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IS THE INGESTION OF SMALL STONES BY DOUBLE-CRESTED CORMORANTS A SELF-MEDICATION BEHAVIOR?

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Abstract. Many birds ingest small stones to aid in the grinding up of coarse food items in the diet. However, small stones are also found in the stomachs of species with diets consisting of soft food items, and may serve to mechanically remove parasites attached to the stomach wall. We sampled Double-crested Cormorants (*Phalacrocorax auritus*) at two sites to examine whether ingestion of small stones served an antiparasite function and whether this behavior differed between the sexes. At a Lake Ontario site, females more often had small stones in their stomachs and were less parasitized by nematodes than were males, and males with small stones had fewer nematodes than males without small stones. We did not find similar patterns in small stone presence or parasitism at a Lake Erie site; however, Lake Erie birds had fewer parasites and lower proportions of birds with small stones. These results suggest that ingestion of small stones by Double-crested Cormorants might indeed serve an antiparasite function.

Key words: cormorant, endoparasitism, lithophagy, nematode, self-medication.

¿Es la Ingestión de Pequeñas Piedras un Comportamiento de Automedicación de *Phalacrocorax auritus*?

Resumen. La mayoría de las aves ingieren piedras pequeñas para ayudarse en la trituration de los alimentos duros. No obstante, también se han encontrado piedras en los estómagos de especies cuya dieta consiste de alimentos blandos, al parecer como mecanismo de remoción de parásitos adheridos a la pared intestinal. En el presente estudio muestreamos a individuos de *Phalacrocorax auritus* en dos sitios, para investigar si la ingestión de piedras tiene una función antiparasitaria y si además, existen diferencias entre sexos. Los resultados indicaron que para el sitio del lago Ontario, las hembras tuvieron más piedras y menos nematodos parásitos que los machos, mientras que los machos que presentaron piedras, tuvieron menos nematodos que los que no tuvieron piedras. No encontramos patrones similares con respecto a la presencia o ausencia de piedras y parasitismo en el sitio del lago Erie. Sin embargo, en este sitio las aves tuvieron menos parásitos y hubo menor proporción de individuos sin

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pedras. Estos resultados sugieren que la ingestión de piedras podría tener una función antiparasitaria.

Animals self-medicate by ingesting or sequestering plant or other materials, either to avoid or to counteract parasitic infections (Huffman 2003). The most well-known examples of self-medication are primates swallowing leaves, ostensibly to rid themselves of gastrointestinal worms (Huffman and Caton 2001), and birds adding specific vegetation to their nests to discourage ectoparasite populations (Gwinner and Berger 2005, Shutler and Campbell 2007). Dust-bathing in birds also might function as self-medication (Ehrlich et al. 1988). Such behaviors appear to have evolved independently in many lineages and throughout the world (Poulin 1995).

Lithophagy, the ingestion of small stones, and the related practice of geophagy, the ingestion of soil and clay, occurs in a variety of mammalian, reptilian, and avian species (Gilardi et al. 1999 and references therein, Wings 2007). Within geophagy, the ingestion of clay is thought to serve the function of nutritional supplementation (Brightsmith and Muñoz-Najar 2004) or neutralization of toxic secondary plant compounds (Gilardi et al. 1999). In comparison, the ingestion of soil has been associated with endoparasitism, but has been interpreted as a therapeutic or medicinal treatment of the ailments resulting from parasitic infections, rather than as a direct control of parasitic infections (Knezevich 1998).

Avian lithophagy appears to reflect diet; seed-eaters often have many small stones in their gizzards, whereas carnivores, omnivores, and frugivores less frequently have small stones and stones are less abundant when present (Gionfriddo and Best 1999). However, the presence of small stones in species with diets consisting of soft items suggests that ingestion of small stones might also serve other functions. For example, in waterbirds, small stones might act to control buoyancy, as suggested for other aquatic animals (Wings 2007).

