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MORPHOMETRIC VARIATION AND POPULATION RELATIONSHIPS OF KRÜPER'S NUTHATCH (*SITTA KRUEPERI*) IN TURKEY

TAMER ALBAYRAK,[1,4] AURELIEN BESNARD,[2] AND ALI ERDOĞAN[3]

ABSTRACT.—We studied the population relationships of Krüper's Nuthatch (*Sitta krueperi*) by capturing 82 individuals using mist nets in six different areas in Turkey during the breeding season, from March to June 2005–2008. Forty-one different morphometric characters were measured. Morphometric characters measured ($\bar{x} \pm$ SD) were: body mass = 13.11 ± 0.88 g, wing length = 74.79 ± 2.35 mm, bill length = 17.65 ± 0.76 mm, and tarsus length = 19.10 ± 0.93 mm, respectively. A stepwise discriminant analysis of four populations retained seven statistically significant measurements: body mass, wing length, length of P 8, alula, bill height, back nail, and left nail. These analyses allowed discrimination among populations. The population in the Aladağlar Mountains differed from others even though it overlapped with the population in the Lütfi Büyük Yıldırım Research Forest, and marginally with the population at Kartalkaya Mountain. Cross validation for the other three populations confirmed large overlap in morphometric characteristics although the population at the Lütfi Büyük Yıldırım Research Forest seemed to be intermediate between populations at Aladağlar Mountains and in the Kazdağları Mountain-Kartalkaya Mountain complex. *Received 12 February 2011. Accepted 12 June 2011.*

The Anatolian Peninsula of Turkey exhibits important biodiversity due to high climatic diversity, topographic variation, and habitat fragmentation. Anatolia is considered to be one of the most important refugia in southern Eurasia during glacial periods (Ciplak 2004). The distribution of sensitive species changed during both the Pliocene and Pleistocene, and contracted during the following interglacial period due to changes in climate (Ciplak 2004). Successive retreats and expansions of glacial ice may have resulted in complex genetic structuring of species isolated in multiple refugia and by postglacial colonization (Howes et al. 2006). Forest vegetation may have survived in several Quaternary glacial refugia in the northern and southern parts of the Anatolian Peninsula (Hughes et al. 2006). Anatolia exhibits substantial altitudinal variation ranging from 0 to 5,137 m asl (1,100 m asl average elevation), which resulted in extensive habitat fragmentation. This topographic relief had an important role in Pleistocene changes by constraining distribution corridors for latitudinal dispersal and providing suitable habitats during altitudinal shifts (Ciplak 2008). Anatolian forests are mainly composed of fragmented areas of *Pinus* spp., *Abies* spp., *Cedrus* spp., and *Picea* spp. (Akman 1995, Albayrak et al. 2010).

Krüper's Nuthatch (*Sitta krueperi*) is an endemic species strictly confined to coniferous forests (Löhrl 1988, Cramp and Perrins 1993, Harrap and Quinn 1996, Hagemeijer and Blair 1997, Matthysen 1998, Albayrak and Erdogan 2005a) whose range is mainly restricted to Anatolia, Lesvos Island, and the Caucasian region (BirdLife International 2004; Albayrak and Erdogan 2005a, b; Albayrak et al. 2006). Krüper's Nuthatch populations, like those of many forest bird species, have been declining in Turkey and Lesvos Island (BirdLife International 2004). The species is categorized as a species of European conservation concern, SPEC 2 by BirdLife International (2004), and near threatened by the IUCN (2010). It is mostly sedentary with limited post-breeding dispersal and seasonal altitudinal movements (Cramp and Perrins 1993, Harrap and Quinn 1996, Handrinos and Akriotis 1997).

We hypothesized this species would be expected to have isolated populations. Our objectives were to: (1) explore potential population diversity and characteristics at the morphometric level in Anatolia, and (2) characterize the little known morphometry of the species.

