

HATCHING ORDER AND SEASONAL TIMING OF DEVELOPMENT PREDICT BILL MORPHOLOGY OF NESTLING AND ADULT LINCOLN'S SPARROWS

KEITH W. SOCKMAN¹

Department of Biology and Curriculum in Neurobiology, University of North Carolina, Chapel Hill, NC 27599-3280

Abstract. Traits under strong directional or stabilizing selection can nonetheless vary considerably if the selective forces impinging on them vary in strength or direction. Although such variation is commonly manifested between unrelated individuals, some of the largest differences occur between siblings because of differences in their developmental experience. This is exemplified by the broods of altricial birds, in which asynchronous hatching produces a competitive hierarchy that influences an individual's developmental rate and fitness. Using Lincoln's Sparrow (*Melospiza lincolnii*), I examined whether the order of hatching or the seasonal timing of development (another factor that influences offspring fitness and estimated as the date of initiation of the clutch from which an individual hatched) predicted variation in bill shape, a trait often under strong natural and sexual selection. I measured individuals as nestlings and as adults for a metric of bill shape (height/width) that is associated with an adult male Lincoln's Sparrow's song performance. I found that an individual's bill shape during the nestling stage positively correlated with its bill shape during the adult stage. Nestling bill shape declined (became wider and flatter in cross-section) with hatching order, and the bill shape of both nestlings and adults declined with the individual's seasonal timing of development. These findings raise the possibility of a trade-off between life stages in the optimization of songbird bill morphology and may explain the persistence of suboptimal bill shapes among adult songbirds in the presence of strong stabilizing or directional selection.

Key words: *beak, developmental constraint, developmental plasticity, hatching asynchrony, laying date, sibling competition.*

El Orden de Eclosión y la Temporalidad Estacional de Desarrollo Predicen la Morfología del Pico de los Pichones y Adultos de *Melospiza lincolnii*

Resumen. Los rasgos bajo fuerte selección direccional o estabilizante pueden, a pesar de esto, variar considerablemente si las fuerzas de selección que actúan sobre ellos varían en intensidad o dirección. Aunque esta variación se manifiesta comúnmente entre individuos no relacionados, algunas de las diferencias más grandes aparecen entre hermanos debido a las diferencias en sus experiencias de desarrollo. Esto se ejemplifica en las camadas de aves altriciales, en las que la eclosión asincrónica produce una jerarquía competitiva que influye en la tasa de desarrollo y la adecuación biológica del individuo. Empleando la especie *Melospiza lincolnii*, examiné si el orden de eclosión o la temporalidad estacional de desarrollo (otro factor que influye en la adecuación biológica de los pichones y que se estima a partir de la fecha de iniciación de la nidada de la cual eclosionó un individuo) predijo la variación en la morfología del pico, un rasgo que usualmente está bajo fuerte selección natural y sexual. Considerando los mismos individuos como pichones y luego como adultos, tomé una medida de la forma del pico (altura/ancho) que está asociada con el desempeño del canto del macho adulto de *Melospiza lincolnii*. Encontré que la forma del pico de los pichones se correlaciona positivamente con la de los adultos. La forma del pico del pichón disminuyó (se volvió más ancha y achatada en la sección transversal) con el orden de eclosión y la forma del pico del pichón y del adulto disminuyó a lo largo de la estación con el tiempo de desarrollo del individuo. Estos resultados sugieren la posibilidad de una solución de compromiso entre estadios de vida en la optimización de la morfología del pico de las aves canoras y puede explicar la persistencia de formas de pico sub óptimas entre los adultos de las aves canoras ante la presencia de fuerte selección estabilizadora o direccional.

INTRODUCTION

Traits under strong directional or stabilizing selection can still vary considerably if the selective forces impinging on them vary (Gibbs and Grant 1987). Such variation is commonly manifested between individuals that are not closely related and therefore diverge genetically (Rowe and Houle

1996). However, some of the strongest differences in selective forces occur between siblings, as exemplified by the broods of altricial birds, in which asynchronous hatching produces a developmental hierarchy that influences an individual's ability to compete with siblings for resources (Mock and Parker 1997). In general, sibling competition can affect growth and survival (reviewed in Sockman et al. 2006), and

Manuscript received 8 March 2011; accepted 16 February 2012.

¹E-mail: kws@unc.edu

the particular competitive environment experienced by any one sibling depends, in large part, on the order in which it hatched (Forbes and Mock 2000, Forbes 2010). Compared to their early-hatched siblings, subordinate late-hatched individuals are less competitive for limited food resources and should benefit from traits that enhance prospects for growth and survival while they are in the nest, even if such traits are suboptimal later in life.

The morphology of the adult songbird's bill is widely known for its variation between individuals and species and for being a major target of selection (Podos 2001, Badyaev et al. 2008, Grant and Grant 2008). Indeed the bill is a bird's most important tool. Adults use it in food acquisition, food handling and manipulation, nest building, defense, grooming, preening, ventilating, thermoregulation, vocalizing, and, in nonprecocial species, in delivering food to their young, which themselves use their bills in begging, defense against siblings, and therefore in procuring their own food (Welty 1975). Because of these many uses of the bill and the influence of bill morphology on an individual's choice of or fit to its foraging niche (e.g., Price et al. 1984, Smith 1987, Grant and Grant 1995, Benkman and Miller 1996, Badyaev et al. 2008), it is clear that bill morphology is under powerful forces of natural selection.

In songbirds, the bill may also be honed by sexual selection. Its morphology affects an individual's feeding performance (Herrel et al. 2005, Badyaev et al. 2008), a behavioral trait of males that females of some species assess when choosing mates (Snowberg and Benkman 2009). Bill morphology and its associated musculature may affect the performance of sexually selected song signals because of the bill's role as part of the vocal tract (Podos 2001, Badyaev et al. 2008). There is also evidence for an indirect effect of bill morphology on vocal performance—one that is mediated through spatial and temporal variation in ecological conditions and the bill's relationship with foraging and energetics (Sockman 2009). It has even been proposed that effects of bill morphology on vocal performance could result in the reproductive isolation necessary for natural selection on bill morphology to lead to population divergence (Badyaev et al. 2008) or sympatric speciation (Podos 2001).

