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RESEARCH ARTICLE

The role of scratching in the control of ectoparasites on birds

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ABSTRACT
Grooming by birds is thought to serve essential anti-parasite functions. While preening has been well studied, little is known about the function of scratching in birds. We conducted a series of experiments to determine the effectiveness of scratching for controlling feather lice (*Columbicola columbae*) on Rock Pigeons (*Columba livia*). First, we used a hobbling technique to impair scratching. After 6 mo, hobbled birds had significantly more lice than controls that could scratch. In addition, lice on hobbled birds were concentrated on the birds' heads and necks (i.e. the regions that birds scratch). Secondly, we tested the role the claw plays in scratching by declawing nestlings. Once mature, declawed pigeons had significantly more lice than control birds with claws. Moreover, lice on declawed birds were concentrated on the head and neck. Next, we tested whether the flange found on the middle claw of many bird species enhances scratching. We experimentally manipulated the flange; however, the number and location of lice on birds without flanges was not significantly different than that on control birds with intact flanges. Finally, we tested whether scratching removes parasites directly or indirectly by "flushing" them onto body regions where they can be preened. When we impaired scratching (with hobbles) and preening (with "bits") we found that scratching no longer reduced the number of lice on birds. Our results indicated that scratching and preening work synergistically; scratching reduces parasite load by flushing lice onto regions of the body where they can be eliminated by preening.

Keywords: behavioral defense, grooming, Ischnocera, lice, Phthiraptera, pigeons, preening

El rol del rascado en el control de ectoparásitos en aves

RESUMEN
Se piensa que el aseo por parte de las aves tiene funciones antiparasitarias esenciales. Mientras que el acicalamiento ha sido bien estudiado, poco se sabe sobre la función del rascado en las aves. Realizamos una serie de experimentos para determinar la efectividad del rascado en el control del piojo de las plumas (*Columbicola columbae*) en *Columba livia*. Primero, usamos una técnica de cojera para impedir el rascado. Luego de seis meses, las aves con cojera tenían significativamente más piojos que las aves control que si podían rascarse. Adicionalmente, los piojos en las aves cojas se concentraron en la cabeza y el cuello de las aves (i.e. las regiones que se rascan las aves). Segundo, evaluamos el rol que juegan las garras en el rascado en polluelos a los que se les sacaron las garras. En la madurez, las palomas sin garras tuvieron significativamente más piojos que las aves control con garras. Más aún, los piojos en las aves sin garras se concentraron en la cabeza y el cuello. Luego, evaluamos si el reborde que se encuentra en la garra del medio de muchas especies de aves mejora el rascado. Manipulamos experimentalmente el reborde; sin embargo, el número y la ubicación de los piojos en las aves sin rebordes no fueron significativamente diferentes del de las aves control con rebordes intactos. Finalmente, evaluamos si el rascado remueve a los parásitos directamente o indirectamente "corriéndolos" hacia regiones del cuerpo donde las aves pueden acicalarse. Cuando impedimos el rascado (con cojera) y el acicalamiento (con "frenos"), encontramos que el rascado dejó de reducir el número de piojos en las aves. Nuestros resultados indicaron que el rascado y el acicalamiento funcionan en sinergia; el rascado reduce la carga parasitaria corriendo a los piojos hacia las regiones del cuerpo donde pueden ser eliminados por acicalamiento.

Palabras clave: acicalamiento, aseo, defensa comportamental, Ischnocera, palomas, Phthiraptera, piojo

INTRODUCTION

Birds and mammals have a variety of defenses against harmful ectoparasites, including behavioral adaptations for avoiding parasites or, if infested, reducing their abundance (Hart 1990, Curtis 2014). Anti-parasite behavior is the host's first line of defense against parasites. For example, birds are known to examine and successfully avoid parasitized nest sites and mates (Bush and Clayton 2018). Birds also engage in so-called maintenance behavior to combat ectoparasites such as lice, fleas, and ticks. Maintenance behavior includes grooming, dusting, sunning, and anointing feathers with formic acid or other substances (Clayton et al. 2010). The most common of these behaviors is grooming, which consists of preening with the beak and scratching with the feet (Clayton and Cotgreave 1994, Cotgreave and Clayton 1994). Preening, whereby birds pull their feathers through the mandibles of the beak, or nibble feathers with the tips of the mandibles (Bush and Clayton 2018), occurs much more frequently and is much better studied than scratching.

