

EVOLUTIONARY GENETICS OF INSULAR ADRIATIC LIZARDS

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The use of electrophoresis to study allozyme variation allows the evolutionary biologist to compare various genetic parameters of populations. In this paper we employ this technique in an analysis of the evolutionary genetics of the genus *Lacerta* in the Adriatic region. We compare genetic similarity among populations and species to obtain a quantitative estimate of relatedness. We compare genetic variability on islands and on the mainland to examine genetic consequences of insularity. We examine the relative contributions of island size, habitat complexity, and various measures of isolation to the determination of the observed levels of heterozygosity. Finally, we attempt to estimate the relative importance of selection and genetic drift, and apply our findings to the question of allelic neutrality.

The Adriatic waters of Yugoslavia are dotted with more than 1000 islands of diverse size and habitat complexity, ranging from rocks barely emergent from the sea to substantial islands measuring hundreds of square kilometers. Lacertid lizards are abundant on most of these islands and the adjacent coastal strip. They have interested evolutionists for two reasons: First, there is a classic case of competitive exclusion (Radovanović, 1956, 1959 cited in Mayr, 1963). Either *Lacerta melisellensis* or *L.*

sicula is found on virtually every island in the Adriatic. The pattern of distribution in places is completely interdigitating, but sympatry is completely absent on islands smaller than several square kilometers. On some large islands both species can be found, but there is usually parapatry or only marginal sympatry. Second, both *L. melisellensis* and *L. sicula* have undergone considerable evolution in size, color, and pattern. Thus Mertens and Wermuth (1960) list 18 subspecies for *Lacerta melisellensis*, and no fewer than 39 for *L. sicula*. The subspecies controversy aside, there is no doubt that some of the insular isolates are very distinctive. For example, there are populations of *L. melisellensis* comprised of large (adult males > 10 grams) jet-black lizards, others of small brown lizards (adult males ~ 5 grams) with a solid green middorsal area, and still others that are highly patterned. In addition to being polytypic, many populations are highly polymorphic for dorsal pattern and for ventral coloration (salmon, cream, yellow, and pale blue may exist in a single population).

Lacerta melisellensis and *L. sicula* appear to be of broad ecological tolerance occurring in a variety of terrestrial situations. They are most often collected in shrub and grassy areas, and also around small stones. A third species enters marginally into the present study. *L. oxycephala* is restricted to large boulders and rock faces on the main-

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land and on the islands. Unlike the other two species, it does not show inter-island variation, nor is it polymorphic within populations. No subspecies have been designated (Mertens and Wermuth, 1960). *L. oxycephala* is broadly sympatric with *L. melisellensis* in the southern portion of the latter's range, and has been found on at least one island in sympatry with *L. sicula* (Nevo et al., 1972). What we now call *L. melisellensis* and *L. sicula* have historically been considered closely related in conventional taxonomic terms (even conspecific), and *L. oxycephala* is considered distant from them (Boulenger, 1920). Recently, Arnold (1973) split the genus *Lacerta* into several genera, leaving *oxycephala* in *Lacerta* and placing *melisellensis* and *sicula* in *Podarcis*. For convenience only, we retain the older nomenclature.

MATERIALS AND METHODS

Species Studied

1) *Lacerta sicula*.—This species occurs in Italy and along the Adriatic coast of Yugoslavia. Its present distribution in northern Yugoslavia suggests that it has been a relatively recent invader, having come from north-eastern Italy around the Adriatic. Its occurrence on some south-central Adriatic islands, and its absence from the Yugoslav mainland at these latitudes suggests direct over-water colonization of these islands (Palagruža, Sušac, Kopište, Pod Kopište) from Italy. *L. sicula* is also present in southern Yugoslavia, its presence there is presumably the result of another over-water colonization. About one-third of the described subspecies are on islands in the eastern Adriatic.

2) *Lacerta melisellensis*.—This is an autochthonous species to the coastal regions of Yugoslavia. Of the 18 named subspecies, one is on mainland Yugoslavia, the remainder are all insular isolates.

3) *Lacerta oxycephala*.—This species is also restricted to Yugoslavia, occurring on the Dalmatian coast and islands.

Populations Sampled

1) *Lacerta sicula*.

a) *L. s. campestris*.—This is one of the more widely distributed races of *L. sicula*. Population samples were collected at Zadar ($n = 30$) and Trogir ($n = 30$), Yugoslavia; and Pescara ($n = 30$), Italy (all mainland). All collections were made within an area < 1 km sq.

b) *L. s. pelagosae*.—This form is endemic to Palagruža ($n = 16$).

c) *L. s. cazzae*.—Three island populations were sampled: Sušac ($n = 34$), Kopište ($n = 19$), and Pod Kopište ($n = 30$).

2) *Lacerta melisellensis*.—(islands only, we were unable to collect a mainland series).

a) *L. m. curzolensis*.—Collected at Žrnovo, island of Korčula ($n = 14$); within an area < 1 km sq.

b) *L. m. lissana*.—Collected at Pasadur, island of Lastovo ($n = 24$) and Pod Mrčaru ($n = 34$), a tiny island near Lastovo; the town of Komiža, island of Vis ($n = 17$), and Greben ($n = 30$), a small island adjacent to Vis; and Biševo ($n = 28$), a moderately large island west of Vis.

c) *L. m. gigas*.—This subspecies is endemic to the tiny island of Mali Paržanj, adjacent to the east coast of Vis ($n = 11$).

d) *L. m. kammereri*.—This subspecies is endemic to the tiny island of Mali Barjak, adjacent to the west coast of Vis ($n = 7$).

e) *L. m. digenea*.—This form is endemic to Svetac, west of Vis and Biševo ($n = 32$).

f) *L. m. melisellensis*.—The nominate race is endemic to Brusnik near Svetac ($n = 28$).

g) *L. m. galvagnii*.—A race endemic to Kamik near Svetac ($n = 20$).

h) *L. m. pomoensis*.—Endemic to the tiny isolated island of Jabuka west of Svetac ($n = 18$).

