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Author(s): Karin Maldonado, Francisco Bozinovic, José M. Rojas, Pablo Sabat

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Within-Species Digestive Tract Flexibility in Rufous-Collared Sparrows and the Climatic Variability Hypothesis

Karin Maldonado^{1,2,*}

Francisco Bozinovic³

José M. Rojas³

Pablo Sabat^{3,4}

¹Instituto de Filosofía y Ciencias de la Complejidad, Los Alerces 3024, Santiago, Chile; ²Departamento de Biología, Facultad de Ciencias, Universidad de La Serena, Casilla 599, La Serena, Chile; ³Center for Advanced Studies in Ecology and Biodiversity, Laboratorio Internacional para el Cambio Global, and Departamento de Ecología, Facultad de Ciencias Biológicas, Pontificia Universidad Católica de Chile, Santiago, CP 6513677, Chile; ⁴Departamento de Ciencias Ecológicas, Facultad de Ciencias, Universidad de Chile, Casilla 653, Santiago, Chile

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ABSTRACT

The climatic variability hypothesis (CVH) states that species are geographically more widespread at higher latitudes because individuals have a broader range of physiological tolerance or phenotypic flexibility as latitude and climatic variability increase. However, it remains unclear to what extent climatic variability or latitude, acting on the phenotype, account for any observed geographical gradient in mean range size. In this study, we analyzed the physiological flexibility within the CVH framework by using an intraspecific population experimental approach. We tested for a positive relationship between digestive-tract flexibility (i.e., morphology and enzyme activities) and latitude and climatic and natural diet variability in populations of rufous-collared sparrows (*Zonotrichia capensis*) captured in desert (27°S), Mediterranean (33°S), and cold-temperate (41°S) sites in Chile. In accordance with the CVH, we observed a positive relationship between the magnitude of digestive-tract flexibility and environmental variability but not latitude. The greatest digestive flexibility was observed in birds at middle latitudes, which experience the most environmental variability (a Mediterranean climate), whereas individuals from the most stable climates (desert and cold-temperate) exhibited little or no digestive-tract flexibility in response to experimental diets. Our findings support the idea that latitudinal gradients

in geographical ranges may be strongly affected by the action of regional features, which makes it difficult to find general patterns in the distribution of species.

Introduction

It has been proposed that variable environments favor the evolution of phenotypic flexibility (Via et al. 1995; Schlichting and Pigliucci 1998). Theoretically, this flexibility would allow a genotype to have a broader tolerance that may facilitate its establishment in novel environments (for a review, see Ghalambor et al. 2007). In turn, the climatic variability hypothesis (CVH; Stevens 1989) posits that broader physiological tolerance of animals allows them to achieve a larger distribution range at higher latitudes as a consequence of increasing climatic variability. Despite the long standing of the CVH, studies attempting to test its predictions using physiological data have been mainly restricted to the analysis of thermal limits among ectothermic species (Sunday et al. 2011 and references therein), in which, moreover, the roles of phenotypic flexibility and acclimation abilities have been underestimated (but see Calosi et al. 2010).

Recently, Naya et al. (2008) tested the CVH with a meta-analytical study using data on rodent intestine length flexibility and found a positive correlation of intestine flexibility with both the number of habitats occupied and latitude. Nevertheless, although the CVH implies that climatic variability and not latitude itself is responsible for physiologically more flexible species, the authors found that latitude is a better predictor of rodents' physiological flexibility than climatic variability per se. The aim of this study is to provide insights into the proximate factors underlying the CVH through an avian interpopulation analysis, specifically to clarify whether latitude or climatic variability is a better predictor of phenotypic flexibility.

Theoretical and experimental studies have demonstrated that animals maintain their digestive performance by modifications in gut size, nutrient transport activity, and hydrolysis rate of tissue-specific digestive enzymes when fed chemically different diets (Karasov 1996). In our study, the amount of both phenotypic flexibility in intestinal enzyme activities and digestive organ flexibility was measured in three populations of *Zonotrichia capensis* from different sites in Chile (desert, Mediterranean, and cold-temperate) that differ in their environmental variability. Then digestive-tract flexibility was related to latitude, climate variability (temperature and rainfall), and the dietary variation that birds experience in the field.

* Corresponding author; e-mail: kmaldonado@ificc.cl.

Because there is a relative ^{15}N enrichment of bird tissues with increasing trophic level, nitrogen-stable isotope ratios ($^{15}\text{N}/^{14}\text{N}$) have been widely used to determine the dietary patterns of animals (Post 2002; Sabat et al. 2009; Ramirez-Otarola et al. 2011). Here we measured the natural diet variation by using $\delta^{15}\text{N}$ signatures from tissues that differ in their turnover rates in order to obtain information about the diet of an animal at different temporal scales. Therefore, rather than estimating the relative position in the food web of birds in different populations, our objective was to estimate seasonal differences in the isotopic niche (sensu Newsome et al. 2007).

