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COVARIATION OF MORPHOLOGICAL AND ALLOZYME FREQUENCY CHARACTERS IN POPULATIONS OF THE RUFOUS-COLLARED SPARROW (*ZONOTRICHIA CAPENSIS*)

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ABSTRACT.—We collected 474 specimens of male Rufous-collared Sparrows (*Zonotrichia capensis*) from 24 sites in northwestern Argentina. Four samples were obtained from each of six habitats: lowland chaco thornscrub, transition forest, montane woodland, montane grassland, Monte desert scrub, and puna high-altitude scrub. Puna birds (*Z. c. pulacayensis*) were differentiated both genetically and morphologically from all other birds in the sample (*Z. c. hypoleuca*). However, the steepness and location of the clines for these two character types were substantially different. An abrupt cline in PGM-1 allozyme frequencies was apparent in the extreme northwest of our study area, while clines in morphological features (primarily body size) were gradual and extended much further southward. Among nonpuna samples (20 sites; *Z. c. hypoleuca*), patterns of intersite differentiation in morphological and allozyme characters were unrelated. Partial Mantel's tests showed that, among these samples, both degree of difference in habitat structure and linear geographic distance are important correlates of among-site morphological differentiation. Similar tests demonstrated that neither of these two environmental factors is related statistically to genetic differentiation among populations. *Received 3 October 1991, accepted 21 November 1992.*

THE STUDY of geographic variation of various types of characters (e.g. morphological, physiological, genetic, behavioral) is central to an understanding of evolutionary processes. Heritable variation is the raw material for natural selection and drift, so the specification of the extent and spatial organization of variation shown by a given species is a fundamental step in the investigation of processes involved in population differentiation. Thus, intraspecific patterns of variation in space can reveal much about a diversity of evolutionary processes, from the structuring of populations and the formation of clines to speciation (e.g. Gould and Johnston 1972, Endler 1977, Zink and Remsen 1986).

Classically, studies of geographic variation considered single-character systems (e.g. morphological). More recently, however, variation in several types of characters has been studied in concert (e.g. Handford and Nottebohm 1976, Sene and Carson 1977, Schnell et al. 1978, Zink 1986, Lagercrantz and Ryman 1990). The various character suites may be subject to different influences and may show different levels or patterns of variation. For example, several studies have shown a lack of concordance between patterns of variation in morphological and genetic characters, which may be due either to differences in selective pressures acting on the different character suites (e.g. Schnell and Selander 1981), or to differences in ability to discern statistical differences (Lewontin 1984). Studies of geographic variation should involve many characters because patterns of variation in space are probably not the result of change in one or a few characters in response to a single environmental variable. Rather, it is more probable that the patterns of variation are due to adaptation of many characters to many different environmental factors (Sokal and Rinkel 1963) or to stochastic factors (e.g. Lynch 1989). To gain a better understanding of the factors potentially influencing population structure within a species, variation in a variety of character types should be documented, recognizing the relative "strengths and weaknesses" of each (Zink 1986).

The Rufous-collared Sparrow (*Zonotrichia capensis*) is well suited for studies of geographic variation in behavioral, morphological and genetic characters because: (1) it is geographically widespread, ranging from southern Mexico to the southern tip of South America (Chapman 1940); (2) it is found in virtually all types of habitats except for closed tracts of forest (Handford 1983, 1985, 1988); (3) it displays a unique system of vocal dialects defined by the rate of delivery of the terminal trill (Nottebohm 1969, 1975, Handford 1981, 1988); and (4) it shows ecological variation in song pitch and duration characters (Handford and Lougheed 1991).

