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SEXUAL DIMORPHISM IN THE SOUTHERN GREY SHRIKE *LANIUS MERIDIONALIS* IN THE CENTRAL WEST OF THE IBERIAN PENINSULA

DIMORFISMO SEXUAL EN EL ALCAUDÓN REAL *LANIUS MERIDIONALIS* EN EL CENTRO-OESTE DE LA PENÍNSULA IBÉRICA

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Sex identification of the different individuals from any species is essential in ecological studies (Lessels & Mateman, 1998). Many bird species have monomorphic plumages, *i.e*. those in which even after careful scrutiny sex identification is complicated, due to the similar appearance of males and females (Green & Theobald, 1989). Morphological measurements are important tools for sex determination in species lacking clear sexual dimorphism (Sweeney & Tatner, 1996; Walton & Walton, 1999). Nevertheless, the overlapping of morphological measurements usually renders satisfactory sex identification in the field impossible (Kavanagh, 1988; Green & Theobald, 1989). The problem of morphological methods is that they are related to mistakes made by different observers, which could affect their final decisions (Barret *et al*., 1989; Jenni & Winkler, 1989). To solve this problem, certain measurements in particular are usually trusted, since they are easy to obtain and highly reproducible, reducing putative errors. The length of the 8th primary feather, together with the wing length, is considered the best measure of a bird's size (Jenni & Winkler, 1989). The length of the 8th primary is highly correlated with wing length and the variation between different observers is reduced by $^2/_3$ against the wing length (Jenni & Winkler, 1989; Pérez-Tris *et al*., 2000). Other studies have suggested that tarsus and keel length are good predictors of body size (Senar & Pascual, 1997), although this measurement could entail some risk for the birds.

Apart from morphological methods, over the past two decades molecular techniques have been shown to be powerful tools in different ornithological studies (Ellegren, 1992; Baker *et al*., 1999). Molecular methods based on DNA analyses allow researchers to identify the sex of birds from blood samples, skins, or feathers in an accurate way.

The Southern Grey Shrike *Lanius meridionalis* is a species recently split from the Great Grey Shrike *Lanius excubitor* (Isenmann & Bouchet, 1993; Isenmann & Lefranc, 1994; Lefranc & Worfolk, 1997; Harris & Franklin, 2000). Unlike its former conspecific the Great Grey Shrike, it shows no sexual dimorphism (Svensson, 1998). However, it is possible to determine its sex using morphological characteristics during the breeding period, such as the existence of the incubation patch in females or the cloacal protuberance in males, or based on their behaviour during the breeding season (Lefranc & Worfolk, 1997; Harris & Franklin, 2000). Here, an attempt was made to find some measurements that would allow separation the sexes of this species in the field using morphological measurements.

Southern Grey Shrikes were trapped in the province of Salamanca (Central-west of the Iberian Peninsula, 5°39' W, 40°57' N) during the years 2000 to 2003. Since this species is considered sedentary (Hernández, 1999), the shrikes were caught throughout the year regardless of season. The birds were trapped using mist nets, bow nets, clap nets and a modified Potter trap (Craig, 1997; Infante & Peris, 2002). For some traps it was necessary to use Domestic Mice *Mus musculus* as baits. The Potter trap was the most frequent trapping method used since it showed the highest efficiency.

Birds were ringed with an official ring from the Spanish Ministry of the Environment and with a combination of coloured metal rings for

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later identification of the individuals. Thus, retrapping was not necessary and sex could be confirmed in most cases by the birds' behaviour.

Six morphological measurements were taken into account: wing length, tail length, bill to skull length, tarsus length, bill height and the length of all the primaries. Primaries were numbered in descending order. Maximum wing length (see Svensson, 1998) was measured using a ruler to the nearest 1 mm, and the lengths of the tail and primary were measured to the nearest 0.5 mm using a precision ruler with the scale starting at the end point of the ruler to be fitted at the base of the feathers (Jenni & Winkler, 1989). Bill to skull length, bill height and tarsus length were measured using a digital precision calliper to the nearest 0.1 mm. Weight was measured using a digital scale to the nearest 0.1 g. To insure the uniformity of the measurements from different individuals, all measurements were taken by O.I., following Svensson (1998) (Barret *et al*., 1989; Jenni & Winkler, 1989). As well as the morphological measurements and weight, blood samples were collected from the brachial vein. Blood was kept at room temperature in Eppendorf tubes containing ethanol (70%).

When possible, sex was determined by the presence of an incubating patch (only females incubate) (Hernández, 1993; Lefranc & Worfolk, 1997) or the cloacal protuberance (males). The behaviour of the colour-ringed birds during their breeding season was also observed in order to infer sex. In this way it was possible to determine the sex of 20 individuals (15 males and 5 females). When it was not possible determine their sex by these means, molecular methods were used. Samples from 23 shrikes (12 males and 11 females) were analysed by the latter method. The sex of some birds was determined using both methods to test the accuracy of behavioural data.