Material and Methods

Animals and Fieldwork

Zonotrichia capensis is distributed between southeast Mexico and Cabo de Hornos, southern South America (Goodall et al. 1951). In Chile, the rufous-collared sparrow is practically ubiquitous, inhabiting both deserts and rain forests (Araya and Millie 2005). The subspecies we studied here, *Zonotrichia capensis chilensis*, has been reported as nonmigratory (Chapman 1940). Moreover, this species has been described as omnivorous because it feeds on mixed diets based on seeds and insects (López-Calleja 1995; Sabat et al. 1998; Ramirez-Otarola et al. 2011).

Nonreproductive adult birds were captured with mist nets during the austral autumn (April–June) in 2008. After capture, birds were weighed to the nearest 0.1 g. The number of individuals collected and the localities were as follows: 16 individuals in Copiapó (27°18'S, 70°25'W), a typical desert scrubland with little rainfall only in winter; 14 in Quebrada de la Plata (33°31'S, 70°50'W), a Mediterranean climate with rainfalls concentrated in the cold winter and high temperatures and low rainfalls in summer; and 16 in Llanquihue (41°16'S, 73°00'W), a southern cold-temperate forest with rainfalls during almost all of the year and lower temperatures (see Table 1). The three capture sites cover a latitudinal range of more than 1,800 km. Historical data for both rainfall and temperatures were obtained from national institutes of information (Dirección Meteorológica de Chile and Centro de Información de Recursos Naturales).

In order to investigate seasonal variability in the natural diet, we measured the isotopic signature ($\delta^{15}\text{N}$) of feathers and blood.

Using the nitrogen isotope ratio of a consumer's tissue to assess the relative position in the food web makes two key assumptions: (1) tissues reflect the isotopic composition of an animal's diet (Hobson and Clark 1992a), and (2) consumers are significantly enriched in ^{15}N relative to food sources (i.e., diet-tissue fractionation). Although fractionation factors may differ significantly among species and tissues, these factors appear to be less variable between feathers and blood (Hobson and Clark 1992b). Consequently, after capturing birds, we obtained both the first primary feather from each bird's left wing and a blood sample (~100 μL) from the brachial vein (placed into heparinized microhematocrit tubes). Because feathers (after growth) are an inert structure and in general their growth is sequential (Campbell and Lack 1985), the isotopic signature of the first primary feather should represent the diet at the beginning of the postnuptial molt. In turn, the blood isotopic composition would correspond to resources assimilated about 14 d before sampling (Hobson and Clark 1992a; Carleton and Martínez del Rio 2005; Bauchinger and McWilliams 2009). Thus, because the *Z. capensis* breeding season is from around the end of October until the end of December (Davis 1971; King 1973, 1974; Moore et al. 2004) and birds were captured during austral autumn, the isotopic composition of these tissues represents the natural diet over two seasons.

Before isotopic signatures were measured, feather samples were cleaned from external contaminants with a 2 : 1 chloroform : methanol solvent (Hobson 1999), and all samples (including blood) were dried to a constant mass at 80°C. After the samples were ground into a fine powder, they were loaded into tin capsules. Nitrogen isotope ratios were measured on a continuous-flow isotope ratio mass spectrometer (Finnigan Delta + XP) with samples burned in a Costech elemental analyzer at the University of Wyoming Stable Isotope Facility. The precision of this analysis was $\pm 0.2\%$. Nitrogen isotope ratios were expressed by using standard delta notation $\delta^{15}\text{N}$ in parts per thousand as

$$\delta^{15}\text{N} = \left(\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right) \times 1,000,$$

where R_{sample} and R_{standard} are the molar ratios of $^{15}\text{N}/^{14}\text{N}$ of the sample and reference, respectively. Samples were referenced against the international standard, atmospheric nitrogen.

Table 1: Localities and climatic variables of different studied habitats of *Zonotrichia capensis* in Chile

	Copiapó	Quebrada de la Plata	Llanquihue
Coordinates	27°18'S, 70°25'W	33°31'S, 70°50'W	41°16'S, 73°00'W
Climate	Desert	Mediterranean	Cold-temperate
Mean annual rainfall \pm SD (mm)	.94 \pm 1.5	28.1 \pm 29.8	164.3 \pm 56.5
CV of rainfall (%)	15.9 ^A	106.04 ^B	34.4 ^C
Mean $T_A \pm$ SD (°C)	15.7 \pm 3.1	13.9 \pm 4.5	9.6 \pm 3.2
CV of temperature (%)	19.7 ^A	32.4 ^A	33.3 ^A

Note. T_A = annual temperature; CV = coefficients of variation. Different superscript letters denote significant differences in the variability among localities after a Levene test.

Animals and Laboratory Work

After completing field procedures, sparrows were transported to the laboratory in Santiago, Chile (33°27'S, 70°42'W). They were kept in individual plastic-mesh cages (35 cm × 35 cm × 35 cm) at constant temperature of 22° ± 2°C and photoperiod of 12L : 12D. During the 1-d habituation period, sparrows were fed with mealworms (larvae of *Tenebrio molitor*), seeds, and water ad lib. After this, one group was acclimated to an insect-based diet of mealworms and the second to a seed-based diet containing commercial bird seed (*Phalaris canariensis*). A vitamin complex was added to the water supplied ad lib. We chose these diets in order to resemble the natural food types of these birds (López-Calleja 1995). The insect-based diet represents a high-protein and low-carbohydrate diet, while the seed-based diet represents a high-carbohydrate and low-protein diet. Diets were provided ad lib. for 7 wk.