3) *Lacerta oxycephala*.—Only 7 individuals were studied, three from Lastovo and two each from Greben and Vis.

As we are not concerned with nomenclature at the subspecific level, the popu-

lations hereinafter are primarily referred to by locality only. Figures 1–4 show collecting sites, and Table 1 presents data on the localities.

Electrophoresis

Specimens were collected and transported alive to the laboratory where they were placed in a -68°C freezer. Before processing for electrophoresis, the head, feet, and skin were removed for morphological studies. After removal of the gut, the body was homogenized in two volumes of grinding buffer (0.1 M Tris, 0.001 M EDTA, and 5×10^{-5} M NADP, pH adjusted to 7.0 with concentrated HCl) and centrifuged with 0.5 ml of toluene at 18,000 r.p.m. for 30 minutes. The supernatant was removed and frozen at -68°C .

Starch gel electrophoresis was carried out with the same proteins and procedures described by McKinney et al. (1972) for 22 loci with the following exceptions: Albumin was not electrophoresed; 6-phosphogluconate dehydrogenase (6 Pgd) was demonstrated with a Tris maleate EDTA buffer, pH 7.4, and isocitrate dehydrogenase (Idh) with a Tris citrate buffer, pH 8.0. Fumarase (Fum) was demonstrated with a Poulik buffer system, pH 8.1, for the tray buffer, and pH 8.6 for the gel buffer.

Heterozygosity estimates are based on actual counts of presumed heterozygotic genotypes. Mean heterozygosity (\bar{H}) is defined as the number of presumed heterozygotic genotypes recorded in a sample divided by the product of the number of individuals and the number of loci surveyed. Polymorphism estimates are based on the number of loci with more than a single allele divided by the total number of loci. In this study a locus is considered polymorphic if any variation was found. In our analyses of genetic variation we primarily use \bar{H} as our dependent variable because it is less sample size dependent than percent polymorphism.

Genetic similarity between populations was estimated with the Rogers coefficient (S_R) (Rogers, 1972). Only 19 loci were

scorable for all populations of all species and S_R calculations are based only on these 19 loci.

RESULTS AND DISCUSSION

Allele frequency data for 20 populations (*oxycephala* treated as one population) are presented in Table 2, and levels of polymorphism and heterozygosity are presented in Table 1. Data on genetic similarity (S_R) appear in Table 3.

1) Taxonomic Comparisons

The similarity coefficients in Table 3 confirm the morphologically based taxonomy for the species level. Within-species similarities range from .84–1.0 among *L. melisellensis* populations and .88–.99 for *L. sicula*. Between species similarities for this pair show no overlap with the above, ranging from .61–.74. *L. oxycephala* is less closely related to *L. melisellensis* and *L. sicula* than the latter are to each other. The mean similarity value is .42 with *sicula* and .44 with *melisellensis*. This confirms the results of a preliminary study (Gorman, 1972).

These data are relevant to the general problem of how similar congeners can be genetically, and are relevant to the question of whether there need be a "genetic revolution" following speciation. Two races of Danish house mice (*Mus musculus*) have average S_R values between them of about 0.50 (Rogers, 1972) and disjunct populations of the lizard *Anolis carolinensis* from Texas and the Bahamas have a value of 0.68 (Webster et al., 1972). Within the relatively tightly-knit *roquet* species group of *Anolis*, between species S_R values are as high as 0.70 for sympatric congeners and as low as 0.37 for the least similar pair in the group (Soulé and Gorman, unpubl. data). Thus *Lacerta melisellensis* and *L. sicula* are more similar to one another than are so-called subspecies of certain mammalian and reptilian species, and even the "distantly related" *L. oxycephala* seems genetically closer to its congeners than are members

TABLE 1. *Biogeographic and electrophoretic data for the Lacerta populations.*

	Area ¹ (sq. km)	Elevation ¹ (m.)	Distance ¹ in km squared (source)	Channel ² depth	Mean heterozygosity	Standard error	Percent ³ loci polymorphic
<i>L. melisellensis</i>							
1) Korčula	279	568	2.89 (mainland)	36	7.14	(1.30)	23
2) Lastovo	53	417	900 (mainland)	66	7.73	(0.94)	32
3) Pod Mrčaru	0.02	16	0.64 (Mrčara); 5.76 (Lastovo)	(66)66	4.48	(0.77)	18
4) Vis	90	587	256 (Hvar)	89	6.44	(1.51)	32
5) Greben	0.06	32	1.0 (Vis)	(89)38	6.08	(0.67)	32
6) Mali Paržanj	0.008	5	0.81 (Vis)	(89)29	2.59	(0.99)	14
7) Mali Barjak	0.006	10	0.36 (Vis)	(89) ?	0.00	(0.00)	0
8) Biševo	5.8	240	16.81 (Vis)	97	3.74	(0.62)	23
9) Svetac	4.6	200	361 (Biševo)	~ 100	2.80	(0.56)	14
10) Brusnik	0.05	30	256 (Biševo); 12.25 (Svetac)	~ 100	3.40	(0.60)	14
11) Kamik	0.01	40	1.69 (Svetac)	~ 100	2.14	(0.54)	5
12) Jabuka	0.01	96	529 (Svetac)	144	0.26	(0.01)	0
<i>L. sicula</i>							
13) Zadar	8.41	(1.14)	27
14) Trogir	12.85	(1.05)	45
15) Pescara	5.88	(0.75)	37
16) Palagruža	0.3	91	2,601.0 (mainland)	~ 110	5.65	(0.89)	21
17) Sušac	4.6	243	9,801 (mainland); 1,764 (Palagruža)	~ 160	2.94	(0.49)	14
18) Kopište	1.0	93	196 (Sušac)	~ 130	4.76	(0.89)	18
19) Pod Kopište	0.05	30	0.36 (Kopište)	(~ 130)20	4.40	(0.46)	14
<i>L. oxycephala</i>							
20) Pooled sample	5.26	(1.55)	21