In many temperate bird species, morphological differentiation is evident without corresponding (detectable) differences in allozyme frequencies (Barrowclough 1983, Zink 1991). However, as Zink (1991) has observed, such temperate populations may not have reached an evolutionary equilibrium, and this may explain the lack of allozyme differentiation. Limited information (e.g. Capparella 1988, Hackett and Rosenberg 1990) suggests that in low-latitude species allozyme differentiation may be more extensive, presumably since they are closer to evolutionary equilibrium (see also Zink et al. 1991). *Zonotrichia capensis* spans 65° of latitude and is primarily a bird of open country. Thus, it is found mostly at high altitudes in tropical latitudes. Therefore, it is not clear which model of variation might apply. Wright (1943, 1978a) suggested that geographic distance may play an important role in population structuring; if isolation-by-distance is important in populations of Rufous-collared Sparrows, then levels of differentiation of allozyme characters between compared populations should be positively related to geographic distance. To the extent that morphological characters reflect underlying genetic differences that are not subject to selection, this would also be true of morphological variation. However, if Rufous-collared Sparrow populations are adapted to some aspects of their environment, then it might be expected that morphological and, possibly, some other aspects of genetic differentiation would be related to the degree of differentiation among habitats.

In a companion article (Lougheed and Handford 1992), we have used the data sets described below to test and reject the genetic-adaptation hypothesis as an explanation for the existence of vocal dialects in northwestern Argentine populations of Rufous-collared Sparrows. In the present paper, we investigate multicharacter variation in the same populations of Rufous-collared Sparrows. Specifically, we address three questions: (1) How do morphological and genetic characters vary in space? (2) How do the patterns of geographic variation exhibited by the two types of character compare? (3) How do geographic patterns relate to geographic distance and environmental characteristics?

MATERIALS AND METHODS

Specimen and data collection.—Approximately 20 male Rufous-collared Sparrows were collected from four sites (arranged in a latitudinal series) in each of six vegetation types: lowland chaco thornscrub, transition forest, montane woodland, montane grassland, Monte desert scrub and puna high-altitude scrub (474 specimens total). A detailed collection protocol and locality map is contained in Lougheed and Handford (1992).

Eight external morphological and 12 skeletal characters were measured using dial and electronic digital calipers, respectively (± 0.1 mm). External characters measured were wing chord, outer rectrix, tarsus length, hind toe plus claw, bill depth, bill width, culmen (measurements as described in Handford 1985, Zink 1986), and bill length from the gape (distance from gape to tip of upper mandible). Skeletal variables measured were partial skull length (from foramen magnum to base of bill), skull width across the bullae, width of the scapula at proximate end, scapula length, coracoid length, humerus length, ulna length, keel depth, sternum length, femur length, width of proximal end of femur, tibiotarsus length, and synsacrum width across acetabula (except for partial skull length, all measures described in Robins and Schnell 1971).

Starch gel electrophoresis, with liver and kidney as source tissues, was used to assay allozyme variation at 20 protein-encoding loci: aspartate aminotransferase, AAT-1, AAT-2; acid phosphatase, ACPH-1; esterase, EST-5; general protein, GP-1, hemoglobin, HB-2; glucose-3-phosphate dehydrogenase, G3PDH; isocitrate dehydrogenase, IDH-1, IDH-2; lactate dehydrogenase, LDH-1, LDH-2; malate dehydrogenase, MDH-1, MDH-2; mannose phosphate isomerase, MPI; peptidase, PEP-A; phosphogluconate dehydrogenase, PGD; phospho-glucomutase, PGM-1, PGM-2; purine nucleoside phosphorylase, PNP; and superoxide dismutase, SOD-1. Details are given in Lougheed and Handford (1992).

Finally, to obtain some measure of vegetation structure, each site was scored independently by both authors for maximum vegetation height, average vegetation height, percent vegetation cover, and percent cover by trees. Scores for each variable ranged from 1 to 5, and were based on photographs, notes, and field experience at each site.

