

## Efficacy of diatomaceous earth at reducing populations of nest-dwelling ectoparasites

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**ABSTRACT.** Nest-dwelling parasites are known to have detrimental consequences for many birds, and thus it may be desirable for management practices to remove these parasites. In addition, manipulation of parasite loads is often necessary for studying host-parasite evolution. While there are various methods that are effective at reducing parasite loads in nests, many of them have shortcomings because they require specialized equipment, are time consuming, or involve toxic substances that may be dangerous to both birds and researchers. I evaluated the efficacy of simple-to-use, non-toxic diatomaceous earth (DE) at reducing ectoparasite loads in nests of Tree Swallows (*Tachycineta bicolor*) over two field seasons. Diatomaceous earth significantly reduced population sizes of bird fleas (*Ceratophyllus idius*) and various species of blow flies (*Protocalliphora* spp.) in both years. Diatomaceous earth appeared to be more effective at reducing the numbers of fleas, possibly because fleas have smaller body size and so are more easily desiccated by the abrasive action of DE. Although DE was effective against both fleas and blow flies, it did not completely rid nests of either type of parasite.

### SINOPSIS. Eficacia de diatomeas terrestres en reducir las poblaciones de ectoparásitos en nidos

Se conoce que los ectoparásitos de nidos tienen consecuencias perjudiciales para muchas aves y como práctica de manejo es deseable poder remover los mismos. Además la manipulación de la carga de parásitos es necesaria para estudiar los aspectos evolutivos de la interacción huésped-parásito. Aunque hay varios métodos para reducir la carga de parásitos en nidos, muchos de estos tienen limitaciones porque requieren de equipo especializado, consumen mucho tiempo o envuelven sustancias tóxicas que pueden ser peligrosas tanto para las aves como para los investigadores. Evalué la relación de diatomeas terrestres, para reducir la carga de parásitos en nidos de *Tachycineta bicolor*, durante dos temporadas de trabajo de campo. La diatomeas redujeron significativamente las poblaciones de pulgas (*Ceratophyllus idius*) y varias especies de moscas (*Protocalliphora* spp.). Las diatomeas parecen ser más efectivas en reducir el número de pulgas, posiblemente porque las pulgas tienen un tamaño corporal menor y por lo tanto son más propensas a la desecación por la acción abrasiva de las diatomeas. Aunque las diatomeas fueron efectivas contra las pulgas y las moscas, no eliminaron completamente a estos ectoparásitos de los nidos.

*Key words:* birds, blow flies, diatomaceous earth, ectoparasites, fleas, *Tachycineta bicolor*, Tree Swallow

Parasites have been recognized as playing a key role in the behavior, ecology and evolution of birds (Clayton and Moore 1997). In particular, nest-dwelling ectoparasites have been shown in numerous studies to have detrimental consequences for their hosts. Such effects have included delayed breeding dates, reduced clutch sizes and hatching success, decreased survival of parents, differential natal dispersal, and abandonment of entire breeding colonies (e.g., Brown and Brown 1986, 1992; Norcross and Bolen 2002; Oppliger et al. 1994; Richner and Tripet 1999; reviewed in Møller 1997). Much of the research that has been performed on the relationship between ectoparasites and birds has used cavity nesting species as models, with many of these studies focusing on species using

artificial nest boxes. Some researchers routinely remove nesting material from nest boxes at the conclusion of the breeding season, leading Møller (1989) to warn that because such practices may result in unnaturally low populations of ectoparasites in nests, some of the conclusions reached in these studies may be erroneous. While clearly a valid concern, the use of nest boxes has also allowed researchers to experimentally manipulate levels of infestations by ectoparasites, which has contributed to a greater understanding of the evolution of both hosts and parasites.