Digestive Enzyme Activities

Birds were killed by CO₂ exposure. After their body mass was measured with an electronic balance (± 0.1 g), birds were dissected abdominally, and the small intestine was removed and carefully cleaned of digesta. The small intestine was weighed in an electronic balance (± 0.0001 g), and its length was measured with a ruler (± 1 mm) and a digital caliper (± 0.01 mm). In order to prepare the small intestine for the enzymatic measurements, it was washed in a 0.9% NaCl solution and immediately stored frozen at -20°C for later enzyme-activity determinations (Martínez del Río et al. 1995; Sabat et al. 1998).

Disaccharidase activity was determined according to the method of Dahlqvist (1964) modified by Martínez del Río (1990). Briefly, 100 µL of tissue homogenate was incubated at 40°C with 100 µL of 56 mmol L⁻¹ of substrate (maltose or sucrose) in 0.1 M Maleate/NaOH buffer, pH 6.5. After incubation of 3 or 5 min for maltase and sucrase, respectively, the reaction was stopped by the addition of 3 mL of stop/develop Glucose-Trinder (Valtek, Santiago, Chile). This solution consisted of 500 mL in 250 mL 0.1 mol L⁻¹ TRIS/HCl, pH 7, plus 250 mL of 0.5 NaH₂PO₄, pH 7. After an incubation period of 18 min at 20°C, the absorbance was measured at 505 nm with a UV-VIS spectrophotometer (Shimadzu UV mini-1240, Tokyo). Aminopeptidase-N assays were done by using L-alanine-*p*-nitroanilide as substrate. The assay began by adding 1 mL of substrate solution (2.04 mmol L⁻¹ of L-alanine-*p*-nitroanilide in 0.2 mol µL⁻¹ NaH₂PO₄/Na₂HPO₄, pH 7) to 100 µL tissue homogenate. The reaction was incubated for 10 min at 40°C and then stopped using 3 mL of ice-cold acetic acid (2 mol L⁻¹). The absorbance was measured at 384 nm. On the basis of their absorbance, standardized intestinal enzymatic activities were calculated. The protein content of small-intestine tissue was measured using bovine serum albumin as standard.

Data Analysis

The possible significant differences among populations in climatic, temperature, rainfall, and natural diet variability were evaluated by Levene's test for the equality of variances with *P* values adjusted according to Bonferroni's method for multiple comparisons (*P* < 0.01). The effect of the diet acclimation on the digestive enzyme activity and the digestive morphology of *Zonotrichia capensis* from different sites were evaluated by using two MANCOVAs. One analysis considered the digestive enzymes aminopeptidase-N, maltase, and sucrase; the other focused on digestive morphology including the small-intestine length/mass and gizzard mass as variables. The body mass of individuals was used as covariate in both analyses. Because the protein content in the homogenate was not statistically different among diets, data of enzyme activities were analyzed as total hydrolytic activity (µmol min⁻¹). Before the analysis, the assumptions of homoscedasticity (by Levene's test), normality, and parallelism were tested. The response variables and the covariate were transformed using log(*x* + 1). The results obtained from the univariate contrasts of variance are also shown in Table 2.

The effect of diet acclimation—namely, the magnitude of the digestive flexibility—was compared among populations by a posteriori estimation of the standardized effect size (SES). SES was estimated using the unbiased estimator of Hedge (*g*) and its asymptotic SE (Kline 2004; Kampenes et al. 2007; Bozinovic et al. 2010). The estimation of SES was obtained by using the adjusted least square mean from MANCOVA and the MANCOVA residual error as variance estimator. The overlap level among asymptotic SEs of populations was used to contrast the magnitude of digestive flexibility. All analyses were carried out using the Statistica 7.0 (StatSoft 2004) statistical package for the Windows operating system.

Results

The greatest variability in rainfall was recorded at medium latitude in the Mediterranean site (Levene's test, *P* < 0.01; Table 1) followed by the cold-temperate Llanquihue and the desert Copiapó (Table 1). The environmental variability in temperature was similar among populations (Levene's test, *P* > 0.01; Table 1).

Regarding variation in the natural diet of birds, significant differences were found in δ¹⁵N between seasons in the Mediterranean site (*t*₁₉ = -2.60, *P* = 0.01) but not in the desert (*t*₁₄ = 1.06, *P* = 0.3) or in the cold-temperate site (*t*₁₄ = -0.96, *P* = 0.35). The coefficient of variation of δ¹⁵N was 5 for the desert, 22 for the Mediterranean, and 13 for the cold-temperate population. Also, the Mediterranean site was the most variable in diet composition, indicated by δ¹⁵N variation (Levene's test, *P* < 0.01) followed by the cold-temperate and the desert site.

