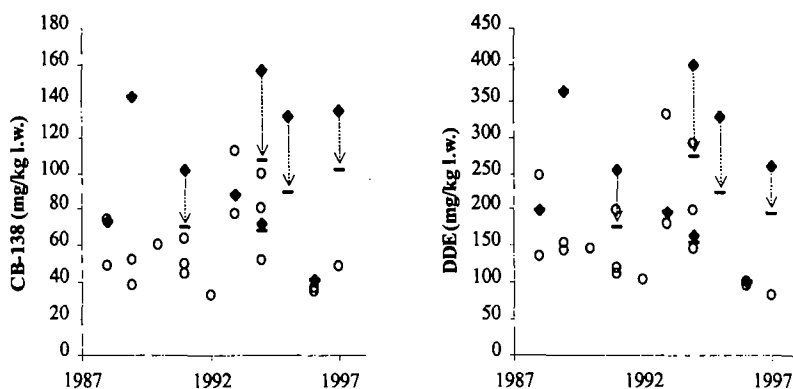


Figure 1. Concentrations of CB-138 and DDE in white-tailed sea eagle eggs from the Swedish Baltic coast. (◆) represent eggs with and (O) without embryonic development. In eggs containing embryos larger than 75 mm the concentration corrected for lipid metabolism are shown with (—).

estimation of the original concentration in the eggs containing embryos larger than 75 mm, can be calculated [original conc. = $(75 \times \text{measured conc.}) / \text{embryo length (mm)}$]. These calculated concentrations are roughly comparable to the residue concentrations in undeveloped eggs. Using these estimated concentrations a significant difference between egg with and egg without embryo is found for CB-138 but not for DDE (Table 1, Figure 1). Either one use estimated or the measured data, the eggs containing embryos have significantly higher PCB concentrations than eggs without embryonic development. It is therefore possible that the embryos died because of lethal concentrations of PCB. If so, this is likely to be a result of the toxic effects of the coplanar PCB congeners.

It can however not be excluded that the embryo mortality is an effect of the dehydration of the eggs. The D_i in embryonated eggs were significantly lower ($p < 0.005$) than the undeveloped. Abnormal dehydration in chicken eggs has been reported to dramatically decrease hatching success (16).

If the embryo mortality is due to PCB, a critical PCB concentration on embryo mortality can be estimated. All eggs with embryonic development had CB-138 concentrations higher than 40 $\mu\text{g/g l.w.}$ (Figure 1). The embryo-egg with the lowest concentration was collected in a nest that also contained a nestling, thus proving that incubation was successfully completed. The concentration of 40 $\mu\text{g CB-138/g l.w.}$ is equivalent to about 300 $\mu\text{g/g}$ total PCB or approximately 100 ng/g of CB-126 ($[\text{CB-126 ng/g}] = 2.54 \times [\text{CB-138 } \mu\text{g/g}]$) (15). These concentrations could be interpreted as the lowest observable effect level (LOEL) for embryo mortality. Recalculating the concentration of CB-126 into TEQ_{WHO} (17) gives 10 ng/g . CB-126 is the major contributor to the total TEQ in WTSE from the Finnish Baltic coast and was reported to be approximately 50 % of the total TEQ_{WHO} in four WTSE samples (18). A rough estimation of our results would then give a LOEL (embryo mortality) value of 20 $\text{ng TEQ}_{\text{WHO/g l.w.}}$. Elliot *et al.* 96 (19) reports a similar LOEL (enzyme induction) for Bald Eagle from the

pacific coast of Canada (12.6 ng TEQ_{WHO}/g l.w.). Other authors have postulated and used a NOEL value for bald eagle and WTSE that is about twenty times lower than our suggested LOEL (5, 6, 18). Twenty times is however the same magnitude as the whole range of measured total PCB concentrations (200-2900 ppm l.w.) in WTSE eggs from the Swedish Baltic coast since the start of environmental contaminant analysis in the late 60's. These egg-concentrations represent eagle females with productivity ranging from normal to zero.