Patterns of geographic variation.—Correlation matrices (PROC NESTED; SAS Institute 1985) were constructed at two levels for both external and skeletal morphological characters: among-site matrices (i.e. analogous to performing correlation analysis on site means, but correcting for within-site variation and covariation) and within-site matrices (i.e. among individuals at each site, averaged over all 24 sites). These matrices then were used in a principal-components analysis, so that within-site and among-site patterns of morphological variation in both external and skeletal characters could be described. A one-way analysis of variance (hereafter ANOVA; PROC ANOVA; SAS Institute 1985) was used to assess the magnitude of within- and among-site variation for each external and skeletal ($\log_e$ transformed) variable. We carried out *a posteriori* tests using Fisher's protected least

TABLE 1. Results of principal-components analyses on among-individual (within-site) and among-site (within-site variation removed) correlation matrices based on eight external characters. Correlations between original variables and derived principal-component (PC) variables. Also included are percentages of total variation explained by each of the derived PC axes.

	Within site				Among site	
Variable	PC1	PC2	PC3	PC4	PC1	PC2
Outer rectrix length	0.60	−0.62	−0.26	0.007	0.90	−0.37
Wing chord length	0.67	−0.48	−0.35	−0.02	0.84	−0.45
Hind toe length	0.39	−0.07	0.68	0.13	0.77	0.24
Tarsus length	0.39	−0.22	0.60	0.34	0.94	0.09
Bill depth	0.53	0.42	−0.34	0.28	0.22	0.82
Bill width	0.44	0.47	−0.16	0.55	0.28	0.72
Gape length	0.56	0.28	0.20	−0.51	0.91	0.04
Culmen length	0.60	0.35	0.04	−0.46	0.95	0.01
Percent of total	28.2	15.7	15.0	12.4	60.6	20.0

squares differences and a Bonferroni-corrected α-level (Abacus Concepts 1990). For comparison with a continental analysis of morphological variation in this species (Handford 1983), general patterns of variation across the study area were displayed in contour plots constructed by distance-weighted least-squares interpolation (Wilkinson 1991) from character site means.

To evaluate levels of among-site genetic differentiation, we calculated Wright's (1978b) fixation coefficients. F_{ST}-values (corrected for sampling error) were calculated for each polymorphic locus separately, and overall (arithmetic means of all values for each F-index), using STEP WRIGHT78 (BIOSYS-1; Swofford and Selander 1981). F_{ST}-values were converted to chi-square statistics testing for heterogeneity of allelic frequencies among subpopulations using the formula outlined by Workman and Niswander (1970; STEP HETXSQ).

Comparisons of patterns of variation.—A matrix of geographic distances between sites (GEO) was calculated using the geographic coordinates for each site, and rounded to the nearest 10 km. A matrix of crude ecological distances (ECO) reflecting differences in habitat structure was constructed as follows: (1) A Spearman rank-correlation matrix was calculated based on all eight habitat variables (four variables scored by each of the two authors). (2) A principal-components analysis was performed on the Spearman correlation matrix. (3) For each site, the scores on principal component 1 (PC1) were calculated based on standardized variables, and the difference between scores for any two sites was considered the ecological distance.

Matrices of Mahalanobis' distances were calculated, using each of the external (EXT) and skeletal (SKE) morphological data sets ($\log_e$-transformed data; PROC CANDISC; SAS Institute 1985), for each site pair. Mahalanobis' distances are a measure of distance between group centroids in multivariate (canonical-variate) space, corrected for correlation among variables (Campbell and Atchley 1981). Finally, Nei's (1978) genetic distances corrected for sample size and a matrix of Rogers' (1972) genetic distances (ROG) were calculated using the SIMDIS procedure in BIOSYS-1. The former distances were computed for purposes of comparison to other published values only, and were not used in any subsequent analyses because of nonmetricity (Evans 1987).