Diet acclimation experiments significantly affected final body mass in two localities (Table 2). Sparrows from the desert and cold-temperate sites acclimated to an insect-based diet tended to have larger body mass than those acclimated to a seed-based

Table 2: Univariate results for a one-way MANCOVA analyzing the effect of diet acclimation on digestive-tract characteristics in rufous-collared sparrows from different localities

Habitats and Diet Acclimation	Digestive Enzyme Activities				Digestive Morphology			
	Body Mass (g)	Aminopeptidase-N ($\mu\text{mol min}^{-1}$)	Maltase ($\mu\text{mol min}^{-1}$)	Sucrase ($\mu\text{mol min}^{-1}$)	Small Intestine Mass (g)	Small Intestine Length (cm)	Gizzard Mass (g)	
Desert:								
Seeds	14.02 \pm .47 ^A	1.53 \pm .21 ^A	5.42 \pm 1.54 ^A	.47 \pm .13 ^A	.38 \pm .06 ^A	12.99 \pm .79 ^A	.61 \pm .16 ^A	
Insects	18.35 \pm .50 ^B	2.42 \pm .36 ^A	19.5 \pm 1.34 ^A	2.7 \pm .42 ^A	.50 \pm .08 ^A	11.95 \pm .77 ^A	.57 \pm .05 ^A	
Mediterranean:								
Seeds	14.92 \pm .75 ^A	.95 \pm .17 ^A	5.74 \pm 1.41 ^A	.7 \pm .26 ^A	.29 \pm .09 ^A	13.14 \pm 1.31 ^A	.54 \pm .10 ^A	
Insects	16.03 \pm 1.17 ^A	2.39 \pm .21 ^B	10.75 \pm 2.84 ^A	.74 \pm .22 ^A	.44 \pm .03 ^B	13.87 \pm 2.22 ^A	.51 \pm .14 ^A	
Cold-temperate:								
Seeds	14.08 \pm .39 ^A	.81 \pm .07 ^A	4.34 \pm .69 ^A	.4 \pm .04 ^A	.40 \pm .07 ^A	14.09 \pm 1.15 ^A	.44 \pm .04 ^A	
Insects	17.77 \pm .75 ^B	2.7 \pm .55 ^B	9.73 \pm 3.82 ^A	.98 \pm .44 ^B	.53 \pm .14 ^A	15.16 \pm 1.57 ^A	.62 \pm .18 ^A	

Note. Body mass, digestive enzyme activities (aminopeptidase-N, maltase, and sucrase), and digestive morphology (small-intestine mass/length and gizzard mass) are reported as mean \pm SD. Different superscript letters denote significant differences ($P < 0.05$) after a univariate test.

