

MORPHOMETRIC SIMILARITIES BETWEEN CENTRAL AND PERIPHERAL POPULATIONS OF THE EUROPEAN SHAG *PHALACROCORAX ARISTOTELIS*

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SUMMARY

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We compared morphometrics and discriminant functions for sexing European Shags *Phalacrocorax aristotelis* in a central (British) population and a peripheral (northwestern Iberia) population within its distribution range, to account for structural differences between populations and sexes. Overall, British shags were structurally larger than Iberian shags, except in their head and bill length. This north–south cline could be attributable to higher food availability or lower ambient temperatures at higher latitudes. Furthermore, Iberian male shags were structurally larger than females for all variables considered, except for bill length, which was similar for both sexes, as in the British population. This suggests that bill length is a conservative trait in the species, being similar between populations and sexes alike. The most parsimonious discriminant function for sexing Iberian shags included only bill depth, and it correctly sexed 92.6% of the original cases, as was the case among British shags. This suggests that the depth of the bill likely plays a relevant sex-specific role in the species. Applying discriminant functions derived from one population to other populations can be problematic. However, we found that the bill depth discriminant function for British shags correctly sexed 90.2% of Iberian shags, supporting the idea that, although British shags are larger than northwestern Iberian shags, differences between sexes are of the same magnitude.

Key words: *Phalacrocorax aristotelis*, sexing, *Prestige*, geographic variation, morphometrics, discriminant analysis

INTRODUCTION

Discriminant function analysis using external measurements has been extensively used to sex seabird species that have similar plumage but that differ in size (e.g. Lo Valvo 2001, Bertellotti *et al.* 2002, Genovart *et al.* 2003). However, a word of caution on the limitation of such discriminant functions in relation to geographic size variation has been raised by a number of authors (Calvo & Bolton 1997, Philips & Furness 1997, Mawhinney & Diamond 1999). Biologic and physical conditions of the foraging areas in the different regions (Granadeiro 1993) and the interaction between natural and sexual selection (Ricklefs & Miller 1999) can lead to differences in morphology and hence render discriminant functions developed in one location inappropriate for other locations. Here we compare morphometrics of European Shags *Phalacrocorax aristotelis* in two extreme cases—a central population (Shetland Islands—Calvo & Bolton 1997) and a population at the species' southernmost limit (northwestern Iberia)—to test whether discriminant functions developed for the central population can be used to sex birds from the peripheral population. We would expect the more northerly shags to be larger (Bergmann's rule) and that discriminant functions developed for the central population would not work well for peripheral populations, where the tendency to

differentiation is higher as a result of reduced gene flow. Although shags can be sexed on voice and behaviour during the breeding season (Baker 1993), it is useful to know the effectiveness of discriminant functions, because shags are often victims of gill netting, oil spills and natural catastrophes (Guyot 1990; Harris & Wanless 1996; Calvo & Bolton 1997; Harris *et al.* 1998). In such cases, sexing dead shags morphometrically is less time-consuming than is sexing by visual inspection of the gonads.

METHODS

Large numbers of shags are difficult to obtain. However, we took advantage of the *Prestige* oil spill, which killed a large number of shags in northwestern Spain in November 2002. Although the seabirds most affected were wintering auks (Camphuysen *et al.* 2002; García *et al.* 2003), many resident shags were also killed, because the area oiled holds two large colonies that total c. 2% of the world population of the species (Velando *et al.* 1999).

Shags do not disperse far from their colonies (Velando *et al.* 2005), and we were thus confident that the carcasses from the oil spill were those of local birds. A total of 340 oiled shags were found during the months immediately after the *Prestige* oil spill

(Velando *et al.* 2005). Of those, 137 birds were frozen in facilities of the regional environmental agency as a sample. These birds were collected along the southern coast of Galicia (northwestern coast of the Iberian Peninsula) by volunteers and staff of the regional government (Xunta de Galicia) in the months following the wreck of the vessel.

