Sexing Carolina Chickadees by Morphometric Measurement Analysis

Michele Grise, Jill M Russell*, Mary Lee Glore, David E Russell

College of Mount St. Joseph, Cincinnati, OH USA 45233
Miami University, Oxford, OH USA 45056

Correspondence Author: * Department of Biology, College of Mount St. Joseph, Cincinnati, OH USA 45056; (513) 244-4783; jill_russell@mail.msj.edu

The purpose of this study was to determine whether a sexually monochromatic species, the Carolina Chickadee, could be accurately sexed using morphometrics. Standard measurements including culmen length, tarsus length, wing chord, and tail length were collected from 52 known male and female Carolina Chickadees mist-netted and banded in southwestern Ohio. Stepwise discriminant analysis of these measurements resulted in the correct classification of sex 72% of the time. These data suggest that this model will aid in sexing Carolina Chickadees in the field, but factors such as inter-bander measurement variability, sample size, and sexing by breeding morphology need to be controlled and further studied.

Introduction

Carolina Chickadees (*Poecile carolinensis*) are members of the family Paridae and year-round residents of central and southern Ohio (Figure 1). The general habitat of the Carolina Chickadee (hereafter, CACH) is wooded areas such as temperate forests near or bordering bodies of water (Mostrom, et al. 2002). Specific habitat requirements include multiple layered forests that provide dead snags or living trees with cavities for nesting (Hamel 1992). Bird populations are often used as environmental indicators because they are more easily surveyed than most other organisms, and changes in their populations may reflect broader environmental changes. A decline in CACH numbers may indicate a decrease in available food, shelter, or breeding habitat. For example, CACH populations were used to look at the effects of deforestation on avian hormone levels, body mass, and caching rates (Lucas et al. 2006). Not only are the numbers of CACH important when looking at

Figure 1: Carolina Chickadee

environmental changes such as climate change and deforestation, but the distribution of male and female birds also provides additional information about the dynamics of the population. For instance if the population is skewed (more males than females or vice versa) it implies differential survivorship of sexes, which would impact breeding and population numbers. Therefore, it is also important to quantify the number of male and female birds when monitoring populations. (DeSante 2010)

Generally, sexing birds does not pose a problem, because males and females of many species have different plumages. However, many species have been categorized as monochromatic to human perception, only to be reclassified as dichromatic as our understanding of differences between bird and human visual acuity have been evaluated—monochromatic to humans is not the same as monochromatic to birds. A study by Eaton (2007) of 166 North American passerine species categorized as sexually monochromatic to human perception showed only 14 of these species to be monochromatic to their avian counterparts. Interestingly, CACH was one of the species confirmed as monochromatic to birds (as well as humans) through their color discrimination model analysis (Eaton 2007). CACH are apparently monochromatic, with plumages of males and females being indistinguishable to the human eye, which makes it difficult to determine the sex of individuals in the field (Pyle 2007). Sex can be determined noninvasively during breeding season by observing the presence or absence of a cloacal protuberance in males or brood patch in females (Twedt 2004), but, during the rest of the year, the sex differences cannot be observed. In other apparently monochromatic species such as Common Terns (*Sterna hirundo*) and Arctic Terns (*Sterna paradisaea*) discriminate function analysis of the bill (culmen, length, breadth, and depth) has been used to determine sex (Coulter 1986; Fletcher and Hammer 2003). Fletcher and Hammer (2003) also showed that sexing terns using biometrics resulted in the highest accuracy, correctly sexing 84% of Arctic Terns and 86% of Common Terns. This study analyzed various body measurements in an attempt to sex CACH when not breeding.

Methods

We examined data collected from 52 known male and female CACH mist-netted and banded in southwestern Ohio between April of 2004 and February of 2009. Sex was determined by the presence of either a brood patch or cloacal protuberance (Pyle 1997). Some individuals originally designated as Unknown Sex at the time of their capture were later recaptured during the breeding season and included in the analysis. Standard body measurements collected from these CACH included: culmen length (CulL), tarsus length (TarL), wing chord (WC), and tail length (TL) (Pyle 1997). Each measurement was made to the nearest 0.1 mm using techniques per Pyle (1997). Culmen was measured

Figure 2: Culmen Measurement

from the anterior edge of the nostril to the bill tip (Figure 2). Tarsus was measured between the intertarsal joint and the distal end of the third leg scale just before the toes (Figure 3). Wing chord was a measurement of the unflattened wing from the carpal joint, or "wrist," to the tip of the longest primary, and tail length is defined as the distance between the tip of the longest rectrix (tail feather) and the point of insertion of the two central rectrices (Pyle 1997). The latter two measurements were made using standard wing rules with

Figure 3: Tarsus Measurement

Table 1: Students t-test

Sex	Calculation	WC	TL	TarL	CulL
F	Mean	59	51.45	16.05	6.89
M		61	54.55	16.66	6.81
F	Standard Deviation	2.25	3.16	0.71	0.36
M		1.18	5.44	1.25	0.55
F	Range	56-63	45-56	14.6-17.7	6.27-7.5
M		60-64	48-73	15 –20.7	6-8.3
	p value	0.003*	0.055†	0.095†	0.189

N= 52. Mean, standard deviation, and range of four body measurements of 19 male (M) and 33 female (F) Carolina Chickadees. * WC significantly different between males and females, $p < 0.05$. † TL and TarL significantly different between males and females, $p < 0.10$.

accuracy to the nearest 1.0 mm.