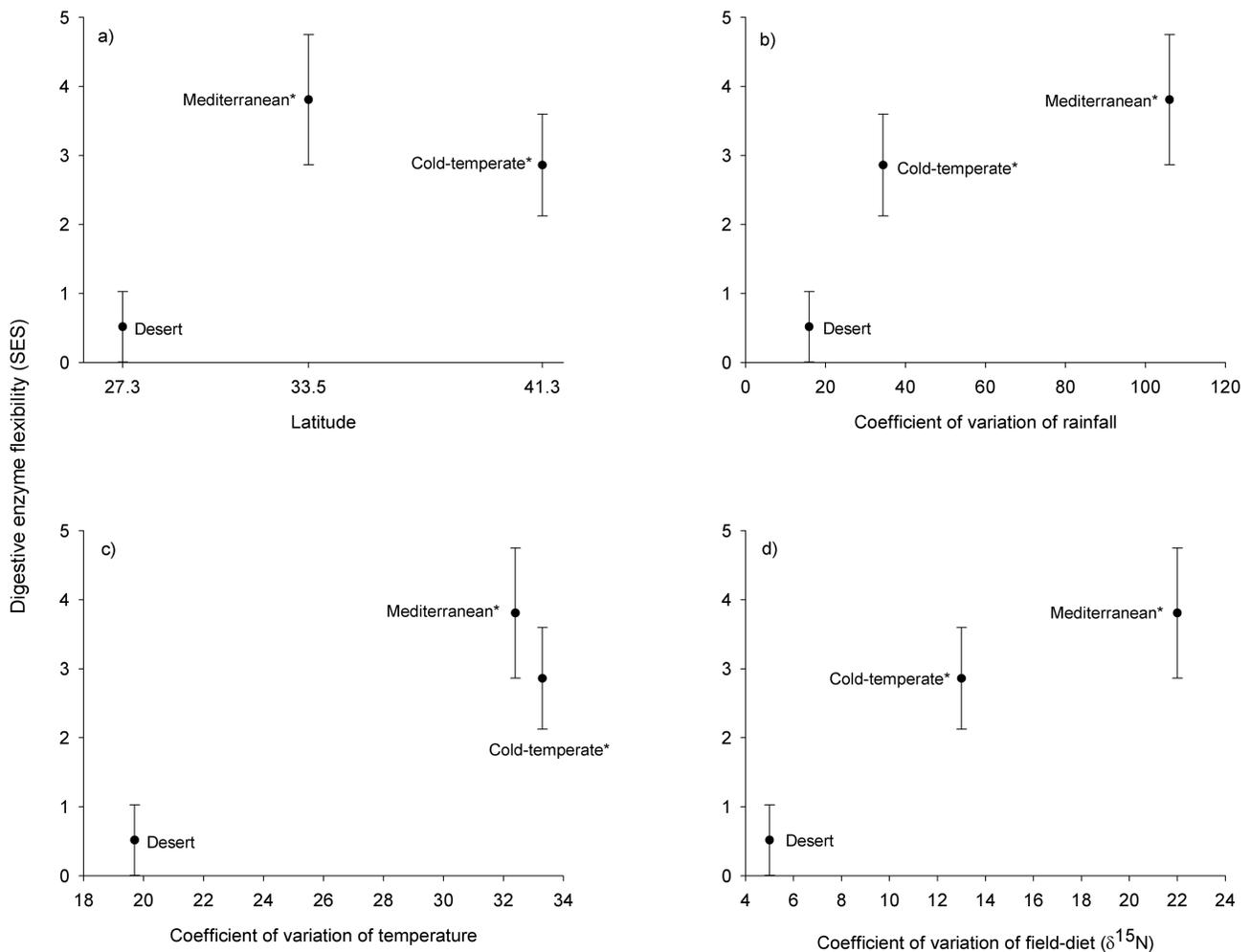


Figure 1. Effect of dietary acclimation on intestinal enzyme activities among three populations of *Zonotrichia capensis* along a latitudinal gradient in Chile. The standardized effect size (SES) or magnitude of digestive enzymatic flexibility is indicated as a function of the coefficient of variation of (a) rainfall, (b) temperature, and (c) natural diet. Error bars represent the asymptotic SE calculated for the effect size. Asterisks denote populations that showed significant differences between diets revealed by MANCOVA ($P < 0.05$).

diet (Table 2). The results of MANCOVA testing for differences in digestive enzyme activities between the diets showed that desert-dwelling birds did not change their overall enzyme activities in response to diet acclimation (Wilks's $\lambda = 0.76$, $F_{3,11} = 1.16$, $P = 0.37$). However, in Mediterranean and cold-temperate populations, changes were seen for some digestive enzymes (Wilks's $\lambda = 0.15$, $F_{3,9} = 16.4$, $P = 0.001$; Wilks's $\lambda = 0.25$, $F_{3,11} = 11.2$, $P = 0.001$ for Mediterranean and cold-temperate sites, respectively). The univariate test revealed that birds from the Mediterranean site significantly increased their aminopeptidase-N activity as a result of an insect-based diet acclimation; in turn, individuals from the cold-temperate site increased their aminopeptidase-N and sucrase activities (Table 2). Moreover, the magnitude of the overall digestive enzyme flexibility (SES) was 3.81, 2.86, and 0.52 for birds from the Mediterranean, cold-temperate, and desert populations, respectively. In turn, neither latitude nor temperature variability

appear to be associated with the phenotypic flexibility of digestive enzyme activities (Fig. 1a, 1c), while the coefficient of variation of both rainfalls and natural diet was associated (Fig. 1b, 1d).

Regarding changes in digestive organ size, MANCOVA tests revealed a significant diet effect on overall digestive morphology only in birds from the Mediterranean site (Wilks's $\lambda = 0.31$, $F_{3,9} = 6.77$, $P = 0.01$). Birds from the desert and cold-temperate populations did not change their digestive morphology in response to dietary acclimation. For the population inhabiting the Mediterranean site, the univariate analysis indicated that birds fed on an insect-based diet had heavier ($F_{3,9} = 17.5$, $P = 0.002$) and longer ($F_{3,9} = 7.44$, $P = 0.02$) small intestines than those fed on a seed-based diet. In addition, the magnitude of the overall phenotypic flexibility (SES) in digestive morphology followed the same pattern as digestive enzyme activities flexibility, with values of 2.35, 1.09, and 0.81 for birds

from the Mediterranean, cold-temperate, and desert sites, respectively. As with digestive enzyme activities, neither latitude nor temperature variability appears to be related to the flexibility of digestive morphology (Fig. 2a, 2c) in contrast to precipitation and natural diet (Fig. 2b, 2d). In summary, birds at the middle latitude inhabiting a highly seasonal Mediterranean site exhibited greater digestive-tract flexibility than those from desert and cold-temperate climates.

Discussion

Contrary to the predictions of CVH, we did not find an association between the magnitude of digestive flexibility and latitude at population level (Figs. 1a, 2a). That is, the greatest digestive-tract flexibility was observed in birds at middle latitudes, which also experienced the highest environmental variability.

From an evolutionary point of view, species or populations from temporally heterogeneous environments may exhibit a more flexible reaction norm (Schlichting and Pigliucci 1998; Pigliucci 2005). This seems to be the case in *Zonotrichia capensis*, because populations from the site with greatest seasonality in rainfall and natural diet also proved to be the most flexible for morphological and enzyme activities (Figs. 1b, 1d, 2b, 2d). By contrast, patterns associated with temperature variability were less clear; the bird population subject to the maximum coefficient of variation of temperature did not show the largest value of digestive-tract flexibility (Figs. 1c, 2c). This may be because temperature variability among populations is not different enough and not statistically significant (Table 1) to account for this relationship.