Despite the importance of bill morphology in influencing a bird's fitness, the proximate sources of individual variation in this trait are largely unknown. I investigated potential sources of variation in bill morphology in Lincoln's Sparrow (*Melospiza lincolnii*), a small migratory songbird that breeds in high-elevation and high-latitude wet meadows of North America (Ammon 1995). The bill shape of an adult male Lincoln's Sparrow correlates with his performance of vocal signals (Sockman 2009). Specifically, under relatively limiting resource conditions, individuals with the bill wider in cross-section (smaller height:width ratio) produce songs with lower trill performance (the ability to rapidly repeat a single syllable type of broad bandwidth) and of shorter duration with fewer syllables and syllable types (phrases), than do individuals with

intermediate bill shapes. Although the mechanism for this relationship is not clear, changing ecological conditions may mediate the relationships between bill morphology, foraging, and energy or time constraints imposed on song production. Furthermore, variation in the trill performance of a male Lincoln's Sparrow predicts the female's preference behavior in a mate-choice context (Caro et al. 2010), as reported in similar studies of the congeneric Swamp Sparrow (*M. georgiana*; Ballentine et al. 2004) and of the domesticated canary (*Serinus canaria*; Draganoiu et al. 2002). This suggests that sexual selection acts on trill performance and, in Lincoln's Sparrow, on the correlated trait, bill shape.

The shape of a songbird's bill changes dramatically as it develops. In nestlings, the bill's cross-sectional shape is relatively wide and flat, a shape that enables a large gape for the procurement of parentally provided food through begging (Fig. 1, top panel) (Illies 1975, Welty 1975). Despite enormous variation between species in adult morphology, the bills of adult songbirds (Fig. 1, bottom panel), in general, are taller and narrower than those of nestlings. This massive transition between life stages raises the possibility that variation in adult bill morphology can be traced to variation in nestling bill morphology and raises the question of what factors drive variation in nestling bill morphology.



FIGURE 1. Variation in bill shape in Lincoln's Sparrows between two life stages, nestling (top panel) and adult (bottom panel).

Of the many factors that influence the development of a nestling bird, food availability imposes some of the strongest selective forces, in that it can determine not only whether or not an individual survives to fledge (reviewed in Sockman et al. 2006) but also the quality of sexual signals it later produces as an adult, if it does survive (Nowicki et al. 2002, Buchanan et al. 2003). Intriguingly, nestling bill morphology is in some species a plastic trait, in that competition between sibling nest-mates can affect the bill morphology of nestlings (Gil et al. 2008) and adults (de Kogel 1997), and presumably this effect is mediated through food acquisition. This leads to the hypothesis that the order in which young hatch affects their bill morphology, as asynchrony affects a nestling's ability to compete with its siblings (see citations above). Additionally, offspring produced late in a breeding season often face worse prospects for growth and survival than their counterparts produced earlier, owing to a tendency for lower-quality parents to delay reproduction and possibly to seasonal decline in food abundance and quality during the race to fuel development, deposit energy stores, and disperse before conditions deteriorate (Daan et al. 1990, Rowe et al. 1994). Given that the fitness effects of both hatching order and seasonal timing of development are mediated, at least in part, by sibling competition or food acquisition during begging and that nestling competition or food acquisition can influence nestling and adult bill morphology, I predicted that nestling bill morphology is associated with hatching order and the seasonal timing of development and that these relationships carry over into the adult life stage, potentially providing a proximate explanation for adult variation in a trait under strong selective forces.

METHODS

With help from field assistants, I studied Lincoln's Sparrows during the breeding seasons of 2006–2009 at Molas Pass, Colorado (37.74° N, 107.69° W). At an elevation of 3250 m, the study site is an open, wet, subalpine meadow approximately 20 ha in area. I have previously described details of the field site, the natural history of the Lincoln's Sparrow, and some materials and methods relevant to this study (Sockman 2008, 2009). Below, I reiterate some of these details.

DATA ON BILL SHAPE, NESTS, HATCHING ORDER, AND SEX

Through the breeding season (early June–late July), field assistants and I captured free-living adults in mist nets or seed-baited Potter traps, sexed them by the presence or absence of a cloacal protuberance, measured their bill height and width at the center of the nares with dial calipers, and then calculated their bill shape, which, hereafter, I define as height/width. This enabled me to analyze sex differences in the bill shape of free-living adults. Other metrics of bill morphology are undoubtedly important in this and other species;

I chose this particular bill metric because it is the one most closely associated with the variation in vocal performance of adult Lincoln's Sparrows described above (Sockman 2009). Although such composite measures can raise concerns in their statistical analysis (Corruccini 1977), a powerful information-theoretic approach revealed that other measures, including height alone and width alone, do not explain variation in vocal performance as efficiently as this composite measure (Sockman 2009).

We found nests, estimated the date of clutch initiation (as the metric for seasonal timing of development), and determined the order in which nestlings hatched by methods previously described (Sockman 2008, 2009). We marked nestlings by clipping a unique toe nail and then by banding them at approximately 7–8 days of age. Ages reported in Results are those of the individual nestling, not necessarily of the brood.

Of 444 nestlings that we banded and released from 2005 to 2008, we recaptured 10 (2.25%) as adults in a subsequent season (2006–2009). One of these fledged in 2005, a year in which we were not yet measuring bill shape. Thus, in this population, philopatry is very low, leaving only nine individuals whose bill shape we recorded as both a nestling and as an adult. Each of these individuals fledged from a different nest. To facilitate this comparison, we also collected and hand-raised many nestlings to adulthood (see below).