To test for congruence (correlation, r) of pattern among certain distance matrices, Mantel and partial Mantel (Smouse et al. 1986) nonparametric tests were used, with 250 iterations per pairwise test ("R"-package; Legendre and Vaudor 1985). Mantel tests assess the independence of pattern between two distance matrices (Mantel 1967, Sokal 1979); in the partial Mantel test, the association of a dependent matrix on both of two predictor matrices is assessed, in each case with the effects of one of the two predictor matrices partialed out. Here, EXT, SKE and ROG are compared with each other using the Mantel test, and each is compared with GEO and ECO as predictors, using partial Mantel tests. Significance of correlations among matrices is assessed by computing t-values (Legendre and Vaudor 1985).

RESULTS

The patterns of correlation between both external and skeletal variables among individuals within sites and among sites (corrected for within-site variation) were very similar. Virtually all correlations were positive and, in general, among-site correlations were greater in magnitude than corresponding within-site correlations, reflecting a lower proportion of error variance (higher "signal-to-noise ratio"). The results of corresponding principal-components analyses are presented in Tables 1 and 2. Only loadings for the axes that cumulatively explained approximately 70% of total variation of the original data sets are reported. In all four analyses, PC1 is primarily a size axis, with all or most of the original variables having com-

TABLE 2. Results of principal-components analyses on among-individual (within-site) and among-site (within-site variation removed) correlation matrices of 12 skeletal characters. Correlations between original variables and derived principal-components (PC) variables. Also included are percentages of total variation from the original data.

	Within site				Among site	
Variable	PC1	PC2	PC3	PC4	PC1	PC2
Skull length	0.53	−0.41	−0.20	0.45	0.89	−0.24
Skull width	0.56	−0.45	−0.10	0.42	0.85	−0.11
Coracoid length	0.79	0.08	0.01	−0.09	0.98	0.11
Scapula length	0.72	−0.11	0.01	−0.23	0.98	0.05
Sternum length	0.61	−0.28	0.13	−0.45	0.96	0.04
Keel depth	0.48	−0.49	0.30	−0.28	0.24	−0.95
Femur length	0.81	0.37	−0.02	0.12	0.98	0.15
Femur width	0.34	0.09	−0.85	−0.29	0.80	−0.16
Synsacrum width	0.49	−0.36	−0.09	−0.01	0.97	0.11
Humerus length	0.84	0.35	0.05	0.06	0.99	0.03
Tibiotarsus length	0.77	0.30	0.18	0.10	0.98	0.19
Ulna length	0.79	0.26	0.12	0.06	0.96	0.01
Percent of total	43.9	10.4	7.9	7.0	81.6	9.1

parably large positive loadings. The among-site PC1 axis for both external and skeletal data sets explained a larger portion of the original variation (60.6 and 81.6%, respectively) than did the within-site PC1 axes (28.2 and 43.9%, respectively), again reflecting a higher signal-to-noise ratio in among-site comparisons. In all analyses, PC2 is a shape axis (i.e. the axes are bipolar, with original variables having both positive and negative loadings; see Tables 1 and 2).

The one-way ANOVAs of sites showed that all morphological characters were significantly heterogeneous over the study area (Table 3). In all cases, it is clear that among-site variation greatly exceeds that within sites, with a major gap in the distributions of virtually all characters separating puna sites from all others.

Contour plots of the variables showed an overall similarity of pattern—a northeast-southwest cline in character values paralleling the prevailing change in altitude. As examples, we show external among-site PC1, wing chord

TABLE 3. ANOVA for all morphological characters (values log-transformed).

