

Behavioral variation and its consequences during incubation for American kestrels exposed to polychlorinated biphenyls

Sheri A. Fisher^a, Gary R. Bortolotti^{a,*}, Kimberley J. Fernie^b, David M. Bird^c, Judit E. Smits^d

^aDepartment of Biology, University of Saskatchewan, 112 Science Pl., Saskatoon, Sask., Canada S7N 5E2

^bEnvironment Canada, 867 Lakeshore Road, Burlington, Ont., Canada L7R 4A6

^cAvian Science and Conservation Centre, McGill University, Ste. Anne de Bellevue, Quebec, Canada H9X 3V9

^dDepartment of Veterinary Pathology, University of Saskatchewan, Saskatoon, Sask., Canada S7N 5B4

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Abstract

We investigated whether polychlorinated biphenyl (PCB) exposure in American kestrels (*Falco sparverius*) influenced incubation behavior and whether altered behavior could lead to poor reproductive success. Captive kestrels were fed a mixture of PCBs (Aroclors 1248:1254:1260) at an approximate daily dose of 7 mg/kg body weight, 1 month prior to pairing and throughout incubation. Behaviors of 23 control and 23 PCB-exposed pairs were monitored throughout incubation using an electronic balance in the nest box. PCB exposure resulted in longer incubation periods and in altered incubation behaviors. Seven of 14 behavioral variables showed some association with treatment, with sex-specific effects largely biased toward disrupted male behavior. For most behaviors, the treatment effect was explained by the delayed clutch initiation induced by PCBs rather than by a direct physiological impact of the contaminants. PCB-exposed pairs with greater attendance to their eggs and better coordination of incubation duties had improved hatching success.

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1. Introduction

Polychlorinated biphenyls (PCBs) are persistent, widespread environmental contaminants. As PCBs bioaccumulate, birds at higher trophic levels of the food chain are particularly vulnerable to PCB exposure (Elliott et al., 1991; Barron et al., 1995; Hoffman et al., 1996a). Historical population declines of raptors have been attributed to contaminant-induced reproductive failure (e.g., Koze and Anderson, 1991; Clark et al., 1998; Valkama and Korpi-maki, 1999). The breeding cycle and associated behaviors that are under hormonal control may be vulnerable to endocrine-modulating substances such as PCBs (Barron et al., 1995; Vos et al., 2000), especially since slight hormonal differences can elicit significant behavioral change (Silver and Ball, 1989; Jacobs and Wingfield, 2000). Behavioral

observations rather than intrusive physiological examinations are increasingly being used and encouraged in toxicological research to identify adverse effects of contaminants (Doving, 1991; Evangelista de Duffard and Duffard, 1996; Clotfelter et al., 2004; Zala and Penn, 2004).

Altered behavior during the breeding season has been observed in birds exposed to organochlorine contaminants, including PCBs. Reduced nest defence behavior occurred in merlins (*Falco columbarius*) (Fyfe et al., 1976). Peregrine falcons (*F. peregrinus*) exposed to dichlorodiphenyldichloroethane displayed clumsy behavior in the nesting scrape, including stepping on the eggs (Nelson, 1976). Egg-destroying behavior occurred in contaminated gray herons (*Ardea cinerea*) (Milstein et al., 1970) and captive mallards (*Anas platyrhynchos*) (Risebrough and Anderson, 1975). McCarty and Secord (1999a) found that tree swallows (*Tachycineta bicolor*) in PCB-contaminated areas built low-quality nests, which were associated with reduced breeding success. Atypical parental behavior has also been suggested

*Corresponding author. Fax: +1 306 966 4461.

E-mail address: gary.bortolotti@usask.ca (G.R. Bortolotti).

to contribute to decreased reproductive success in cormorants (*Phalacrocorax carbo*) contaminated with PCBs, chlorinated dioxins, and dibenzofurans (Van Den Berg et al., 1995).

Incubation behavior can be disrupted by sublethal exposure to environmental pollutants (Fry, 1995), including a lack of behavioral synchrony between the sexes which normally ensures that eggs are not left unattended (Koenig, 1982). Unattended nests are exposed to greater risk from predation, and embryos could die if exposed to a suboptimal thermal environment (Webb, 1987). Because the cooling rate of an unattended egg exceeds the heating rate when parents return, absences greater than about 1 min are also energetically wasteful to the parent (Drent, 1973). Exposure of parent birds to PCBs has been linked to lengthened and more frequent recesses, prolonged incubation, and increased incidence of nest abandonment (Peakall and Peakall, 1973; Fox et al., 1978; McArthur et al., 1983; Kubiak et al., 1989; McCarty and Secord, 1999b; Bustnes et al., 2001).