Peripheral blood samples were drawn by wing vein puncture and stored in 70% ethanol. DNA was extracted using the Chelex 100 method (Walsh *et al*., 1991) and was used in a 15 µl PCR reaction containing 0.2 µM of primers 2550F (5'-GTTACTGATTCGTCTACGAGA-3') and 2718R (5'-ATTGAAATGATC-CAGTGCTTG-3') (Fridolfsson & Ellegren, 1999), 100 µM of each dNTP, 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 1.5 mM $MgCl_2$ and 0.5 U Taq polymerase (Biotools). Shrike sex was determined based on the size difference between the CHD1Z and CHD1W introns (Fridolfsson & Ellegren, 1999).

Based on the known sex (inferred from behaviour and the molecular method) the morphological measurements of males and females were compared using t-tests that compared the average of both groups (Sokal & Rohlf, 1995). Before this, the F-test was used to determine whether the samples were taken from a population with similar variance (Fowler & Cohen, 1999). Statistical calculations were carried out using Statistica and SPSS programs.

The morphological measurements recorded here did not differ from those reported by other researchers (Cramp & Perrins, 1993; Panov, 1995; Lefranc & Worfolk, 1997; Harris & Franklin, 2000), except for tail length, which in this population seemed to be longer and bill-to-skull length, which tended to be shorter (Table 1). However, in other studies (Cramp & Perrins, 1993; Lefranc & Worfolk, 1997; Harris & Franklin, 2000), these measurements were obtained from museum skins, while in this study they were taken from living birds. In the skins of Great Grey Shrikes Kuczyński *et al*. (2003) observed a mean shrinkage of *ca*. 5 %, depending on the measurement. This could be the reason why living birds tended to have longer tails.

No statistically significant differences were found between the biometric measurements of males and females of the Southern Grey Shrike (Table 2). Therefore, morphological measurements did not allow differentiation between the sexes. However, on average most primary feathers (excluding the first, ninth and tenth primaries) tended to be longer in males than in females. Nevertheless, a strong overlap in the measurements was detected, which prevents differentiation between sexes (Table 3).

In a review of the determination of sexual size dimorphism in birds, Greenwood (2003) suggested an index called m/f (i.e. the ratio between the mean, m, of the male trait and the mean, f, of the female trait). This index was used for the length of the 8th primary. The sexual dimorphism m/f ratio for 8th primary length was 1.01, a value close to unity, showing that there is no sexual dimorphism in this species regarding the length of the 8th primary. The same was the case for wing length (1.01).

TABLE 1

Biometric measurements in mm. shown by other researchers and our results. In the study population, the tail seems to be longer and bill-to-skull length seems to be shorter. In brackets the average and *n* = sample size.
[Medidas biométricas en mm. de otros estudios y propias. En la población estudiada, la cola es más larga y la longitud entre el pico y el cráneo más corta. Entre paréntesis la media y n = *tamaño muestra.]*

	Cramp & Perrins (1993)	Panov (1995)	Lefranc & Worfolk (1997)	Svensson (1998)	Harris & Franklin (2000)	This Study
Wing Length *[Longitud alar]*	102-112 (106.6) *n* = 16	102-116	102-112 (107)	103-113	102-112 (106.6) *n* = 16	102-113 (107.41) *n* = 35
Bill to skull *[Longitud pico-cráneo]*	23.2-25.3 (24.2) *n* = 15		23-25		23.2-25.3 (24.2) *n* = 15	16.06-21.59 (19.20) *n* = 35
Tail *[Cola]*	101-118 (109.5) *n* = 16		101-118	105-116 *n* = 23		104-129 (115.96) *n* = 35
Tarsus *[Tarso]*	29.4-31.3 (30.0) *n* = 17	29.5-31				28.13-36 (30.81) *n* = 35

TABLE 2

Biometric measurements of male and female Southern Grey Shrikes. *n* = sample size and n.s.: not significant. *[Medidas biométricas de machos y hembras de Alcaudón Real*. n = *tamaño muestral y n.s.: no significativo.]*

Variable	Males *[Machos]* (*n* = 21)	Females *[Hembras]* (*n* = 14)	*P*
Wing *[Ala]*	107.55 ± 2.2	107.22 ± 2.6	n.s.
Tail *[Cola]*	116.8 ± 3.5	114.58 ± 6.6	n.s.
Bill to Skull *[Longitud Pico a Cráneo]*	19.51 ± 1.6	18.95 ± 0.9	n.s.
Bill Height *[Altura del Pico]*	8.81 ± 0.3	8.82 ± 0.4	n.s.
Tarsus *[Tarso]*	31.05 ± 2.2	30.46 ± 1.4	n.s.
Wing/Tail *[Ala/Cola]*	0.92 ± 0.1	0.93 ± 0.1	n.s.
Wing/Tarsus *[Ala/Tarso]*	3.47 ± 0.3	3.52 ± 0.2	n.s.

