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Mercury and organochlorine content of Dipper *Cinclus cinclus* eggs in south-west Ireland: trends during 1990–1999

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"Capsule": *A baseline for organochlorines in Irish freshwater ecosystems has been established.*

Abstract

A sample of 124 deserted/infertile dipper (*Cinclus cinclus*) eggs was collected in south-west Ireland during six seasons (1990–1994 and 1999) and analysed for mercury and organochlorines. Mercury was detected in three of the 6 years but no trend was observed. DDT occurred above the limit of detection in only two eggs, one each in 1993 and 1994. In contrast, two derivatives of DDT (DDE and TDE) were found much more commonly. DDE occurred in over 87% of eggs, while TDE contamination was less widespread and reached a maximum of 60% in 1993, with none detected in 1991 or 1999. There was no trend in occurrence of DDE or TDE across years. PCB contamination was present consistently over the 10-year period, with little change in the proportion of contaminated eggs collected over time. In all years, the lowest recorded occurrence of total PCBs was 69% expressed on a formulation basis as Arochlor 1254. PCB congeners 138, 153 and 180 dominated. The congener pattern was similar across years with the exception of 1990 when congener 118 dominated. A distinct trend of high values of contaminants was observed in 1990 and 1993. In 1990, PCB 118 was the dominant contaminant, while in 1993, HEOD, DDE, PCB 138 and PCB 153 were primary contributors to the observed result. Other contaminants (HCB, PCB 101, gamma-HCH, PCB 170 and PCB 180) showed little obvious patterns between years and occurred at relatively low levels. No pattern was observed in contaminants when eggs were grouped according to river or altitude. These data provide a baseline for organochlorine contamination levels in Irish freshwater ecosystems against which future trends can be assessed.

1. Introduction

The dipper (*Cinclus cinclus*) occurs discontinuously across Europe, Asia, North Africa, and Britain and Ireland (Tyler and Ormerod, 1994). They are common on fast-flowing rivers and streams, preferring sites with good water quality, and with walls and bridges for nesting and roosting (Buckton and Ormerod, 1997). They are exclusively riverine and feed predominantly on aquatic invertebrates and sometimes fish, although fish have not been found in their diet in Ireland (Taylor and O'Halloran, 1997). Therefore, they are useful biological indicators (Sorace et al., 1999). True migration and altitudinal movements occur in some parts of their range, although most populations only perform post-juvenile dispersal (Tyler and Ormerod, 1994; O'Halloran et al., 2000).

Because dippers have a limited post-juvenile dispersal rate and are highly site faithful thereafter (O'Halloran et al., 2000), they are suitable indicators of freshwater acidification and of organochlorine and heavy metal contamination (Ormerod and Tyler, 1990). No age effect has been reported on the accumulation of metals in dippers, but sex differences in zinc and copper levels have been found (Nybø et al., 1996). Persistent organochlorine compounds bio-magnify in aquatic food chains and accumulate in the tissues of top predators, where adverse effects can be measured. Dipper eggs are considered good indicators of spatial and temporal patterns of organochlorine pesticides due to maternal transfer (Ormerod and Tyler, 1994; Barron et al., 1995; Kallenborn et al., 1998; Scharenberg and Ebeling, 1998; Braune et al., 2001). Organochlorine contamination has historically been related to a lowering of breeding success in

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many bird species (Kubiak et al., 1989). This is primarily mediated during the egg stage of breeding (Nygård, 1999), and has contributed to a decline in the range of many species throughout the world. For example, poor hatching success in some species has been related to elevated levels of PCBs in eggs (Dirksen et al., 1995). DDE is widely reported to impact on reproductive success through eggshell thinning (Cooke, 1973; Lundholm, 1987; Wiemeyer et al., 1993). Ormerod and Tyler (1992) reported moderate eggshell thinning in relation to increasing DDE in dipper eggs from Scotland, Wales and Ireland, but with only slight evidence of toxic effects.