To experimentally manipulate the number of ectoparasites in nests, researchers have often treated nests with chemical fumigants such as organophosphates or carbamates. While these chemical fumigants have had the desired results, there is some concern over their potential ef-

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fects not only on the birds but also on the researchers applying these treatments (Jackson 1985). Many of these chemicals are known to be toxic or disrupt normal endocrine functions (Fairbrother 1996; Yamamoto et al. 1996), and their repeated use may result in the development of resistance by the ectoparasites. Alternatively, some researchers have successfully removed ectoparasites by subjecting nests to heat treatments (Richner and Tripet 1999) or physically removing individual parasites (Whitworth and Bennett 1992). These methods are also effective, as well as safe, but their use may be precluded in many field situations due to logistical constraints. As a result, there is a need for a fast, effective, and safe method of removing parasites from bird nests that does not require specialized equipment or electrical power, and therefore one that is usable in most field studies.

Parasites within bird nests are of interest to ornithologists and evolutionary ecologists, but insects living in other mediums are also of great practical importance. For example, in many parts of the world, stored crops are damaged by insect pests (Stathers et al. 2002). In general, many crop producers are hesitant to use synthetic chemical pesticides and instead have begun to utilize naturally occurring inert dusts such as diatomaceous earth to control insects in stored crops (Stathers et al. 2002). Diatomaceous earth (DE) consists of fossilized diatoms and is made up of almost pure amorphous silicon dioxide. It is stable, produces no toxic residues, and has been used in commercial water purification and as a feed supplement to clear internal gastro-intestinal parasites in cattle and pets (Allen 1972; Korunic 1998). Moreover, DE has been shown to be non-toxic for mammals (Quarles 1992) and presumably also is non-toxic for avian species. It operates either by abrading the exoskeleton of the insect, or by absorbing waxy fats and lipids from the cuticle of the insect; in both cases the death of the insect results as a consequence of water loss (Ebeling 1971).

Besides being effective at reducing insect pests in stored crops, DE holds great potential to be an effective treatment for reducing parasite loads in nests of birds. Although some researchers and operators of nest box programs use DE as means to reduce parasite loads in nests (T. L. Whitworth, pers. comm.), to my

knowledge its efficacy has not been evaluated in birds. The goal of this study was to investigate how DE performed against two common types of ectoparasite, blow flies and bird fleas, in the nests of Tree Swallows (*Tachycineta bicolor*). Given that ectoparasites may have detrimental consequences for offspring survival and mortality in many species (Møller 1997), the efficiency of a non-toxic insecticide for reducing ectoparasite loads is of interest not only to researchers studying host-parasite evolution, but also to those individuals operating nest box programs for population enhancement. I also investigated whether parasite loads were affected by attributes of the nest itself, in particular the amount of feathers and other nesting material that parent swallows brought into the nest boxes.

## METHODS

I studied Tree Swallows breeding in artificial nest boxes during 2001 and 2002. The study area was located near Prince George, British Columbia, Canada (53°N, 122°W), and consisted of grazed pasture land interspersed with small wetlands, surrounded by a mosaic of forested lands in various stages of regeneration following logging. Approximately 190 nest boxes were mounted on fence posts and there was little variation among boxes in habitat as all were located in pastures. I cleaned all old nesting material from boxes prior to the start of this study. In 2001, the owner of the nest boxes had placed several centimeters of clean wood shavings in the bottom of 22 of the 33 nest boxes I studied. These shavings originated from aspen (*Populus tremuloides*), a species in which the wood is not known to have insecticidal properties (Johnson et al. 1995). No wood shavings were used in 2002.