As mentioned in the “Introduction,” latitude seems to be a better predictor of the phenotypic flexibility than temperature variability (Naya et al. 2008). In this intraspecific study, although both the variability in rainfall and natural diet were not

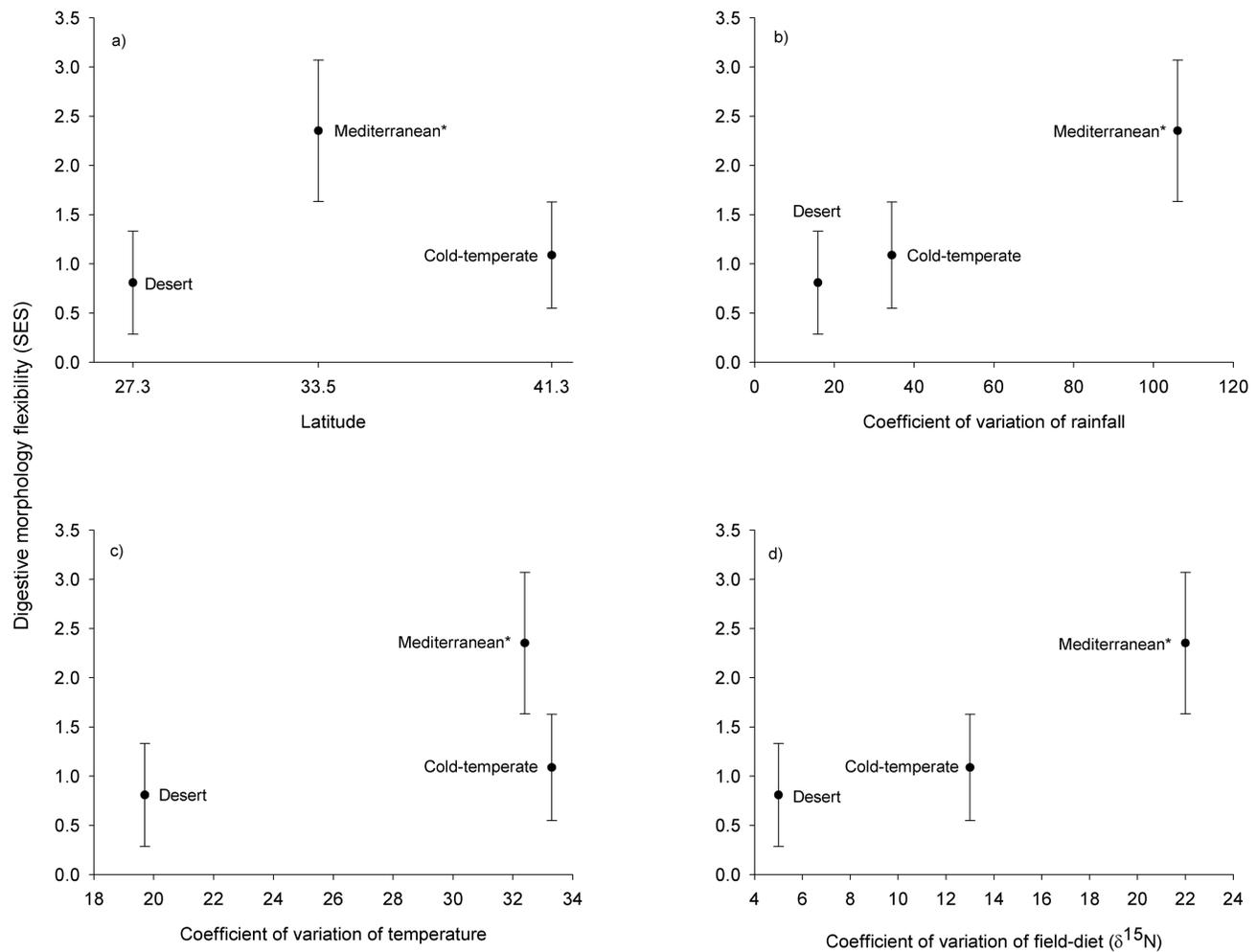


Figure 2. Relationship between the magnitude of digestive morphology flexibility and the coefficient of variation of (a) rainfall, (b) temperature, and (c) natural diet. See Figure 1 for details. Asterisks denote populations that showed significant differences between diets revealed by MANCOVA ($P < 0.05$).

correlated with latitude, they appear to be better predictors of digestive flexibility than latitude or even temperature variability. In general, recent studies testing CVH have used only temperature as the explanatory variable for the animals' physiological tolerance (e.g., Gaston and Chown 1999*b*; Addo-Bediako et al. 2000; Pither 2003; Naya et al. 2008). Nonetheless, a number of studies have shown that there are environmental factors more highly correlated with range size that are not linked to latitude (Hawkins and Diniz-Filho 2006). The limited studies considering the role of other environmental factors and other physiological traits than those related to thermal tolerance emphasize the need for additional effort to incorporate possible alternative variables to provide a better understanding of the proximal factors underlying the CVH. In addition, our findings support the idea that latitudinal gradients in geographical ranges may be strongly affected by the action of regional features (Gaston and Chown 1999*a*; Fortes and Absalão 2010). Consequently, different species that share the same localities might be more similar in their physiological tolerance than expected from the latitudinal ranges of their own species, perhaps as an adaptive response to local conditions. This fact makes it difficult to find a general pattern in the distribution of species. Thus, we stress the importance of considering the existence of interpopulation differences in future studies of the CVH.