Pollutant loads in biological systems increased worldwide through the earlier part of the 20th century (Thompson et al., 1992), leading to a decline in the range of many bird species. The banning of production and use of compounds containing organochlorines in many parts of the world has resulted in a decline in environmental contamination in more recent times (Bignert et al., 1995; Braune et al., 2001). There has been an associated recovery in populations of some bird species (Mason et al., 1997; Donaldson et al., 1999; Newton et al., 1999). Despite the banning of DDT in many countries, elevated concentrations of DDE (one of its main derivatives) are still responsible for reduced reproductive success in some bird species (Pearce et al., 1979; Custer et al., 1999). However, the levels observed in recent years are generally considered insufficient to cause a significant reduction in reproductive success (Fasola et al., 1998; Bishop et al., 1999). However, Ormerod et al. (2000) reported some impacts on breeding biology in dippers from a PCB point source. Although birds' eggs provide a useful measure of pollutant levels and trends, in many parts of the world routine environmental monitoring does not take place (Barrett et al., 1996; Braune et al., 2001; Goutner et al., 2001).

Despite the known oestrogenic risks caused by organochlorines, including PCBs, few data exist on their occurrence or patterns in Irish freshwater ecosystems. A recent review (Dempsey and Costello, 1998) noted no adverse effects on reproductive biology of Irish wildlife populations, with the exception of marine molluscs. Here we present an examination of patterns and trends of organochlorines and mercury in the eggs of dippers collected in south-west Ireland over a 10 year period (1990–1999). Ormerod and Tyler (1992) and O'Halloran et al. (1993) have previously reported on the eggs collected in 1990 and 1991.

2. Study area and methods

Infertile and deserted eggs (total 124) were collected under licence from rivers in south-west Ireland as follows: 1990 (17), 1991 (42), 1992 (20), 1993 (15), 1994 (20) and 1999 (10) (Fig. 1). Eggs were collected during the course of ongoing monitoring of dipper populations (O'Halloran et al., 1999) and were stored at 4 °C prior to analysis at the Monks Wood Laboratory of the Centre for Ecology and Hydrology, UK.

2.1. Sample analysis

Each egg was analysed separately for mercury (Hg) and organochlorines. Egg contents were analysed for mercury using cold vapour atomic absorption spectrophotometery (CVAAS) following acid digestion. A five point multi-level calibration was run prior to each batch of samples and the top standard and blank repeated every five samples to allow for re-scaling of the calibration curve. Within each batch, a sample of chicken egg was spiked with a known amount of analyte, and analysed in the same way as the dipper eggs. Certified reference material (BCR 186-Trace Elements in Lypholised pig kidney) was used. Recovery of spiked mercury averaged at 70.5% and the lower limit of detection was 0.03 μg/g. Data acquisition was done via a Solar 969 AAS (Thermounicam) and results are expressed in ppm dry weight for mercury, and ppm lipid for all other contaminants.

The following organochlorines were analysed using gas chromatography with electron capture detector:

p,p′-DDE (1,1′- (2,2-dichloroethenylidene) bis (4-chloro)-benzene)
p,p′-TDE (1,1′- (2,2-dichloroethylidene) bis (4-chloro)-benzene)
p,p′-DDT (1,1′- (2,2,2-trichloroethylidene) bis (4-chloro)-benzene)
HCB (hexachlorobenzene)
gHCH (1,2,3,4,5,6-hexachlorocyclohexane, Gamma isomer) (lindane)
HEOD (1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-exo-1,4-endo-5,8-dimethanonaphthalene) (aldrin and dieldrin)
Total PCBs (polychlorinated biphenyls)
Individual PCB congeners (8, 18, 28*, 31*, 52, 77*, 101, 118, 126*, 128*, 138, 149*, 153, 169*, 170, 180). (*In 1990 and 1991 the PCB congeners annotated were not analysed)

Total PCB refers to the polychlorinated biphenyl concentrations expressed on a formulation basis as Arochlor 1254. It is obtained by summation of all peaks present in the chromatogram which correspond to peaks present in Arochlor 1254 after elimination of known organochlorine pesticides. Consistency in analytical accuracy was achieved using internal standards at intervals of 25 determinations to assess the recovery for each organochlorine. The results of the spiked recovery