In June and July 2004, shags were thawed (<24 hours) for visual inspection of gonads. Heavily-oiled shags were not considered. From corpses for which gonad inspection was impossible ($n = 23$), a small sample of muscle tissue was taken to be molecularly sexed (see Genovart *et al.* 2003 for further details). Before inspection of gonads, the following external measurements were taken: culmen length (CL), minimum bill depth (BD), tarsus length (TL), wing length (maximum flattened chord, WL), ulna length (UL), tail length (TAL) and total head plus bill length (HL). All measurements were taken by the same researcher using a digital calliper to the nearest 0.01 mm, except for wing length, which was measured with a stopped wing-rule to the nearest 1 mm. A total of 114 shags (38 males and 76 females) were successfully sexed, aged and measured. Although measurements were taken from previously frozen carcasses, we compared them with measurements from British shags subjected to the same preservation procedure. We thus assumed that we did not introduce any bias with this procedure. The female:male bias in the sample has been explained by a larger number of females being at sea at the time of the oil spill, because males were already in the colonies defending territories (Martínez-Abraín *et al.* 2006). There was, furthermore, no evidence that the sample of male shags was from a subset of smaller-size shags (based on measurements made at colonies—AV unpub. data).

From the overall sample of 114 individuals, we created two subsets with roughly 50% of the shags in each. The first subset ($n = 54$) was used to develop the discriminant functions, and the second subset ($n = 60$) was used to validate the resulting functions. All carcasses were classed in two age classes: adults (birds with an all-black plumage) and immatures (birds with some brownish or whitish plumage on wings or belly). However, both age classes were pooled, because they are considered to be morphologically similar except in tail length (Calvo & Bolton 1997). This variable was not considered in the analysis despite the fact that we did not find any statistically significant differences between the age classes after checking the 95% confidence interval of the difference between means (−0.56 to 0.30).

A cut-off point for separating males and females based on discriminating scores was obtained by fitting a logistic curve, through nonlinear regression, to the values of the discriminant scores and the posterior probability that an individual score was likely to belong to one or the other sex group. The fit of the logistic curve provides the two constants (β_0 and β_1) of the logistic curve equation, which are specific for each analysis, and the cut-off point (x) is obtained from the equation for a probability value of 0.5.

The magnitude of the difference between morphometric characters of central and peripheral populations, as well as within populations, was evaluated using the Cohen's d , because the only information available to us from British shags was the mean and standard deviation:

$$d = \frac{\mu_1 - \mu_2}{\sqrt{\sigma_1^2 + \sigma_2^2}/2}$$

where a value of 0.2 (i.e. 20% effect) is typically considered small; 0.50, medium; and 0.80 large (Cohen 1988).

RESULTS

Morphometrics

British shags were larger than Iberian shags in all variables considered except for head and bill length (CL: $d=0.18$; HL: $d=0.26$; WL: $d=1.12$; BD: $d=0.64$). The largest difference attained was in bill depth. Similar results were obtained for the British population (Table 1).

Discriminant functions

The variable showing a larger correlation coefficient with the discriminant function was BD (0.72), followed by TL (0.64) and UL (0.61). The WL (0.38), HL (0.30) and especially CL (−0.003) had almost no effect on the discriminant function and were removed from later analyses. The initial discriminant function,

$$D = 0.931BD + 0.367TL + 0.070UL - 0.007WL - 0.007HL - 0.049CL - 37.35 [1]$$

correctly explained 94.4% of the original cases (Wilk's lambda = 0.319, $\chi^2 = 56.039$, $P < 0.001$).

TABLE 1
Comparison (mean \pm standard deviation) between culmen length (CL), bill depth (BD), wing length (WL) and head length (HL) of male and female European Shags *Phalacrocorax aristotelis* from Great Britain (Calvo and Bolton 1997) and the northwestern Iberian Peninsula (present work)

	Galicia (northwestern Iberian Peninsula)				Shetland Islands (United Kingdom)			
	Males	Females	Total	d Score ^a	Males	Females	Total	d Score ^a
CL	62.1 \pm 2.90	62.22 \pm 3.71	62.2 \pm 3.45	0.03	62.9 \pm 2.35	62.70 \pm 2.69	62.75 \pm 2.59	0.05
HL	132.06 \pm 5.72	134.75 \pm 4.13	133.92 \pm 4.79	0.53	135.19 \pm 3.94	131.39 \pm 3.70	132.75 \pm 4.19	0.99
WL	266.9 \pm 6.54	258.05 \pm 5.78	261.0 \pm 7.35	1.43	275.64 \pm 4.86	265.91 \pm 5.04	268.91 \pm 6.71	1.96
BD	10.4 \pm 0.74	8.93 \pm 0.55	9.4 \pm 0.93	2.25	11.03 \pm 0.62	9.51 \pm 0.53	9.98 \pm 0.89	2.63

^aThe Cohen d score is shown for comparing the magnitude of the difference between sexes within both populations (see text). Sample size, Shetland Island: $n = 335$ females for all variables, except for HL, where $n = 103$; $n = 149$ males, except for HL, where $n = 57$. Sample size, Galicia: $n = 82$ females, except for HL, where $n = 35$; $n = 41$ males, except for HL, where $n = 24$.

