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Determining sex of South Island saddlebacks (*Philesturnus carunculatus carunculatus*) using discriminant function analysis

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Abstract Male and female South Is saddlebacks (*Philesturnus carunculatus carunculatus*) have monomorphic plumage characteristics and are not easily distinguishable. In this study, we developed discriminant functions to classify males and females using birds of known sex from Ulva I. Three discriminant functions using tarsus length, body mass, and a combination of tarsus length and body mass could classify birds with 90% accuracy.

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Keywords Ulva Island; South Island saddleback; *Philesturnus carunculatus*; sex determination; discriminant function analysis

INTRODUCTION

Identifying the sex of individuals in a population is fundamental to many detailed ecological, behavioural, and evolutionary studies, which rely on known numbers of each sex to examine parameters such as survival, mating behaviour, and effective population size. Discriminant function analysis is commonly used to distinguish males from females in avian species with monomorphic plumage characteristics or where the physical measurements of the sexes overlap. The technique statistically distinguishes between adults of known sex by creating a linear function of several measurements (Phillips & Furness 1997; Bertellotti *et al*. 2002; Genovart *et al*. 2003; Quintana *et al*. 2003).

There are no obvious differences between the sexes of many of New Zealand's threatened endemic bird species (e.g., Jenkins & Veitch 1991; Renner & Davis 1999; Armstrong 2001; Flux & Innes 2001; Setiawan *et al*. 2004). The South Is saddleback (*Philesturnus c. carunculatus*) shows sex-specific behaviour during courtship and incubation, but no sexual dimorphism in plumage coloration. It is time-consuming to look for rare reproductive behaviours during the breeding season, and impossible otherwise, but it is necessary to distinguish the sexes to interpret ecological data and it is especially important for assessing sex ratios in translocated populations where the individuals can be measured but the sex is seldom known. Male and female North Isl saddlebacks (*P. c. rufusater*) can be distinguished by tarsus length (Jenkins & Veitch 1991), but there is no comparable information for the South Is taxon (*P. c. carunculatus*). Body size is known to vary geographically in several species (Renner & Davis 1999). In saddlebacks, the presently-recognized subspecies have, amongst other features, noticeably different songs and juvenile plumage (Heather & Robertson 1996). A separate disciminant function for South Is saddlebacks is therefore desirable. Below we describe discriminant functions using external measurements to distinguish the sexes in South Is saddlebacks.

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METHODS

As part of a larger study, in 2000-2005 adult and yearling ("jackbird") saddlebacks on Ulva I (Paterson Inlet, Stewart Is) were caught in mistnets and each banded with a unique combination of colour bands. Males and females were identified during subsequent breeding seasons, based on behavioural observations consisting of courtship feeding by males and incubation behaviour by females.

The study was conducted under permits from the Department of Conservation and the University of Otago Animal Ethics Committee. Birds were captured and measured between Jul and Mar (Fig. 1). Wing length (maximum unflattened chord) was measured with a stopped wing ruler to the nearest ±1 mm and birds were weighed to ± 1g with spring scales. Tarsus length (intra-articular notch at the proximal end of the tarsometatarsus to the middle of the midtarsal articulation; Fig. 2), bill length (exposed culmen), bill depth (at nares), and bill width (at nares) were measured with Vernier callipers to ± 0.1 mm. Two measurements were typically taken from each side of the body and averaged. Over the course of the study, 10 different observers measured the males (n = 24) and females (n = 37) included in these analyses. Field methodology changed over the course of the study to reduce the time birds were handled , so some measurements were not obtained for all birds. Sample sizes therefore varied for each discriminant function examined.

Data analysis

Measurements were compared for birds of known sex using the discriminant function analysis (DFA) option in SPSS v.13.0. Before DFA analyses, assumptions of homogeneity of the within-group (i.e. sex) covariance matrix and deviation from multivariate normality were tested and met (*Box's M* = 0.018, approximate $F_{1, 9155}$ = 0.018, P = 0.89). The usefulness of each variable and combinations of variables in the DFA was then evaluated with *Wilks's Lambda*, a statistic that decreases as discriminatory power increases. The accuracy of the discriminant function was determined by the proportion of adults of known sex correctly classified using all of the individuals in the analysis and then by a jackknifing procedure that repeatedly removed a single individual and classified it based on the discriminant function generated for the remaining birds (SPSS v.13.0).

