

Supplementary Material

Climatic stability and genetic divergence in the tropical insular lizard *Anolis krugi*,
the Puerto Rican “Lagartijo Jardinero de la Montaña”

Javier A. Rodríguez-Robles, Tereza Jezkova, and Manuel Leal

Molecular Ecology 19(9):1860–1876 (2010)

doi: 10.1111/j.1365-294X.2010.04616.x

Supplementary Material, Table S2. Redundant and contracted haplotypes of *Anolis krugi*. The first column indicates all the haplotypes that were redundant with other mitochondrial types (second column) and/or that represent an ancestral haplotype to which one or more other haplotypes were contracted using the star contraction method (see Materials and Methods). Haplotypes in bold appear on the maximum likelihood tree (Fig. 2). The third column indicates the satellite haplotypes that were contracted to the ancestral mitochondrial type listed on the first column.

Haplotype	Redundant haplotypes	Contracted haplotypes
AB 2-7	SL 45-160	—
AB 2-8	Ca 9-34, Ju 26-86, Mb 31-111, Mb 31-112, Mb 31-113, SL 45-165, Yb 52-193	AB 1-1, AB 1-2, AB 1-4, AB 1-6, AB 2-9, AB 2-10, AB 2-11, AB 2-12, Cd 11-36, Cg 12-42, Ju 26-87, Ju 26-89, Ju 26-90, Ju 26-91, Ju 26-92, Mb 31-114, Mb 31-118, SJ 44-154, SJ 44-155, SJ 44-157, SL 45-158, SL 45-159, SL 45-161, Yb 52-192, Yb 52-196
Ad 3-14	Ad 3-18	—

Supplementary Material, Table S2. Continued.

Haplotype	Redundant haplotypes	Contracted haplotypes
Ad 3-15	Ad 3-20	—
Ca 8-29	SL 45-163, Yb 52-201	—
Cd 11-39	—	Cd 11-37, Cd 11-40
Co 15-57	Fl 17-60, Fl 18-62	Ba 7-27, Ci 13-43, Ci 13-46, Ci 13-47, Co 15-58, JD 25-81, JD 25-82, Or 40-146, Or 41-147
Gu 21-68	Gu 21-70	—
Is 23-73	Is 24-77	—
Is 24-75	Is 24-79	—
La 27-94	La 27-95, La 27-96, La 27-97, La 27-98, LM 28-101, LM 28-103, LM 28-106, LM 28-107	LM 28-102, LM 28-104, Ma 30-110, SS 46-167, SS 46-168, SS 47-171, Ut 49-177, Ut 49-178, Ut 50-181, Ut 50-182, Ut 50-183

Supplementary Material, Table S2. Continued.

Haplotype	Redundant haplotypes	Contracted haplotypes
La 27-99	La 27-100	—
Mb 31-114	—	Mb 31-115
Mb 31-117	Yb 52-200	—
Mo 32-120	Mo 32-121, Mo 32-123, Mo 32-126	—
Mo 32-127	Mo 32-128	—
SJ 44-154	SJ 44-156	—
SL 45-158	SL 45-164	—
SS 46-167	—	La 27-93, La 27-99
VA 51-185	VA 51-189	—
VA 51-188	VA 51-190	—
Yb 52-196	—	Ju 26-88

Supplementary Material, Table S2. Continued.

Haplotype	Redundant haplotypes	Contracted haplotypes
Yc 53-202	Yc 53-203, Yc 53-204	—
Vector 1	—	Cd 11-38, Cg 12-41, Yb 52-194
Vector 2	—	Na 39-141, TA 48-174, VA 51-185
Vector 3	—	Ci 13-45, Co 15-55
Vector 4	—	Co 15-52, Co 15-53, Fl 16-59, VA 51-186, VA 51-187
Vector 5	—	Is 24-74, Is 24-75, Is 24-76, Is 24-78, Mo 32-119, Mo 32-120, Mo 32-122, Mo 32-124, Mo 32-125, Mo 32-130, SS 46-169
Vector 6	—	Ad 3-14, Ad 3-21, Gu 21-69, Gu 21-71, Yc 54-206, Yc 54-211
Vector 7	—	VA 51-188, VA 51-191