METHODS

Study Site.—We selected six different areas (Fig. 1) locally occupied by Krüper's Nuthatch in Turkey: Aladağlar Mountains (ALA, 930 m asl; red pine [*Pinus brutia*] forest), Alanya (ALY, 1,300 m asl; red pine forest), Lütfi Büyük Yıldırım Research Forest (BUK, 650 m asl; red

[1] Department of Biology, Faculty of Science and Art, Mehmet Akif Ersoy University, Burdur, Turkey.

[2] EPHE, Ecologie et Biogéographie des Vertébrés, Centre d'Ecologie, Fonctionnelle et Evolutive, Montpellier, France.

[3] Faculty of Science and Art, Department of Biology, Akdeniz University, Antalya, Turkey.

[4] Corresponding author; e-mail: albayraktamer@gmail.com

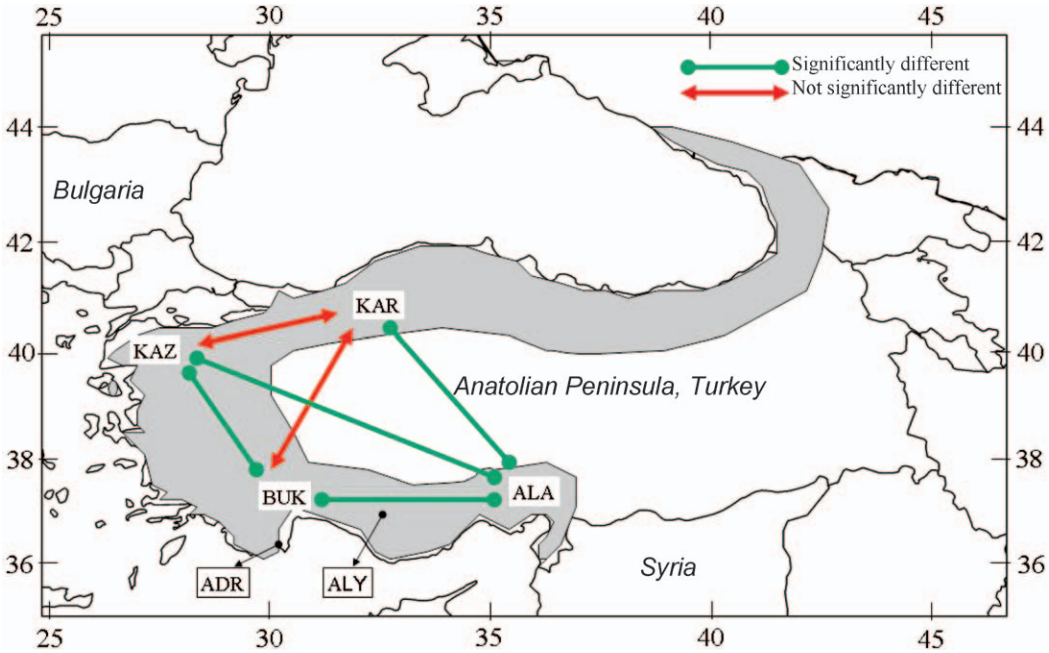


FIG. 1. Study sites in the Anatolian Peninsula, Turkey and significant differentiation of Krüper's Nuthatch populations. World distribution is in gray.

pine forest), Adrasan (ADR, 200 m asl; red pine forest), Kazdağları Mountain (KAZ, 800 m asl; black pine [*P. nigra*] forest), and Kartalkaya Mountain (KAR, 1,300 m asl; black pine forest) to collect morphometric data. The areas were generally mountainous with natural vegetation, rising from sea level to 2,500 m asl. All capture sites were in mature coniferous forests. The shortest distance was between ADR and BUK (81 km) and the longest was between ALA and KAZ (783 km; Fig. 1).

Bird Census.—Surveys for nuthatches were conducted during the breeding season between March and June 2005–2008 to understand possible gender differences (breeding females can be identified from brood patches). In total, 78 m (2×12, 2×10, 2×8, and 3×6 m) of mist nets were set and playback of calls was used to attract nuthatches; 82 individuals were captured. Only males were expected to be attracted by playback of calls but five individuals captured had brood patches and were probably females. It is not possible to separate most adult males from adult females based on plumage characteristics although there is limited information in the literature (Cramp and Perrins 1993). These five individuals were excluded from the population comparison analysis since they may differ from males and were too few to be analyzed separately.