		Sum of squares		Mean square		
Variable	df	Among	Within	Among	Within	F^a
Outer rectrix length	23,437	1.58	0.74	0.069	0.002	40.42
Wing chord length	23,450	1.51	0.46	0.066	0.001	64.77
Tarsus length	23,447	0.56	0.49	0.024	0.001	22.36
Hind toe length	23,450	1.04	0.73	0.045	0.002	27.63
Bill depth	23,450	0.10	0.53	0.004	0.001	3.56
Bill width	23,450	0.07	0.57	0.003	0.001	2.44
Bill gape length	23,450	0.56	0.75	0.024	0.002	14.53
Culmen length	23,448	0.62	0.71	0.027	0.002	16.91
Skull width	23,448	0.08	0.17	0.004	0.000	9.50
Skull length	23,449	0.10	0.15	0.004	0.000	13.19
Coracoid length	23,448	0.30	0.34	0.013	0.001	17.27
Scapula length	23,435	0.30	0.42	0.013	0.001	13.81
Sternum length	23,448	0.30	0.37	0.008	0.001	10.31
Keel depth	23,445	0.09	0.74	0.004	0.002	2.38
Synsacrum width	23,444	0.69	0.54	0.030	0.001	24.87
Femur width	23,450	0.26	0.25	0.011	0.001	20.51
Femur length	23,447	0.73	0.26	0.032	0.001	54.42
Tibiotarsus length	23,409	0.51	0.25	0.022	0.001	36.67
Humerus length	23,444	0.69	0.54	0.030	0.001	24.87
Ulna length	23,409	0.51	0.25	0.022	0.001	36.67

[a] All values $P < 0.001$.

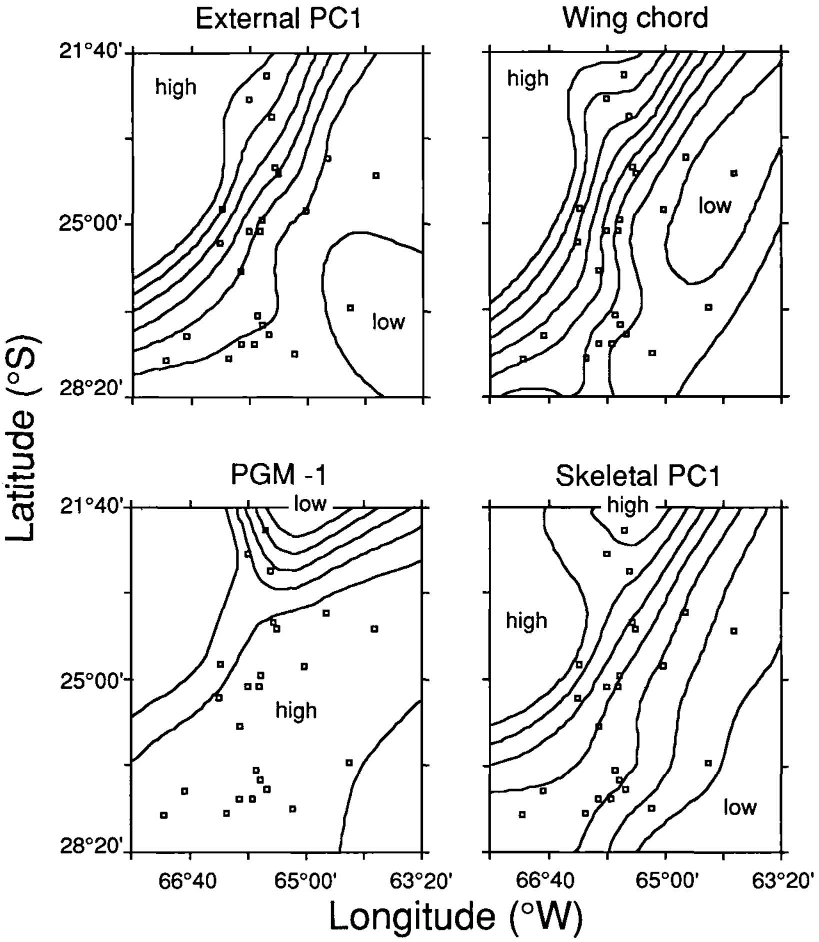

Fig. 1. Contour plots of variation in among-site external PC1 scores, wing chords, among-site skeletal PC1 scores, and frequencies of the PGM-1 "c" allele. Wing-chord site means vary from 65.1 to 78.9 mm., and PGM-1-c allele frequencies from 1.00 to 0.175. Sample sites indicated by open squares.

and skeletal among-site PC1 (Fig. 1). The largest birds are found in the northern portion of the study area, corresponding to high-altitude puna scrub vegetation. Size grades through intermediate-sized birds in the pre-Andean ranges and foothills to the smallest birds in the lowland chaco thornscrub vegetation to the southeast.