A second discriminant function including only the variables with a large correlation coefficient (i.e. after removing CL, WL and HL),

$$D = 0.938BD + 0.363TL + 0.058UL - 39.64, [2]$$

explained correctly 92.6% of the original cases (Wilk's lambda = 0.328, $\chi^2 = 56.347$, $P < 0.001$). The correlation coefficients of each variable and the discriminant function were 0.736 for BD, 0.654 for TL and 0.624 for UL in this case.

Given the stronger correlation between BD and the discriminant function, we finally obtained a single-variable function for bill depth,

$$D = 1.441BD - 14.106, [3]$$

which again correctly explained 92.6% of the original cases (Wilk's lambda = 0.473, $\chi^2 = 38.515$, $P < 0.001$). Single-variable functions for TL (Wilk's lambda = 0.533, $\chi^2 = 32.427$, $P < 0.001$) and UL (Wilk's lambda = 0.556, $\chi^2 = 30.239$, $P < 0.001$) only explained 85.2% of the original cases. Function 3 thus had a discriminant power equal to function 2 and only slightly less than function 1, but it was more parsimonious and simpler to use. To validate the single-variable function for bill depth (function 3), we used a different set of shags not included in the original analyses ($n = 60$), and we obtained a correct classification for 93.3% of the cases (93.3% of the males and 93.3% of the females; Wilk's lambda = 0.353, $\chi^2 = 59.803$, $P < 0.001$).

From the fitting of our logistic curve, the cut-off point for discriminating between males and females was 0.34. Discriminant scores larger than the cut-off point corresponded to males and scores smaller than the cut-off point corresponded to females (Philips & Furness 1997).

The single-variable discriminant function for bill depth developed for British shags led to a correct classification of 90.2% of Iberian shags.

DISCUSSION

The fact that Iberian shags had larger heads than British shags is surprising. This difference reflected the fact that, in our sample, female Iberian shags had longer head lengths than the males did. This finding is probably an artefact attributable to the small sample size.

Male Iberian shags were larger than the females in all variables measured except CL, as was also found among shags affected by the *Braer* oil spill in Shetland (Calvo & Bolton 1997). This conservatism in the length of the culmen between the sexes within and between populations could reflect stabilizing selection acting upon a trait directly linked to the foraging ecology of the species. Culmen growth was similar in male and female chicks (Velando *et al.* 2000). Modal prey size in the environment around colonies may act as a selective pressure leading to similar CLs in males and females and among birds of different populations within the same biogeographic region. In this sense, Velando & Freire (1999) found that the modal prey size of Iberian shags was similar in all prey species regardless of prey size availability.

The fact that the single-variable discriminant function for BD developed for British shags led to a correct classification of *c.* 90% of Iberian shags suggests that the magnitude of the difference between sexes was similar between populations. Hence, British shags were

larger than Iberian shags irrespective of sex. This could be because more food was available at higher latitudes, or it could reflect a response to lower ambient temperatures (Bergmann's rule).

The fact that BD alone offered good predictive power for sexing shags corroborates the high predictive power of that morphometric variable in other seabird species such as Balearic Shearwaters *Puffinus mauretanicus* (Genovart *et al.* 2003), Cory's Shearwaters *Calonectris diomedea* (Granadeiro 1993, Lo Valvo 2001) or Kelp Gulls *Larus dominicanus* (Torlaschi *et al.* 2000). Even when CL is similar between sexes, such as in the case of shags, bill depth is significantly different. This dimorphism in bill depth could reflect either the effects of natural selection acting on niche segregation at foraging or sexual selection, with females choosing more robust males.

Discriminant functions developed for a central population of shags were used safely to sex shags from a peripheral population. However, other seabird species, or even other peripheral populations of shags, could behave differently, and it is advisable that the validity of functions be checked in every case.

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