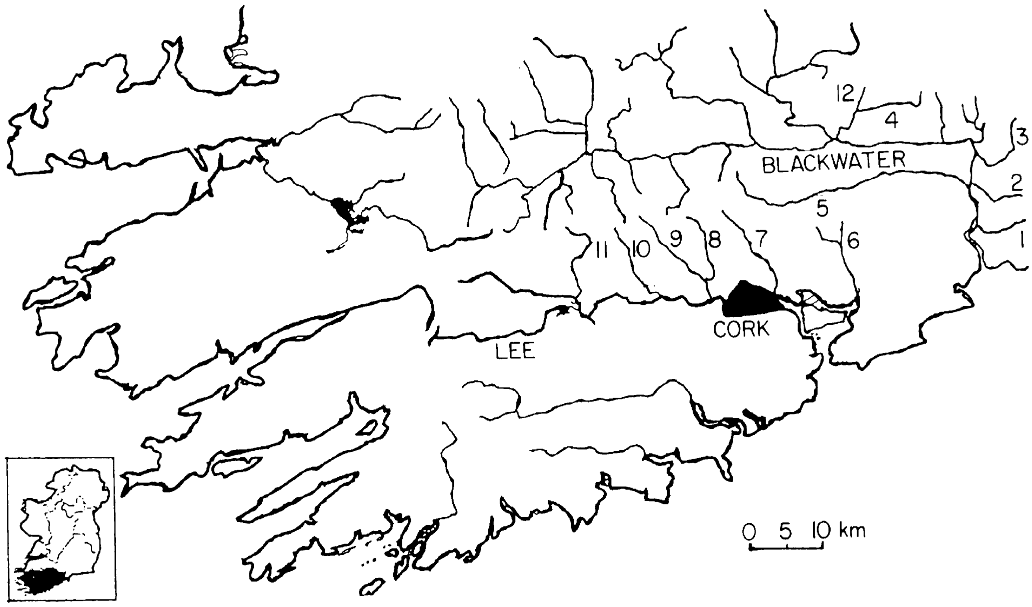

Fig. 1. Map of collection locations of dipper eggs in south-west Ireland; 1 = Licky, 2 = Goish, 3 = Finisk, 4 = Araglin, 5 = Leamlara, 6 = Owennacurra, 7 = Glashaboy, 8 = Martin, 9 = Shournagh, 10 = Dripsey, 11 = Laney, 12 = Douglas (after Smiddy et al., 1995).

samples ranged from 60–120%, with the exception of congeners 8 and 101 which were slightly higher (Wienburg et al., 1999). The recovery of organochlorines under investigation in this study were as follows: DDE, 82%; TDE, 88%; DDT, 77%; HCB, 79%; gamma-HCH, 80%; HEOD, 56%; PCB congeners, 86–144%. The minimum detectable level of organochlorines was 0.01 μg/g in lipid. The mean percent lipid concentration and the mean percent moisture content in dipper eggs was 6.3 (S.E. = 0.9) and 81.6 (S.E. = 0.4) respectively.

2.2. Data analysis

All infertile and deserted eggs were collected. Where more than one egg was collected from a nest and analysed, one was randomly selected for statistical analysis. Data analysis are summarised as follows:

(a) Percentage occurrence (at levels greater than the detection limit) of each contaminant.
(b) Because of the usual log-normal distribution of data, the values should be presented as geometric mean. However, since the difference between arithmetic and geometric means were minimal and the variance within each parameter low, the data are presented as arithmetic mean. Where levels of a contaminant were below detection limits, eggs were given an arbitrary value equal to the minimum detectable value (0.01 μg/g) after Ormerod et al. (2000).

Ordination analysis was used to examine the overall pattern of contaminant levels in the eggs across different rivers, sites and years. Following examination of the length of the ordination axes, using detrended correspondence analysis (< 2.5 S.D.), a principal components analysis (PCA) was performed on contaminant data, using the CANOCO (version 4) ordination programme (ter Braak and Smilauer, 1998). One-way ANOVAs were performed on axis 1 and axis 2 scores for samples grouped according to year, altitude and river, to test for significant differences in the ordination between the different levels of these factors.

3. Results

3.1. Percentage occurrence

Mercury was detected in 3 of the 6 years during this study (Fig. 2) and no trend in occurrence was observed. DDT occurred above the limit of detection in only two

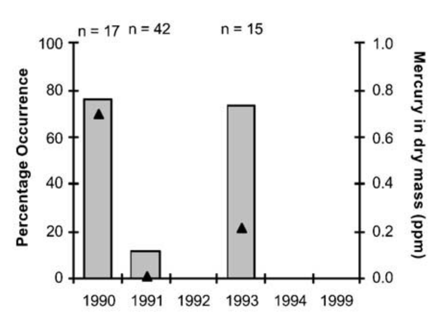

Fig. 2. Percentage occurrence of dipper eggs contaminated with mercury at levels above the limit of detection (vertical bars), and mean (▲) levels in contaminated eggs (ppm in dry weight).

eggs, one each in 1993 and 1994, with very low levels of 0.74 and 0.18 ppm in lipid respectively (Fig. 3). By comparison, two derivatives of DDT (DDE and TDE) were found much more commonly (Fig. 3). DDE occurred in over 87% of eggs while TDE contamination was less widespread and reached a maximum of 60% in 1993, with none detected in 1991 or 1999. There was no directional trend in occurrence of TDE across years (Fig. 3).