Forty-one different morphometric characters based on Svensson (1992) were measured by the same observer (TA) in all populations: body mass, wing length, tail length, bill length (BL), bill width at nostril (BW), bill height at nostril (BH), length of bill apex to head back (LBaHb), length of nostril to bill apex (LNBa), length of black on forehead (LbF), alula, tarsus, four nails of left foot, all 10 primaries and nine secondaries of the left wing, and all six rectrices from the left side. Digital calipers and a digital balance were used for taking measurements. All individuals were released after measurement at their capture location.

Data Analyses.—We performed a stepwise discriminant analysis to explore the level of morphologic differentiation among Krüper's Nuthatch populations. We retained only 15 characters subjectively supposed to be the most discriminating among populations, and for which sufficient individuals were measured. Populations ALY and ADR had few individuals with complete mea-

TABLE 1. Descriptive statistics of morphometrics of Krüper's Nuthatch, Anatolian Peninsula, Turkey.

Character	*n*	Mean ± SD	Min	Max
Body mass, g	72	13.11 ± 0.88	11.17	14.80
Wing , mm	77	74.79 ± 2.35	65.32	80.00
Tail, mm	75	37.45 ± 2.06	31.04	42.00
BL, mm	77	17.65 ± 0.76	15.47	19.23
BW, mm	76	4.55 ± 0.34	3.78	5.51
BH, mm	76	3.87 ± 0.24	3.32	4.68
LBaHb, mm	66	36.18 ± 0.90	34.09	38.36
LNBa, mm	77	12.64 ± 0.68	11.14	14.87
Alula, mm	66	18.55 ± 1.22	13.62	22.00
Tarsus, mm	76	19.10 ± 0.93	16.59	21.50
Back nail, mm	70	6.50 ± 0.34	5.56	7.34
Left nail, mm	62	4.04 ± 0.36	3.46	4.93
Mid. nail, mm	63	4.63 ± 0.39	3.95	6.36
Inner nail, mm	63	3.78 ± 0.36	3.10	4.71
LbF, mm	58	14.25 ± 1.98	9.86	19.66

surement data, and were excluded from the analysis. The discriminant analysis was performed on four populations totaling 60 individuals.

Our data set indiscriminately incorporated adult males and females, and possibly some juveniles; we suspected that size could be a confusing factor in the discriminant analysis. We thus focused on shape analysis (morphology instead of morphometry) to remove the potential effect of size and avoid misinterpretation of the results. Size differences were removed using a bicentering transformation on the log of the measurements. Two individuals in this transformation were linked by a multiplying constant and became identical (Gower 1976). This approach is classically used to study morphology instead of morphometry (Yoccoz 1993). Bicentering was performed using the library Ade4 (function bicenter.wt) (Chessel et al. 2004) and R 2.10.1 (R Development Core Team 2009).

The data set was subjected to stepwise discriminant analysis to ascertain which morphologic traits best discriminated among the four populations. All traits that remained in the model when the stepwise selection process stopped were significantly discriminating among populations. The stepwise discriminant analysis was performed using SAS/STAT® software (PROC STEPDISC) (SAS 1985). PROC DISCRIM (SAS 1985) was used to obtain the discriminant function. The function was validated by a leave-one-out cross validation procedure. The leave-one-out cross-validation procedure in discriminant analysis consists of recalculating the disciriminant function several times, based on all birds except the one which is classified using the estimated function. Each classified bird in this procedure was allocated to the group with the nearest centroid, and the proportion of correctly allocated individuals was calculated (Ripley 1996). A graphical representation of the classification was also obtained using library Ade4 in R 2.10.1 (R Development Core Team 2009).