Birds, especially top consumers, are hosts to a variety of nematode species (Owre 1962) that are commonly found attached to the stomach wall and which cause large ulcerations and deplete host resources (Huizinga 1971). It is possible that ingestion of small stones is a form of self-medication in birds. Stones might act to physically remove parasites attached to the stomach wall through mechanical abrasion, as has been proposed for pinnipeds by Hamilton (1933) and Emery (1963), and as has been observed occurring through hispid leaf-eating in primates—studies have reported large numbers of nematodes in the feces of individual primates after they consume hispid leaves (Huffman et al. 1996). Furthermore, leaf-eating is observed more often in individuals that are more heavily parasitized (Huffman et al. 1996, 1997), suggesting that these primates are actively combating the parasitic infection.

Double-crested Cormorants are aquatic birds that are predominantly piscivorous (Hatch and Weseloh 1999). Fish are more easily digested than seeds and grains; therefore, we might not expect to find small stones in the stomachs of most Double-crested Cormorants because of their reduced need for mechanical abrasion in digestion. However, Double-crested Cormorants are common hosts for a variety of parasites transferred trophically, including *Helminthes* (Huizinga 1971, Robinson et al. 2008). Therefore, they are a good model species to study whether the ingestion of small stones might serve a self-medication purpose.

We first addressed how often small stones were found in Double-crested Cormorants. However, the primary objective of this study was to determine if male Double-crested Cormorants (*Phalacrocorax auritus*) differed from females in the likelihood of having small stones in their stomachs, because a previous study found male-biased parasitism by *Contracaecum* spp. nematodes at a Lake

Ontario site (Robinson et al. 2008). Additionally, male vertebrates often have more parasites than females (Poulin 1996, Zuk and McKean 1996, Schalk and Forbes 1997). We hypothesized that small stones help to mechanically remove nematodes from the stomach wall or prevent their attachment. Thus, we predicted that either the more heavily infected males would have small stones to combat the established nematodes or have fewer small stones and higher parasite burdens. We also examined whether individual Double-crested Cormorants of either sex with small stones had fewer nematodes than individuals without such stones. Lastly, we considered possible alternative reasons for the ingestion of small stones, including a digestive function, a hydrostatic (buoyancy control) function, and accidental ingestion lacking a purpose.

METHODS

Double-crested Cormorants were collected in May of 2006 from Lake Ontario at Presqu'île Provincial Park (43°60'N, 77°43'W), Brighton, Ontario, and in May of 2008 from Lake Erie at Middle Island (41°41'N, 82°41'W). We started noting small stone presence or absence after 99 birds had been processed from a larger sample of 263 Double-crested Cormorants collected from Lake Ontario (Robinson et al. 2008). All subsequent birds were noted for presence or absence of small stones, giving a subsample of 164 adult Double-crested Cormorants from Lake Ontario, and a sample of 193 Double-crested Cormorants from Lake Erie. We individually bagged each gastrointestinal tract, which was then frozen at -40°C at the National Wildlife Research Centre (NWRC). The gastrointestinal tracts remained frozen until dissection (methods detailed by Forbes et al. 1999).

Birds were sexed by gonadal inspection and weighed to the nearest 0.1 g. The proventriculus and stomach region were examined for small stones and nematodes. Nematodes, if found, were washed in avian saline (0.85%) and enumerated. All nematodes (possibly two *Contracaecum* spp.) attached to the proventriculus and stomach walls were collected and representatives were deposited with the Canadian Museum of Nature (details given by Robinson et al. 2008).

Stomach contents were removed and small stones, if present, were separated from digestive contents (i.e., whole or partially digested fish and fish bones). All small stones were washed, air-dried, and counted. We described the shapes of the stones, based on the category characteristics of Best and Gionfriddo (1991), from three birds collected in 2006 and from 25 birds collected in 2008. Maxima for linear dimensions of diameter, width, and height of each stone were measured to the nearest 0.1 mm with digital calipers. We collectively weighed all stones found per stomach to the nearest 0.0001 g using a Mettler AE 166 analytical balance (Mettler Toledo, Inc., Columbus, Ohio) and calculated the percentage of total body mass represented by the stones. We also recorded other pertinent data, and performed other analyses, for our Lake Erie birds, which were collected and processed after Lake Ontario birds. We recorded whether there were unattached, partially digested *Contracaecum* spp. in the intestine and weighed the digesta separately from small stones to the nearest 0.1 g.