HCB, gHCH and HEOD were found less frequently in sampled eggs than DDE. However, HEOD was widespread and occurred in all years. The occurrence of this compound peaked during 1993 and 1994 and decreased in 1999 (Fig. 4). The occurrence of HCB increased between 1990 and 1994 but it was absent in 1999, while gHCH showed no trend in occurrence and was absent in 1990 and 1993 (Fig. 4).

PCB contamination was present consistently throughout the ten-year period, with no change in the proportion of contaminated eggs collected over time (Fig. 4). The lowest recorded occurrence of PCBs was 69% in 1991 (Fig. 4). PCB congeners 138, 153 and 180 dominated (Fig. 5). The congener pattern was similar across years with the exception of 1990 when congener 118 dominated (Fig. 5).

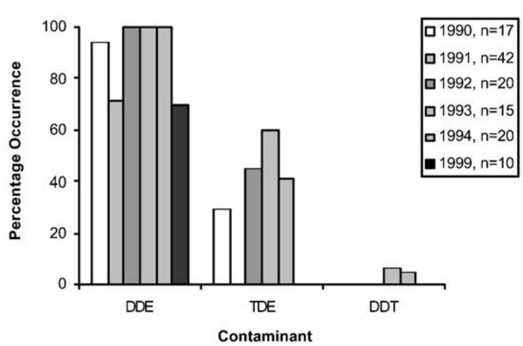

Fig. 3. Percentage occurrence of dipper eggs contaminated with DDT and its derivatives (DDE and TDE) at levels above the limit of detection.

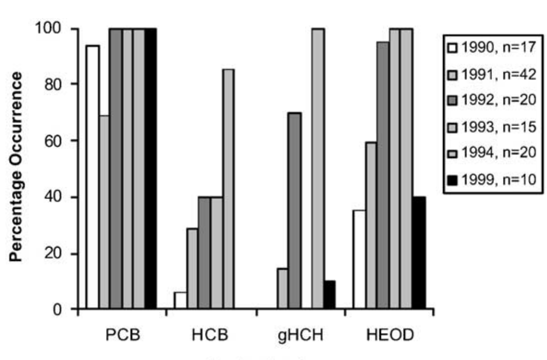

Fig. 4. Percentage occurrence of dipper eggs contaminated with total PCBs and other organochlorines (HCB, gHCH and HEOD).

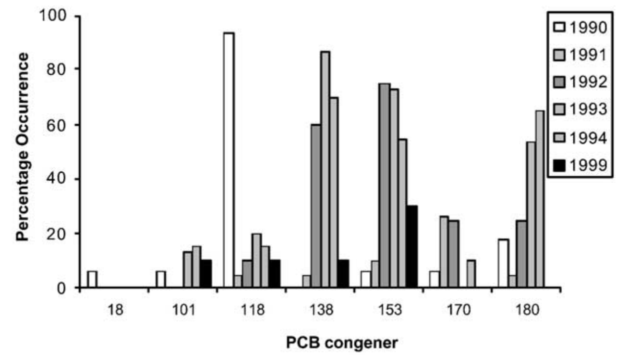

Fig. 5. Percentage occurrence of dipper eggs contaminated with selected PCB congeners.

3.2. Patterns in contaminant concentrations

Contaminants in dipper eggs could be grouped broadly into three categories according to their temporal patterns across years (Fig. 6). The first group of contaminants were all most abundant in 1990 and relatively rare thereafter. They included Hg, PCB 18, TDE and PCB 118. The second group showed increasing levels of abundance from 1990 to a peak in 1993 and then declining levels thereafter. They included HEOD, DDE, PCB 138 and PCB 153. The third group showed little clear temporal pattern and included HCB, PCB 101, gHCH, PCB 170 and PCB 180 (Fig. 6).