In 2007 and 2008, we measured nestling bills (height/width at the center of the nares and the length of the culmen), beginning when the oldest nestling in each nest was 4 days old and again near the end of the nestling stage. At the high elevation of this study site, the weather can be very cold and wet, even in the summer. Because of this and high rates of nest predation and abandonment, we minimized time spent at nests collecting data, particularly at hatching, when nestlings were most vulnerable. Therefore, we have no bill-shape measures at hatching. In 2008, we collected a small blood sample from nestlings for sex identification (because, unlike the reproductive adults described above, nestlings are sexually monomorphic), as described below. From 2006 to 2008, we collected a subset of broods ($n = 21$), measured the nestlings' bills (except in 2006), and hand fed them for approximately 2 weeks, until they were feeding independently, ad libitum. Nestling Lincoln's Sparrows often fledge prematurely if handled at 8 days of age. Occasionally, they fledge naturally (unprovoked by handling) at this age. Because we wanted to maximize the duration of a natural developmental experience, we waited as long as possible (until the brood was 8 days of age) before collecting broods (the birds are much more difficult to catch as fledglings). However, to prevent premature fledging, we measured bills at 7 days of age in broods that we did not plan to collect. Once the collected birds had reached adulthood several months later, we measured their bills again (or for the first time in broods hatched in 2006). For sexing, we also collected a blood sample from birds we had not previously sampled

(i.e., those hatched in 2006 and 2007). We extracted DNA from each blood sample with an InstaGene matrix (Bio-Rad Laboratories, Hercules, CA) and then identified the sex by the method of Griffiths et al. (1998).

STATISTICAL ANALYSES

For analyses with hatching order as a predictor, I used only those nests in which all eggs hatched, and I defined hatching order categorically—as first, middle, or last—because this definition dissociates hatching order from brood size, which, in Lincoln's Sparrow, typically varies from three to five nestlings. For more explanation of this approach, see Sockman (2008). Sample sizes vary with analysis because of variation in available data. For example, sample sizes with hatching order as a predictor are smaller than those without because I lack hatching-order data for some samples.

The high nestling mortality alluded to above resulted in variation between broods in number of nestlings and between nestlings in the number of measurements recorded. In addition, the data set consisted of a combination of fixed and hierarchically structured random effects, each of which may differ from the others in its correlation structure (e.g., observations at ages 4 and 7 days nested within individuals and individuals nested within broods). Therefore, I used a mixed-model framework (with the software Stata IC 10.0 for the Macintosh, Stata Corporation, College Station, TX), which is readily amenable to unbalanced data sets with combinations of fixed and hierarchically structured random effects (Burton et al. 1998, Goldstein et al. 2002, Rabe-Hesketh and Skrondal 2005). These models use *z*-tests, which assume a standard normal sampling distribution under the null hypothesis that a coefficient equals 0. They have the advantage of estimating parameters with maximum-likelihood procedures (I used restricted maximum likelihood), which are often more accurate and more powerful than the traditional least-squares estimates used in analyses of variance and other linear models (e.g., Goldschmidt and Timm 2003, Whitman 2003, Orton and Lark 2007). I followed the advice of Schielzeth and Forstmeier (2009) in modeling random effects as both random coefficients (with unstructured covariance matrices) and random intercepts when levels of the predictor varied between units of my random effect and as only random intercepts when they did not. For additional information on applying this mixed, multi-level modeling approach to morphological measures of individuals clustered within broods, see Sockman et al. (2008).

RESULTS

A nestling's bill shape positively predicted its bill shape as an adult. Specifically, the higher the value of an individual's bill shape (i.e., its height/width value) when it was a free-living nestling, the higher it was later when it matured into a laboratory-housed adult (Fig. 2, top panel). To assess the statistical reliability of this relationship, I used nestling bill shape at