¹Data on island area, elevation, and distance obtained from *The Yugoslav Coast, Guide Book and Atlas*, Yugoslav Lexicographical Institute, Zagreb, 1966; and the following nautical charts: Jabuka-Vis, 1:100,000, #100-22; Sibenik-O. Lastovo, 1:200,000, #153; both published by Hidrografski Institut Jugoslavenske ratne mornarice, Split; and Promontorio del Gargano to Ortona, 1:167,520, #H.O. 3952, U.S. Navy Hydrographic Office. Elevations of Mali Paržanj and Mali Barjak are estimates, as they do not appear on the charts.

²Channel depths in meters based upon hydrographic charts listed above. The depths in parentheses are those of the parent island.

³Estimates of polymorphism rounded to nearest whole number.

TABLE 2. (Continued).

Locus and allele	Population: Code number, abbreviated name, and sample size																				
	1 Kor 14	2 Las 24	3 Pdm 34	4 Vis 17	5 Gre 36	6 Mlp 11	7 Mlb 7	8 Bis 28	9 Sve 32	10 Bru 28	11 Kam 20	12 Jab 18	13 Zad 30	14 Tro 30	15 Pes 30	16 Pala 30	17 Suš 30	18 Kop 34	19 Pdk 16	20 Oxy 7	
<i>Idh-2</i>	a	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	.86	
	b14
<i>Es-1</i>	a02
	b23	.61	.19
	c	.28	.98	1.0	.06	.18	..	.02
	d	.5114
	e	.21	.02	..	.71	.21	1.0	.96	.86	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
<i>Es-2</i>	a	X	X	X	X	X	X	X	X	X	X	X	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	X
<i>Es-3</i>	a	1.0
	b	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	..
	c	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	..
<i>Es-4</i>	a	1.0	.6690	..	.30
	b0750
	c	..	.34	.93	.91	.60	1.0	.70	1.0	1.0	1.0	1.0
	d38	.73	.63	.69	.75	.78	.50
	e62	.27	.37	.31	.04
	f	1.0
<i>Gp-1</i>	a	1.0	.98	1.0	1.0	1.0	1.0	1.0	1.0	..
	b02
	c	1.0
	d	1.0	1.0	.77	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
<i>Gp-2</i>	a	1.0	1.0	1.00	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	.97	.97	1.0	1.0	1.0	1.0	1.0	..
	b03	.03
	c	1.0
<i>Gp-3</i>	a	1.0	1.0	1.00	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	..
	b
	c	1.0
<i>Gp-4</i>	a	.96	.33	.68	X	X	X	X	X	X	X	X	..
	b	.04	.67	.32	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	X	X	X	X	X	X	X	X	..

The following loci were fixed identically in all 20 populations: *Mdh*, *Ipo*, *Ldh-2*, *Fum*.

Locality abbreviations are as follows: 1) Kor (Korčula), 2) Las (Lastovo), 3) Pdm (Pod Mrčaru), 4) Vis (Vis), 5) Gre (Greben), 6) Mlp (Mali Paržani), 7) Mlb (Mali Barjak), 8) Bis (Biševo), 9) Sve (Svetac), 10) Bru (Brusnik), 11) Kam (Kamnik), 12) Jab (Jabuka), 13) Zad (Zadar), 14) Tro (Trogir), 15) Pes (Pescara), 16) Pala (Palagruža), 17) Sus (Sušac), 18) Kop (Kopište), 19) Pdk (Pod Kopište), 20) Oxy (*L. oxycephala* mixed localities).

TABLE 3. Rogers' (1972) coefficients of genetic similarity for all the *Lacerta* populations. The population number key is given in Table 1. Decimals are omitted from the values, which should be multiplied by 10^{-2} .

Kor 1	Las 2	Pdm 3	Vis 4	Gre 5	Mip 6	Mib 7	Bis 8	Sve 9	Bru 10	Kam 11	Jab 12	Zad 13	Tro 14	Pes 15	Pala 16	Suš 17	Kop 18	Pdk 19	Oxy 20
	92	88	88	89	92	86	89	90	86	84	86	71	66	71	67	70	70	72	46
		91	90	92	91	88	90	90	87	85	88	67	67	67	64	70	69	69	43
			89	88	84	88	87	92	86	85	88	71	64	70	68	74	73	73	46
				95	94	96	96	95	95	94	96	65	66	66	64	66	66	67	44
					94	93	95	91	91	89	93	66	67	66	64	67	66	67	43
						94	96	91	91	90	94	64	65	64	62	65	65	66	43
							97	95	96	96	100	63	63	63	61	64	63	64	43
								95	95	94	97	64	65	64	63	65	65	66	44
									94	92	96	67	63	67	65	68	67	69	47
										93	96	67	67	67	65	68	67	68	41
											97	63	64	64	62	64	64	65	43
												63	64	63	61	64	64	64	43
													90	95	91	94	93	94	44
														89	88	89	88	88	41
															92	94	94	95	43
																92	91	91	40
																	99	97	41
																		97	41
																			42

within a tightly-knit species group in another genus of lizards.