3.3. PCA analysis

Eigen values of the first two axes of the principal components analysis were high (0.453 and 0.183 for axis 1 and 2 respectively), explaining 45.3 and 63.3% of the cumulative percentage variation for the first two axes. Fig. 7A shows sample scores (each 'sample' being an individual egg) of the PCA of axis 1 plotted against axis 2, labeled for the different years. Fig. 7B shows species scores (each 'species' being a separate PCB contaminant) of axis 1 plotted against axis 2.

The results of one-way ANOVAs performed on axis 1 and axis 2 scores for sample scores grouped according to river, year and altitude are shown in Table 1. There was no significant difference ($P > 0.05$) in sample scores on axis 1 and axis 2 for either river or altitude. However, year was significant for both axis 1 and axis 2, demonstrating that this was the dominant factor distinguishing between sample scores.

There were two clear patterns of PCB congener contamination levels in eggs for different years (Fig. 7A). The first (horizontal) axis of the ordination distinguished between PCB congener contamination in eggs in 1993 relative to other years and the second (vertical) axis distinguished between PCB congener contamination in eggs in 1990 relative to other years. Contaminants with high levels associated with 1993 eggs

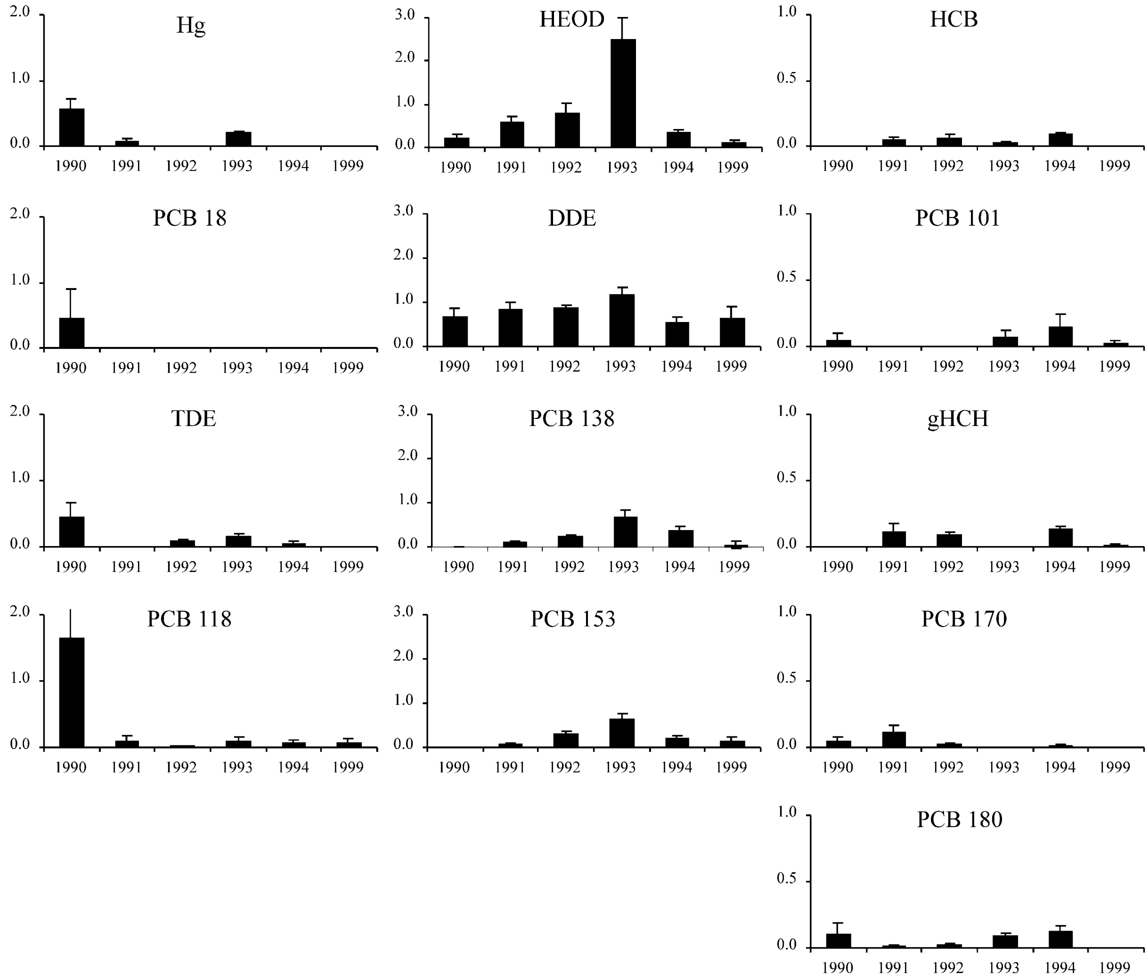

Fig. 6. Mean levels (±S.E.) of Hg, PCB congeners and selected chlorinated pesticides in dipper eggs (all years and rivers).

were HEOD, DDE, PCB 138 and PCB 153 (Figs. 6 and 7). The single contaminant with high levels associated with 1990 eggs was PCB 118 (Fig. 7B).