The effectiveness of preening for controlling ectoparasites has been demonstrated experimentally (Clayton et al. 2010). For example, preening has been impaired using poultry "bits," which are small C-shaped pieces of metal or plastic inserted between the upper and lower mandibles of the beak (Figure 1A). Bitted Rock Pigeons *(Columba livia)* experience dramatic increases in ectoparasites, such as feather lice (Clayton 1991, Clayton and Tompkins 1995, Clayton et al. 1999, 2005) and hippoboscid flies (Waite et al. 2012). While beaks are first and foremost adaptations for feeding, some components of beak morphology are adaptations for parasite control. Many species have a small overhang on the beak's upper mandible that is critical for the control of ectoparasites (Clayton and Walther 2001, Clayton et al. 2005). The mandibular overhang is tiny, averaging only 1.5 mm in length in pigeons; when the overhang is removed, louse loads increase dramatically (Clayton et al. 2005). The beak overhang also plays a role in the control of parasites on Darwin's finches (Villa et al. 2018) and other songbirds (Bush and Clayton 2018).

Although preening is the primary form of grooming in birds (Clayton et al. 2005, Waite et al. 2012, Bush and Clayton 2018), scratching with the feet is also common (Clayton 1991). Virtually all species of birds scratch (Simmons 1957), yet the function of scratching remains unclear. Scratching may serve to control ectoparasites on body regions birds cannot preen, such as the head and neck (Clayton 1991). But scratching may also serve other adaptive functions. For example, several bird species have been observed transferring uropygial oil from their beaks to their feet, followed by head-scratching (Simmons 1961). This behavior presumably helps spread oil onto feathers that are inaccessible to the bill (Simmons 1961). Scratching undoubtedly also relieves itching of the skin due to a variety of factors.

Comparative analyses suggest that scratching compensates for inefficient preening in species of birds with long, unwieldy beaks. Clayton and Cotgreave (1994) compared the grooming behavior of birds with different bill lengths (corrected for overall body size). Long-billed taxa, such as toucans and hummingbirds, averaged 16.2% of their grooming time scratching, while relatively short-billed taxa, such as falcons and woodpeckers, averaged just 2.3% of their grooming time scratching. Phylogenetically independent comparisons confirmed that birds with long beaks spend significantly more time scratching than birds with short beaks, consistent with the hypothesis that birds compensate for inefficient preening by scratching.

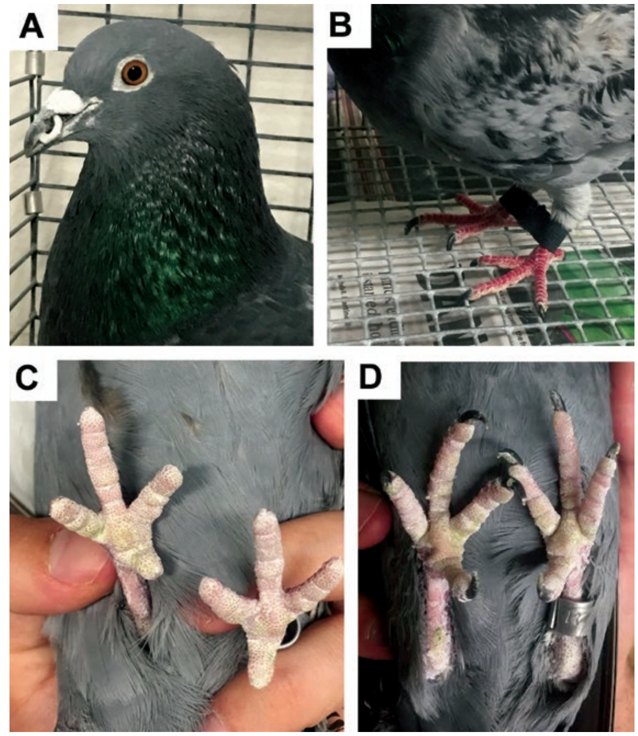

FIGURE 1. Rock Pigeon fitted with a flexible, plastic bit to impair preening ability (**A**). An example of a flexible, elastic hobble used to impair scratching ability (**B**). The feet of a declawed pigeon (**C**) compared to a control bird with intact claws (**D**).

Anecdotal evidence suggests that scratching is effective in controlling avian ectoparasites. Birds with a deformed or missing foot often have large numbers of lice and eggs restricted to the head and neck, which cannot be scratched while standing on the remaining good leg (Clayton 1991, Bush and Clayton 2018). Although these observations suggest that scratching may control ectoparasites, it is possible that birds with deformed or missing feet are in poor general condition, which might contribute to the higher parasite loads. The purpose of the current paper is to describe the results of 4 experiments designed to rigorously test the hypothesis that scratching is an effective anti-parasite behavioral defense. In the first experiment, we prevented captive Rock Pigeons from scratching by hobbling their feet, then monitoring parasite loads over time. Next, we specifically tested the role of claws in effective scratching by declawing captive nestling Rock Pigeons and monitoring their parasite loads over time.