In experimental studies dose response curves of CB-126 (LD50) in chicken and cormorant eggs show approximately one order of magnitude difference in sensitivity between individual eggs (9,10), i.e. a factor of ten in concentrations from where the first embryo dies to 100% embryo mortality. Thus if there is a LOEL of 300 ppm total PCB in WTSE as suggested in this study, one would expect females with a wide range of higher concentrations that still reproduce normal. Within the material of eggs sampled at the Swedish Baltic coast, eggs with total PCB concentrations as high as 1400 ppm l.w. have been collected from eagle pairs with normal reproduction. Thus, founding like those are not contradictory to the postulated LOEL value in this study.

References

1. Helander, B., Olsson, M. and Reutergårdh, L. *Holarct. Ecol.* **1982**, *5*, 349-366.
2. Wiemeyer, S. N., Lamont, T. G., Bunck, C. M., Sindelar, C. R., Gramlich, F. J., Fraser, J. D. and Byrd, M. A. *Arch. Environ. Contam. Toxicol.* **1984**, *13*, 529-549.
3. Wiemeyer, S. N., Bunck, C. M. and Stafford, C. J. *Arch. Environ. Contam. Toxicol.* **1993**, *24*, 213-227.
4. Colborn, T. J. *Toxicol. Environ. Health* **1991**, *33*, 395-453.
5. Giesy, J. P., Bowerman, W. W., Mora, M. A., Verbrugge, D. A., Othoudt, R. A., Newsted, J. L., Summer, C. L., Aulerich, R. J., Bursian, S. J., Ludwig, J. P., Dawson, G. A., Kubiak, T. J., Best, D. A. and Tillitt, D. E. *Arch. Environ. Contam. Toxicol.* **1995**, *29*, 309-321.
6. Bowerman, W. W., Giesy, J. P., Best, D. A. and Kramer, V. J. *Environ. Health Perspect.* **1995**, *103*, 51-59.
7. Blus, L. J., Wiemeyer, S. N. and Bunck, C. M. *Environ. Pollut.* **1997**, *95*, 67-74.
8. Lillie, R. J., Cecil, H. C., Bitman, J. and Fries, G. F. *Poult. Sci.* **1975**, *54*, 1550-1555.
9. Brunström, B. and Andersson, L. *Arch. Toxicol.* **1988**, *69*.
10. Larson, J. M., Karasov, W. H., Sileo, L., Stromberg, K. L., Hanbidge, B. A., Giesy, J. P., Jones, P. D., Tillitt, D. E. and Verbrugge, D. A. *Environ. Toxicol. Chem.*, **1996**, *15*, 553-559.
11. Bowerman, W. W., Kubiak, T. J., Holt, J. B., Evans, D. L., Eckstein, R. G., Sindelar, C. R., Best, D. A. and Kozie, K. D. *Bull. Environ. Contam. Toxicol.* **1994**, *53*, 450-457.
12. Helander, B. *SNV report 1386*, **1982**, ISBN 91-7590-141-2.
13. Helander, B., Olsson, A., Bignert, A., Litzén, K., Asplund, L. and Bergman, Å. *Organohalogen Compounds* **1998**, *39*.
14. Jensen, S., Reutergårdh, L. and Jansson B. *FOA Fish Tech. Pap.* **1983**, *212*, 21-33.
15. Helander, B., Olsson, A., Bergman, A., Litzén, K., Asplund, L. and Bignert, A. *Publ. Abstr.*, 18th Annual Meeting, Society of Environmental Toxicology and Chemistry, San Francisco, CA, **1997**.
16. Davis, T. A. and Ackerman, R. A. *J. Exper. Zool. Suppl.* **1987**, *1*, 357-364.
17. Ahlborg, U. G., Becking, G. C., Birnbaum, L. S., Brouwer, A., Derks, H. J. G. M., Feeley, M., Golor, G., Hanberg, A., Larsen, J. L., Liem, A. K. D., Safe, S. H., Schatter, C., Waern, F., Younes, M. and Yrjänheikki, E. *Chemosphere* **1994**, *28*, 1049-1067.
18. Koistinen, J., Koivusaari, J., Nuuja, I., Vuorinen, P. J., Paasivirta, J. and Giesy, J. P. *Environ. Toxicol. Chem.* **1997**, *16*, 1533-1544.
19. Elliott, J. E., Norstrom, R. J., Lorenzen, A., Hart, L. E., Philibert, H., Kennedy, S. W., Stegeman, J. J., Bellward, G. D. and Cheng, K. M. *Environ. Toxicol. Chem.* **1996**, *15*, 782-793.