Beginning in early May, nest boxes were visited every two days until egg laying commenced (late May to early June), and were then visited daily until laying was complete. Once incubation had begun, I sequentially allocated nests to either a control or treatment group. Allocations occurred in an alternating fashion such that as laying was completed at two nests, one would be assigned to the control group while the other would become part of the treatment group. Therefore, nests were initially distributed among control and treatment groups evenly

with regard to breeding phenology. Nests that were depredated, abandoned or that failed before young were 12 d old (see below) were excluded from analyses; in total, data were collected from 55 nests during this study (2001, 18 control, 15 treated with DE; 2002, 14 control, 8 treated with DE). At treatment nests, I removed all eggs from the nest box on day 10 of incubation and lightly dusted the nesting material with approximately 2 g of commercial insecticide that contained diatomaceous earth. The dust was gently worked into the nesting material and then the eggs were placed back in the nest. Different insecticides were used during incubation in each year of the study. In 2001, I used Drione Crawling Insect Killer (Wilson Laboratories, Dundas, Ontario, Canada) that contained 38.12% diatomaceous earth as well as 0.2% pyrethrins and 1.0% piperonyl butoxide. Pyrethrins are insecticides derived from the flowers of chrysanthemum and have low toxicity for birds and mammals, and piperonyl butoxide acts as a synergist (Jackson 1985). During the incubation period in 2002, Insectigone (Chemfree Environment Inc., Kirkland Quebec, Canada) earwig killer, containing 80% silicon dioxide was used. At control nests, eggs were removed and replaced in a similar manner as treatment nests, and nesting material was also handled in a comparable manner to treatment nests.

Nests were also visited near the predicted date of hatching to determine date of emergence of the first nestling and the number of eggs that successfully hatched. When nestlings were 6 d old (hatching date = day 0), I repeated the insecticide application at treatment nests; however, in both years only Insectigone diatomaceous earth was used. Nestlings were removed prior to treatment and placed back after nests were dusted. Nestlings were also removed and replaced in a similar manner at control nests. At all nests, I banded each nestling with a standard aluminum leg band when they were 12 d old. Tree Swallows fledge between 18 and 22 d old (Robertson et al. 1992).

Between 22 and 24 d after hatching (i.e., after young had fledged), all nesting material was carefully removed from nest boxes and placed in individual plastic bags. Material was collected only from nests where young survived to at least 12 d of age; if nests fail before this, then there is insufficient time for larval devel-

opment of blow flies (*Protocalliphora* spp.). Nests were maintained at room temperature in the lab for a period of at least 2 wks to allow all adult *Protocalliphora* to emerge from pupae. Nests were then frozen for 24 h, transferred to paper bags, and dried in an oven at 80°C. overnight to kill any fleas or *Protocalliphora* that remained alive. Material from each nest was weighed and then teased apart to count ectoparasites. Total number of adult fleas and total number of pupae or pupal cases of *Protocalliphora* were determined for each nest. In addition, the mass of wood shavings (2001 only) and feathers in each nest was also quantified.

**Statistical analysis.** I tested whether treating nests with diatomaceous earth affected the number of fleas or *Protocalliphora* using analysis of covariance. Parasite number was the dependent variable, while treatment and year were categorical independent variables. As covariates in these models, I used hatching date (where 1 January = 1), brood size at day 12, mass of feathers, and mass of all nesting material. I also included all possible two-way interactions. All interactions and terms, except treatment, that did not approach significance ( $P > 0.10$ ) were iteratively removed from the models and the analyses repeated to obtain the most parsimonious models. To meet assumptions of normality, both number of fleas and *Protocalliphora* were log-transformed before analysis. As the goal of this study was to evaluate the efficacy of diatomaceous earth, I was also concerned that the treatment of nests during incubation in 2001 may have affected the results because pyrethrins were present in the insecticide as well as diatomaceous earth. However, nests during incubation probably contained relatively few ectoparasites. Pyrethrins also have a short half-life and quickly degrade in both air and light, so such a bias seemed unlikely. Nonetheless, if such a bias was present, then a significant interaction between year and treatment would have been expected.