The within-species similarities suggest some relationships that depart from present taxonomy. The populations of Lastovo and

Vis are placed in the same subspecies (*L. m. lissana*) and the Korčula form is placed in another subspecies (*L. m. curzolensis*). However, the S_R data indicate that the Lastovo population is slightly more similar to Korčula than it is to Vis, whereas the populations most similar to Vis are on its fringing islands, and on all the islands west of Vis extending to Jabuka (Figs. 2, 3). Thus at least six designated subspecies are more similar electrophoretically to *L. m.*

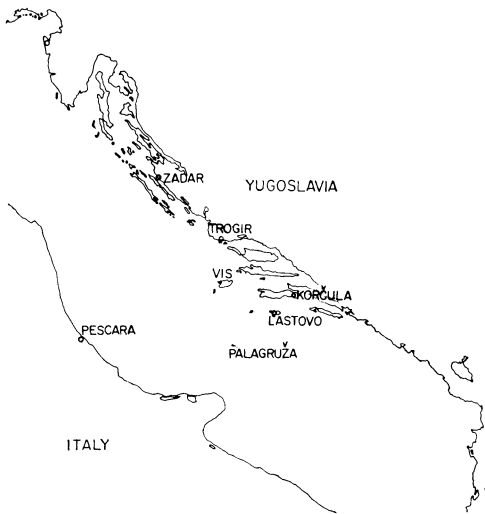


FIG. 1. The Adriatic coast. Mainland collecting sites and some of the major islands are indicated.

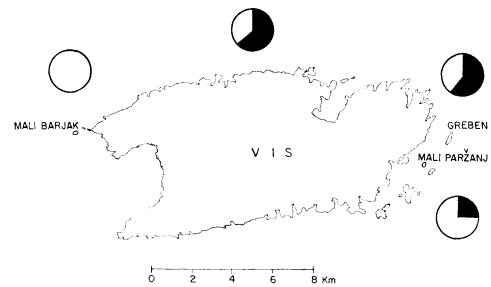


FIG. 2. Vis and its fringing islands. Black areas of pie diagrams indicate percent heterozygosity for populations of *L. melisellensis*; complete black would represent 10%.

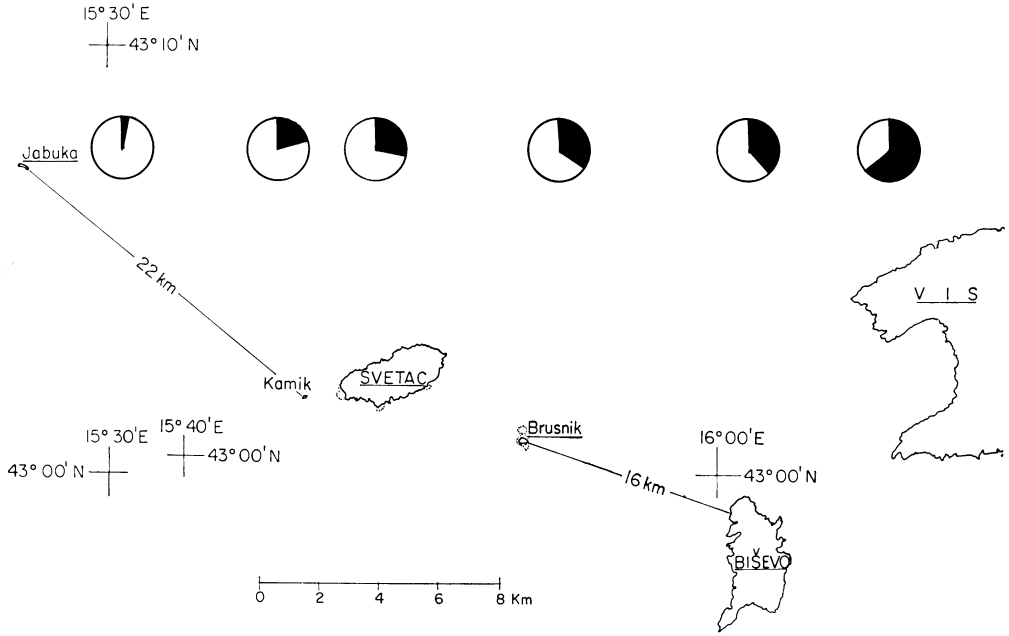


FIG. 3. Vis to Jabuka transect. For proper scaling, several islands had to be displaced, crosses indicate true latitudes and longitudes. Island sizes are drawn to scale. True minimum distances between islands are indicated where displacement would be misleading. Black areas of pie diagrams represent percent heterozygosity for populations of *L. melisellensis*; complete black would represent 10%.