Effect of sample size on the performance of SAMOVA

We explored patterns of genetic divergence among sampling localities of *Anolis krugi* using the program SAMOVA 1.0 (Spatial Analysis of Molecular Variance; Dupanloup *et al.* 2002; <http://web.unife.it/progetti/genetica/Isabelle/samova.html>). Because SAMOVA uses F_{ST} statistics (which measure genetic differentiation among subpopulations), its performance is negatively affected by small sample sizes (i.e. small number of individuals per population; I. Dupanloup, pers. comm.). To determine how small sample size affects the performance of SAMOVA, we performed a sensitivity study by conducting the analyses five times, with minimum sample sizes per population of 4, 5, 6, 7, and 8 individuals.

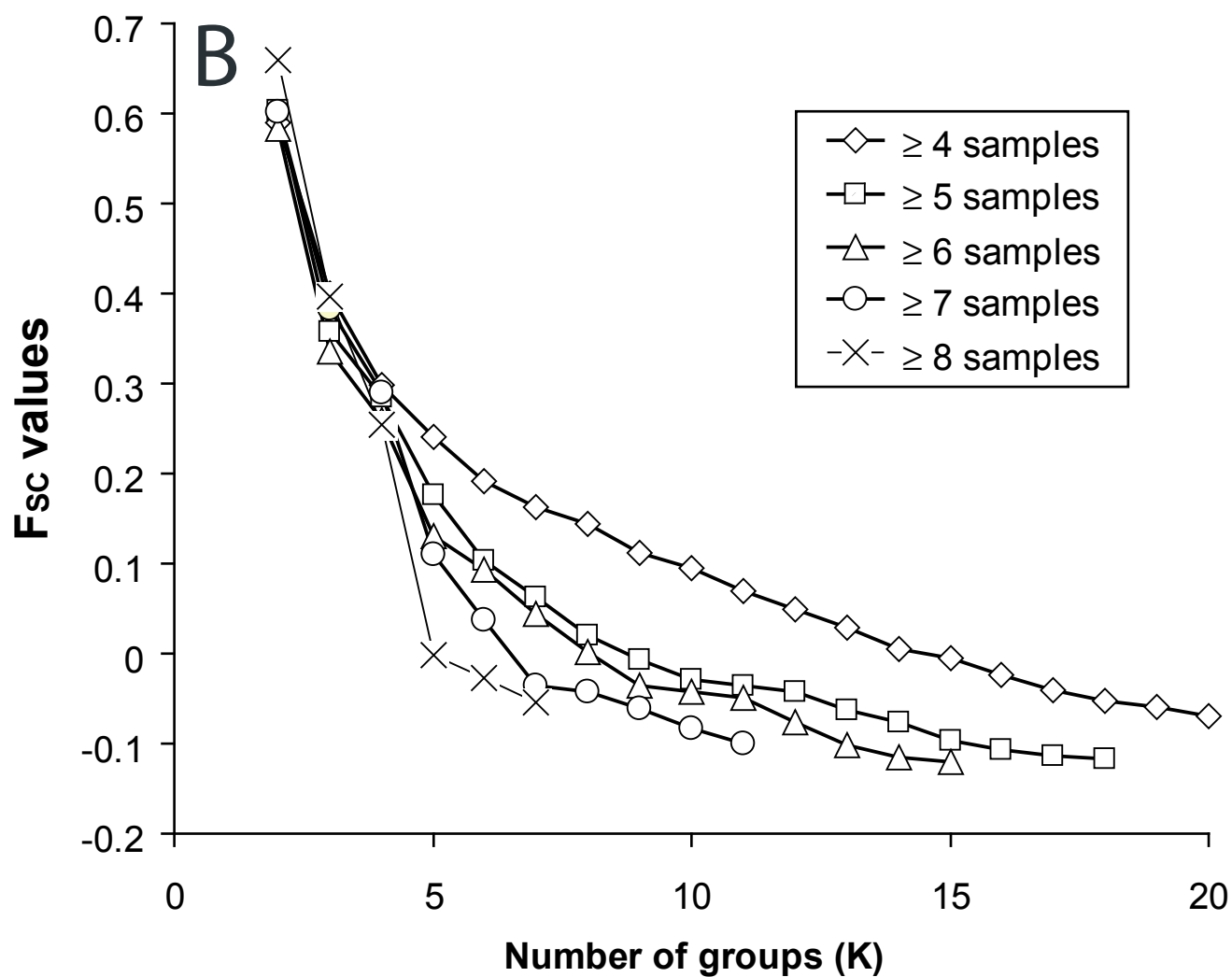