There were two other potential biases in my data set. First, some nests in 2001 had a layer of wood shavings in the bottom of the nest box. Although it is unlikely that the aspen shavings were detrimental for ectoparasites, I investigated this potential bias by excluding the nests from 2001 where wood shavings were present and re-analyzing the data. I also analyzed the data from 2001 only and included the presence

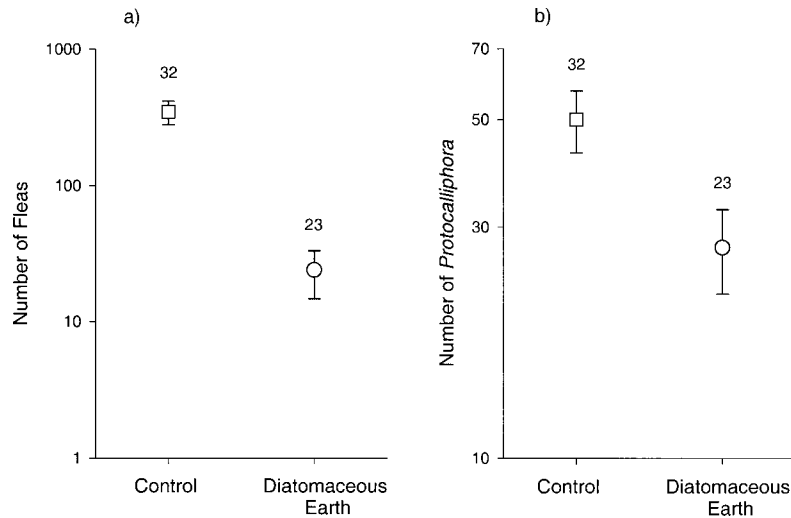


Fig. 1. Number ( $\pm$  SE) of (a) fleas (*Ceratophyllus idius*) and (b) blow flies (*Protocalliphora* spp.) recovered from nests of Tree Swallows that were treated either with diatomaceous earth or acted as a control. Note that numbers of fleas and *Protocalliphora* are plotted on a logarithmic scale. Sample sizes above error bars refer to number of nests.

or absence of wood shavings as a factor in the model. Second, even though all nest boxes were thoroughly cleaned at the end of 2001, it was also possible that parasite loads of nests in 2002 could have been affected by the presence of the insecticide from treatment that occurred during the previous year. Of the 22 nests studied in 2002, only four had been treated with insecticide in 2001. To ensure that these four nests did not alter my results, I also analyzed the data after excluding these nests from the data set.

Analyses were performed using SPSS (Norusis 2000). Means ( $\pm$  1 SE) are presented and two-tailed tests were considered significant at the 0.05 level.

## RESULTS

I detected one species of bird flea (*Ceratophyllus idius*) and at least four species of blow fly (*Protocalliphora braueri*, *P. occidentalis*, *P. bennetti*, and *P. rugosa*) in nests of Tree Swallows. The abundance of fleas in a nest was positively related to the number of *Protocalliphora* ( $r = 0.39$ ,  $N = 55$ ,  $P < 0.01$ ). There were significantly fewer fleas in nests treated with diatomaceous earth than in control nests ( $F_{1,53} = 50.16$ ,  $P < 0.001$ ; Fig. 1). Diatomaceous earth had a similar effect for *Protocalliphora*, with fewer being detected in nests treated with DE

than in control nests ( $F_{1,52} = 15.17$ ,  $P < 0.001$ ; Fig. 1). In addition, the number of *Protocalliphora* increased significantly with increasing brood size ( $F_{1,52} = 16.91$ ,  $P < 0.001$ ; Fig. 2). Year, hatching date, mass of feathers, and mass of nesting material, as well as interactions, did not contribute significantly to statistical models for data on either fleas or *Protocalliphora*.

When I excluded data for four nest boxes from 2002 that were treated in 2001, results were similar for both numbers of fleas (treatment:  $F_{1,49} = 43.17$ ,  $P < 0.001$ ) and *Protocalliphora* (treatment:  $F_{1,48} = 15.91$ ,  $P < 0.01$ , brood size at day 12:  $F_{1,48} = 17.54$ ,  $P < 0.001$ ). This suggests that residual effects of insecticide treatment in 2001 were not influencing my results.