STATISTICAL ANALYSES

We used chi-square tests to compare the percentage of males and females with small stones, and the percentage of both sexes with small stones in 2006 (Lake Ontario) and 2008 (Lake Erie). We used median tests to determine whether the presence or absence of small stones was associated with differences in median numbers of *Contracaecum* spp. nematodes in either sex at either site. We used median tests because *Contracaecum* spp. abundance

was not normally distributed. We used Spearman rank correlations to determine the relationship between the mass of small stones and the mass of digesta in the stomach. We also compared the presence or absence of small stones between birds that had empty stomachs and birds that had digesta remaining, using a chi-square test. We report median values with their associated 25% and 75% interquartile ranges. We used JMP® statistical software (SAS Institute, Inc., Cary, North Carolina) for all statistical analyses. Results were considered significant at $P < 0.05$.

RESULTS

Twenty-seven (17%) Lake Ontario birds had small stones compared to 25 (13%) Lake Erie birds. Small stones ranged in size from maximum diameters of 1.0 mm to 20.4 mm, maximum widths of 1.0 mm to 16.6 mm, and maximum heights of 0.4 mm to 10.9 mm. Total mass of all small stones per stomach ranged from 0.003 g to 15.1 g, and ranged from 0.0002% to 0.7% of body mass, with an average of 0.15% of body mass. Most small stones were considered rounded or well-rounded according to the shape categories described by Best and Gionfriddo (1991).

Lake Ontario females were more likely to have small stones in their stomachs (16 of 65 [25%] females had small stones) than Lake Ontario males (12 of 99 [12%] males had small stones; Pearson $\chi^2 = 4.4$, $P = 0.04$). Lake Ontario males with small stones had fewer *Contracaecum* spp. nematodes (median of six nematodes with an interquartile range of 0–14 nematodes) than males with no small stones (median of 19 nematodes with an interquartile range of 4–42 nematodes; $Z = -2.3$, $P = 0.02$). Lake Ontario females that had stones did not differ in median abundance of *Contracaecum* spp. nematodes from females that lacked stones (median of 12 versus 10 nematodes, respectively; interquartile ranges were 4–32 and 2–24 nematodes, respectively; $Z = -0.1$, $P = 0.61$).

We found no difference in the presence of small stones in Lake Erie females (nine of 73 [12%] had small stones) compared to males (16 of 120 [13%] had small stones; Pearson $\chi^2 = 0.04$, $P = 0.84$). For Lake Erie birds, there was no relationship between presence or absence of small stones and median abundance of *Contracaecum* spp. nematodes (males: median of one versus two nematodes, respectively; interquartile ranges were 0–8 and 0–10 nematodes, respectively; $Z = 0.1$, $P = 0.89$; females: median of 0 versus 0 nematodes, respectively; interquartile ranges were 0–5 and 0–4 nematodes, respectively; $Z = -0.1$, $P = 0.89$). The prevalence of infection by *Contracaecum* spp. nematodes was not significantly different between Lake Erie females (34 of 73 [47%] were infected) and males (69 of 120 [58%] were infected; Pearson $\chi^2 = 2.2$, $P = 0.14$). Lake Erie males and females also did not differ in the abundance of *Contracaecum* spp. nematodes; males had a median abundance of one nematode (interquartile range = 0–8 nematodes) and females had a median abundance of 0 nematodes (interquartile range = 0–5 nematodes; $Z = -1.2$, $P = 0.25$).

Seventy of 193 Lake Erie birds (36%) had *Contracaecum* spp. nematodes in the intestine; however, there was no relationship between the absence or presence of small stones and the occurrence of *Contracaecum* spp. nematodes in the intestine for males or females (males: Pearson $\chi^2 = 0.1$, $P = 0.83$; females: Pearson $\chi^2 = 0.1$, $P = 0.82$). There also was no relationship between the mass of small stones per stomach and the mass of digesta for Lake Erie males or females (males: Spearman $Rho = 0.4$, $P = 0.14$; females: Spearman $Rho = -0.6$, $P = 0.12$). The proportion of birds with stones that had empty stomachs and the proportion of birds with stones that had some digesta remaining (six of 25 birds [24%]

had empty stomachs, 19 of 25 birds [76%] had digesta remaining) were indistinguishable from proportions of birds without stones that had empty stomachs versus some digesta remaining (Pearson $\chi^2 = 1.5$, $P = 0.23$).