Like beak morphology, claw morphology may be adapted for controlling parasites. Many birds have a flange on the inner edge of the middle claw of each foot (Morgan 1925; Appendix Table 2). This flange may increase the effectiveness of scratching for controlling ectoparasites. We conducted a third experiment in which we removed this flange from the middle claw on each foot of captive

Rock Pigeons and monitored parasite loads over time. In a fourth and final experiment, we tested whether scratching removes lice directly, or flushes them onto regions where they can be preened. For this final experiment we compared the parasite loads of hobbled and non-hobbled birds, all of which had preening impaired with bits.

METHODS

Experiments were conducted in 2017 and 2018, and all procedures and behavioral manipulations were approved by our Institutional Animal Care and Use Committee and a university veterinarian. Adult Rock Pigeons were captured using walk-in traps baited with grain at several sites in Salt Lake City, Utah, USA. Birds were housed individually in 30 × 30 × 56 cm wire mesh cages in our animal facility with ad libitum pigeon mix, grit, and water. We randomly assigned each bird to an experimental or control treatment. Experimental and control cages were interspersed on racks in our animal rooms. Plexiglas barriers were placed between the cages to prevent lice from transferring between birds in adjacent cages. Cages were cleaned weekly. Throughout the experiments, birds were maintained on a 12-hr photoperiod at room temperature.

Prior to the start of each experiment, we fumigated each bird with ethyl acetate (Clayton and Drown 2001) to kill hippoboscid flies and other ectoparasites that may already have been on the birds. Because the ethyl acetate procedure does not kill lice eggs, which are glued to the feathers, some birds retained small populations of the 2 common species of feather lice: *Columbicola columbae* and *Campanulotes compar* (Insecta: Phthiraptera: Ischnocera). These species have very similar life cycles and they both feed solely on host feathers and dead skin (Nelson and Murray 1971). Both species are controlled mainly by preening (Clayton et al. 2005). To be certain that every bird in each experiment had lice, we experimentally infested all birds with 25 *C. columbae*. The lice used in these experiments came from "donor" pigeons that were also wild-caught in Salt Lake City. Donor bird lice were anesthetized with a stream of CO_2, then removed from the bird and immediately transferred to recipient birds (Moyer et al. 2002). At the end of each experiment, birds were euthanized using cervical dislocation.

Experiment 1: Hobbling Experiment

The goal of this experiment, which lasted 6 mo, was to test whether scratching plays a role in the control of feather lice, particularly on body regions that birds cannot preen, such as the head and neck. The experiment was initiated with 40 wild-caught adult Rock Pigeons randomly assigned to the experimental and control groups. Birds in the experimental treatment were fitted with 3.5-cm hobbles made of black elastic bands (Figure 1B). The hobbles prevented birds from scratching their heads and necks, but did not prevent them from walking around in the cage. Control birds were exposed to the same level of handling; they were briefly fitted with hobbles, but these were removed later the same day. We checked for side effects of hobbling on the condition of experimental birds by monitoring their general behavior and body mass. There were no apparent side effects of hobbling (Table 1).

During the latter half of the experiment we measured the amount of time birds spent preening and scratching using the instantaneous scan sampling method (Altmann 1974). Specifically, we noted whether each bird was preening or scratching at 5-min intervals over 2-hr observation sessions split evenly between morning (0900–1100 hours), midday (1200–1400 hours), and late afternoon (1600–1800 hours). Each observation session began with a 15-min acclimation period, during which the observer sat motionless in full view of the birds. We defined preening as touching the plumage with the bill. We defined scratching as touching the plumage with the foot. We made a total of 270 instantaneous observations per bird.

We also monitored the distribution of lice on pigeons using Clayton and Drown's (2001) visual examination method every 2 mo. We examined the following 5 regions of each bird for timed intervals: head and neck (30 s), right wing (60 s), tail (60 s), keel (30 s), and back and rump (60 s). At the end of the experiment, we weighed all birds to check for any effect of hobbling on body mass. We then euthanized each bird and quantified louse loads using the "body washing" method of Clayton and Drown (2001), which accounts for ~99% of the lice on individual birds.

TABLE 1. Mean (± SE) body mass, preening, and scratching for birds in each experiment.