The goal of our statistical analysis was to derive the best gender prediction strategy based on the four morphometric measurements WC, TL, TarL, and CulL. For each measurement, the standard deviation, mean, range, and a Student's t-test were calculated. The four Students t-tests were performed to answer the question: "Is there enough evidence from the data to conclude that there is a difference between the mean measure for females and the mean measure for males?" If the *p*-value is small, then we say there is evidence of a difference. Usually, if the *p*-value is < .05, we call the result significant.

We used logistic regression (SPSS 16.0) to model the classification of the gender of the birds. The model predicts *p* the probability that a bird is female given the morphometric measurements of: CulL, TarL, WC, and TL. The statistical model for the analysis is that the logarithm of the odds of being female is linear: $\log(p/[1-p]) = \beta_0 + \beta_1 WC + \beta_2 TarL + \beta_3 CulL + \beta_4 TL$, where $(p/[1-p])$ are the odds that a bird is female given the 4 morphometric measurements. Logistic regression calculates the β's that best predict gender as female if *p* is greater than 0.5 and as male if *p* is less than 0.5 using a Maximum Likelihood method. This equation is our discriminant equation D. When $p>0.5$, the discriminant

Table 2: Logistic Regression Using the 3 Significant Measurements

Sex	Estimate (β)	Significance
WC	-.594	.007
TarL	-.728	.115
CulL	1.892	.044
Intercept	35.713	.013

Removing TL from the model, the remaining three measurements are significant and these numbers were used in the ultimate model equation: D = 35.713-0.594WC-0.728TarL+1.892CulL.

equation D > 0. So, $D = \beta_0 + \beta_1 WC + \beta_2 TarL + \beta_3 CulL + \beta_4 TL$. Therefore, for a given bird, if D is greater than 0, then the predictive probability that the bird is female is over ½, and we predict that the bird is female; otherwise, we predict that the bird is male.

Results

The average female WC was significantly shorter than average male WC at the 5% significance level, and the average TL and average TarL were both significantly shorter than the average male TL and average TarL at the 10% significance level. The average CulL was not significantly different among the genders (Table 1). In a stepwise discriminant analysis, WC, CulL, and TarL contributed most to the discrimination by sex, and TL did not improve discrimination significantly. Therefore, TL was removed from the model, resulting in the ultimate model of D=35.713-0.594WC-0.728TarL+1.892CulL (Table 2). The discriminant equation was applied to 52 birds with known sex and correctly predicted gender 72% of the time (Table 3).

Principal Component Analysis was also applied to the data. The first component was not significant and ultimately the discriminate equation was very similar to the component analysis model. This model added complexity but did not significantly improve the success of predicting gender.

Discussion

There are no known morphological characteristics

Table 3: Discriminant Function Results

	Predicted Sex		
Observed Sex	Female	Male	Percentage Correct
Female	26	7	79
Male	8	11	60
Overall Percentage			72

Prediction of sexes using the ultimate model D = 35.713 - 0.594WC - 0.728TarL + 1.892CulL. The model correctly predicted gender in females 79% of the time and in males 60% of the time. (N= 52: 19 male and 33 female).

that separate male and female Carolina Chickadees. Early research on related taxa, such as Black-capped Chickadees (hereafter, BCCH), used breeding behavior to determine sex (Odum 1941). However, these behavioral determinants are useless when observing large populations of non-breeders and individuals outside breeding season, or when handling birds at banding stations. Discriminant analysis using three body measures can dramatically improve the accuracy of year-round determination of BCCH sex over univariate or behavior methods (Desrochers 1990). Furthermore, once developed, the discriminant function and score can be a useful sexing tool for field research and banding stations. If we could develop a model/equation that field biologists could use, then they would be able to sex CACH in the field using morphometrics and not have to take tissue samples and examine DNA. Our data suggest that a number of factors make the development of a significant discriminant function for CACH in southwestern Ohio particularly challenging.

For example, BCCH are present in the northernmost counties of Ohio (Peterjohn 2001), yet, annually, some BCCH individuals move southward, and, periodically, large southward irruptions occur during winters of high food stress (Peterjohn 2001). Between the northern tier and central counties of Ohio is an intergrade zone between BCCH and CACH. Evidence from our bird-banding stations in southwestern Ohio suggests that as much as 15% of the chickadee population matches BCCH wing and tail length criteria (Russell in review).

Additionally, CACH show weak geographic variation among four subspecies; P.c. extima, carolinensis, atricapilloides, and agilis. P.c. extima (formerly extimus) occurs north of a rough line drawn from central North Carolina to western Tennessee, and P.c. carolinensis populations are south of that line (Pyle 1997). P.c. extima is listed as the representative subspecies in southwestern Ohio, although little information is available and no published account exists of the actual subspecies in this area. P.c. extima and P.c. carolinensis might intergrade in southwestern Ohio. Nine individuals (18%) in the current study have tail lengths characteristic of P.c. carolinensis. Another factor contributing to the difficulty of using a discriminant function analysis of morphometric measurements to sex CACH in the current study is inter-bander variability. It was impossible to determine which bander collected each individual measurement used in this study. Tarsus length in particular is quite variable among bird banders (Descrochers 1990), and future morphometric data should either be collected by only one bander or adjusted for interbander variability among mean tarsal lengths. This form of morphometric evaluation of sex produced a higher resolution (72%) than earlier studies in which discriminant analysis determined sex in only 12 to 70% of individuals (Cooper 1996). Yet, the correct assignment of sex in 72% of the individuals in this study is not a high enough percentage to provide confidence to the field biologist. Although this form of analysis provides additional information for evaluating CACH, correctly determining sex must be improved to be useful to the field researcher. Therefore, another study of morphometrics taken by a single bird bander would provide better resolution of the analysis. In the meantime, incorporating the use of DNA to sex non-breeding individuals will greatly improve the evaluation of discriminant function analysis.

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The Counties of Ohio