Models were developed using data from only 2001 to investigate whether the presence of wood shavings influenced parasite numbers. Nests with diatomaceous earth had significantly fewer fleas than control nests ( $F_{1,30} = 30.53$ ,  $P < 0.001$ ), but I could not detect any effect attributable to the presence or absence of wood shavings (number in nests with wood shavings,  $272.3 \pm 74.3$ ; without wood shavings,  $210.2 \pm 109.2$ ;  $F_{1,30} = 2.41$ ,  $P = 0.13$ ). Results for *Protocalliphora* in 2001 were similar to those using the entire data set; parasite numbers increased with brood size at age 12 d ( $F_{1,29} =$

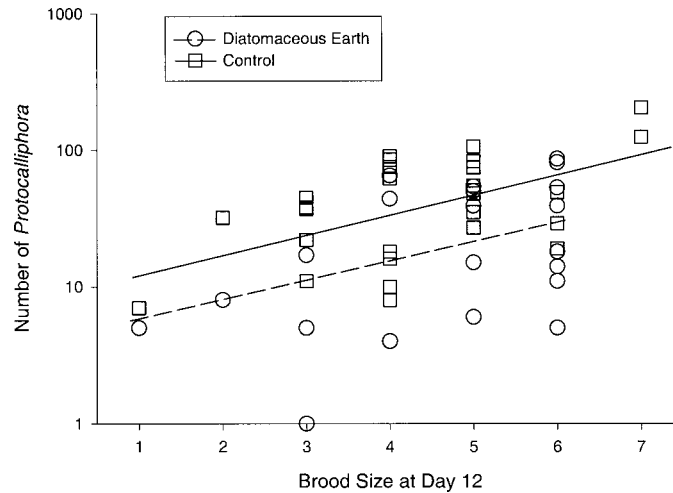


Fig. 2. Relationship between brood size when nestlings were 12 d old and number of *Protocalliphora* recovered from nests of Tree Swallows treated with diatomaceous earth or that acted as controls. Number of *Protocalliphora* is plotted on a logarithmic scale.

20.60,  $P < 0.001$ ), and there were fewer parasites in nests with diatomaceous earth ( $F_{1,29} = 13.64$ ,  $P = 0.001$ ). The presence or absence of wood shavings, however, did not significantly influence abundance of *Protocalliphora* found in nests (number in nests with wood shavings,  $49.8 \pm 8.7$ ; without wood shavings,  $39.9 \pm 12.8$ ;  $F_{1,29} = 0.02$ ,  $P = 0.90$ ). When I excluded nests with wood shavings from the entire data set, the results did not change. Both numbers of fleas ( $F_{1,31} = 37.28$ ,  $P < 0.001$ ) and *Protocalliphora* ( $F_{1,30} = 5.96$ ,  $P = 0.02$ ) were significantly lower in DE-treated nests, and numbers of *Protocalliphora* also increased as brood size at day 12 became larger ( $F_{1,30} = 6.13$ ,  $P = 0.02$ ). Collectively, these results suggest that the presence of wood shavings did not confound my initial results showing the effectiveness of treating nests with DE to reduce populations of parasites.

## DISCUSSION

My results showed that diatomaceous earth is an effective treatment that reduces populations of fleas and *Protocalliphora* in nests of Tree Swallows. Although DE was effective for both types of parasites, it appears that it reduced numbers of fleas more than it did *Protocalliphora* (Fig. 1). Korunic (1998) suggested that in general, DE was more detrimental for insects

with relatively small body size, possibility because they have a larger surface area to volume ratio and so lose a larger relative amount of water compared to larger insects. Flanders (1941) also suggested that insects that are better able to recover lost water will be less affected than those that must metabolize water from their food sources. Both fleas and blow flies are obligate blood feeders, and the larger body size of *Protocalliphora* larvae may allow them to replenish lost water faster and more efficiently compared to adult fleas. It has also been reported that DE is more effective against insects that have relatively rough or setaceous surfaces, probably because such surfaces are more likely to retain DE particles and so cause greater damage to the insect's cuticle (Korunic 1998). Any of these explanations would be sufficient to account for the larger effect of DE on fleas, but they are not mutually exclusive and further research is needed to adequately address this issue.