Experiment	Treatment (n)	Mass	% time preening	% time scratching
Hobbling	Hobbled (17)	364.55 ± 8.05	13.71 ± 1.02	0.07 ± 0.03
	Not Hobbled (19)	371.70 ± 8.44	12.37 ± 0.87	0.23 ± 0.05
Declawing	Claws Removed (15)	336.93 ± 10.53	8.44 ± 0.94	0.48 ± 0.15
	Not Removed (15)	330.13 ± 8.74	8.56 ± 1.20	0.26 ± 0.11
Flange	Flange Removed (20)	359.64 ± 5.58	11.06 ± 0.87	0.17 ± 0.08
	Not Removed (20)	353.70 ± 8.35	10.75 ± 0.63	0.22 ± 0.10
Hobbling/Bitting	Hobbled (17)	368.47 ± 8.13	13.11 ± 1.54	0.10 ± 0.05
	Not Hobbled (19)	379.92 ± 8.46	11.56 ± 1.36	0.41 ± 0.13

Four birds were excluded from the dataset prior to analysis. One of these birds died over the course of the experiment. Another bird chipped its bill, which interferes with effective preening (Clayton et al. 2005). The remaining 2 birds were subjected to errors during the washing process, meaning that final estimates of their louse loads would have been unreliable. Following the exclusion of these 4 birds, the dataset consisted of 17 hobbled and 19 control birds.

Experiment 2: Declawing Experiment

The goal of this experiment, which lasted 4 mo, was to test whether claws are integral to the effectiveness of scratching on ectoparasites. Birds for this experiment were bred in captivity from wild-caught Rock Pigeons. Thirty birds were used; these consisted of 13 pairs of siblings and 2 more pairs of unrelated birds of similar age. The members of each pair were randomly assigned to declawing and control groups. We surgically removed the claws of the nestlings in the declawing treatment within 24 hr of their hatching from the egg (Figure 1C). This procedure, which took less than a minute to perform, did not elicit any reaction from the nestlings and did not affect their subsequent growth (Table 1). Birds in the control group were handled similarly, but instead of removing claws, we pinched all of the toes of each bird (Figure 1D).

Before continuing the experiment, we waited until all of the birds were at least 9 mo of age, which is adulthood for Rock Pigeons (Johnston and Janiga 1995). Birds were housed under the same conditions as described in the hobbling experiment. We infested each bird with 25 *C. columbae* and conducted visual examinations of the lice every 2 mo. During the latter half of the experiment, which lasted 4 mo, we collected scan sampling data, as described in the hobbling experiment, with ~180 observations per bird. At the end of the experiment, we euthanized birds and quantified their lice using the washing method (Clayton and Drown 2001).

Experiment 3: Flange Removal Experiment

The goal of this experiment, which lasted 4 mo, was to test whether the middle toe flange plays a role in the control of feather lice when birds scratch. The experiment included 40 wild-caught adult Rock Pigeons randomly assigned to the experimental and control groups. Methods were similar to those used in the hobbling experiment except that experimental birds had the flange on the middle toe of each foot (Figure 2) harmlessly removed with a sandstone Dremel tool. Birds in the control group were sham-dremeled with a buffing wheel that removed no tissue. The dremeling (and buffing) procedure was repeated about once a week to prevent regrowth of the flange over the course of the experiment. We used the same infestation method, behavioral data collection, microhabitat examination, and washing methods as described for the declawing experiment.

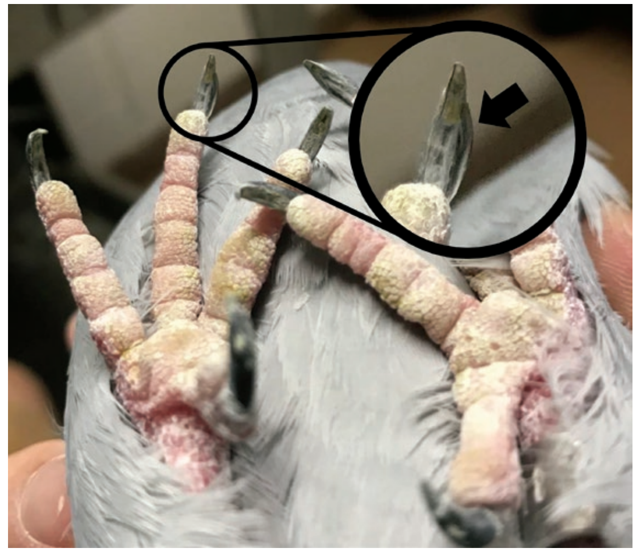

FIGURE 2. The flange on the middle claw of a Rock Pigeon.

Experiment 4: Hobbling/Bitting Experiment

The goal of this experiment, which lasted 4 mo, was to test whether scratching controls lice directly, or indirectly by interacting with preening. The experiment was initiated with 40 wild-caught adult Rock Pigeons randomly assigned to the experimental and control groups. All 40 birds had their preening impaired with bits (Figure 1A). Twenty of the birds, chosen at random, were also hobbled throughout the experiment, as described earlier; the remaining 20 birds were not hobbled beyond the first day. We used the same infestation method, behavioral data collection, microhabitat examination, and washing methods as described above.