DISCUSSION

Our results suggest that ingestion of small stones by Double-crested Cormorants might protect them from reaching high levels of infestation by *Contracaecum* spp. nematodes. We found that females from Lake Ontario more often had small stones and were less parasitized by *Contracaecum* spp. nematodes. Males were more variable in their degree of parasitism, but males from Lake Ontario with small stones had fewer *Contracaecum* spp. nematodes than males without small stones. We found no such sex biases in presence of small stones or in levels of parasitism in Lake Erie birds; however, birds from Lake Erie had many fewer parasites and lower proportions of birds with small stones overall. Perhaps the use of small stones for self-medication by Double-crested Cormorants occurs when parasitism levels are high and the host is especially sensitive to the energy-depleting nature of nematodes (Huizinga 1971).

Three alternative hypotheses for the ingestion of small stones include mixing of food items in the stomach, buoyancy control, and accidental ingestion. We found a lack of small stones in most birds and no relationship between small stones and digesta, suggesting that small stones are not necessary for digestion in Double-crested Cormorants. If small stones served a function in mixing digesta or breaking down hard pieces such as fish bones, we would expect to see a relationship between digesta and small stones. Siegel-Causey (1990) found a relationship between small stone mass and mass of crustacean stomach contents in Rock Shags (*Strictocarbo magellanicus*), but not for any other cormorant or shag species studied.

The ingestion of small stones was proposed by Wings (2007) to fulfill antibuoyancy functions. Double-crested Cormorants are pursuit divers and therefore need to adjust their buoyancy while foraging. For crocodiles and seals there is suggestive evidence that they might ingest stones to reduce buoyancy (Wings 2007 and references therein); however, in our study, small stones totaled <1% of a birds' body mass. For small stones in the stomach to influence buoyancy, the total stone mass must exceed the buoyancy changes induced by changes in lung volume during dives, which is commonly >3% of body mass in vertebrates with more "complex" lungs (Wings 2007). Therefore, given that Double-crested Cormorants can expel air from their plumage to increase diving ability (Hatch and Weseloh 1999), it does not seem probable that Double-crested Cormorants are using small stones to increase buoyancy control.

It is possible that ingestion of small stones is accidental while foraging for prey. Double-crested Cormorants forage in benthic habitats and have been reported to probe bottom sediments for prey (Hatch and Weseloh 1999). Interestingly, males probe bottom sediments more often than females (Grémillet et al. 1998), which means that males should be more likely to ingest small stones. However, females from Lake Ontario had more small stones than males, suggesting that the presence of small stones in females might be due to deliberate ingestion. Furthermore, Siegel-Causey (1990) suggested that the maneuvering of fishes by shags and cormorants for easier swallowing would dislodge any small stones, reducing the likelihood of their accidental ingestion. However, we cannot rule out the possibility that prey fishes ingest and therefore harbor these small stones.

Double-crested Cormorant breeding colonies are littered with small stones that have been egested in pellets (Hatch and Weseloh

1999). As such, the small stones seen in stomachs represent the net difference between ingestion and egestion rates, up to the time of sampling. A variety of factors, such as time spent foraging, time spent at the colony (where regurgitation occurs), and even colony disturbance during each sampling period (projectile vomiting is a defense against intruders) could influence the retention of small stones, and thus presence or absence of stones in stomachs at the time of sampling. We collected pellet samples to verify the egestion of small stones and to determine if nematodes were also egested. We collected 13 pellets from the Lake Erie site; eight pellets contained small stones of similar shape and size as found in stomachs. We did not find any nematodes in these pellets. We did not observe males or females ingesting small stones; therefore, the higher proportion of small stones in females at the Lake Ontario site might have occurred because females were more likely to have retained stones up to the time of sampling than males.

Although our results are highly suggestive of a self-medication function for the ingestion of small stones in Double-crested Cormorants, they do not unequivocally refute other potential functions of small stone ingestion or egestion, and further investigation is needed.

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