RESULTS

Measurements of male and female South Is saddlebacks overlapped but generally males were larger than females for all measurements (Table 1; Fig. 3). In the DFA, tarsus length, body mass, and wing length all performed well as single variables, especially tarsus and body mass, which individually assigned birds correctly 90% of the time (Table 2). The 3 discriminant functions that used a combination of variables had greater discriminatory power than single variables (as indicated by lower *Wilks's Lambda* Λ values), however they tended to assign birds incorrectly during jack-knifing procedures, probably as a result of smaller sample sizes (Table 2). Consequently, we identified 3 discriminant functions that had a high degree of discriminatory power and accuracy during jack-knifing. The 1st 2 functions used single variables (tarsus length, body mass); the 3rd used tarsus length and body mass together.

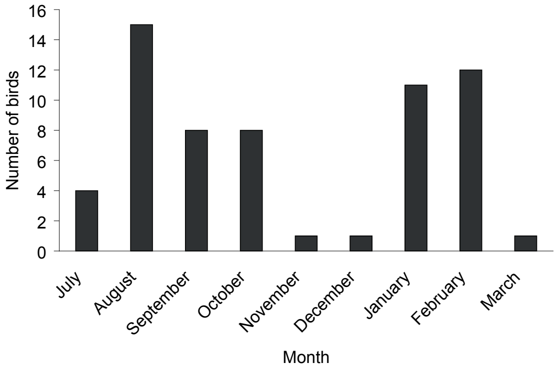

Fig. 1 Months of capture for South Is saddlebacks (*Philesturnus c. carunculatus*) used to develop discriminant function.

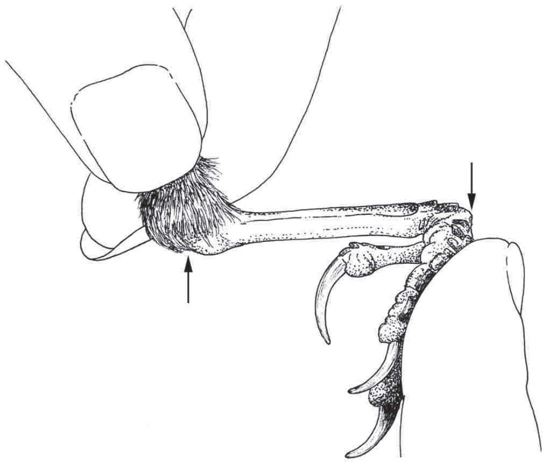

Fig. 2 Tarsus length measurement.

Table 1 Standard measurements (lengths, mm; mass, g; mean ± SD, n, range) of South Is saddlebacks (*Philesturnus c. carunculatus*) on Ulva I, Stewart Is group.

	Sex	
Variable	Male	Female
Bill length	34.68 ± 1.90	34.62 ± 2.30
	11, 31.0-38.2	28, 30.9-39.8
Bill depth	9.01 ± 0.71	8.61 ± 0.71
	15, 7.2-10.2	30, 6.9-9.5
Bill width	6.69 ± 0.33	6.46 ± 0.41
	13, 6.1-7.4	25, 5.7-7.5
Wing length	97.39 ± 1.86	94.47 ± 3.90
	14, 94.38-100.5	25, 86.0-105.0
Tarsus length	42.23 ± 1.44	39.46 ± 1.48
	24, 39.0-44.0	37, 33.4-41.9
Body mass	84.05 ± 4.99	73.15 ± 4.21
	22, 75.5-94.0	36, 64.0-83.0

All birds can be sexed using their discriminant scores, but the probability that an individual is

Table 2 Accuracy in sexing South Is saddlebacks (*Philesturnus c. carunculatus*) provided by discriminant function analysis and denoted by *Wilks's Lambda* (Λ), using single variables and the 3 best combined functions. *, not significant.

		% correct assignment					% correct (jack-knifing)		
Variable	Λ	Males	*n*	Females	*n*	Over all	Males	Females	Over all
Tarsus length	0.53	83.3	24	94.6	37	90.2	83.3	94.6	90.2
Body mass	0.41	86.4	22	91.7	36	89.7	86.4	91.7	89.7
Wing length	0.84	71.4	14	80.0	25	76.9	71.4	80.0	76.9
Bill length*	1.00	45.5	11	57.1	28	53.8	9.1	28.6	23.1
Bill depth*	0.93	73.3	15	43.3	30	53.3	73.3	43.3	53.3
Bill width*	0.92	69.2	13	56.0	25	60.5	69.2	56.0	60.5
Tarsus + body mass	0.36	90.9	22	91.7	36	91.4	90.9	91.7	91.4
Tarsus + wing + all bill	0.35	85.7	7	100.0	16	95.7	85.7	87.5	87.0
Tarsus + mass + wing + all bill	0.30	85.7	7	100.0	16	95.7	85.7	87.5	87.0