Four birds were excluded from the data set prior to analysis. One bird had lost its bit during the experiment and the hobbles on 3 other birds proved to be too tight, leading to minor swelling of the legs that could conceivably affect scratching behavior. Following the exclusion of these 4 birds, the data set consisted of 17 hobbled and 19 unhobbled birds, all of which were bitted.

Statistical Analyses

For each of the 4 experiments we normalized the number of lice on each bird by log-transforming the number of lice recovered from body washing (ln(total number of lice + 1)). We used 2-tailed t-tests to compare the number of lice, rates of behavior, and body masses of experimental and control birds. To compare the microhabitat distribution of lice between experimental and control birds, for each bird we tallied the number of lice for each body region in each visual examination and used a generalized linear mixed-effects model (GLMM) with a binomial distribution. Treatment and individual visual examination results were fixed effects; band number (bird ID) was a random effect. We conducted analyses in JMP 13.0 (SAS Institute, Cary, North Carolina, USA) and R (R Core Team 2016) using the *lme4* library (Bates et al. 2015).

RESULTS

Experimental infestation with *C. columbae* was successful: all birds (n = 142) across all 4 experiments had *C. columbae* when checked 2 mo following the start of each experiment. Thirty-five of the birds (25%) also had *Campanulotes compar*, but with a mean intensity of only 12.03 individuals of this species per infested bird. By comparison, the mean intensity of *C. columbae* was 178.43 across the 35 birds that had both species of lice. Four of the 142 birds in the overall study also had very small numbers of parasitic mites *(Dermanyssus gallinae*); 3 of these birds had a single mite each, and a fourth bird had 6 mites. No other parasites were found on any of the birds in the study.

Experiment 1: Hobbling Experiment

Hobbling had no significant effect on the body mass of birds (t-test, t = 0.61, df = 34, P = 0.544; Table 1) or preening rates (t-test, t = 1.00, df = 35, P = 0.324; Table 1). In contrast, hobbling was very effective at preventing scratching (t-test, t = 2.72, df = 35, P = 0.010; Table 1). Only 3 hobbled birds were ever observed to scratch (a single time each, by dipping the head down to the level of the feet). Hobbled birds had significantly more lice than non-hobbled birds (t-test, t = −2.80, df = 35, P = 0.009; Figure 3A). Lice on the hobbled birds were also significantly more common on the head and neck than lice on non-hobbled birds (GLMM, z = −2.83, P = 0.005; Figure 3B).

Experiment 2: Declawing Experiment

Declawing had no significant effect on the body mass of birds (matched pairs t-test, t = 0.77, df = 14, P = 0.452; Table 1) or preening rates (matched pairs t-test, t = 0.09, df = 14, P = 0.923; Table 1). Unlike hobbling, it had no significant effect on scratching rate, although there was a trend for birds without claws to scratch more than birds with intact claws (matched pairs t-test, t = −1.87, df = 14, P = 0.082; Table 1). Despite this trend, declawed birds had significantly more lice than control birds (matched pairs t-test, t = −2.70, df = 14, P = 0.017; Figure 4A). Lice on declawed birds were also significantly more common on the head and neck than lice on control birds with intact claws (GLMM, z = −2.84, P = 0.005; Figure 4B).

Experiment 3: Flange Removal Experiment

Flange removal had no significant effect on body mass (t-test, t = 0.59, df = 39, P = 0.558; Table 1), preening rate (t-test, t = 0.28, df = 39, P = 0.778; Table 1), nor scratching rate (t-test, t = 0.43, df = 39, P = 0.673; Table 1). Moreover, flange removal had no significant effect on the number of lice on birds (t-test, t = −0.80, df = 39, P = 0.428; Figure 5A), nor did it have any effect on the number of lice on the head and neck of birds (GLMM, z = −0.37, P = 0.714; Figure 5B). In summary, flange removal had no significant effect on any of the parameters we measured.

Experiment 4: Hobbling/Bitting Experiment

The hobbling/bitting treatment had no significant effect on the body mass of birds (t-test, t = 0.98, df = 35, P = 0.336; Table 1), nor did it affect preening rates (t-test, t = −0.75, df = 35, P = 0.456; Table 1). Birds in this experiment, all of which were bitted, preened about the same amount as birds in the first experiment, none of which were bitted. Thus, although bitting impaired the ability of birds to remove lice, it did not alter their rate of preening. As in the first experiment, hobbling was effective at preventing scratching (t-test, t = 2.17, df = 35, P = 0.040; Table 1). Moreover, as in the first experiment, 3 hobbled birds were observed scratching (a single time each) by lowering their heads to the level of their feet.