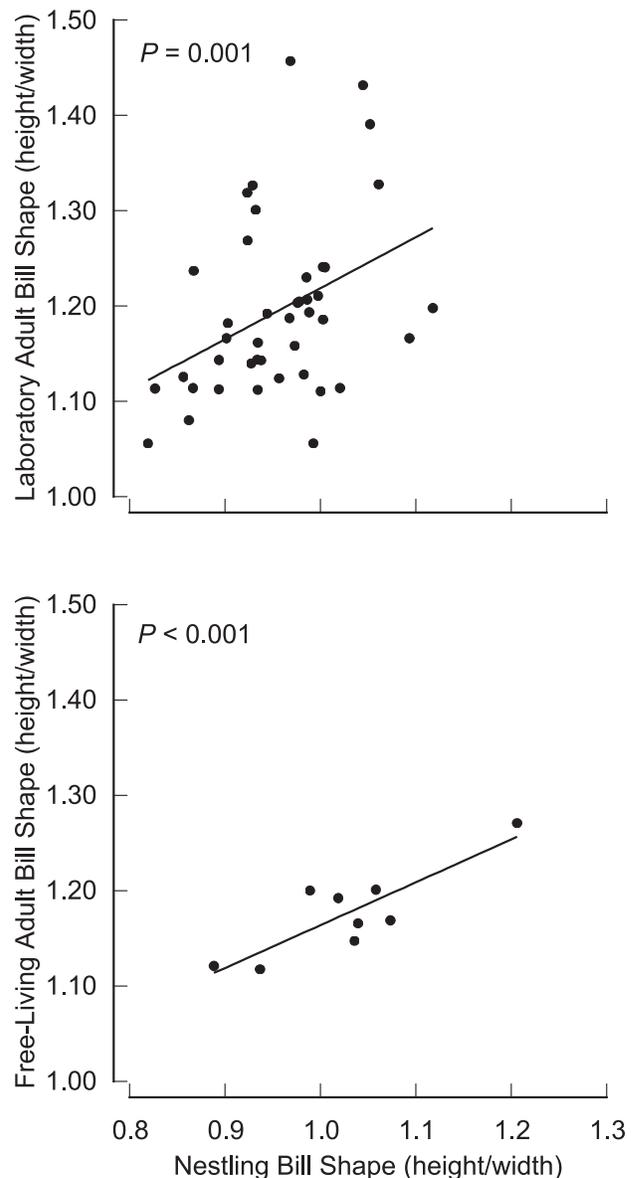


FIGURE 2. In Lincoln's Sparrow, the shape of a nestling's bill predicts the bill's shape when the bird is an adult. All nestlings were free-living in their natural environment when measured. Some were then collected and raised to adulthood in the laboratory (top panel), whereas others were not collected but instead captured free-living on the study site as adults a year or more later (bottom panel). *P* values are for the effects of nestling bill shape on adult bill shape from mixed-effects models explained in the text. Prediction lines are from linear regressions.

8 days of age ($z = 3.19$, $P = 0.001$) as a predictor of the response adult bill shape and nested individual nestling ($n = 42$) within brood ($n = 21$) as a random intercept and as a random coefficient on nestling bill shape.

The bill shape of hand-reared birds might not follow natural patterns of development because of the artificial

environment of the laboratory. Moreover, perhaps the correlation between the bill shape of nestlings and that of adults described above (Fig. 2, top panel) would disappear under more natural conditions. Thus it would be ideal to compare the bill shapes of nestlings and adults living free in their natural environments. This comparison requires a bird be measured as a nestling and in a subsequent year when it is a free-living adult. For only four of the nine individuals available for this comparison (see Methods) did I know hatching order, and for only six did I know date of clutch initiation. Consequently, I was not able to evaluate the relationship between either of these variables and the bill shape of free-living adults. Using a general linear model, however, I was able to determine if a nestling's bill shape predicted that when it was a free-living adult. If I had a nestling's bill-shape measurements at multiple ages (e.g., 4 and 7 days), I used the latter measurement. For a few nestlings, however, I had only a measurement at 4 days of age, which I used instead. As indicated below, there is no detectable change in this metric from day 4 to day 7, so it should not matter which I used. I found that the bill shape of nestlings ($z = 4.31$, $P < 0.001$, $n = 9$ individuals) strongly predicted their bill shape once they had matured into adults, living free in their natural environment (Fig. 2, bottom panel). One caveat is that it appears that this correlation could have been driven primarily by the single individual with the highest bill-shape values as both a nestling and as an adult. However, removal of this individual from the analysis did not change the conclusion that the bill shape of free-living nestlings predicted their bill shape when they matured into free-living adults ($z = 2.17$, $P = 0.03$, $n = 8$ individuals).

Nestling bill shape declined with both hatching order (Fig. 3) and date of clutch initiation (Fig. 4, left panel). To assess the statistical reliability of these relationships, I simultaneously analyzed the effects of the predictors date of clutch initiation, hatching order, and nestling age (because I used repeated measurements of nestlings at ages 4 and 7 days). I expanded hatching order into a dummy-variable set to model the contrast between the first- and last-hatched and the independent contrast between the middle and last hatched. I nested the two measurement observations (at ages 4 and 7 days) ($n = 62$) within nestling ($n = 43$) as a random intercept and random coefficient on age, and I nested nestling within brood ($n = 21$) as a random intercept and a random coefficient on hatching order. The declines in bill shape from the first to the last hatched ($z = 2.96$, $P = 0.003$), from the middle to the last hatched ($z = 2.11$, $P = 0.035$), and with date of clutch initiation ($z = -2.49$, $P = 0.013$) were all statistically reliable. I observed no change in bill shape from age 4 to 7 days ($z = 0.65$, $P > 0.2$), but this is not necessarily surprising, given that this metric is a ratio of two size measurements. Although height and width each increased from day 4 to day 7 (data not shown), they must have increased proportionally to one another.

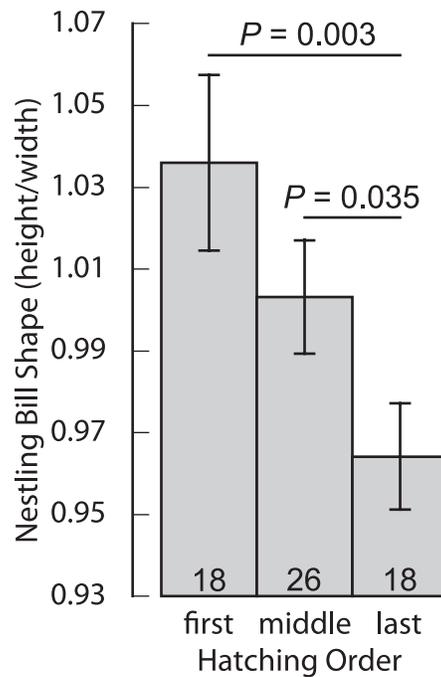


FIGURE 3. Order of hatching predicts bill shape of nestling Lincoln's Sparrows. Values are means (\pm SE) that were adjusted to the means of other variables (age, date of clutch initiation) in the statistical model. The number of birds for each sequential category (first, middle, last) is indicated at the base of each bar. P values are for independent contrasts indicated in the graph and specified by the mixed-effects models explained in the text.

The findings above raised the question of the specificity of these relationships and thus what other aspects of nestling morphology might have been associated with hatching order and date of clutch initiation. To examine this, I used the same model as above, except with culmen length as the response variable. I found no evidence that culmen length changed with date of clutch initiation ($z = 1.32$, $P = 0.18$), the contrast between the first- and last-hatched ($z = 0.68$, $P > 0.2$), or the contrast between the middle- and last-hatched ($z = 0.47$, $P > 0.2$). However, culmen length increased from day 4 to day 7 ($z = 11.03$, $P < 0.001$).