Our study subject, the American kestrel (*Falco sparverius*), is a biparental incubator (Balgooyen, 1976; Bortolotti and Wiebe, 1993). We were able to monitor breeding pairs for continuous periods of time throughout incubation, whereas many previous studies of contaminated birds have used infrequent nest checks or monitoring for only short segments (Peakall and Peakall, 1973; Kubiak et al., 1989; Harris et al., 1993; Custer et al., 1998; Bustnes et al., 2001). In addition, and unlike some studies, we could easily differentiate males from females, enabling us to reliably investigate sex-specific effects. Complete data on reproductive performance also allowed us to investigate the consequences of variation in incubation. We predict that pairs with greater attendance to their eggs, and better coordination of their incubation duties, should have greater hatching success.

2. Materials and methods

2.1. Study animals

This study took place at the Avian Science and Conservation Centre of McGill University, Canada, using American kestrels of known pedigree and age. Birds were randomly designated control (CTL) ($n = 25$ pairs) or PCB-exposed (PCB) ($n = 25$ pairs) and were placed in flight pens ($6 \times 6 \times 2.5$ m) segregated by sex and treatment. The kestrels were fed ad libitum on their typical diet of day-old cockerels.

Based on the PCB congeners found in the eggs and tissues of wild birds from the Great Lakes region (Braune and Norstrom, 1989; Clark et al., 1998) and PCB residue levels found in wild prey species of kestrels (Environment Canada, unpublished data), a dosing regime that would generate environmentally relevant levels of PCBs was calculated (Fernie et al., 2000). Chronic dietary exposure began 1 month before pairing on 18 March 1998 and

continued until the end of the incubation period of each pair (range 79–117 days of exposure; mean 95 days). A mixture of Aroclors 1248:1254:1260 (1:1:1 by weight; Monsanto, St. Louis, MO, USA) was dissolved in safflower oil at a concentration of 4.85 mg/g total PCB. As kestrels show a preference for consuming the heads of cockerels (I. Ritchie, pers. comm.), 100- μ l aliquots of either PCB dosing mixture or plain safflower oil were injected into the heads of frozen–thawed day-old cockerels to be fed to the PCB-exposed or control groups, respectively. The birds consumed approximately 7 mg/kg body weight/day (Fernie et al., 2000; Drouillard et al., 2001). The total PCB residue levels from the kestrel eggs averaged 34.1 μ g/g (geometric mean) on a whole-egg wet-weight basis (Fernie et al., 2000). These levels are comparable to those found in the eggs of wild raptors exhibiting decreased reproductive success (Hoffman et al., 1996b; Clark et al., 1998; Valkama and Korpimäki, 1999).

Genetically unrelated birds, all with previous breeding experience, were paired on 21 April 1998. The pairs were placed into outdoor breeding pens ($2.3 \times 0.9 \times 3.6$ m), which contained rope and wooden perches, a one-way glass window for observation, and a nesting box.

2.2. Monitoring of incubation behavior

The incubation period was defined as the time from the completion of the clutch to the hatching of the first egg or until 28 days after clutch completion for pairs that failed to hatch any egg. A custom-built electronic balance system was used to quantify behavior. The monitoring system consisted of a wooden incubation box ($20.3 \times 17.8 \times 10.2$ cm) mounted onto a balance. Electrical impulses proportional to the mass of the incubation box were sent from the balance to a microchip once every minute, 24 h per day. Twenty electronic balances were randomly assigned to PCB-exposed or control pens. Every 5 days during the incubation period balances were switched between pens of the same treatment group to increase sample size (PCB $n = 23$; CTL $n = 23$). One PCB-exposed pair did not complete a clutch and was not monitored, while another PCB-exposed pair abandoned their clutch. Two control pens were not monitored to spread the effort equally between treatments. When pairs were not being electronically monitored, a wooden incubation box with the same dimensions was placed in the nest box. The bottom of the incubation box was filled with 2–4 cm of wood shavings. A door on the back of the nest box facilitated nest checks and removal of balances. Nest boxes were checked daily, and eggs were numbered to identify laying sequence. Ten days after clutch completion, eggs were candled to determine fertility and one egg was removed from each pair for PCB residue analysis (see Fernie et al., 2000). Another fresh but nonviable kestrel egg was put into the box to maintain the original clutch size.

Female kestrels were heavier than their mates, allowing us to use mass to determine the sex of the bird in the nest

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