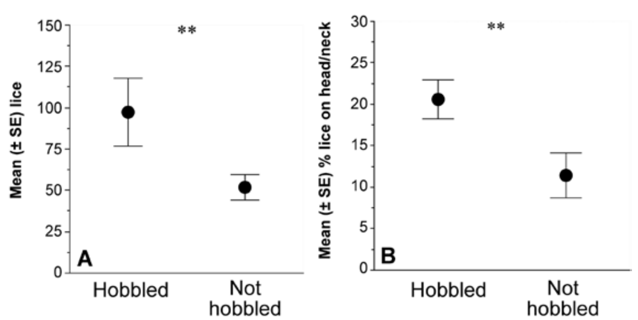

FIGURE 3. (**A**) Abundance of lice on hobbled birds (n = 17) vs non-hobbled birds that could scratch (n = 19). (**B**) Percent of lice on the head and neck of hobbled birds vs. non-hobbled birds that could scratch. Hobbled birds had about twice as many lice, overall, and twice as many lice on the head and neck as birds that were not hobbled. **P < 0.01.

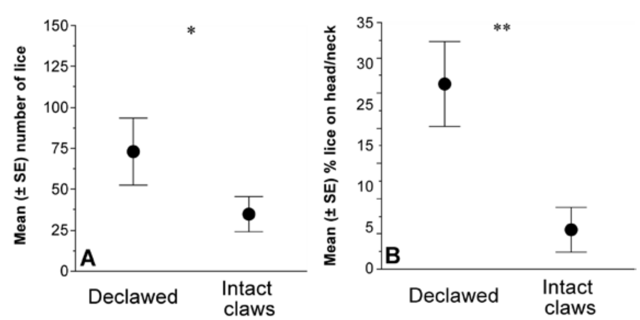

FIGURE 4. (**A**) Abundance of lice on declawed birds (n = 15) vs. birds with intact claws (n = 15). (**B**) Percent of lice on the head and neck of declawed birds vs. birds with intact claws. Birds without claws had more than twice as many lice, overall, and about 5-fold as many lice on the head and neck as birds with intact claws. *P < 0.05, **P < 0.01.

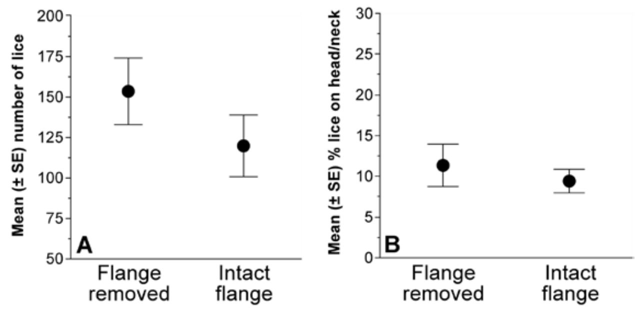

FIGURE 5. (**A**) Abundance of lice on birds without flanges (n = 20) vs. birds with intact flanges (n = 20). (**B**) Percent of lice on the head and neck of birds without flanges vs. birds with intact flanges. Birds without flanges had similar numbers of lice as those with intact flanges, and lice on birds with their flanges removed were not concentrated on the head and neck.

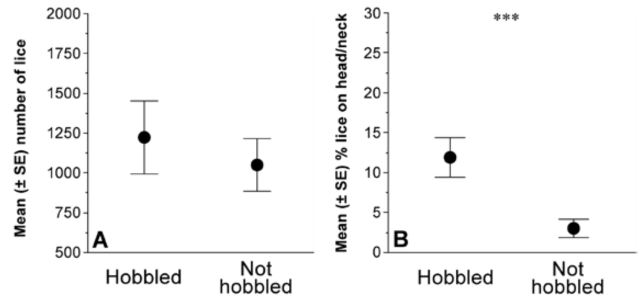

FIGURE 6. (**A**) Abundance of lice on hobbled/bitted birds (n = 17) vs. non-hobbled/bitted birds that could scratch (n = 19). (**B**) Percent of lice on the head and neck of hobbled/bitted birds vs. non-hobbled/bitted birds that could scratch. Hobbled/bitted birds had similar numbers of lice as non-hobbled/bitted birds but these lice were much more concentrated on the head and neck compared to birds that were not hobbled. ***P < 0.001.

In contrast to the first hobbling experiment, hobbled/bitted birds did not have more lice than control birds that could scratch but not preen (t-test, t = –0.18, df = 35, P = 0.855; Figure 6A). Similar to the first hobbling experiment, lice on hobbled/bitted birds were significantly more common on the head and neck than lice on non-hobbled/bitted birds (GLMM, z = –6.62, P < 0.001; Figure 6B). These results show that, although lice are still disturbed by scratching, they are not killed, nor removed by scratching on birds with impaired preening.