Given that nestling bill shape was associated with both hatching order (Fig. 3) and date of clutch initiation (Fig. 4, left panel) and that a nestling's bill shape positively correlated with its bill shape as an adult (Fig. 2), it follows that an individual's bill shape as an adult should be associated with its hatching order and the date its egg was laid. Using a mixed-effects model with individual ($n = 70$) nested within brood ($n = 30$) as a random intercept, I found that adult bill shape declined with date of clutch initiation ($z = -4.37$, $P < 0.001$) (Fig. 4, right panel), indicating that the seasonal timing of development predicted adult bill shape to some extent. Unfortunately, I knew hatching order for only a small number of nestlings

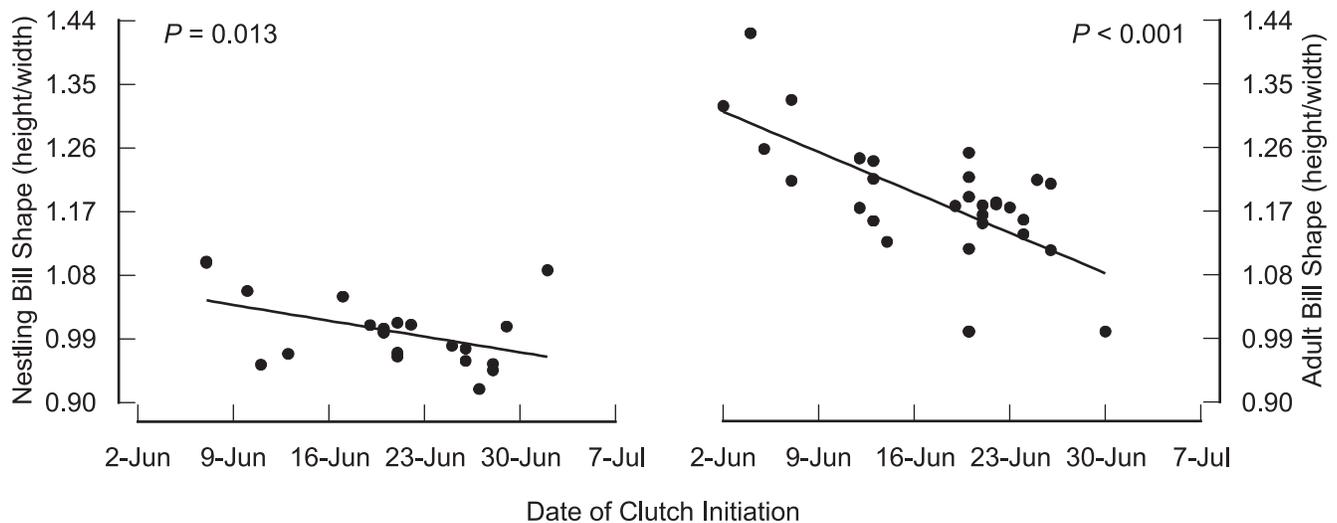


FIGURE 4. Seasonal timing of development (date of clutch initiation) predicts the bill shape of nestling (left panel) and adult (right panel) Lincoln's Sparrows. Each point corresponds to the value for a single brood, which is the mean of each brood's nestling-mean. There are two broods with identical (overlapping) values of 20 June and 1.00 in the right panel. P values are for the effects of date on bill shape from mixed-effects models explained in the text. Prediction lines are from linear regressions.

that we collected and raised to adulthood, precluding a meaningful analysis with this predictor.

Graham et al. (2011) found a seasonal decline in the percentage of Lincoln's Sparrow nestlings that were male. Thus, if the sexes differ in bill shape, then the relationship between date of clutch initiation and bill shape described above could be explained by effects of date of clutch initiation on the offsprings' sex. Additionally, the relationship between hatching order and bill shape could also be explained by effects of hatching order on the offsprings' sex. To analyze whether hatching order or date of clutch initiation affected sex in this particular sample of offspring, I used a mixed-effects logistic regression, nesting individual ($n = 31$) within brood ($n = 16$) as a random intercept and random coefficient on hatching order and found no evidence that either hatching order (contrast between first and last hatched: $z = 0.71$, $P > 0.2$; contrast between middle and last hatched: $z = 0.76$, $P > 0.2$) or date of clutch initiation ($z = -1.02$, $P > 0.2$) affected the offsprings' sex. Also, nesting individual within brood as a random intercept and a random coefficient on sex, I analyzed the relationship between sex and bill shape at age 4 days ($n = 75$ individuals nested within 29 broods) and 7 days ($n = 38$ individuals nested within 15 broods) and in laboratory-housed adults ($n = 73$ individuals nested within 32 broods) and found no evidence that males differed from females in bill shape (each age: $|z| \leq 1.22$, $P > 0.2$) (Fig. 5). In an additional effort to find a relationship between bill shape and sex, I took advantage of a much larger set of data on bill shapes of free-living adults that I did not use in the analyses above because I did not measure the birds as nestlings. Again, I found no evidence for a sex difference in bill shape ($z = 0.93$, $P > 0.2$, $n = 266$ individuals) (Fig. 5).

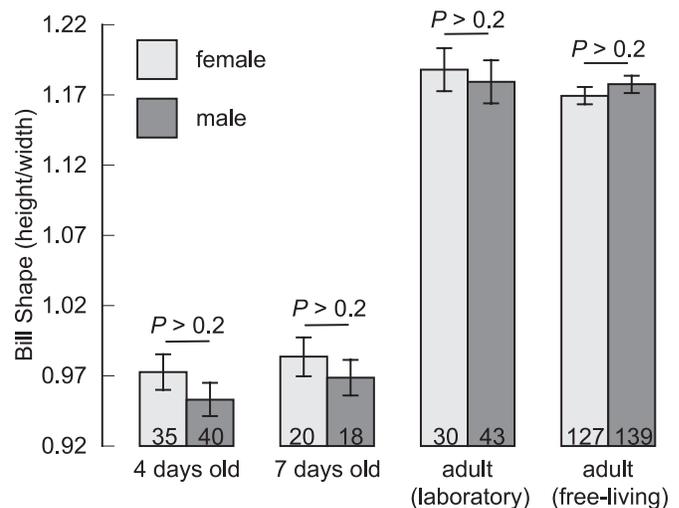


FIGURE 5. Relationship between sex and bill shape (mean \pm SE) at multiple life stages in the Lincoln's Sparrow. The number of birds is indicated at the base of each bar. P values are for the effects of sex on bill shape from mixed-effects models explained in the text.