RESULTS

Morphometric Characterization.—We found a small statistically significant differentiation between individuals without a brood patch and with a brood patch for bill length, bill width, and P 7 (*t*-test; $P < 0.05$). We recorded no measurements for brood patch individuals due to the low sample size ($n = 5$, probably females). Mean (± SD), range, and sample size for the 15 morphometric measurements used in the discriminant analysis (Table 1), and measurements of primaries, secondaries, and tail feathers varied (Table 2). The longest primary was P 6 and longest secondary was S 2 (Table 2).

Population Relationships.—The stepwise discriminant analysis retained only seven measurements allowing discrimination among populations based on body mass, wing length, length of P 8, alula, BH, back nail, and left nail (Table 3). The plot of the first two canonical variables indicates small differences among populations as the overlaps are great (Fig. 2). The discriminant analysis correctly classified 50 of the 60 birds, i.e., 83% (Table 4). However, the cross validation

TABLE 2. Measurements (mm) of primary, secondary, and tail feathers of Krüper's Nuthatch, Anatolian Peninsula, Turkey.

Primary length					Secondary length					Tail length				
	n	Mean ± SD	Min	Max		*n*	Mean ± SD	Min	Max		*n*	Mean ± SD	Min	Max
10th	61	18.77 ± 1.52	15.0	22.0	1st	64	53.65 ± 2.23	46.0	58.0	6th	18	37.83 ± 3.88	25.0	44.0
9th	65	50.86 ± 1.78	47.6	57.0	2nd	64	53.70 ± 1.68	50.0	57.0	5th	18	38.72 ± 2.30	33.0	44.0
8th	65	57.82 ± 1.92	54.0	65.0	3rd	64	52.95 ± 1.96	48.0	58.0	4th	18	38.53 ± 1.56	35.0	42.0
7th	64	58.89 ± 1.93	52.5	64.0	4th	63	51.19 ± 2.80	40.0	55.0	3rd	18	37.92 ± 2.09	35.0	43.0
6th	65	60.05 ± 1.91	55.0	64.0	5th	63	48.79 ± 2.19	42.0	55.0	2nd	17	37.35 ± 2.12	35.0	42.0
5th	65	59.75 ± 2.19	50.0	65.0	6th	60	45.45 ± 2.95	32.0	53.0	1st	18	36.36 ± 1.71	34.0	40.0
4th	65	56.22 ± 2.13	49.0	59.0	7th	60	40.90 ± 2.41	31.0	45.0					
3rd	65	53.92 ± 2.25	45.0	58.0	8th	62	31.76 ± 2.58	22.0	37.0					
2nd	61	53.11 ± 2.45	44.0	58.0	9th	57	23.46 ± 1.91	19.0	30.0					
1st	62	52.92 ± 2.03	47.0	57.0										

procedure correctly classified only 30 birds, i.e., 50% (Table 4), which is low. The ALA population is quite different from the others even though it overlaps with population BUK and marginally with population KAR. Cross validation confirmed these results by correctly assigning 75% of the birds. Cross validation of the three other populations confirmed the large overlap in morphological characteristics, although population BUK was intermediate between population ALA and the KAZ-KAR complex.

DISCUSSION

The morphometric charateristics of Krüper's Nuthatch are poorly known. Only measurements of wing length, tail length, bill length, tarsus, and body mass, which are similar to our results, have been recorded from small numbers of individuals (Cramp and Perrins 1993, Roselaar 1995, Harrap and Quinn 1996). Differentiation among Krüper's Nuthatches is not known although a few measurements for both males and females were given by Cramp and Perrins (1993) and Harrap and Quinn (1996). We compared measurements of morphometric characteristics among populations using males that were captured using playback of calls during the breeding season and which did not exhibit a brood patch. There was slight dimorphism in the species, and verification of the measurements of males and females now depends on molecular identification of individuals.

TABLE 3. Stepwise discriminant analysis of morphometric characteristics of Krüper's Nuthatch, Anatolian Peninsula, Turkey.