DISCUSSION

The results of our first experiment show that scratching is effective in reducing the number of ectoparasites on birds. Pigeons that were hobbled, and thus unable to scratch, had nearly twice as many lice as those without hobbles (Figure 3A). Moreover, lice on hobbled birds were more common on the head and neck, where birds cannot preen. These results are consistent with studies of scratching in mammals. For example, mice prevented from scratching had many more lice than mice that could scratch normally (Bell et al. 1962). Lice in the Bell et al. (1962) study were similarly concentrated in regions that the mice could not groom orally, such as the head and neck. In summary, our results show that, as in the case of mammals, scratching helps control ectoparasites on birds. These results are striking because unlike mammals, which spend as much as 40% of their time scratching (Bolles 1960), the birds in our study spent <1% of their time scratching.

Our second experiment shows that claws are necessary for the parasite-control function of scratching. Although birds without claws spent nearly twice as much time scratching as birds with claws (Table 1), they had twice as many lice as birds with claws (Figure 4A). Moreover, birds without claws had a higher proportion of lice on the head and neck (Figure 4B), showing that it is the presence of claws, not scratching behavior per se, that reduces the number of lice on the head and neck. Although all bird species possess claws of some kind on their toes (Stettenheim 2000), claw morphology has been studied mainly in relation to locomotion and foraging (Pike and Maitland 2004, Csermely and Rossi 2006, Fowler et al. 2009). It would be fascinating to compare the functional morphology of different claw types in relation to parasite control among different species of birds.

To this end, our third experiment tested whether the flange on the middle claw of pigeons (Figure 2) enhances the effectiveness of scratching for parasite control. Pigeons scratch predominantly with the flanged middle claw (G. B. Goodman et al. personal observation). Nevertheless, birds with flanges removed did not have significantly more lice than birds with intact flanges (Figure 5A). Moreover, there was no difference in the number of lice on the head and neck of birds with and without flanges (Figure 5B). Thus, the flange appears to play no role in the control of feather lice on pigeons. These results are consistent with those of Clayton and Walther (2001), who tested for a relationship between foot and claw morphology and louse species richness and abundance among 52 species of Peruvian birds, but found no significant correlations. The flange may serve another function. For example, scratching with the flanged claw may help pinfeathers on the head emerge from sheaths more rapidly as they develop. This hypothesis could be tested by removing the flange before a molt cycle and then quantifying how long it takes for the feathers to emerge.

Our fourth and final experiment was designed to get at the mechanism by which scratching helps control ectoparasites. Does scratching remove lice directly, or does it simply flush them into the range of preening? The fourth experiment was a repeat of the first experiment, but with birds that were bitted and thus could not preen; half of the birds were also hobbled, while the other half was not hobbled. As in the case of the first experiment, birds with hobbles scratched much more than birds without hobbles (Table 1). Moreover, birds with hobbles had more lice on the head and neck than birds without hobbles (Figure 6B).

In contrast to the first experiment, however, birds with and without hobbles did not differ significantly in their overall number of lice (Figure 6A). This was a striking result, considering that birds in the fourth experiment averaged more than 1,000 lice, due to the lack of preening, compared to means of less than 100 lice on birds in the first experiment. Birds in the fourth experiment had an order of magnitude more lice because they wore bits that impaired preening.

The disruptive effect of scratching is demonstrated by its highly significant effect on the microhabitat distribution of lice in the first, second, and fourth experiments (Figures 3B, 4B, and 6B). However, the inability of scratching alone to remove lice is demonstrated by the lack of a significant difference in the number of lice on hobbled and non-hobbled birds (Figure 6A). In contrast, the synergistic effect of scratching and preening was demonstrated by the lower number of lice on birds that could both scratch and preen in Experiment 1 (Figure 3A). Thus, the result of the first, second, and fourth experiments, taken together, show that scratching with claws helps control lice by flushing them onto regions of the body where they can then be removed by preening.

It is not uncommon for different defenses against parasites to work together. For example, in mammals, the itch sensation is often triggered by acquired immune responses to ectoparasites (Owen et al. 2010, Palm et al. 2012, Mack and Kim 2018). Once the itch sensation is triggered, the host scratches these regions and removes parasites that triggered the response. Our study provides another example of how multiple defenses can interact. In this case, scratching interacts with preening to control parasites that birds cannot reach by preening alone.