Finally, we note that the effect of climatic variability on the size of a species' range through its effect on individual phenotypic flexibility may be critical in the context of global climate change. One of the central tenets of ecology and biogeography is that climate influences the geographic range of species (Gaston and Blackburn 2000). Indeed, global climate change has been shown to affect the distribution of many species, becoming one of the greatest threats to biodiversity. Many studies have shown that the average global temperature—a popular metric for summarizing the state of global climate—has increased by 0.7°C over the past century, and future projections show a substantial acceleration (Walther et al. 2002). Nevertheless, theoretical models as well as empirical data (Houghton et al. 2001) have concluded that climate change also affects climatic variability. Most current studies and analyses of global change have focused on mean values, paying less attention to the role of fluctuations in the climatic variables (but see Terblanche et al. 2010). According to our results, experimental studies testing not only the effect of changes in means but also in climatic variability on phenotypic traits may be relevant at different evolutionary, ecological, and physiological scales (Travis and Futuyma 1993). Future comparative studies focused on the sensitivity of populations to climatic variability may reveal important effects on biodiversity that have perhaps been underestimated in the context of global climate change.

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Literature Cited

- Addo-Bediako A., S.L. Chown, and K.J. Gaston. 2000. Thermal tolerance, climatic variability and latitude. *Proc R Soc B* 267: 739–745.
- Araya M.B. and H.G. Millie. 2005. *Guía de Campo de las Aves de Chile*. Editorial Universitaria, Santiago.
- Bauchinger U. and S. McWilliams. 2009. Carbon turnover in tissues of a passerine bird: allometry, isotopic clocks, and phenotypic flexibility in organ size. *Physiol Biochem Zool* 82:787–797.
- Bozinovic F., J.M. Rojas, K. Maldonado, P. Sabat, and D.E. Naya. 2010. Between-population differences in digestive flexibility in the olivaceous field mouse. *Zoology* 113:373–377.
- Calosi P., D.T. Bilton, J.I. Spicer, S.C. Votier, and A. Atfield. 2010. What determines a species' geographical range? thermal biology and latitudinal range size relationships in European diving beetles (Coleoptera: Dytiscidae). *J Anim Ecol* 79:194–204.
- Campbell B. and E. Lack. 1985. *A Dictionary of Birds*. Poyser, Calton, UK.
- Carleton S.A. and C. Martínez del Rio. 2005. The effect of cold-induced increased metabolic rate on the rate of ¹³C and ¹⁵N incorporation in house sparrows (*Passer domesticus*). *Oecologia* 144:226–232.
- Chapman F.M. 1940. The post-glacial history of *Zonotrichia capensis*. *Bull Am Mus Nat Hist* 77:381–438.
- Dahlqvist A. 1964. Method for assay of intestinal disaccharidases. *Anal Biochem* 7:18–25.
- Davis J. 1971. Breeding and molt schedules of the rufous-collared sparrow in coastal Perú. *Condor* 73:127–146.
- Fortes R.R. and R.S. Absalão. 2010. The latitudinal and bathymetric ranges of marine fishes: a global analysis to test the application of Rapoport's rule. *Mar Ecol* 31:483–493.
- Gaston K.J. and T.M. Blackburn. 2000. *Pattern and Process in Macroecology*. Blackwell Science, Oxford.
- Gaston K.J. and S.L. Chown. 1999*a*. Elevation and climatic tolerance: a test using dung beetles. *Oikos* 86:584–590.
- . 1999*b*. Why Rapoport's rule does not generalize. *Oikos* 84:309–312.
- Ghalambor C.K., J.K. McKay, S.P. Carroll, and D.N. Reznick. 2007. Adaptive versus non-adaptive phenotypic plasticity and the potential for contemporary adaptation in new environments. *Funct Ecol* 21:394–407.
- Goodall J.D., A.W. Johnson, and R.A. Philippi. 1951. *Las Aves*