Step	Variable	Wilks' λ	*P*
1	Body mass	0.72	0.0003
2	Back nail	0.60	<0.0001
3	P 8	0.52	<0.0001
4	Wing	0.38	<0.0001
5	Alula	0.33	<0.0001
6	BH	0.29	<0.0001
7	Left nail	0.26	<0.0001

Geographical distance and habitat fragmentation are considerable among populations in the Anatolian Peninsula. Our results indicate there may be exchange among populations, possibly because of the existence of suitable corridors, especially between BUK and the KAR-KAZ complex. Krüper's Nuthatch is probably capable of long distance movement, and it is unlikely that only isolation by distance can produce differentiation among populations. We detected some differentiation among populations in the data set. The ALA population is quite different from the three other populations even if it overlaps with population BUK and marginally with the population in the KAR-KAZ complex.

This could be the result of the existence of both southern and northern refugia during glacial periods which were partially reconnected after isolation by distance. ALA and BUK are both in the southern refugia and overlap in morphologic characters was observed between them. The KAR-KAZ complex is localized in the northern part of Anatolia and populations are probably sufficiently close to exchange some individuals. ALA is distant from these two northern populations. These two complexes are also separated by habitat fragmentation and high mountains, which are unoccupied by the species (Fig. 1).

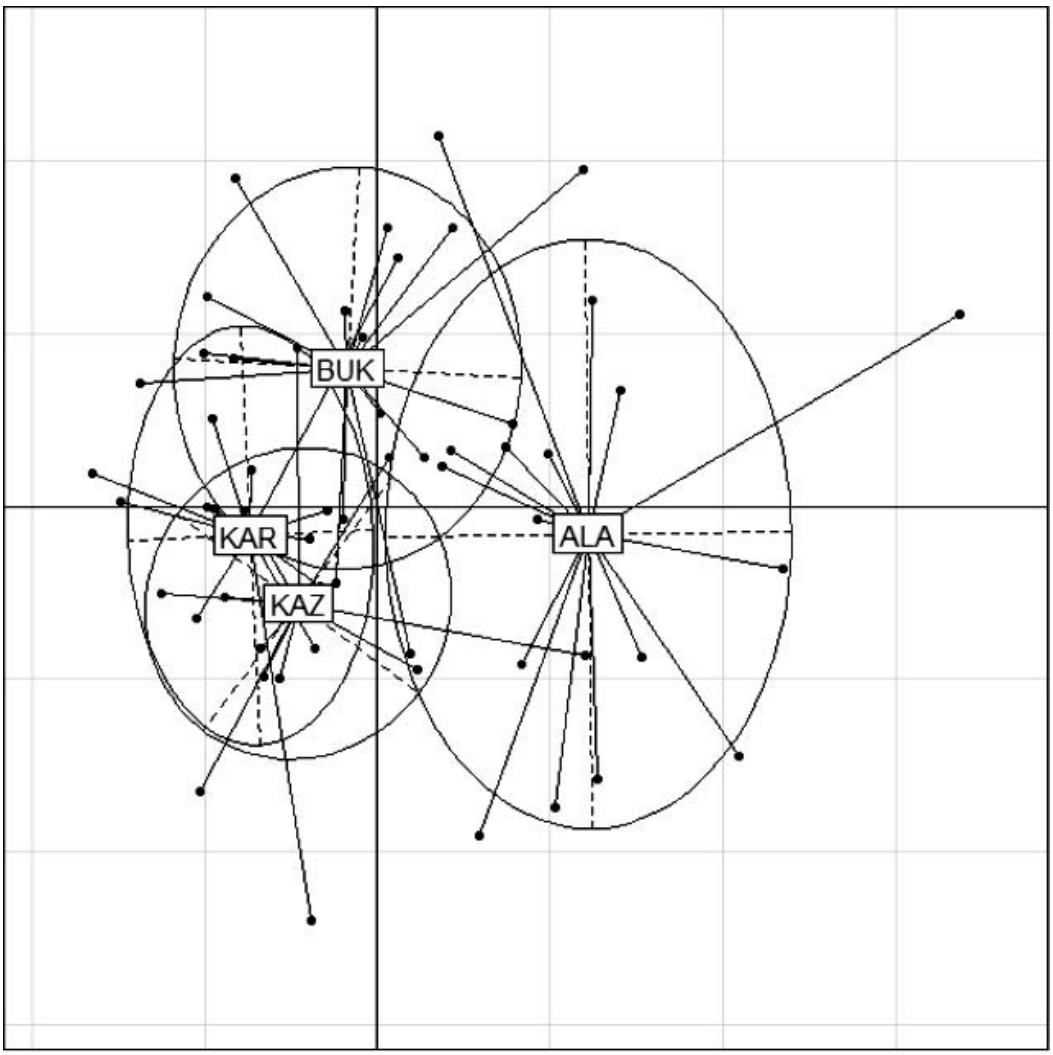


FIG. 2. Scatter plot of canonical scores from the discriminant analysis of Krüper's Nuthatch populations, Anatolian Peninsula, Turkey.

The BUK population is classified as intermediate between the KAR-KAZ complex and ALA. This agrees with both the refugia hypothesis of later partial reconnection and the hypothesis of isolation by distance only. The Great Reed Warbler (*Acrocephalus arundinaceus*) population in the Eurasian breeding range of the species has two clades that have colonized from two separated refugia in Europe and the Middle East (Hansson et al. 2008). The forest types of KAR and KAZ were black pine while the other populations occupied red pine forest. These forest types may also have affected the morphometrics independently of isolation due to long distances or refugia disconnection between locations ALA and the KAR-KAZ complex (min = 440 km; Fig. 1).