While DE was effective, it did not completely rid nests of either type of parasite. The lack of complete eradication, and the possible reduced success of DE in controlling *Protocalliphora*, may be a consequence of the fact that DE has detrimental effects not only on these specific insects, but also on other insects that are their natural enemies, such as parasitoids. Stathers et al. (2002) found that in some cases,

insect pests in stored crops were actually more abundant in DE-treatments than in controls, and they attributed these differences to reductions in populations of predators of the pests. Predators and parasitoids may be killed outright by DE, or in the presence of DE may spend more time grooming than they do seeking hosts or prey (Perez-Mendoza et al. 1999). While complete eradication of nest-dwelling parasites may be a goal of some studies, in some cases it is probably undesirable to completely eliminate all parasites from nests. If all parasites are removed from nests, then a potentially important selective pressure may also inadvertently be removed. These benign environments may promote the retention of certain traits in birds that could be maladaptive in more natural environments (see Heath et al. 2003).

Some individuals who monitor nest-box breeding populations of birds often use wood shavings to line boxes. This practice is especially common when species do not bring nesting material into boxes, such as occurs for example with falcons (e.g., Bortolotti 1994). Some of my nest boxes in 2001 had a layer of wood shavings lining the bottom, but my results suggest that this did not influence ectoparasite loads. The benefits of providing this nesting substrate, for example in terms of reducing damage to eggs, appear to outweigh any detrimental effects that might have been associated with ectoparasite loads. Tree Swallows typically build a grass nest and then line the cup with feathers (Robertson et al. 1992). While these materials might be expected to provide a safe refuge for ectoparasites, or conversely may protect nestlings from exposure to ectoparasites (Winkler 1993), I could detect no influence of these variables on either numbers of fleas or *Protocalliphora*.

I detected a significant increase in the number of *Protocalliphora* with increasing brood size (Fig. 2). Whitworth and Bennett (1992) reported that a relatively few individual *Protocalliphora* larvae can completely exsanguinate nestlings in a short period of time. Therefore, if there are more nestlings within a nest, there are more resources for larvae to exploit, allowing larger population sizes of the parasite. It is unclear why a similar relationship between brood size at day 12 and the number of fleas was not detected. However, given that fleas consume much less blood than *Protocalliphora*,

it is probable that they are less affected by the number of hosts within a nest; other forms of regulation may be more important for determining population sizes of fleas.

In summary, DE is an effective method to reduce populations of parasites in the nests of birds (Fig. 1). Given that DE has been approved in several countries as an additive in food and grain products (Korunic 1998), it is also a safe alternative to many commonly used chemical insecticides; however, because DE acts through desiccation, some caution is necessary. Walther and Clayton (1997) used a series of experiments designed to evaluate various methods of recovering ectoparasites directly from the plumage of living birds, and showed that powders containing DE can be detrimental if applied directly to the plumage of a bird. They demonstrated that when birds were experimentally wetted that it took significantly longer for the plumage of birds treated with dusts containing DE to dry compared to control birds and those treated with other types of dusts (Walther and Clayton 1997). This phenomenon is probably the result of DE removing oils from the birds' plumage, and treated birds may thus be more susceptible to exposure if they encounter wet, inclement weather soon after being dusted (Walther and Clayton 1997). While caution should be exercised when using any type of treatment, it is also important to note that Walther and Clayton's (1997) experiments were designed for a different purpose than my study, and they used a "dust-ruffling" technique where dusts were worked directly into the plumage of the birds. Both nestling and parent birds in DE-treated nest boxes are unlikely to be affected to the same extent, as the DE was worked into the nesting material and not onto the birds themselves. Diatomaceous earth is therefore unlikely to cause the detrimental effects that were identified by Walther and Clayton (1997).

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