4. Discussion

Ormerod and Tyler (1994) showed that individual dipper eggs could be used successfully to represent contaminant levels within entire clutches. In this study, while all eggs were taken and no distinction could be made as to the order in which eggs were laid, a single egg was randomly selected for the purpose of analysis. Although the sampling method employed here is non-random, it represents the standard method of egg collection for investigations of contaminant levels. Results are therefore comparable to other studies. Birds' eggs are widely used to monitor contaminant levels in a range of environments (Albanis et al., 1995; Aurigi et al., 2000; Braune et al., 2001), and they are particularly useful in riverine ecosystems (Kallenborn et al., 1998).

4.1. Contamination levels between 1990 and 1999

Although mercury occurred above the limit of detection in only 3 of the 6 years in this study (Fig. 2), during two of those years over 70% of eggs were contaminated. Despite the sporadic occurrence of mercury, a decrease in mean levels in contaminated dipper eggs was observed between 1990 and 1993, after which time it was entirely absent. These levels of mercury were higher than the mean values obtained in black-legged kittiwakes (*Rissa tridactyla*), though lower than fulmars (*Fulmarus glacialis*) and thick-billed murres (*Uria lomvia*) in Arctic Canada (Braune et al., 2001). A decreasing trend in mercury levels in birds' eggs has recently been reported in parts of Europe (Mason et al., 1997; Nygård, 1999) with no temporal trends reported in other areas (Barrett et al., 1996). Although there was no apparent trend in the extent of mercury pollution in eggs in this study, a decrease in contaminant levels was observed. Only during the first year of this study, 1990, did mercury levels in any individual egg exceed 3 ppm,