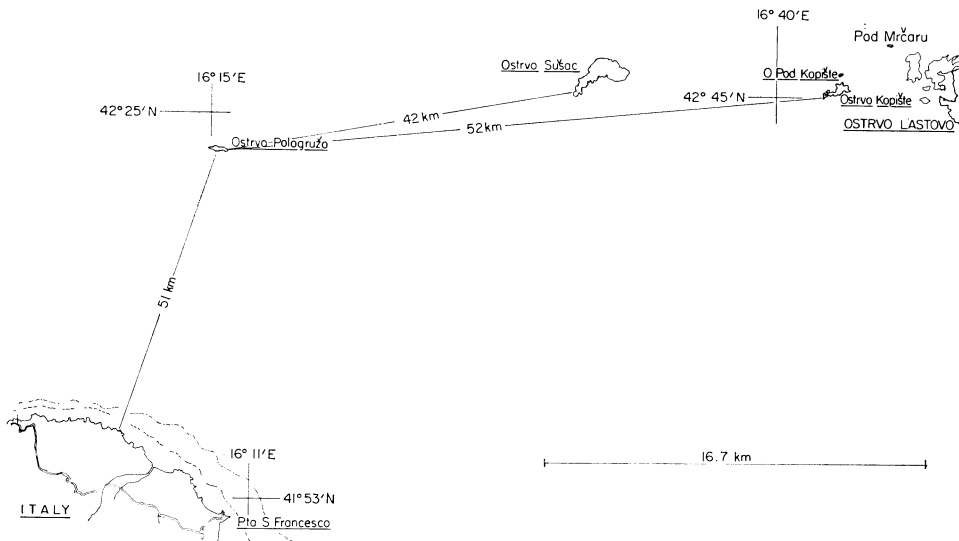


FIG. 4. A portion of the Italian mainland and islands inhabited by *L. sicula*. In addition, two islands that have *L. melisellensis* are shown (Lastovo, Pod Mrčaru). For proper scaling, some islands had to be displaced. Crosses indicate true latitude and longitude. True minimum distances are indicated where displacement would be misleading. Land mass sizes are drawn to scale.

lissana from Vis than is the latter to the same so-called subspecies from Lastovo.

The *L. sicula* comparisons generally confirm our expectations while illuminating some problems. First, the Sušac, Kopač, and Pod Kopač populations are very similar. The island of Palagruža, a possible stepping stone, is situated between these islands and the Italian mainland (Fig. 4), yet the Sušac-Kopač group seems more closely related to Pescara in Italy and Zadar in northern Yugoslavia than to the Palagruža or the nearby Trogir population. This strengthens the supposition that these populations were founded by Italian *sicula*. It also suggests that the Palagruža colonization may have been independent of the Sušac-Kopač colonization and not a stepping stone for the latter.

2) Interspecific Introgression?

The small island of Pod Mrčaru lies very close to Lastovo (Fig. 4). There are two striking peculiarities about the genetics of the Pod Mrčaru *melisellensis* population. First is the relative lack of similarity to Lastovo (Table 3). The S_R value is the lowest among the fringing island-parent island comparisons (see below). Second, from Table 2 we see that the Pod Mrčaru population has three presumed alleles that are unique among *melisellensis* populations (and unique alleles are uncommon among the populations studied).

Two of these alleles are electrophoretically identical with alleles found in *L. sicula* (*Est-4^b* and *Gp-1^a*), and one (*Gp-1^c*) is found in *oxycephala* which is sympatric with *melisellensis* on Pod Mrčaru. At the latitude of Lastovo, Pod Mrčaru is at the species border. All islands to the west have *L. sicula*, islands to the east have *L. melisellensis*. The evidence is circumstantial, but hybridization and introgression are strongly implicated. It is perhaps more than coincidence that the S_R values between *melisellensis* and *sicula* are highest for Pod Mrčaru *melisellensis* compared with *sicula* from Pod Kopač, the nearest *sicula* island (Table 3).

L. melisellensis and *L. sicula* are ecologically similar. If there is occasional immigration, then hybridization and introgression might also be encountered occasionally. It is less likely that *melisellensis* and *oxycephala* would hybridize, for they are broadly sympatric, ecologically distinct, and presumably have well established isolating mechanisms. It is, of course, not unlikely that the "shared" allele in this case is not the same protein, but merely has the same electrophoretic mobility.

Little is known about the relationship between genetic distance and successful hybridization in reptiles. The best documented case is the hybridization of *Anolis trinitatis* and *A. aeneus* on Trinidad. Although the F_1 is predominantly sterile (Gorman, et al., 1971), we now have several examples of backcrossed individuals, indicating that introgression is possible (Gorman and Yang, 1975). Their similarity coefficient is 0.72, which is about the same as the most similar *Lacerta melisellensis*-*L. sicula* populations (Gorman and Soulé, unpublished data).

3) Amounts of Genetic Variation

A) *Mainlands vs. islands*.—Our collecting efforts for mainland *Lacerta* were successful only with *L. sicula*, which were extremely abundant at mainland sites. The mainland populations of *L. sicula* in Yugoslavia and Italy are highly polymorphic and heterozygous for the systems we surveyed. Half the loci at Trogir and about a third at Pescara were polymorphic. The lowest percent polymorphism on the mainland (Zadar) was greater than the maximum for an island population (Palagruža). Heterozygosity was greatest at the three mainland localities, and lowest on the four islands. Mean heterozygosity for the three mainland populations (giving equal weight to each population) is 9.05%, while mean heterozygosity for the four island populations is 4.33%. Palagruža was only slightly lower than Zadar in polymorphism and just below Pescara in heterozygosity (Table 1).

Highest variability was found in *sicula*

from Trogir. The distribution of *sicula* along the Yugoslav coast has a gap that begins just south of Trogir and extends south for several hundred kilometers. A separate invasion from Italy probably accounts for the southern populations; thus the gap is not believed to be the result of extinction, and the Trogir population is presumably near the moving front of an invasion. One might be surprised to find that this peripheral mainland population is the most variable of all populations sampled. Power (1971), however, studied morphological variation in Brewer's blackbird and found that a recently invaded region had birds with as much variation as birds from the original range. What little evidence exists, therefore, suggests that successful overland range expansions need not be accompanied by genetic depauperization at or near the periphery.

Although we lack mainland *melisellensis*, the largest *melisellensis* islands (Korčula, Lastovo, and Vis) might be considered approximations of the mainland ecological condition. This is almost certainly true for Korčula which is less than 2 km from the mainland and is separated by a shallow water channel. If this is so, it appears that there is slightly less variability in "mainland" *melisellensis* than mainland *sicula*. We also note that variability is highest in these three largest *melisellensis* islands (plus Greben, see Fringing Islands, below) and considerably lower on the smaller islands. Thus the pattern is similar for both *melisellensis* and *sicula*.