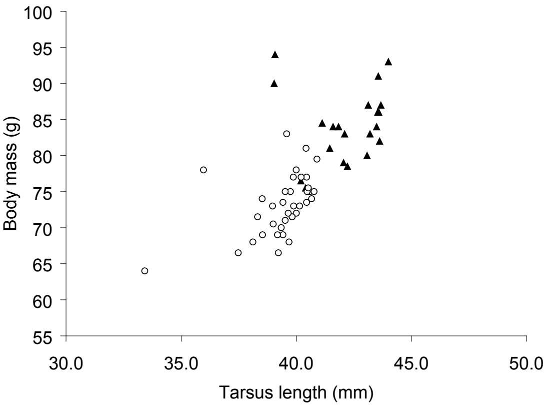

Fig. 3 Tarsus length and body mass measurements for male and female South Is saddlebacks (*Philesturnus c. carunculatus*). Filled triangles, males; open circles, females.

assigned correctly varied. The probability (*P*) that an individual belongs to a particular group is related to its discriminant score (D) by a logistic relationship:

$$P = 1 / (1 + e^{k*(D+c)}),$$

where *P* and D are obtained from the statistical output (in our case SPSS) , and k and c were obtained by performing a non-linear regression.

The logistic curve means that individuals with discriminant scores at or very near the cutoff values have *c.*50% probability of being correctly classified whereas birds with values slightly above or below the cutoff values have an exponentially greater probability of being assigned correctly. The following 3 discriminant functions can be used to sex South Is saddlebacks, and supplementary equations can be used to calculate the probability that a bird is male (probability bird is female = 1 – probability of being male).

D1 = -27.742 + 0.684 (tarsus length)

Birds with a D1 < 0.20 (or tarsus length < 40.85 mm) were classified as females, otherwise as males.

[Probability of being male = 1/(1 + exp(-1.898*(D1 – 0.202)))]

D2 = -17.096 + 0.221 (body mass)

Birds with a D2 < 0.29 (or body mass < 78.6 g) were classified as females, otherwise as males.

[Probability of being male = 1/(1 + exp(-2.410*(D2 – 0.291)))]

D3 = -25.714 + 0.321(tarsus length) + 0.165 (body mass)

Birds with a D3 < 0.324 were classified as females, otherwise as males.

[Probability of being male = 1/(1 + exp(-2.688*(D3 – 0.324)))]

DISCUSSION

Male and female South Is saddlebacks may be distinguished with >90% accuracy using tarsus length or body mass, or both measurements together. We recommend tarsus length as the most reliable variable because the tarsus is fully grown at fledging and thereafter is constant in size (Jenkins & Veitch 1991). Body mass is known to vary seasonally in many birds (Lima 1986; Moreno 1989) and so the 2 discriminant functions involving body mass may be contentious. However, body mass can be used for discriminant functions if it is measured at the same stage of the season and if variation among years is limited (Phillips & Furness 1997). Body mass in South Is saddlebacks accurately distinguished the sexes using data across several months (12 Jul - 4 Mar), and body mass data for North Is saddlebacks show that the lowest monthly mean for males (75.2 g) is greater than the highest monthly mean for females (69.1 g; Jenkins & Veitch 1991). These data suggest that body mass varies more between the sexes than within the sexes and therefore may be a reliable method for distinguishing males from females at any period of the year. However, we have no data for body mass outside our measurement period and data for some months within our measurement period are limited (Fig. 1). Tarsus length alone may be the most conservative method for sexing South Is saddlebacks.

Wing and bill measurements may also contribute significantly to the power of discriminant functions that use a combination of variables. Including wing and bill measurements produced low values for *Wilks's Lambda Λ* and a high degree of accuracy (95.7%) without resorting to the jackknifing procedure. However, jack-knifing reduced accuracy to 87%, probably because sample sizes were small. We were unable to determine the usefulness of wing and bill measurements but believe that larger sample sizes may show them to be effective.

Sexing birds using DFA may have limited application for populations that show geographical variation or measurements that are strongly biased by observers (Phillips & Furness 1997; Renner & Davis 1999). Following a population bottleneck in 1964, all South Is saddleback populations have been established by translocation and all individuals are descended from 36 birds (Merton 1975; Hooson & Jamieson 2003). If differential survivorship of the translocated individuals is related to morphology (e.g., Brown & Brown 1998), then separate discriminant functions may be necessary for individual populations. If not, there may be little geographic variation in the morphology of South Is saddlebacks .

Observer bias is most important when a discriminant function is created using measurements by a single observer and then applied to new individuals measured by a different observer (Phillips & Furness 1997). In contrast, discriminant functions using measurements by a large number of observers can be very accurate (Fox *et al*. 1981). Our discriminant function used measurements by 10 different observers for 61 saddlebacks. Given the large number of observers, our discriminant function should be less sensitive to observer bias and hence be applicable to measurements made by other observers.

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