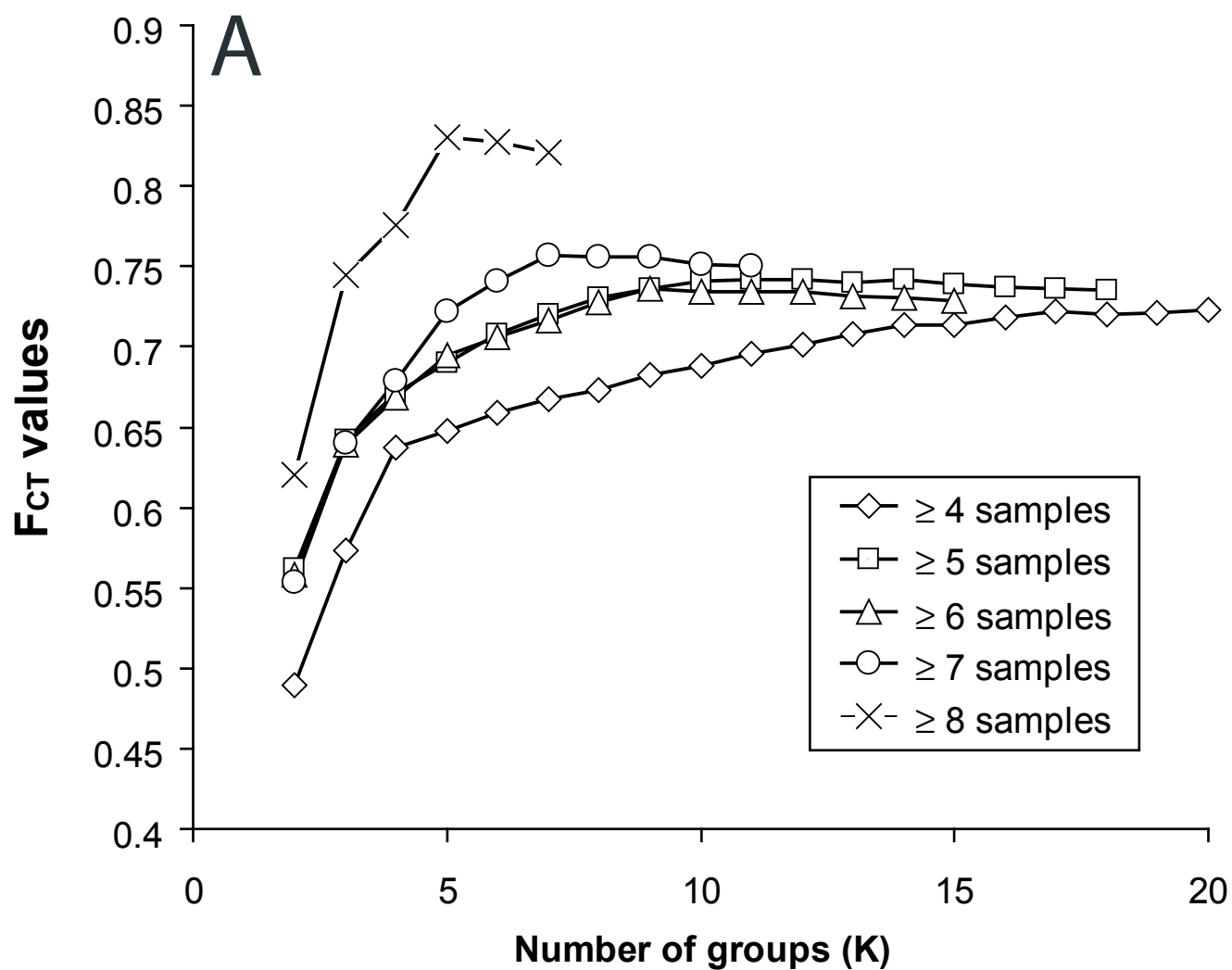
Figure S1 illustrates the effect of minimum number of samples per locality on F_{CT} and F_{SC} values. For each value of K , a smaller minimum sample size (n) resulted in lower F_{CT} values and higher F_{SC} values, whereas larger minimum sample sizes resulted in higher F_{CT} and lower F_{SC} values. We anticipated these results, because with increasing minimum sample sizes, more sampling localities (i.e. populations) are excluded from the analysis, and consequently the geographic and (often) the genetic distances among the remaining populations increase. Also as expected, the number of groups (K) at which F_{CT} values (degree of differentiation among groups) peaked approximately corresponded to F_{SC} values of zero.