DISCUSSION

In multiple species, an adult's bill morphology may influence its choice of or fit to its foraging niche (Price et al. 1984, Smith 1987, Grant and Grant 1995, Benkman and Miller 1996, Badyaev et al. 2008) and may also affect its vocal sexual signaling (Podos 2001, Badyaev et al. 2008, Sockman 2009). Here, I report for the Lincoln's Sparrow that nestling bill shape (height/width) predicts adult bill shape, that the order in which an

individual hatches relative to its sibling nest mates predicts nestling bill shape, and that the seasonal timing of a nestling's development predicts its bill shape as both a nestling and as an adult. This suggests that developmental conditions can serve as a proximate source of variation in adult bill shape.

At first pass, it seems possible that my results could be explained by an interaction between sex differences in bill shape and the effects of hatching order and seasonal timing of development on the offsprings' sex. Indeed, in some species (reviewed in Komdeur and Pen 2002), the offsprings' sex can vary with hatching order (e.g., Badyaev et al. 2002) and seasonal timing of development (e.g., Daan et al. 1996, Graham et al. 2011). In my study, however, I was unable to find a relationship between these variables nor did I find a sex difference in the bill shape of either nestlings or adults (Fig. 5), making it unlikely that the results I report here could have been caused by such an interaction. This lack of sex differences may seem surprising given the probability that this trait is under sexual selection. However, sexual monomorphism in traits under sexually antagonistic selection is not uncommon and may be explained by any of several phenomena, including unresolved intra-locus sexual conflict, temporal variation in selection, and selection on the portion of phenotypic variation that is not heritable (Cox and Calsbeek 2009).

The results presented here raise questions about the proximate mechanisms for variation in nestling bill morphology. Although traits need not be heritable to be targets of selection, one possibility is that bill shape is genetically inherited from the parents. Indeed, it is the heritability of bill morphology in Darwin's finches that has formed much of the basis for how natural selection might act on morphological traits to drive rapid speciation in that group (Grant and Grant 2008). Perhaps nestling Lincoln's Sparrows are inheriting their parents' bill shapes and that the frequencies of alleles influencing this trait change seasonally. I know too little as yet about the parentage of the nestlings I studied for a meaningful analysis of bill-shape heritability. Still, although it is certainly plausible that genetic inheritance may be driving the effect of seasonal timing of development (Fig. 4), it seems unlikely as a mediator of the hatching-order effects (Fig. 3), simply because the parentage of last-hatched chicks is unlikely to differ in genome type from that of first- and middle-hatched nest mates. Instead, the relationship between hatching order and bill shape in Lincoln's Sparrow (Fig. 3) is probably a product of developmental experience, and the relationship between seasonal timing of development and bill shape (Fig. 4) may be as well.

I was unable to measure bill shape immediately after hatching, so it is not clear if the relationships between hatching order and bill shape and between seasonal timing of development and bill shape arose *before* hatching, perhaps through the differential allocation of resources or compounds (e.g., hormones) to the developing embryo, or *after* hatching, perhaps through the response of nestlings to food acquisition or competition. To investigate each of these possibilities, one

could begin by examining whether and how eggs differ relative to the seasonal timing of their production or to the order in which they are laid, which, in Lincoln's Sparrow, almost perfectly mirrors the order in which they hatch (Sockman 2008). Across a diversity of bird species, females routinely produce eggs that, in relation to laying order or seasonal timing of production, vary in size and their maternal allocation of yolk steroids and other yolk components (reviewed by Sockman et al. 2006). The size of an egg is often associated with the size of the nestling hatching from it (Williams 1994), and yolk androgens can influence the mass of the musculus complexus (hatching muscle) (Lipar and Ketterson 2000) and nestling growth (for review, see Sockman et al. 2006). I have been unable to find a reliable relationship between egg size and laying order in the Lincoln's Sparrow (Sockman 2008), and for no species am I aware of a maternally deposited factor, be it an androgen or otherwise, that influences the bill morphology of the nestling hatching from the egg. Still, the possibility that some aspect of the egg affects the bill shape of the nestling hatching from it merits investigation.

Although differences between eggs are certainly one plausible, pre-hatching basis for the relationships I observed, food acquisition or competition during the nestling stage is a viable candidate for the post-hatching mediation of these relationships. Zebra Finches (*Taeniopygia guttata*) reared in experimentally reduced broods have taller bills as adults than do those reared in experimentally increased broods (de Kogel 1997). Presumably, sibling competition is less in smaller broods, which results in more food for the nestlings in smaller broods than in larger broods. A similar phenomenon occurs in the Spotless Starling (*Sturnus unicolor*); when competition within a brood is experimentally elevated and when food conditions worsen, nestlings develop wider gapes (Gil et al. 2008). In short, there is a reasonable foundation for examining the hypothesis that food acquisition or competition during the nestling stage affects bill shape.