Of the 20 resolved isozymes, 12 were polymorphic in at least one of the 24 study sites (see Lougheed and Handford 1992). Corrected F_{ST}-values ranged from 0.000 (for EST-6, MDH-1, MDH-2, and PGD) to 0.344 (PGM-1; Table 4). The mean corrected F_{ST}-value across all loci was 0.119. Corrected chi-square tests revealed that allozyme frequencies for three loci (GP-1, MPI, and PGM-1) were significantly heterogeneous among sites (Table 4). Not surprisingly, when all loci were considered together, there was significant heterogeneity across the study area.

Mantel tests showed that the patterns of variation among external, skeletal and allozyme were significantly concordant ($P < 0.001$). Partial Mantel tests (Table 5) showed that a significant amount of variation in the three dependent matrices (EXT, SKE and ROG) was explained by both the GEO and ECO predictor matrices, while controlling for the effects of one of them.

DISCUSSION

Variation of morphological characters.—Positive correlations between morphological characters among individuals within sites indicate that,

TABLE 4. F_{ST}-values (corrected for sampling error; Wright 1978b) for entire study area (subpopulation refers to site, regardless of vegetation) based on all polymorphic loci. Chi-square statistics for heterogeneity of allelic frequencies also indicated (Workman and Niswander 1970).

Locus	F_{ST}	X^2	df
AAT-1	0.005	7.1	46
EST-6	0.000	0.0	23
GP-1	0.045	81.9***	46
IDH-1	0.014	35.4	69
IDH-2	0.012	20.7	46
MDH-1	0.000	0.0	46
MDH-2	0.000	0.0	23
MPI	0.061	111.7***	46
PEP-A	0.020	35.6	46
PGD	0.000	0.0	92
PGM-1	0.344	888.8***	69
PGM-2	0.031	54.2	46
Overall	0.119	1235.4***	

*, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

locally, birds vary primarily in size. The correlations between morphological characters among sites indicate that, across the study area, variation also is primarily in size. The principal-components analyses economically describe this finding: in all analyses, the first axes are unipolar (primarily size dimensions; Tables 1 and 2). However, there is substantial shape information carried by the among-site PC1 in the analysis of external characters, with loadings ranging from over 0.90 (bill length, tarsus and tail length) to less than 0.30 for bill width and depth. Handford (1983) has shown that on a continental scale PC-1 is bipolar, conveying much shape variation.

Morphological characters showed great intersite variability as indicated by the highly significant F-values resulting from ANOVA (Table 3). Although the various characters show different relative amounts of among-locality character variability, as indicated by the among-group mean squares from the ANOVAs, much of the effect is generated by the major separation between puna sites from all others. *A posteriori* tests indicated that puna birds are the only significantly distinct group.

The spatial distribution of morphological character variation (Fig. 1) is described most simply as a southeast to northwest cline in overall size between the subspecies *hypoleuca* and the Bolivian high-altitude subspecies *pulacayensis* (Chapman 1940). This corresponds directly with Handford's (1983) finding that high-Andean individuals are substantially larger and longer-winged than those from the lowlands. Chapman (1940) suggested that the change from *hypoleuca* to *pulacayensis* takes place clinally between 2,000 and 3,000 m in the front ranges of the Andes, citing a series of specimens taken from a transect near the center of our study area. Our morphological results are consistent with this interpretation; all characters show smooth southeast-northwest clines. Such large, long-winged high-altitude (and high-latitude; Handford 1983) birds are migratory, sometimes extensively so, and their long wings no doubt reflect an adaptation to this migratory propensity. However, our allozyme results (below) indicate a very different clinal pattern, suggesting that "true" *pulacayensis* are only found on the Bolivian plateau to the north of our area.