The shapes of the curves of the F_{CT} and F_{SC} plots for five, six, and seven samples per locality were similar (Fig. S1). We determined by visual inspection of these plots that five individuals per population was a threshold that allowed us to present conservative results, without having to exclude an excessive number of populations. We emphasize that this threshold should not be extrapolated to other datasets. Instead, we recommend that when genetic surveys include populations with a

relatively small number of individuals, researchers empirically assess how F_{CT} and F_{SC} values vary as a function of sample size, and rely on those findings to determine the threshold that will allow them to include in the SAMOVA analyses as many populations as possible, without sacrificing too much accuracy.

Additionally, in this study we used F_{SC} values (indicators of genetic differentiation among populations within groups) as an alternative to F_{CT} values (indicators of differentiation among groups) to identify the number of maximally differentiated groups of populations of *Anolis krugi*. Traditionally, researchers have used F_{CT} values as indicators of maximum differentiation, with the number of maximally differentiated groups corresponding to the highest F_{CT} value. However, it is often difficult to determine precisely when F_{CT} values peak. Our analyses suggest that F_{SC} values can be used to identify more objectively the number of maximally differentiated groups in a species. The F_{SC} values approach zero as the genetic variation within populations approximates the degree of variation within groups. When F_{SC} values become negative, the variation within populations is larger than the variation within groups (which can occur when populations within a group share haplotypes (e.g. the Eastern phylogroup) or when the populations that compose a group have haplotypes belonging to two or more divergent clusters). Consequently, when F_{SC} values equal zero, additional splitting of populations is undesirable, and therefore the number of maximally differentiated populations has been identified.

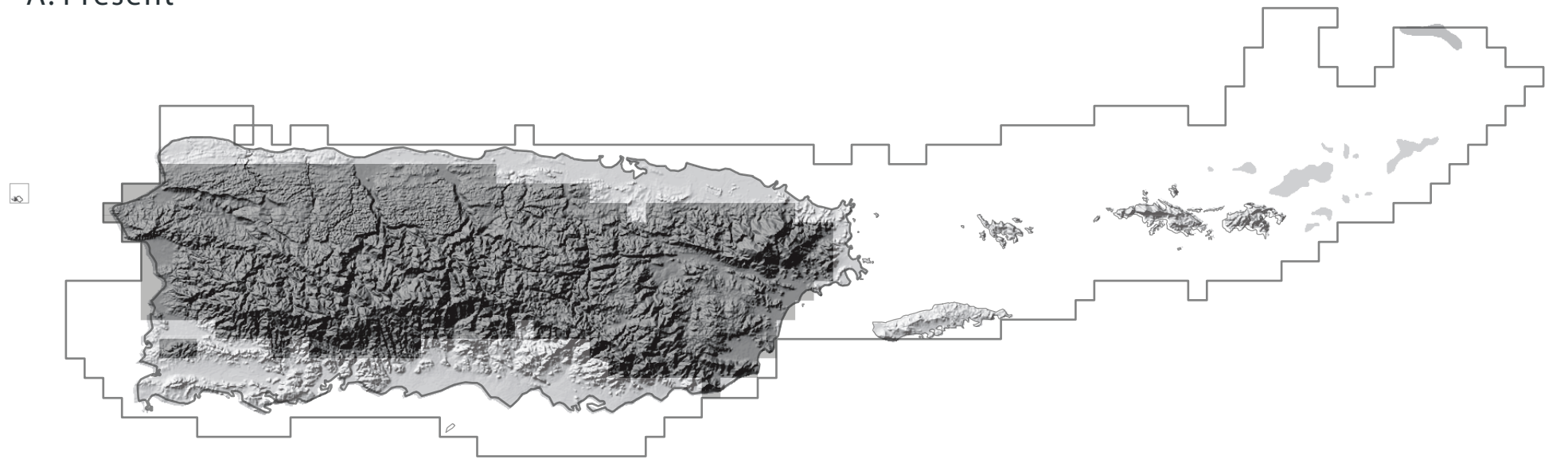
Supplementary Material, Figure S1. Effect of sample size (i.e. minimum number of individuals per population) on (A) F_{CT} values (indicators of the proportion of total genetic variance due to differences among groups of populations), and on (B) F_{SC} values (indicators of differentiation among populations within groups).