The possibility that sibling competition or food acquisition regulates the bill shape of nestlings and adults raises a hypothesis about the ultimate basis for variation in this naturally and sexually selected trait. If a flatter, wider bill means a larger gape for the ingestion of larger food boluses (Illies 1975, Wiebe and Slagsvold 2009) or means a larger target or stronger signal for parents delivering food, a nestling might benefit from a wider bill if it is facing problems with food acquisition or competition or if it is predisposed toward these problems by virtue of its hatching later than its nestmates or at an unfavorable time (see Slagsvold and Wiebe 2007, Wiebe and Slagsvold 2009 for possible scenarios in which this might occur). An analogous phenomenon occurs in spadefoot toads (*Spea* spp.), where competition between two species for limited resources during development drives the development of alternative phenotypes that reduces the competition (Pfennig et al. 2006). However, as explained earlier, in adult Lincoln's Sparrows wider bills are associated with poorer performance of vocal sexual signals (Sockman 2009). Therefore,

there may be a trade-off between life stages in the optimization of bill morphology, and the optimal resolution of that trade-off may depend on the circumstance of an individual's development. Nestlings otherwise handicapped by hatching later in their brood or later in the season may benefit from a wider, flatter bill more than do nestlings hatching earlier in their brood or earlier in the season. The idea that developmental variation in food acquisition drives variation in the morphology of an adult songbird's bill is consistent with the results of this and other studies that reveal this form of phenotypic plasticity (de Kogel 1997, Gil et al. 2008). However, at this point, this idea is merely a hypothesis. It is also possible that the wider bill of later-hatched nestlings is not adaptive but instead due to a developmental constraint imposed by nutritional deprivation or some other factor.

Here, I have shown that developmental conditions associated with sibling competition and food availability can predict the developmental and adult value of a trait, bill morphology, that, in many bird species, is under strong selective pressure. This raises the possibility that variation between life stages in a trait's optimal state constrains the optimization of the trait at any one life stage.

ACKNOWLEDGMENTS

I thank the many individuals on my field crew who helped collect the data. I also thank S. P. Caro, E. B. Graham, and K. G. Salvante for the molecular sexing and D. W. Pfennig for comments on a previous draft of the manuscript. This study was supported in part by National Institutes of Health grant R01 NS055125 and a University of North Carolina award from the R. J. Reynolds fund.

LITERATURE CITED

- AMMON, E. M. 1995. Lincoln's Sparrow (*Melospiza lincolni*), no. 191. In A. Poole and F. Gill [EDS.], *The Birds of North America*. Academy of Natural Sciences, Philadelphia.
- BADYAEV, A. V., G. E. HILL, M. L. BECK, A. A. DERVAN, R. A. DUCKWORTH, K. J. MCGRAW, P. M. NOLAN, AND L. A. WHITTINGHAM. 2002. Sex-biased hatching order and adaptive population divergence in a passerine bird. *Science* 295:316–318.
- BADYAEV, A. V., R. L. YOUNG, K. P. OH, AND C. ADDISON. 2008. Evolution on a local scale: developmental, functional, and genetic bases of divergence in bill form and associated changes in song structure between adjacent habitats. *Evolution* 62:1951–1964.
- BALLENTINE, B., J. HYMAN, AND S. NOWICKI. 2004. Vocal performance influences female response to male bird song: an experimental test. *Behavioral Ecology* 15:163–168.
- BENKMAN, C. W., AND R. E. MILLER. 1996. Morphological evolution in response to fluctuating selection. *Evolution* 50:2499–2504.
- BUCHANAN, K. L., K. A. SPENCER, A. R. GOLDSMITH, AND C. K. CATCHPOLE. 2003. Song as an honest signal of past developmental stress in the European Starling (*Sturnus vulgaris*). *Proceedings of the Royal Society of London B* 270:1149–1156.
- BURTON, P., L. GURRIN, AND P. SLY. 1998. Extending the simple linear regression model to account for correlated responses: an introduction to generalized estimating equations and multi-level mixed modeling. *Statistics in Medicine* 17:1261–1291.
- CARO, S. P., K. B. SEWALL, K. G. SALVANTE, AND K. W. SOCKMAN. 2010. Female Lincoln's Sparrows modulate their behavior in response to variation in male song quality. *Behavioral Ecology* 21:562–569.
- CORRUCCINI, R. S. 1977. Correlation properties of morphometric ratios. *Systematic Zoology* 26:211–214.
- COX, R. M., AND R. CALSBEEK. 2009. Sexually antagonistic selection, sexual dimorphism, and the resolution of intralocus sexual conflict. *American Naturalist* 173:176–187.
- DAAN, S., C. DIJKSTRA, AND J. M. TINBERGEN. 1990. Family planning in the kestrel (*Falco tinnunculus*): the ultimate control of covariation of laying date and clutch size. *Behaviour* 114:83–116.
- DAAN, S., C. DIJKSTRA, AND F. J. WEISSING. 1996. An evolutionary explanation for seasonal trends in avian sex ratios. *Behavioral Ecology* 7:426–430.
- DE KOGEL, C. H. 1997. Long-term effects of brood size manipulation on morphological development and sex-specific mortality of offspring. *Journal of Animal Ecology* 66:167–178.
- DRAGANOIU, T. I., L. NAGLE, AND M. KREUTZER. 2002. Directional female preference for an exaggerated male trait in canary (*Serinus canaria*) song. *Proceedings of the Royal Society of London B* 269:2525–2531.
- FORBES, S. 2010. Family structure and variation in reproductive success in blackbirds. *Behavioral Ecology and Sociobiology* 64:475–483.
- FORBES, S., AND D. W. MOCK. 2000. A tale of two strategies: life-history aspects of family strife. *Condor* 102:23–34.
- GIBBS, H. L., AND P. R. GRANT. 1987. Oscillating selection on Darwin's finches. *Nature* 327:511–513.
- GIL, D., E. BULMER, P. CELIS, AND I. LÓPEZ-RULL. 2008. Adaptive developmental plasticity in growing nestlings: sibling competition induces differential gape growth. *Proceedings of the Royal Society of London B* 275:549–554.
- GOLDSCHMIDT, L., AND N. H. TIMM. 2003. A comparison of noniterative generalized least squares and iterative maximum likelihood estimators when testing hypotheses in random coefficient growth curve models. *Communications in Statistics—Simulation and Computation* 32:297–318.
- GOLDSTEIN, H., W. BROWNE, AND J. RASBASH. 2002. Partitioning variation in multilevel models. *Understanding Statistics* 1:223–231.
- GRAHAM, E. B., S. P. CARO, AND K. W. SOCKMAN. 2011. Change in offspring sex ratio over a very short season in Lincoln's Sparrows: the potential role of bill development. *Journal of Field Ornithology* 82:44–51.
- GRANT, P. R., AND B. R. GRANT. 1995. Predicting microevolutionary responses to directional selection on heritable variation. *Evolution* 49:241–251.
- GRANT, P. R., AND B. R. GRANT. 2008. How and why species multiply: the radiation of Darwin's finches. Princeton University Press, Princeton, NJ.
- GRIFFITHS, R., M. C. DOUBLE, K. ORR, AND R. J. G. DAWSON. 1998. A DNA test to sex most birds. *Molecular Ecology* 7:1071–1075.
- HERREL, A., J. PODOS, S. K. HUBER, AND A. P. HENDRY. 2005. Bite performance and morphology in a population of Darwin's finches: implications for the evolution of beak shape. *Functional Ecology* 19:43–48.
- ILLIES, J. 1975. Schlüsselreize fürs gemüt. *Kosmos* 8:324–330.
- KOMDEUR, J., AND I. PEN. 2002. Adaptive sex allocation in birds: the complexities of linking theory and practice. *Philosophical Transactions of the Royal Society of London Series B* 357:373–380.
- LIPAR, J. L., AND E. D. KETTERSON. 2000. Maternally derived yolk testosterone enhances the development of the hatching muscle in the Red-winged Blackbird *Agelaius phoeniceus*. *Proceedings of the Royal Society of London B* 267:2005–2010.
- MOCK, D. W., AND G. A. PARKER. 1997. The evolution of sibling rivalry. Oxford University Press, Oxford, England.