Variation in allozyme frequencies.—The overall F_{ST}-value (0.119) and its associated chi-square statistic indicates significant differentation among sites in allozyme frequencies (Table 4). For natural populations of birds, this level of differentiation is exceptionally high (Barrow-

TABLE 5. Pattern of variation in the dependent matrix A compared to that in the predictor matrix B, while controlling for the effects of matrix C, using partial Mantel tests (Smouse et al. 1986). Matrix correlations (r) and test statistics (t) for all 24 sites (left), and the 20 nonpuna sites (right).

Matrix			24 sites		20 sites	
A	B	C	r	t	r	t
EXT	GEO	ECO	0.52	6.176*	0.33	3.589*
EXT	ECO	GEO	0.54	8.894*	0.34	4.644*
SKE	GEO	ECO	0.52	5.641*	0.31	3.434*
SKE	ECO	GEO	0.40	6.577*	0.24	3.302*
ROG	GEO	ECO	0.35	3.513*	−0.05	−0.467
ROG	ECO	GEO	0.19	3.643*	0.00	0.053

*, $P < 0.01$.

clough 1983, Evans 1987), although not the highest reported (e.g. for *Empidonax difficilis* populations, uncorrected $F_{ST} = 0.153$; Johnson and Marten 1988). Barrowclough (1983) reported average F_{ST}-values among populations within species for five vertebrate classes: fish, 0.114 (9 species); amphibians, 0.383 (15 species); reptiles, 0.304, (3 species); mammals, 0.230 (25 species); and birds, 0.022 (5 species). The F_{ST}-values reported here for Rufous-collared Sparrows, then, greatly exceed most avian values, and are comparable to some intraspecific comparisons within other vertebrate classes.

Although corrected F_{ST} and chi-square statistics indicate that 3 of the 12 loci (Table 4) are significantly heterogeneous, PGM-1 is the most strongly differentiated (corrected $F_{ST} = 0.344$). A contour plot of the common allozyme of PGM-1 (Fig. 1) suggests a steep cline involving the three most northerly puna sites. Here, the frequency of the common allele drops abruptly from a frequency near 1.0 to less than 0.20. Nei's (1978) and Roger's (1972) genetic distances between the northernmost puna site and all nonpuna sites (average Nei's distance = 0.031 ± SE of 0.0050) exceed most other intraspecific comparisons of avian populations. In birds, Nei's (1978) genetic distance averages 0.0024 ± 0.0028 ($n = 113$ comparisons) among local populations, 0.0048 ± 0.0049 ($n = 86$) among subspecies and 0.044 ± 0.0221 ($n = 71$) among species. For all other (nonpuna) intersite comparisons, genetic distance values are within the range reported for other intraspecific local population comparisons for birds (see also Baker et al. 1990, Zink 1986). A change of such magnitude over such a small distance is uncommon in allozyme studies of avian species (but see Barrowclough and Gutierrez 1990).

The three most northerly puna sites are at altitudes in excess of 3,500 m at the southern end of the Bolivian/Peruvian high plateau. This suggests to us that, despite the broad clines in morphological characters across our study area, prototypical high-plateau *pulacayensis* barely enters Argentina, and that the location of the *hypoleuca/pulacayensis* transition inferred by Chapman (1940) is mistaken. According to our present view, virtually our whole study region would be seen as a zone of introgression between two highly differentiated groups (quite probably not coincident with described subspecies): "*pulacayensis*" of the high plateau (altiplano) and "*hypoleuca*" of the Argentine/Paraguayan plains. If this is the case, then clearly we are dealing with a nonequilibrium situation, and the F_{ST}-value of 0.119 represents deep historical subdivision of the species range. If we restrict attention to nonpuna populations as an approximation to "*hypoleuca*" populations (which may be closer to evolutionary equilibrium), we find an average F_{ST}-value of 0.017. This value appears to be typical of intraspecific comparisons in birds (Barrowclough 1983).