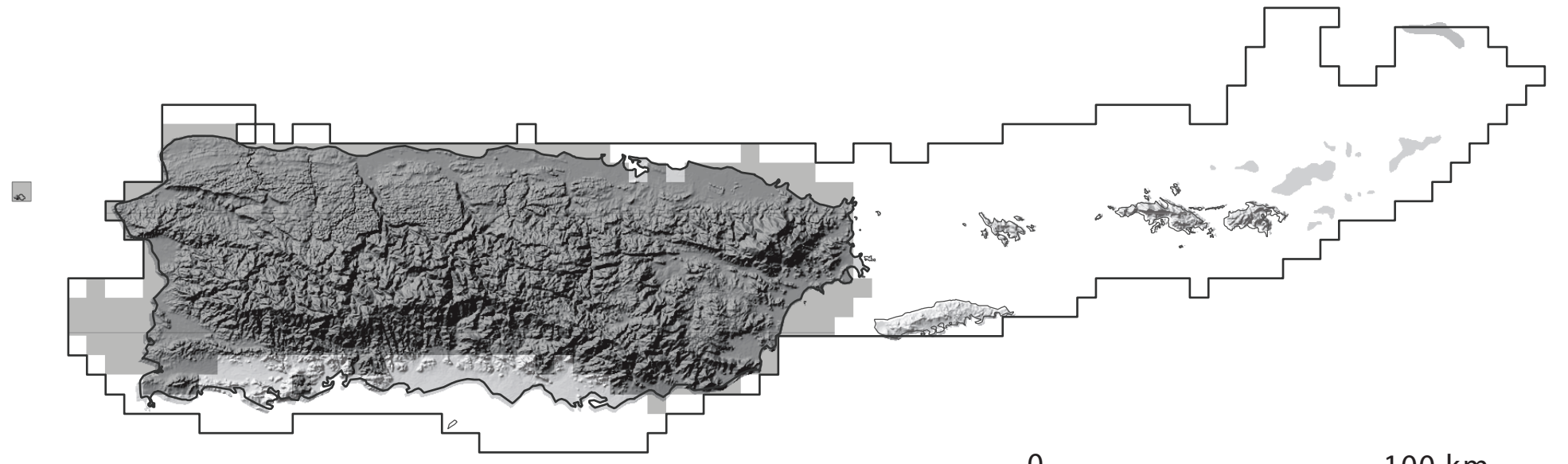
Supplementary Material, Figure S2. Ecological niche models for *Anolis krugi* for the (A) current climatic conditions (30 sec resolution) in Puerto Rico, and for the climatic conditions during the last glacial maximum (2.5 min resolution) estimated using the (B) CCSM and (C) MIROC models. The current shoreline of Puerto Rico and adjacent islands is depicted in light grey, whereas the approximate shoreline of the Puerto Rican Bank during the last glacial maximum (adjusted to the resolution of 2.5 min) is shown in black. The shading represents habitat modeled as suitable for *A. krugi*.



A. Present



B. Last Glacial Maximum (CCSM)



C. Last Glacial Maximum (MIROC)