These results are consistent with the rest of the data pertaining to the averages calculated for other parts of the body using the *t* test. The same results were obtained using PC. The three first axes explain 77% of the variance. Thus, it is concluded that there is no evidence of sexual dimorphism in this species.

According to some authors (Harvey & Bradbury, 1991; Owens & Hartley, 1998), species that are apparently monogamous and display parental care by both parents, such as the Southern Grey Shrike for example, are often highly dimorphic. However, this was not the case in the Southern Grey Shrike. The answer could lie in the social mating system of this species. Shrikes are strongly territorial, defending a territory of between 10 and 25 ha. in the case of the nominal subspecies (Lefranc & Worfolk, 1997), and males do not need to show a conspicuous plumage to attract females because they use impaled prey for such purposes (Yosef & Pinshow, 1989). The distance between nests in this species, on average 96 m (Hernández, 1993), is too large for a territorial passerine, so extra-pair copulations probably occur only rarely. Males of the Southern Grey Shrike help to raise their chicks, so they are prevented from searching for other mates. In contrast, other species of smaller body size and territory, such as the Red-backed Shrike *Lanius collurio*, have sexually dimorphic plumage (Owens & Hartley, 1998). A possible explanation for this phenomenon is that smaller species often breed in loose colonies, with smaller distances between

Table 3

Overlapping in the measurements that prevents a differentiation between the sexes of the Southern Grey Shrike using biometrical methods. *n* = sample size and n.s.: not significant.
[Solapamiento de las medidas entre machos y hembras que impide la diferenciación sexual en el Alcaudón Real usando métodos biométricos. n = *tamaño muestra y n.s.: no significativo.]*

Primary number *[N.º de Primaria]*	Males *[Machos]* (*n* = 19)	Females *[Hembras]* (*n* = 14)	*P*
1st	38.22 ± 2.4	38.64 ± 2.0	n.s.
2nd	67.86 ± 2.3	67.59 ± 1.9	n.s.
3rd	80.61 ± 2.2	79.71 ± 2.6	n.s.
4th	82.95 ± 2.4	82.00 ± 2.9	n.s.
5th	83.63 ± 2.0	82.87 ± 2.6	n.s.
6th	79.37 ± 2.1	78.87 ± 2.8	n.s.
7th	75.39 ± 2.1	74.82 ± 2.1	n.s.
8th	73.63 ± 2.1	72.92 ± 2.5	n.s.
9th	72.08 ± 3.1	72.22 ± 2.0	n.s.
10th	71.53 ± 1.8	71.63 ± 1.6	n.s.

the nests -19 m on average- and hence defend territories no larger than 2 ha, as is the case of Red-backed Shrikes. Since the distance between nests is smaller, the copulation rate between individuals from different pairs may be greater (Fornasari *et al*., 1994). In fact, extra-pair copulations have frequently been reported in Red-backed Shrikes (Lefranc & Worfolk, 1997).

In addition, birds with abundant food in their territories do not need to migrate, and this abundance may favour monogamous relationships (Gill, 2003). This, similar migrating species, such as the Great Grey Shrike, tend to find partners before they arrive in their breeding territories. In order to achieve this, males must attract females using their plumage (since shrikes do not sing) and their larger body size (as compared with females) and hence show sexual dimorphism, even though in this species it is quite subtle (Svensson, 1998). The Southern Grey Shrike is sedentary (Hernández, 1999) and attracts females by impaling prey throughout its territory (Yosef & Pinshow, 1989).

However, the main reason for the sex-related variation in body size in different bird species seems to be due to different breeding behaviours (social or territorial) and to the provisioning pattern in parental care. Currently, molecular methods allow researchers to detect a high percentage of extra-pair copulations in monogamous species (Gill, 2003). Owens & Hartley (1998) reported that sexual selection is tightly related to an increase in extra-pair copulations, and sexual dimorphism in size and in plumage would be linked to different features of the breeding behaviour. Sexual dimorphism in size is related to high levels of polygamy, while dimorphism in plumage is linked to extra-pair copulations. Regarding Southern Grey Shrikes, only the subspecies *aucheri* has been reported to include polygamous individuals (Yosef & Pinshow, 1988) but no extra-pair copulations have been observed for the nominal subspecies, supporting the absence of sexual dimorphism in this new species (Andersson, 1994; Owens & Hartley, 1998). Further studies using paternity analyses should be carried out.

RESUMEN.—*Se presenta la ausencia de dimorfismo sexual en el Alcaudón Real mediante el uso de biometría en aves vivas, conociendo el sexo previamente mediante características morfológicas, conducta o técnicas moleculares. La ausencia de este dimorfismo puede radicar en el comportamiento territorial de la especie, con un territorio en torno a 25 ha. Otras especies, como el Alcaudón Dorsirrojo Lanius collurio con un territorio que no supera las 2 ha, presentan dimorfismo sexual acusado y alto número de cópulas fuera de la pareja. Éstas no han sido observadas en el Alcaudón Real lo que puede haber provocado la ausencia de dimorfismo sexual.*

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