- NOWICKI, S., W. A. SEARCY, AND S. PETERS. 2002. Brain development, song learning and mate choice in birds: a review and experimental test of the "nutritional stress hypothesis." *Journal of Comparative Physiology A* 188:1003–1014.
- ORTON, T. G., AND R. M. LARK. 2007. Estimating the local mean for Bayesian maximum entropy by generalized least squares and maximum likelihood, and an application to the spatial analysis of a censored soil variable. *European Journal of Soil Science* 58:60–73.
- PFENNIG, D. W., A. M. RICE, AND R. A. MARTIN. 2006. Ecological opportunity and phenotypic plasticity interact to promote character displacement and species coexistence. *Ecology* 87:769–779.
- PODOS, J. 2001. Correlated evolution of morphology and vocal signal structure in Darwin's finches. *Nature* 409:185–188.
- PRICE, T. D., P. R. GRANT, H. L. GIBBS, AND P. T. BOAG. 1984. Recurrent patterns of natural selection in a population of Darwin's finches. *Nature* 309:787–789.
- RABE-HESKETH, S., AND A. SKRONDAL. 2005. Multilevel and longitudinal modeling using Stata. Stata Press, College Station, TX.
- ROWE, L., AND D. HOULE. 1996. The lek paradox and the capture of genetic variance by condition dependent traits. *Proceedings of the Royal Society of London B* 263:1415–1421.
- ROWE, L., D. LUDWIG, AND D. SCHLUTER. 1994. Time, condition, and the seasonal decline of avian clutch size. *American Naturalist* 143:698–722.
- SCHIELZETH, H., AND W. FORSTMEIER. 2009. Conclusions beyond support: overconfident estimates in mixed models. *Behavioral Ecology* 20:416–420.
- SLAGSVOLD, T., AND K. L. WIEBE. 2007. Hatching asynchrony and early nestling mortality: the feeding constraint hypothesis. *Animal Behaviour* 73:691–700.
- SMITH, T. B. 1987. Bill size polymorphism and intraspecific niche utilization in an African finch. *Nature* 329:717–719.
- SNOWBERG, L. K., AND C. W. BENKMAN. 2009. Mate choice based on a key ecological performance trait. *Journal of Evolutionary Biology* 22:762–769.
- SOCKMAN, K. W. 2008. Ovulation order mediates a trade-off between pre-hatching and post-hatching viability in an altricial bird. *PLoS One* 3:e1785.
- SOCKMAN, K. W. 2009. Annual variation in vocal performance and its relationship with bill morphology in Lincoln's Sparrows, *Melospiza lincolnii*. *Animal Behaviour* 77:663–671.
- SOCKMAN, K. W., P. J. SHARP, AND H. SCHWABL. 2006. Orchestration of avian reproductive effort: an integration of the ultimate and proximate bases for flexibility in clutch size, incubation behaviour, and yolk-androgen deposition. *Biological Reviews* 81:629–666.
- SOCKMAN, K. W., J. WEISS, M. S. WEBSTER, V. TALBOTT, AND H. SCHWABL. 2008. Sex-specific effects of yolk-androgens on growth of nestling American Kestrels. *Behavioral Ecology and Sociobiology* 62:617–625.
- WELTY, J. C. 1975. *The life of birds*. W. B. Saunders, Philadelphia.
- WHITMAN, C. S. 2003. Accelerated life test calculations using the method of maximum likelihood: an improvement over least squares. *Microelectronics Reliability* 43:859–864.
- WIEBE, K. L., AND T. SLAGSVOLD. 2009. Parental sex differences in food allocation to junior brood members as mediated by prey size. *Ethology* 115:49–58.
- WILLIAMS, T. D. 1994. Intraspecific variation in egg size and egg composition in birds: effects on offspring fitness. *Biological Reviews* 68:35–59.