

Organochlorines, Reproductive Impairment and Declines in Bald Eagle *Haliaeetus leucocephalus* Populations: Mechanisms and Dose-response Relationships

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ABSTRACT

This paper re-analyses some published data on organochlorine residues, reproductive impairment, and population declines in Bald Eagles in the USA and Canada. DDE showed the strongest associations with both eggshell thinning and reduced productivity; in a stepwise regression model, no other chemical residue contributed significantly to the explained variances. However, PCBs and oxychlordane could not be conclusively separated from DDE within this data set. S-shaped dose-response functions yielded better fits to the data than other functions tested. These conclusions are preliminary and are being tested further on this and another data set. Both reproductive impairment (caused by DDE) and excess adult mortality (caused by HEOD and perhaps by DDT) appear to have contributed to regional population declines in Bald Eagles, but evidence for the relative importance of these factors is circumstantial at best.

INTRODUCTION

This paper presents a preliminary analysis of data on the relationships between organochlorine contaminants, reproductive impairment, and declines in populations of the Bald Eagle (*Haliaeetus leucocephalus*) in North America. The following issues are addressed:

1. What are the quantitative relationships between eggshell-thinning and organochlorine levels?
2. What are the quantitative relationships between reproductive impairment and organochlorine levels?
3. Is eggshell-thinning the primary mechanism through which organochlorines caused reproductive impairment?
4. Is reproductive impairment the primary mechanism through which organochlorines caused population declines?

Issues (1-3) are addressed by analysis of dose-response relationships, using multiple regression techniques to separate the effects of the various organochlorine contaminants. Issue (4) is

addressed by review of historical data on the occurrence of eggshell-thinning, reproductive impairment, adult mortality, and local population declines. These analyses are ongoing and more detailed results will be published elsewhere.

Geographical and Historical Background

The Bald Eagle is confined to North America, nesting widely throughout the USA and Canada and in Baja California, Mexico (AOU 1983). Eggshell-thinning had occurred in Florida birds by 1947-48 and in Texas birds by 1949-50 (Anderson & Hickey 1972). Reproductive impairment was first reported in Florida in 1947 (Broley 1958) and became widespread during the 1950s and 1960s (Sprunt 1963; Stickel 1966; Sprunt & Ligas 1966; Abbott 1967; Postupalsky 1971; Wiemeyer *et al.* 1972, 1979; Grier 1972; Sprunt *et al.* 1973). By 1970, a number of local populations in the lower 48 states of the USA and in southern Canada had been markedly reduced or extirpated (Broley 1958; Howell 1963; Abbott 1967; Postupalsky 1971; Grier 1972; Sprunt *et al.* 1973; USDI 1974; Kiff 1980); populations in Alaska and parts of western and northern Canada were generally unaffected. A survey conducted in 1973 suggested that about 1,000 pairs were then nesting in the lower 48 states of the USA, plus between 30,000 and 55,000 birds in Alaska (USDI 1974). Grier (1972) described the Canadian population as "relatively large but mostly unknown"; about 500 pairs were being studied in 1969. Several studies have shown inter-relationships between eggshell-thinning, reproductive impairment, population declines, and levels of contamination with DDE and other organochlorines (Postupalsky 1971; Wiemeyer *et al.* 1972; 1979; Sprunt *et al.* 1973; Grier 1974). Following reductions in uses of DDT and other pesticides, concentrations of DDE and other organochlorines began to decline in Bald Eagle eggs in the late 1960s, and reproductive success began to improve in the mid-1970s (Grier 1982; Wiemeyer *et al.* 1984). Bald Eagles are now being re-introduced into several areas and populations are believed to be increasing (U.S. Fish & Wildlife Service, unpublished data), although little systematic information on recent population levels and trends has been published.

MATERIALS AND METHODS

Extensive studies of Bald Eagles were conducted in the period 1960-1979 by the U.S. Fish & Wildlife Service, the National Audubon Society, the Canadian Wildlife Service, and many state and provincial wildlife agencies and individuals. For the analysis reported in this paper, I use data on 126 Bald Eagle eggs collected in 15 states of the USA during 1974-1979 and reported on in part by Wiemeyer *et al.* (1984); additional data on productivity were provided by S.N. Wiemeyer.