Anatolian refugia are known to have affected the evolution of structured populations in several groups including grasshoppers (Ciplak 2008), small mammals (Castiglia et al. 2009), and a passerine bird (Hansson et al. 2008). For example, the *Antrastes* genus of Orthoptera arose from an ancestral stock during the Pliocene era in northwestern Anatolia whose range later expanded and contracted under the effects of the ice ages (glacial and interglacial periods, respectively)

TABLE 4. Classification results for the discriminant analysis (original) of Krüper's Nuthatch, Anatolian Peninsula, Turkey; 85 and 50% of selected original grouped cases are correctly classified.

		Predicted group membership				
Population		ALA	BUK	KAZ	KAR	Totals
ALA	85%	14 (87.50)	1 (6.25)	1 (6.25)	0 (0.00)	16 (100)
	50%	12 (75.00)	2 (12.50)	1 (6.25)	1 (6.25)	16 (100)
BUK	85%	1 (6.25)	12 (75.00)	2 (12.50)	1 (6.25)	16 (100)
	50%	3 (18.75)	8 (50.00)	2 (12.50)	3 (18.75)	16 (100)
KAZ	85%	0 (0.00)	1 (7.14)	12 (85.71)	1 (7.14)	14 (100)
	50%	2 (14.29)	4 (28.57)	5 (35.71)	3 (21.43)	14 (100)
KAR	85%	0 (0.00)	2 (14.29)	0 (0.00)	12 (85.71)	14 (100)
	50%	0 (0.00)	7 (50.00)	2 (14.29)	5 (35.71)	14 (100)

(Ciplak 2004). The explanation for the present findings regarding differentiation of nuthatch populations may be isolation of subpopulations during the glacial period and ongoing habitat fragmentation. There may have been a minimum of two different refugia, southern and northern, for Krüper's Nuthatch during the glacial periods due to the topographic structure of Anatolia. The extent of isolation may increase with ongoing fragmentation of breeding habitats due to logging, deforestation, etc.

Our study did not fully answer the question why Krüper's Nuthatch exhibit morphologic differentiation in Turkey. Our results agree with the already observed pattern of isolation by distance, and also the existence of northern and southern refugia during recent glacial periods. Molecular markers can provide more information about the biogeographic history of this species and can also assess the relationship among isolated subpopulations. Our results should be useful in conservation applications and future research. The population diversity of Krüper's Nuthatch at present is small, but still detectable, and could be used to identify and design protected areas for subpopulations and help with conservation efforts.

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