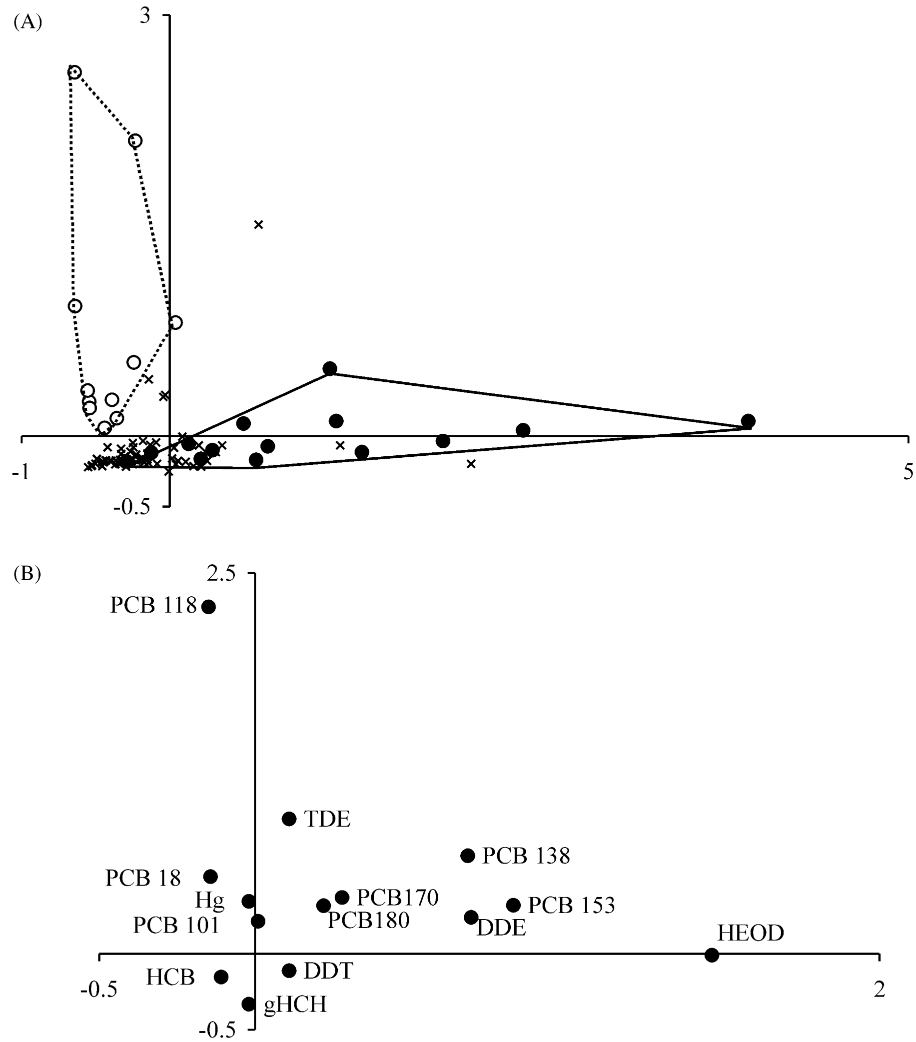

Fig. 7. (A) PCA plot of axis 1 sample scores against axis 2 sample scores for PCB congeners in dipper eggs. A dotted line polygon is drawn around samples for 1990 (○) and a solid line polygon is drawn around samples for 1993 (●). Samples for all other years are shown by the symbol ×. (B) PCA plot of axis 1 'species' scores against axis 2 'species' scores for PCB congeners in dipper eggs.

the level reported to affect the breeding success of merlins (*Falco columbarius*; Newton and Haas, 1988). This once-off high value is thought to have been derived from a grain store containing seeds with mercury dressing. Currently, no data are available on the sub-lethal effects of mercury in dippers. Only one dipper egg collected in Wales was found to be contaminated with mercury, and the level (0.57 ppm dry weight, Ormerod and Tyler, 1990) was in line with the mean values found

Table 1
Summary of one-way ANOVA performed on PCA axis 1 and axis 2 sample scores, grouped according to river, year and altitude

	Axis 1			Axis 2		
Factor	DF	*F*-Ratio	*P*-value	DF	*F*-Ratio	*P*-value
River	15, 94	0.502	0.932	15, 94	1.022	0.442
Year	5, 96	11.846	<0.001	5, 96	11.516	<0.001
Altitude	2, 96	1.207	0.304	2, 96	0.830	0.439

in this study (Fig. 6). During 1990, mercury levels in dipper eggs were reported to be higher in Ireland than in Scotland or Wales (Ormerod and Tyler, 1992). However, after 1993 mercury was entirely absent from Irish eggs.

DDT was found in only two of the 124 eggs examined in this study. DDT (mostly its derivatives DDE and TDE) residues have been found in the eggs of aquatic birds in Scotland, the Netherlands, Italy and Norway (Ormerod and Tyler, 1992; Dirksen et al., 1995; Fasola et al., 1998; Kallenborn et al., 1998). DDE was commonly found in this study, occurring in over 87% of all eggs analysed (Fig. 3). Even with this high frequency of occurrence, mean DDE levels rarely exceeded 1 ppm in lipid in any year. There was some variation in DDE levels across years (1990–1999) and concentrations ranged from below the limit of detection to over 3 ppm (Fig. 6). Although persistent levels of DDE were recorded in this study, a significant long term decline has been reported for other European bird species, with the steepest declines generally occurring before 1990 (Newton et al., 1993; Fasola et al., 1998). DDE levels in merlin eggs in Britain during the 1970s and 1980s were an order of magnitude greater than those found in this study (Newton and Haas, 1988). Levels were also lower than those reported for dipper eggs in Scotland and Wales between 1988 and 1990 (Ormerod and Tyler, 1992), and most likely represent persistent background levels.

TDE is a less commonly studied metabolite of DDT, and little data exist on its occurrence in birds' eggs. In this study, it was less widespread than DDE and although a decline was found with time, this was most likely the result of a single peak in contamination levels in 1990 (Figs. 3 and 6). With the exception of 1990, TDE levels varied from below the limit of detection to <1.0 ppm in lipid.