B) *Fringing islands and the "small island effect."*—Our data allow us to compare the genetic variation in fringing island and "parent" island populations. We define a fringing island as a small island that probably has been disconnected by eustatic sea level rise from the adjacent larger island within the recent geological past, as evidenced by shallow channel depths. In all, seven fringing island populations associated with four parent islands were sampled. The only *L. sicula* fringing population studied was Pod Kopašće, a small island (0.05 sq.

km) only 0.6 km from Kopašće. As shown in Table 1, the heterozygosity estimates are virtually the same; the Kopašće population is polymorphic at an additional locus. Kopašće has two unique alleles (*6-Pgd^b*, *Gp-3^c*) not found in any of the other *Lacerta* populations. Pod Kopašće had a rare allele *6-Pgd^a* that was absent on Kopašće, but present in all other *L. sicula* populations, and might be assumed to be present, but rare on Kopašće. Otherwise, they were fixed for the same alleles, or they shared polymorphisms.

We studied three fringing populations in the vicinity of the large island of Vis: Mali Barjak, Mali Paržanj and Greben. All are within one kilometer of Vis. The two Malis are truly tiny ("mali" means small in Serbo-Croatian), each less than 0.01 sq. km, and they are relatively flat. Greben is not only larger (0.75 sq. km), but much more diverse topographically, rising moderately steeply on the west side, and dropping precipitously on the east. Mali Barjak was the only population sampled with zero variability, and Mali Paržanj was very low compared to Vis (although not as low as some of the most remote islands). Our sample sizes were too small on these islands to account for many of the possible rare polymorphisms, but there can be little doubt that the low heterozygosity is real and significantly different from Vis.

Greben has less than 1% the land area of Vis, is slightly farther from Vis than the two tiny islands, yet had the same variability as Vis. Some rare alleles not in our Vis sample were found on Greben (*Pgm-1^c*, α *Gpd^b*), but one of these appears in the Lastovo sample, the other in Biševo, and it is not unlikely that they would be found in Vis with additional sampling.

The two small islands of Brusnik and Kamik might be considered fringing islands of Svetac. However, neither of their lizard populations show particularly strong affinity with Svetac (Table 3). The Brusnik population is equal to Svetac in polymorphism and higher in heterozygosity. It is considerably larger than Kamik (0.05

vs. 0.01 sq. km) and is shown on the hydrographic charts to be surrounded by extremely shallow water, so that in relatively recent times Brusnik was perhaps twice as large as at present. The Kamik population shows reduction in polymorphism compared to Svetac and at loci (*Pgm-2* and *Got-1*), it is fixed or predominant for alleles that are relatively rare on Svetac.

Jabuka is not a fringing island. It is isolated and surrounded by deep water and almost certainly has not had recent connection with the other islands. Thus the low variability on Jabuka is in accord with the results for the other very small island populations, but other factors, such as founder effect, might be the real cause of low variation.

The fringing island of Pod Mrčaru has lower variability than Lastovo, but more than we might expect from its size. However, introgression with *sicula*, as stated above, might well account for the elevated levels of heterozygosity and polymorphism.

From these results it appears that we can define a "small island effect" in which variability is markedly reduced from the parent island. This effect is limited to tiny islands indeed (0.01 sq. km and less) and is not seen on islands as small as 0.05 sq. km. A similar phenomenon has been found for *Anolis cristatellus* on the fringing islands of Puerto Rico and the Virgin Islands (Soulé and Gorman, unpubl. data). We have no rigorous estimates of population size on such tiny islands, but based on collecting experience our estimates range from 100 to about 1000.

We have considered three explanations for the severe reduction in genetic variation on tiny islands: (1) Genetic drift is likely and its impact would be even greater if in the past the population went through a bottleneck, such as would occur during prolonged drought; (2) extinction of the original relict population followed by over-water colonization by one or a few individuals from the large island; (3) strong directional selection on very small islands.

We dislike the second explanation because we have calculated elsewhere (Soulé et al., in prep.) that colonies established by a single, fertilized female should, on the average, retain about 34% of the polymorphisms in the source population, and these small island populations do not. Unfortunately, the data do not permit us to distinguish between the drift and selection hypotheses.

C) *Retained polymorphisms: Are the allozymes neutral?*—Although genetic drift might account for the possible loss of alleles in the smallest fringing populations, this in no way implies that the alleles are behaving neutrally. In this section we try to estimate the proportion of polymorphic loci for which alleles could be effectively neutral. The major difficulty in such analyses was succinctly stated by Lewontin and Krakauer (1973): "Any observed distribution of gene frequencies over space or time, if considered to be in a steady state can be explained by a suitable choice of N , m and μ , with s being made arbitrarily small. . . ." For example, if many populations are sampled and the gene frequencies are everywhere very similar this can be attributed to strong, uniform selection throughout the range. On the other hand, a neutral model can be applied by assuming that the effective migration between neighboring populations is of the order of one individual per generation (Wright, 1951). Lewontin and Krakauer resolve this conundrum by pointing out that there is one feature of the drift-migration-neutrality hypothesis that produces a different result than does a selection model: "While natural selection will operate differently for each locus and each allele at a locus, the effect of breeding structure is uniform over all loci and all alleles." That is, if for some loci there is spatial or temporal gene frequency heterogeneity, while for others there is uniformity in gene frequencies, and therefore, all loci together form a heterogeneous or bimodal distribution, then it is safe to assume that the neutral model is not a sufficient explanation. In other words, if drift, migra-