Scratching often occurs immediately before or after bouts of preening (Burtt and Hailman 1978). This pattern is consistent with scratching displacing parasites onto regions where preening or oral grooming (in mammals) can kill or remove them. This mechanism is consistent with behavioral observations in mammals (Bell et al. 1962, Duboscq et al. 2016). For example, prairie dogs (*Cynomys* spp.) often switch between scratching and oral grooming in rapid succession, with oral grooming thought to be targeted at individual parasites (Eads et al. 2017).

Our study is also interesting in light of the morphology and behavior of lice that live primarily on the head and neck of birds. Avian "head lice" have large, triangular heads with expanded temple regions that support large muscles attached to the mandibles (Clay 1949, Bush et al. 2010). These lice also have a rostral groove on the head that helps anchor them to feather barbs (Clayton et al. 2015). This unusual mechanism for holding onto feathers may have evolved in response to scratching-mediated selection. This hypothesis could be tested by measuring the amount of force needed to remove head lice, compared to wing or body lice. Wing lice that primarily exploit other microhabitats have a less pronounced head groove, or it is absent entirely (Johnson et al. 2012). Wing and body lice have evolved other morphological and behavioral adaptations to escape from preening (Clayton et al. 2003, Johnson et al. 2005, Bush et al. 2010, 2019; Johnson et al. 2012). For example, wing lice escape by inserting between the barbs of flight feathers, and by having cryptic coloration (Bush et al. 2019).

In conclusion, our results show that scratching is an important anti-parasite defense of birds. Claws, but not the flange on pigeon claws, are necessary for scratching to be effective. Finally, scratching functions by flushing lice onto regions that can be preened. It is very much the interaction of preening and scratching that controls lice on the head and neck of birds.

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Author contributions: G.B.G., S.E.B., and D.H.C. designed the experiment. G.B.G. and M.C.K. collected the data. G.B.G. and S.E.B. analyzed the data. G.B.G. wrote the paper with input from S.E.B. and D.H.C. All authors agree to be held accountable for the content therein and approve the final version of the manuscript.

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Appendix Table 2. Occurrence of flanges on the middle claws of 53 study skins of birds representing 25 species in 24 genera (20 families, 13 orders). All skins were part of the Natural History Museum of Utah collection. Classifications follow the AOS checklist (Chesser et al. 2019). Occasionally, the scientific name on the specimen differed from name recognized by the AOS; in these cases, the name, as written on the specimen, is provided in brackets.

Order	Family	Species	Number examined	Number with flange on both feet	Number with flange on one foot
Anseriformes	Anatidae	Tundra Swan, *Cygnus columbianus*	2	2	0
		Greater White-fronted Goose, *Anser albifrons*	2	2	0
		Snow Goose, *Anser caerulescens*	2	1	1
		Canada Goose, *Branta canadensis*	2	2	0
Galliformes	Odontophoridae	Northern Bobwhite, *Colinus virginianus*	3	1	1
Gruiformes	Rallidae	Virginia Rail, *Rallus limicola*	2	2	0
Charadriiformes	Scolopacidae	Western Sandpiper, *Calidris mauri* [*Ereunetes mauri*]	2	2	0
	Laridae	Herring Gull, *Larus argentatus*	2	2	0
Procellariiformes	Hydrobatidae	Leach's Storm-Petrel, *Hydrobates leucorhous* [*Oceanodroma leucorhoa*]	2	2	0
Suliformes	Phalacrocoracidae	Double-crested Cormorant, *Phalacrocorax auritus*	2	2	0
Accipitriformes	Accipitridae	Red-tailed Hawk, *Buteo jamaicensis*	2	2	0
		Golden Eagle, *Aquila chrysaetos*	2	2	0
Strigiformes	Strigidae	Great Horned Owl, *Bubo virginianus*	2	2	0
Piciformes	Picidae	Northern Flicker *Colaptes auratus* [*Colaptes cafer*]	2	2	0
Falconiformes	Falconidae	Peregrine Falcon, *Falco peregrinus*	2	2	0
Passeriformes	Corvidae	American Crow, *Corvus brachyrhynchos*	2	0	2
	Hirundinidae	Purple Martin, *Progne subis*	2	2	0
	Troglodytidae	House Wren, *Troglodytes aedon*	3	2	0
	Turdidae	American Robin, *Turdus migratorius*	2	2	0
	Sturnidae	European Starling, *Sturnus vulgaris*	2	2	0
	Passerellidae	Abert's Towhee, *Pipilo aberti*	3	0	1
		Song Sparrow, *Melospiza melodia*	2	2	0
	Icteridae	Red-winged Blackbird, *Agelaius phoeniceus*	2	2	0
	Cardinalidae	Summer Tanager, *Piranga rubra*	2	1	1