Procedures for compiling and averaging data for each egg follow those of Wiemeyer *et al.* (1984), with minor modifications. Where more than one egg was collected from a clutch, arithmetic means of eggshell-thickness and geometric means of residue levels are used. Unlike Wiemeyer *et al.*, however, I use data from the same "breeding area" in different years; thus, the statistical unit is the clutch (breeding attempt) rather than the pair (breeding area). Data available for most clutches included the following:

1. Eggshell-thickness of sample egg(s); this was normalized to the regional mean for eggshells collected before 1947 (Anderson & Hickey 1972), to minimize confounding effects of geographical variation (Wiemeyer *et al.* 1984).
2. Productivity (estimate of chicks raised per nesting attempt): this was based on most cases on a 3-to 5-year average in the same breeding area (Wiemeyer *et al.* 1984).
3. Chemical residues in sample egg(s): original measurements were adjusted to yield estimates of concentrations in parts per million (ppm), wet weight, in fresh egg contents (Wiemeyer *et al.* 1984). Chemicals analyzed for included DDE, DDT + DDD (combined because of variable inter-conversion during incubation), HEOD (dieltrin), heptachlor epoxide, oxychlordane, *cis*-chlordane, *trans*-nonachlor, mirex, PCBs and total mercury.

I investigated the statistical associations between productivity, eggshell-thickness, and chemical residues using stepwise multiple regression techniques. The statistical model used is the following:

$$f(Y) = a + b_1D(x_1) + b_2D(x_2) + \dots, (1)$$

where Y is the dependent variable (eggshell-thickness or productivity); f(Y) is a transformation of Y; x_1, x_2, \dots are independent variables (residue concentrations) selected in stepwise fashion by the program; $D(x_1), D(x_2), \dots$ are dose-response functions; and b_1, b_2, \dots are regression coefficients.

The procedure followed was to select a form of the dose-response function for the first variable (x_1) so as to maximize its contribution to the explained variance (R^2), before proceeding to the second variable, and so on. The regression procedure thus yields information on the dose-response functions $D(x)$ as well as on the relative "importance" of the various independent variables.

To select the form of the dose-response functions, I used two procedures. The *parametric* procedure involved systematic exploration of various parametric functions for $f(Y)$ and $D(x)$ until R^2 was maximized. The functions explored included logarithmic and power functions, and the S-shaped function defined by

$$S_{cn}(x) = 1/(1 + x^n/c^n). \quad (2)$$

In all cases studied, this S-shaped function provided a better fit to the data than any other parametric function explored; the parameters c and n and the transformation $f(Y)$ were estimated by an iterative maximization procedure which usually converged rapidly. The *non-parametric* procedure used the "ACE" algorithm described by Breiman and Friedman (1985). This algorithm selects non-parametric functions for $f(Y)$ and for each of the $D(x)$ such that R^2 is maximized; the only constraints on the functions $f(Y)$ and $D(x)$ are that they should be locally smooth and (an optional constraint selected for this application) monotonic. The ACE algorithm was applied in a stepwise manner to provide information on the relative "importance" of the independent variables in explaining the variance in the dependent variable.

Causes of death in Bald Eagles found dead in the USA since 1960 have been investigated in a continuing program of the U.S. Fish & Wildlife Service (see footnotes to Table 1).

RESULTS

Eggshell Thickness

Complete data on eggshell thickness and chemical residues were available for 76 eggs. Applying the parametric regression procedure, the first independent variable selected was DDE, and the best-fitting dose-response function was the function $S_{cn}(DDE)$ (see equation 2 above) with $f(Y) = \ln(\text{normalized eggshell thickness})$ and with parameter values $c = 4.5$ ppm, $n = 4.5$; the explained variance R^2 was then equal to 0.270. Almost as good a fit was obtained, however, if PCBs were selected as the first independent variable: the best-fitting functions and parameters were $f(Y) = \ln Y$, $c = 10.3$ ppm, $n = 4.5$; $R^2 = 0.241$. The stepwise regression procedure selected DDE as the first independent variable and PCBs as the second independent variable, but the addition of PCBs yielded only a non-significant increase in R^2 to 0.337. No other chemical residue variable, if included in the regression equation as either second or third independent variable, would have further increased the value of R^2 by as much as 0.02.

The non-parametric ACE procedure yielded the best fit for PCBs ($R^2 = 0.337$) and almost as good a fit for DDE ($R^2 = 0.306$). When both these independent variables were included, R^2 increased to 0.354 and DDE showed a stronger association with eggshell thickness than did PCBs. No other chemical residue variable increased this value of R^2 by more than 0.01 when included as a third independent variable.

Productivity

Complete data were available on productivity, eggshell thickness, and chemical residues for 62 clutches. Analysis of these clutches showed only insignificant associations between productivity and eggshell thickness ($R^2 = 0.014$ using parametric transformations, 0.048 using ACE). Hence, the sample size was expanded to 97 by incorporating data from 35 clutches with data on productivity but not on eggshell thickness.