tion and neutrality explain the constancy of gene frequencies at the uniform loci, then selection, acting differently in different places or times, must account for those loci which have heterogeneous gene frequencies. Alternatively, if drift is assumed to account for the loci which have heterogeneous gene frequencies, then uniform selection must explain the loci which are geographically or temporally uniform. Lewontin and Krakauer tested for heterogeneity in the gene frequency distributions for a given locus by estimating the parameter F_e , the "effective inbreeding coefficient." The estimate is:

$$\hat{F}_e = \frac{s_p^2}{\bar{p}(1-\bar{p})}$$

when \hat{F}_e = estimate of effective inbreeding, s_p^2 = variance of one of the two alternate alleles from population to population, and \bar{p} = mean frequency of the allele over the ensemble of populations. Next, the ratio of the observed variance over all loci to the theoretical variance (see Lewontin and Krakauer, 1973), s_p^2/σ^2 , is calculated. A high and statistically significant ratio is taken as evidence for heterogeneity of the \hat{F}_e values and for selection at some of the loci.

We chose the most homogeneous set of populations—those on Vis, its fringing island, Greben, Mali Paržanj and Mali Barjak, and the population on Biševo, which, though more than 4 km from Vis, was connected to it about 10,000 to 15,000 years ago. During the last glaciation all of these islands were part of a single, emergent land mass. Sea level reached its present height about 5000 years ago. The best guess, then, about the ages of these populations is that they have been isolated for at least 1000 to 2000 generations. To include populations such as those on the more distant islands of Svetac, Brusnik, Kamik and Jabuka, might weaken the analysis because there is a distinct likelihood that they have not been so recently connected to Vis, hence their lizards might not be samples of the same original gene pool.

We analyzed seven polymorphic loci. The Vis population was scored as monomorphic at one of these loci (α -*Gpd*), but we expect that the rare allele is actually present at a low frequency because of its occurrence on both Greben and Biševo. The same is probably true for the rare alleles in many of the populations. The presence or absence of rare alleles is sampling error and should not seriously affect the results.

The results are given in Table 4. The ratio of the observed variance of \hat{F}_e to the expected variance is $.06981/.01177 = 5.93$ ($P < .001$). (In calculating σ^2 we set $k = 2$, a conservative test. Degrees of freedom were determined as per Lewontin and Krakauer.) Some loci, at least, apparently are selected. Aside from the data in Table 4, chi-square analyses (Snedecor, 1956, p. 227) suggest that only two or three loci are significantly heterogeneous geographically, the two esterases and possibly *Got-1*. The null hypothesis is accepted in five of the seven cases (P for rejection 0.01). The results are as follows: *Pgi*, $\chi^2 = 3.70$, $P > 0.40$; *Got-1*, $\chi^2 = 10.15$, $0.05 > P > 0.02$; *Got-2*, $\chi^2 = 1.16$, $P > 0.80$; α *Gpd*, $\chi^2 = 1.87$, $P > 0.70$; *Pgm-2*, $\chi^2 = 4.17$, $P > 0.30$; *Es-1*, $\chi^2 = 94.65$, $P < 0.01$; *Es-4*, $\chi^2 = 44.53$, $P < 0.01$. The alleles at the other loci comprise a "conservative" set.

Three interpretations of this result are possible. First, neither set of alleles is neutral; the geographically unstable loci manifest the effects of different selection regimes on different islands, whereas the conservative loci are everywhere being stabilized by the same balancing selection forces. Second, alleles at the geographically unstable loci are relatively neutral and are drifting, whereas the conservative loci are responding to selection as in the first interpretation. The third possibility is that there is sufficient gene flow between islands to swamp the effects of selection differentials for most loci; the other loci, the two esterases and possibly *Got-1*, are diverging in the face of the gene flow because they are

TABLE 4. *Estimates of effective inbreeding coefficients for seven polymorphic loci.*

Population	Locus and allele frequency								
	<i>Pgi</i> ^c	<i>Got-1</i> ^c	<i>Got-2</i> ^d	<i>aGpd</i> ^c	<i>Pgm-2</i> ^a	<i>Es-1</i> ^b	<i>Es-1</i> ^c	<i>Es-1</i> ^e	<i>Es-4</i> ^c
Vis	.97	.91	.97	.97	.79	.23	.06	.71	.91
Greben	.96	1.0	.99	1.0	.85	.61	.18	.21	.60
Mali Paržanj	1.0	1.0	1.0	1.0	.91	.19	0	.81	.10
Mali Barjak	1.0	1.0	1.0	.98	1.0	0	0	1.0	1.0
Biševo	1.0	.98	1.0	1.0	.88	0	.02	.96	1.0
s_p^2	.00038	.00125	.00017	.00020	.00603	.0622	.00572	.10067	.14792
\hat{F}	.02753	.07064	.02142	.02020	.0597	.38027	.11603	.52065	.73696

responding to very strong and very different selection pressures.

The last interpretation is the least probable. It is difficult for us to accept that lizards are migrating across the channels at the rate of one per generation (the generation time is about one per year), the rate required to maintain effective panmixis in semi-isolated colonies (Wright, 1951; Maruyama, 1970). The three islets are uninhabited and are rarely visited by man. The actual rates of immigration are more likely to be two orders of magnitude less than that required for panmixis.

Both the first two interpretations require non-neutrality at most of the loci. Circumstantial evidence, therefore, weighs in favor of a selectionist interpretation of the patterns, at the majority, but definitely not all of the loci. The esterase alleles, especially, could well be effectively neutral.