- de Chile, su Conocimiento y sus Costumbres. Platt establecimientos gráficos, Buenos Aires.
- Hawkins B.A. and J.A.F. Diniz-Filho. 2006. Beyond Rapoport's rule: evaluating range size patterns of New World birds in a two-dimensional framework. *Glob Ecol Biogeogr* 15:461–469.
- Hobson K.A. 1999. Stable-carbon and nitrogen isotope ratios of songbird feathers grown in two terrestrial biomes: implications for evaluating trophic relationships and breeding origins. *Condor* 101:799–805.
- Hobson K.A. and R.G. Clark. 1992a. Assessing avian diets using stable isotopes. I. Turnover of ^{13}C in tissues. *Condor* 94:181–188.
- . 1992b. Assessing avian diets using stable isotopes. II. Factors influencing diet-tissue fractionation. *Condor* 94:189–197.
- Houghton J.T., Y. Ding, D.J. Griggs, M. Noguer, P.J. van der Linden, and D. Xiaosu. 2001. *Climate Change 2001: The Scientific Basis: Contributions of Working Group I to the Third Assessment Report of the Intergovernmental Panel on Climate Change*. Cambridge University Press, Cambridge.
- Kampenes V.B., T. Dybå, J.E. Hannay, and D.I.K. Sjøberg. 2007. A systematic review of effect size in software engineering experiments. *Inf Softw Technol* 49:1073–1086.
- Karasov W.H. 1996. Digestive plasticity in avian energetics and feeding ecology. Pp. 61–84 in C. Carey, ed. *Avian Energetics and Nutritional Ecology*. Chapman & Hall, New York.
- King J.R. 1973. The annual cycle of the rufous-collared sparrow *Zonotrichia capensis* in three biotopes in north-western Argentina. *J Zool (Lond)* 170:163–188.
- . 1974. Notes on geographical variation and the annual cycle in Patagonian populations of the rufous-collared sparrow *Zonotrichia capensis*. *Ibis* 116:74–83.
- Kline R.B. 2004. *Beyond Significance Testing*. American Psychological Association, Washington, DC.
- López-Calleja M.V. 1995. Dieta de *Zonotrichia capensis* (Emberizidae) y *Diuca diuca* (Fringillidae): efecto de la variación estacional de los recursos tróficos y la riqueza de aves granívoras en Chile Central. *Rev Chil Hist Nat* 68:321–331.
- Martínez del Rio C. 1990. Dietary, phylogenetic, and ecological correlates of intestinal sucrase and maltase activity in birds. *Physiol Zool* 63:987–1011.
- Martínez del Rio C., K.E. Brugger, J.L. Rios, M.E. Vergara, and M. Witmer. 1995. An experimental and comparative study of dietary modulation of intestinal enzymes in European starlings (*Sturnus vulgaris*). *Physiol Zool* 68:490–511.
- Moore I.T., H. Wada, N. Perfito, D.S. Busch, T.P. Hahn, and J.C. Wingfield. 2004. Territoriality and testosterone in an equatorial population of rufous-collared sparrows, *Zonotrichia capensis*. *Anim Behav* 67:411–420.
- Naya D.E., F. Bozinovic, and W.H. Karasov. 2008. Latitudinal trends in digestive flexibility: testing the climatic variability hypothesis with data on the intestinal length of rodents. *Am Nat* 172:E122–E134.
- Newsome S., C. Martínez del Rio, D.L. Phillips, and S. Bearhop. 2007. A niche for isotopic ecology. *Front Ecol Environ* 5: 429–436.
- Pigliucci M. 2005. Evolution of phenotypic plasticity: where are we going now? *Trends Ecol Evol* 20:481–486.
- Pither J. 2003. Climate tolerance and interspecific variation in geographic range size. *Proc R Soc B* 270:475–481.
- Post D.M. 2002. Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology* 83:703–718.
- Ramirez-Otarola N., C. Narváez, and P. Sabat. 2011. Membrane-bound intestinal enzymes of passerine birds: dietary and phylogenetic correlates. *J Comp Physiol B*, doi: 10.1007/s00360-011-0557-3.
- Sabat P., S. Gonzalez-Vejares, and K. Maldonado. 2009. Diet and habitat aridity affect osmoregulatory physiology: an intraspecific field study along environmental gradients in the rufous-collared sparrow. *Comp Biochem Physiol A* 152:322–326.
- Sabat P., F. Novoa, F. Bozinovic, and C. Martínez del Rio. 1998. Dietary flexibility and intestinal plasticity in birds: a field and laboratory study. *Physiol Zool* 71:226–236.
- Schlichting C.D. and M. Pigliucci. 1998. *Phenotypic Evolution*. Sinauer, Sunderland, MA.
- Stevens G.C. 1989. The latitudinal gradient in geographical range: how so many species coexist in the tropics. *Am Nat* 133:240–256.
- Sunday J.M., A.E. Bates, and N.K. Dulvy. 2011. Global analysis of thermal tolerance and latitude in ectotherms. *Proc R Soc B* 278:1823–1830, doi:10.1098/rspb.2010.1295.
- Terblanche J.S., C. Nyamukondiwa, and E. Kleynhans. 2010. Thermal variability alters climatic stress resistance and plastic responses in a globally invasive pest, the Mediterranean fruit fly (*Ceratitidis capitata*). *Entomol Exp Appl* 137:304–315.
- Travis J. and D.J. Futuyma. 1993. Global change: lessons from and for evolutionary biology. Pp. 251–263 in P.M. Kareiva, J.G. Kingsolver, and R.B. Huey, eds. *Biotic Interactions and Global Change*. Sinauer, Sunderland, MA.
- Via S., R. Gomulkiewicz, G. De Jong, S.M. Scheiner, C.D. Schlichting, and P.H. Van Tienderen. 1995. Adaptive phenotypic plasticity: consensus and controversy. *Trends Ecol Evol* 10:212–217.
- Walther G.R., E. Post, P. Convey, A. Menzel, C. Parmesan, T.J.C. Beebee, J.M. Fromentin, O. Hoegh-Guldberg, and F. Bairlein. 2002. Ecological responses to recent climate change. *Nature* 416:389–395.