Zonotrichia capensis is clearly one of the most polytypic bird species in the New World (Chapman 1940, Handford 1985) and shows substantial genetic differentiation (this study, Lougheed et al. unpubl. data, Zink et al. 1991). Capparella (1988) suggested that species of Neotropical birds generally show greater levels of genetic differentiation than temperate-zone birds, an observation that has been supported by other studies (e.g. Hackett and Rosenberg, 1990). He favored the so-called "riverine-barrier" hypothesis of Sick (1967), which contends that the development of rivers in the Amazon basin dissected once-contiguous areas of habitat, leading to reduced gene flow among fragmented populations of Amazonian forest birds. Although the level of genetic differentiation found in Rufous-collared Sparrows is consistent with Capparella's general observation, Sick's riverine-barrier hypothsis would not appear to be appropriate. Rufous-collared Sparrows are an open-country species and are not found in continuous closed tracts of forest. At equatorial latitudes this species is found in higher-altitude, open habitats, and its range extends south to Tierra del Fuego; thus, it is primarily a subtropical and temperate rather than a tropical species.

Covariation among characters sets.—There was significant concordance in patterns of EXT, SKE, and ROG. The correspondence between the EXT and SKE matrices is unremarkable, but the significant association of both of these with ROG is worthy of note. Zink (1988) found no significant relationship between allozyme and morphological distances in populations of Brown Towhees (*Pipilo fuscus* complex) or in Fox Sparrows (*Passerella iliaca*; Zink 1991). Barrowclough (1983) noted that, generally in birds, morphological differentiation among populations is often evident without a similar level of differentiation of allozyme frequencies (although

Lewontin [1984] has cautioned that the statistical power for discriminating population differences is different for these two classes of characters). Lack of concordance between patterns of morphological and allozyme variation might be evidence for differences in evolutionary rates of different types of traits (e.g. Berlocher and Bush 1982, Gorman and Kim 1976, Schnell et al. 1978, Schnell and Selander 1981, Sene and Carson 1977, Turner et al. 1979, Zink 1986).

Environmental correlates of variation.—Partial Mantel tests (Table 5) indicate that differentiation in external, skeletal and allozyme characters is concordant with the geographic dispersion of the sites, suggesting that isolation-by-distance may be an important, perhaps sufficient factor in explaining among-site differentiation (Wright 1943, 1978a). However, there was equally close correspondence between variation in external, skeletal and allozyme characters and ecological differentiation among sites. This suggests that there may be regional character patterning due to ecological factors.

Covariation with puna sites eliminated.—Because puna sites are so markedly different from other sites, both morphologically and genetically, it is possible that the Mantel comparisons based on all 24 sites are nonrepresentative with respect to variation in the nonpuna region. For example, any conclusions regarding isolation-by-distance are confounded by the fact that all puna scrub sites were in the northwest of the study area and, as we have seen, possibly attributable to another subspecies. Thus, we compared distance matrices using only the 20 nonpuna sites. In comparing EXT, SKE and ROG, only EXT and SKE showed (highly significant) association; there was no congruence with ROG. Partial comparisons of EXT, SKE and ROG with GEO and ECO left the strong association of SKE and EXT with both ECO and GEO intact. However, ROG showed no significant concordance with either GEO or ECO. Thus, in contrast with the overall analysis (all 24 sites), concordance of pattern between morphological and genetic distances is not evident. The significant Mantel tests based on 24 sites simply reflect the inclusion of the highly distinctive puna specimens. It would appear, then, that there are no simple patterns attributable to isolation-by-distance among the 20 nonpuna sites in allozyme characters, nor is there differentiation related to habitat, independent of geographic distance. Morphological variation, on the other hand, remains equally strongly associated with geographical distance and habitat differences among the 20 nonpuna sites.

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