Applying the parametric regression procedure to the data from these 97 clutches, the first independent variable selected was DDE. The best fit was obtained with $f(Y) = Y^{1.4}$ and with $D(x) = S_{cn}(DDE)$ with $c = 3.8$ ppm and $n = 20$; the resulting value of R^2 was 0.272. The next best fit was obtained with oxychlordan ($R^2 = 0.191$). Addition of oxychlordan to the model as second independent variable (after DDE) increased R^2 by only 0.009. No other chemical residue variable increased R^2 by as much as 0.01 when added as second or third independent variable. Addition of eggshell-thickness as second or third independent variable (within the subset of 62 clutches with available data) increased R^2 by only 0.009.

The non-parametric ACE procedure also selected DDE as the first independent variable ($R^2 = 0.280$) and oxychlordane as the second ($R^2 = 0.244$ when tested by itself, $R^2 = 0.380$ when added as second independent variable after DDE). No other chemical residue variable yielded a value of R^2 greater than 0.19 when tested by itself, or increased R^2 by more than 0.07 when added as second independent variable after DDE. Addition of eggshell thickness as second or third independent variable (within the subset of 62 clutches with available data) increased R^2 by only 0.049.

Adult mortality

Table 1 summarizes data on 747 Bald Eagles found dead in the USA between 1960 and 1981 and autopsied and/or analysed by the U.S. Fish & Wildlife Service (for references see footnotes to Table 1). About 33 of these birds appeared to have died from poisoning by HEOD (dieldrin), 9 from poisoning by thallium, and 5 from poisoning by other organochlorines (2 attributed primarily to endrin, 1 to DDT + DDD, and 2 to DDE + PCBs). Based on criteria developed subsequently (Blus *et al.* 1983; Stone & Okoniewski 1987), several others may have died from combined effects of chlordane-group pesticides (heptachlor epoxide, oxychlordane, chlordane and/or nonachlor, in some cases augmenting or adding to the effects of HEOD).

DISCUSSION

Factors affecting eggshell-thinning and reproductive impairment

Wiemeyer *et al.* (1984) have already presented an analysis of associations between organochlorine contaminant levels, eggshell thickness and productivity, based on the same set of data. However, Wiemeyer *et al.* used only one mathematical transformation of the data (logarithmic transformation of the chemical residue variables), used different statistical techniques (simple correlation coefficients and principal components analysis), and did not investigate the relationship between productivity and eggshell thickness. The analysis summarized in this paper is consistent with several of the major conclusions drawn by Wiemeyer *et al.*:

1. Both eggshell thickness and productivity in Bald Eagles are strongly negatively correlated with residue levels of several organochlorine contaminants.
2. DDE is the contaminant most strongly associated in both cases, explaining about 30% of the variance in eggshell thickness and 28% of the variance in productivity.
3. Certain other organochlorines, notably DDT + DDD, PCBs, and oxychlordane, also show associations with eggshell thickness and productivity which approach in strength those of DDE.

The preliminary results reported in this paper suggest several additions or modifications to these conclusions:

1. In all cases investigated, S-shaped dose-response functions of the form indicated by equation (2) yielded better fits to the data than the logarithmic transformations used by Wiemeyer *et al.*
2. The non-parametric dose-response functions generated by the ACE algorithm provided better, but only slightly better, fits to the data than the best parametric functions investigated.
3. Although DDE showed the strongest statistical association with reduced eggshell thickness, PCBs were almost as strongly associated; the effects of DDE and PCBs cannot be distinguished within this data set.
4. Although DDE showed the strongest statistical association with reduced productivity, oxychlordane also showed a strong association; the effects of DDE and oxychlordane were not clearly distinguished within this data set.
5. After selecting an appropriate dose-response function for DDE, no other chemical residue variable contributed significantly to explaining the variance in either eggshell thickness or productivity.
6. Eggshell thinning was not significantly associated with reduced productivity, either before or after controlling for the effects of DDE on productivity.

All these conclusions should be regarded as hypotheses for further testing. Ongoing work includes refining these hypotheses with more intensive analysis of the same data set, sensitivity analysis, and testing the hypotheses with an independent set of data on Bald Eagles generated in field studies in the period 1963-1973 (Krantz *et al.* 1970; Postupalsky 1971; Wiemeyer *et al.* 1972; Sprunt *et al.* 1973; Weekes 1974; Whitfield *et al.* 1974; Grier 1974).