The occurrence of HCB, gHCH (lindane, formerly used in forestry) and HEOD (dieldrin, used in sheep dip and seed dressings) was investigated. Although the impact of these compounds on bird populations declined during the 1980s, they nonetheless represent contaminants introduced through human activity (Newton and Haas, 1988). These organochlorines were found in lower proportions of sampled eggs than was DDE. However, the occurrence of HEOD in dipper eggs in south-west Ireland was widespread, presumably due to high sheep numbers (8–11 million animals in the country; Morgan and O'Toole, 1992). The incidence of this compound increased from 1990, peaked during 1993 and 1994, and decreased again in 1999 (Fig. 4). Of all organochlorines analysed, HEOD was found in the highest concentrations, and these varied considerably between years (Fig. 6). HEOD levels, particularly in 1993 (Fig. 7B) heavily influenced the overall pattern of organochlorine contamination. HEOD was commonly used as a sheep dip in Ireland up to the early 1980s, when its use was banned. However, the possibility of persistent residues and illicit use resulting in continuing or sporadic contamination of surface water exists (O'Halloran et al., 1993). It is interesting to note that the high values in 1993 were recorded in three separate rivers 20–30 km apart. Therefore, it is likely that reasons for the increased level in that year are perhaps related to environmental conditions (e.g. temperature or some biological activity) leading to the release of HEOD into surface waters. These results suggest an increase in the presence of this contaminant in riverine ecosystems in south-west Ireland above background levels during this time.

Lindane (gHCH), an organochlorine considered unlikely to have toxic effects on birds (Blus et al., 1985), occurred only occasionally in dipper eggs in this study. Levels were also relatively low (<1 ppm in lipid, Fig. 6) with the exception of one egg sampled in 1991. This organochlorine was widely used in the forest industry to combat pine weevils up to the 1990s, when its use was discontinued. However, because of its persistent nature it is likely to occur in the environment for a number of years. HCB was relatively more widespread and showed an increase in percentage occurrence from less than 10% in 1990 to over 80% in 1994 (Fig. 4). This increasing trend was mirrored by levels of HCB, the mean concentration of which peaked also in 1994 at 0.08 ppm in lipid (Fig. 6).

PCB residues at sub-lethal levels are believed to disrupt normal reproduction in birds (Barron et al., 1995). PCBs were present in almost all dipper eggs sampled in this study (Fig. 4). The widespread occurrence of PCBs (and some other organochlorines) with no apparent trend between stream headwaters and downstream areas, suggest that in Ireland there are no point sources of these pollutants, but rather a widespread low level of contamination. Although two of the most toxic PCB congeners (PCB 77 and PCB 126) were looked for in 4 of the 6 years, they were not detected. Concentrations of PCBs showed a consistent spread, generally <2 ppm in lipid. These levels, although persistent and showing no sign of decrease, are markedly lower than levels reported for merlin eggs in Norway during the 1970s and 1980s (Nygård, 1999). Levels of PCBs in dipper eggs in south-west Ireland were lower than those reported for eggs collected in Scotland and Wales during the 1990 breeding season (Ormerod and Tyler, 1992; Ormerod et al., 2000). A downward trend in total PCB contamination during the past few decades has been reported (Bignert et al., 1995; Barrett et al., 1996; Braune et al., 2001).

The dominant congeners in dipper eggs collected during this study were PCBs 138, 153 and 180, with PCBs 101, 118 and 170 also making a significant contribution to the total burden. The contribution of PCBs 138, 153 and 180 declined in 1999, with this year differing markedly in their signatures from all other years. These congeners are also the most commonly occurring in other bird species (Mason et al., 1997). Interestingly, Ormerod et al. (2000) also reported high levels of PCBs 138, 153 and 180 in dipper eggs in Wales. They suggested that Arochlor 1260, a common constituent of transformer and capacitor oils, was responsible for this pattern. Whether this is also the case in Ireland is unknown. The high level of PCB 118 in 1990 is also interesting and it had a major influence on the pattern of congeners found (Figs. 6 and 7B), though its frequency of occurrence has been much lower in recent years (Fig. 5).

Despite the widespread presence of organochlorines (and mercury) in eggs, with no evidence of an obvious point source of pollution, there appears to be no significant impact on dipper populations. A recent analysis (O'Halloran et al., 1999) showed no major change in dipper populations in the last decade in this study area.

However, what is not clear is what effect chronic exposure to such mixtures of pollutants will have on aquatic birds such as dippers in the long term. As Ireland embarks on considering incineration for waste management, the prospect of increased organochlorine burdens in the environment will grow. This possible input coupled with continued and cumulative exposure may lead to measurable effects in the future. The data presented here will provide a baseline essential to the tracking of future changes in contaminant loads in river systems in south-west Ireland.

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