The most unexpected of the conclusions listed above is No. 6, that eggshell thinning is not associated with reduced productivity, although each is associated independently with residue levels of DDE. One possible explanation of this result is suggested by the dose-response functions derived during the analysis: the dose-response function for the effect of DDE on productivity is very steep, with most of the effect occurring between DDE = 2.5 ppm and DDE = 5 ppm. The dose-response function for the effect of DDE is flatter (resulting from the lower value of the parameter n), and DDE = 5 ppm corresponds to an average reduction in eggshell thickness of only about 10%. Thus, this analysis suggests that eggshell thinning may be a parallel symptom of DDE poisoning rather than a mechanism of reproductive impairment. This hypothesis will be tested in the ongoing work.

Factors causing population decline

Data cited in this paper show that in the period 1947-1981, Bald Eagles suffered from both reproductive impairment (caused primarily by DDE) and pesticide-induced mortality (attributable most frequently to HEOD). Both phenomena would have contributed to the local population declines that were observed in the 1950s and 1960s. However, it is very difficult to identify the relative importance of these two phenomena (Nisbet 1988). One possible approach is to match the temporal and geographical patterns of population decline with information on temporal and geographical patterns of use of DDT, dieldrin, and other pesticides. However, temporal patterns of population decline in Bald Eagles were poorly documented, and the areas that were most seriously affected (the Great Lakes, the Mississippi Valley, the east coast of the USA, and California) were areas where both DDT and dieldrin were heavily used in the 1950s (Nisbet 1988). Another approach is to investigate whether the population declines took place more quickly than can be explained by failure of recruitment alone (Nisbet 1988). Available population models for Bald Eagles (Grier 1982) remain unvalidated, but it seems likely that this species (like many other large birds) has low recruitment rates, high adult survival rates, and slow population turnover. At least three populations that were studied in the 1950s showed marked declines before 1960. A population in west Florida showed a sharp decline in 1957-58 and a further decline by 1962 (Broley 1958; Sprunt 1963). A population in east Florida declined by two-thirds between 1951 and 1961, mainly after 1956 (Howell 1963). A population on the California Channel Islands was extirpated by the end of the 1950s (Kiff 1980). These declines took place within 9-14 years after the onset of breeding failures. It is unlikely that such large declines in breeding populations could have resulted from failure of recruitment alone, so it seems probable that excess adult mortality was significant also. However, none of the areas involved was one where heavy use of dieldrin was documented in the 1950s (Nisbet 1988); the population declines appear more closely associated with uses of DDT (especially in California) and may conceivably have resulted from DDT poisoning. HEOD-induced mortality of Bald Eagles appears to have peaked in the period 1968-1973 (Table 1) and was probably associated with direct discharges of dieldrin into waterways from mothproofing plants (Nisbet 1988). In any event, it seems likely that both reproductive impairment and excess adult mortality contributed to local population declines in the Bald Eagle, that both DDT and dieldrin (and other pesticides) were causative factors, and that the bans on these pesticides led to the species' recovery that began in the mid-1970s.

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TABLE 1. Deaths of Bald Eagles attributed to poisoning by organochlorine compounds in the USA, 1960-1981.

Years	No. of birds analysed	Deaths attributed to poisoning by HEOD	Other organochlorines	References ^a
1960-62	6	ND ^b	-	1, 2
1963	9	-	-	1, 2
1964-65	40	1	-	1, 2, 3
1966-67	43	1	1 (DDT+DDD)	4, 5
1968	26	7	-	4, 5
1969-70	39	6	1 (DDE+PCBs)	6
1971-72	37	4	1 (DDE+PCBs) ^c	7
1973-74	86	4	-	8
1975-77	168	5	2 (endrin)* ^d	9
1978-81	293	5	-* ^e	10

^a 1, Coon *et al.* 1970; 2, Stickel *et al.* 1966; 3, Reichel *et al.* 1969a; 4, Reichel *et al.* 1969b; 5, Mulhern *et al.* 1970; 6, Belisle *et al.* 1972; 7, Cromartie *et al.* 1973; 8, Prouty *et al.* 1977; 9, Kaiser *et al.* 1980; 10, Reichel *et al.* 1984.

^b ND, not analysed for; -, no deaths attributed to these compounds.

^c Also 9 birds killed by thallium poisoning in 1971.

^d *, 2 or more birds contained residues of chlordane-group compounds high enough to have caused death, either alone or in combination with HEOD; this group of compounds was not fully analysed for in previous periods.

^e Also 17 birds killed by lead poisoning.