One source of bias in the above analysis is uncorrected. Many of the loci were fixed in the smaller island populations (for the common allele in all cases). Hence, the \hat{F}_e

estimates for these loci are artificially low, the rare allele being present in only two of the five populations for the first four alleles in Table 4. Nevertheless, for three of these four cases, the frequencies at the rare alleles in the two populations where they were detected are virtually identical. In contrast, if, for esterases, one chooses at random pairs of populations where the allele is neither fixed nor absent and examines the frequencies, the gene frequency differences will average about 0.31.

D) Biogeographic correlates of variation.—Here we address one of the central problems of contemporary population genetics—the determinants of genetic diversity in populations. A perusal of Figure 3 and Table 1 will show that the populations of *L. melisellensis* on the more remote islands have lower \bar{H} values. This simple pattern is, however, confounded by a negative correlation between distance and island area; in general, the more remote an island, the smaller it is. Hence, the smaller an island the lower the \bar{H} value. Which is more important, distance or size? Multiple regression is a statistical tool for determining the importance of individual variables in such multivariate problems. The independent variables (IV's) we chose to test were:

A = area of the island;

A' = log of area of the island;

E = maximum elevation of the island;

D^2 = the distance squared between the island and the mainland or, in the

TABLE 5. *Correlation coefficients from multiple regression analyses. The coefficients above the diagonal are from analyses III, IV and V, those below from analysis II.*

	A	A'	D^2	M	E	\bar{H}
A	—	.66	-.42	-.83	—	.63
A'	.66	—	-.25	-.91	—	.91
D^2	-.07	.37	—	.21	—	.08
M	-.77	-.48	.13	—	—	-.92
E	.78	.93	.29	-.60	—	—
\bar{H}	.61	.67	.24	-.78	.72	—

TABLE 6. Coefficients of multiple correlation, R^2 , from four stepwise multiple regression analyses of the biogeographical correlates of genetic variation of *Lacerta melisellensis*. Boldface IV's indicate a negative partial regression coefficient.

Populations	Dependent Variable					
	\bar{H}	\bar{H}	\bar{H}	P		
	II					
All excluding	M	0.61**				
Pod Mrčaru	MD²	0.72**				
	MD²A'	0.76*				
	MD²A'A	0.78*				
	MD²A'AE	0.79				
	III ¹		IV ^{1,2}			
Non-fringing only	M	0.85**	A'	0.83*	A'	0.68*
	MD²	0.92*	A'D ²	0.93*	A'D ²	0.82
	MD²A'	0.97*	A'D ² A	0.95	A'D ² A	0.86
					A'D²AM	0.87

¹ E not included in analysis.

² M not included in analysis.

* $P < 0.05$.

** $P < 0.01$.

case of a small island, distance squared to the likeliest source island (see Table 1);

M = channel depth: the sea level drop in meters that would create a land bridge between the island and the mainland.

The first three IV's are estimates of island size and, by correlation, habitat diversity (see below), while the latter two IV's are estimates of isolation. We use D^2 rather than distance itself because if colonization plays any role, the rate is probably a decreasing exponential function of distance (MacArthur and Wilson, 1967). We use M because it may be correlated with the time since the population was isolated by rising eustatic sea levels following the last glaciation.

Five analyses were performed. The first four used \bar{H} as the dependent variable; the fifth used P . Analysis I employed all of the *L. melisellensis* populations. Analysis II was like the first except that the Pod Mrčaru population was deleted because its \bar{H} estimate is probably a result of inter-specific hybridization. In analyses III, IV and V, all of the fringing island populations

were deleted because they are probably recent isolates of the source population, and are therefore an additional source of heterogeneity. In analysis IV the variable M was deleted. The product-moment correlation coefficients among the variables are given in Table 5. *L. sicula* populations could not be treated in a similar manner because the sample size of islands is too small (fewer than the number of "independent" variables).

The results excepting analysis I are given in Table 6. The first and second analyses give very similar results, although removing Pod Mrčaru gives slightly higher coefficients of multiple correlation, R^2 values.

An impressive improvement in R^2 values occurs upon removal of the fringing populations (analyses III, IV and V). Virtually all of the variance of \bar{H} values is explained by the three variables M , D^2 , and A' , with M alone accounting for 85% of the variance. In this stepwise design, M is the best predictor of heterozygosity. A' , however, is virtually as good, and the combination $A'D^2$ gives a slightly higher R^2 than MD^2 . When P is the dependent variable, A' is the best predictor and $A'D^2$ again is the best two-variable combination.

Interpretation is hazardous, especially because of the small sample size. We are encouraged, nevertheless, by the similarity between these results and those from an analogous study of side-blotched lizards (*Uta*) on islands in the Gulf of California, Mexico (Soulé and Yang, 1973). In the former study we found that the same two-variable combination, $A'D^2$, gave the highest R^2 values. Equally good was the combination MD^2 where M was the number of sympatric, confamilial lizard species. In both the *Uta* and *Lacerta* studies A' accounted for about 75 to 85% of the variance in heterozygosity or polymorphism, and D^2 accounted for about 10%.

Our hypothesis for interpreting the *Uta* results (which we have called the "time-divergence theory of variation") was that (1) genetic variation is lost as a consequence of directional selection at rates proportional to average evolutionary rates; (2) evolutionary rates of island reptiles everywhere seem to be inversely proportional to island size, and the reason for this is that (3) the relative ecological distinctness of an island in comparison to the mainland is much greater on small islands than on larger ones. Specifically, we argued that the ecological dissimilarity, or relative simplicity, of an island compared to the mainland can be rather crudely estimated by the log of island area, or by the number of sympatric confamilial species. The former estimates distinctness, we argued, because the log of habitat diversity might be expected to increase linearly with the log of area in an analogous fashion to the species-area relationship. In qualitative terms, the small islands in both the Gulf of California and the Adriatic are exceedingly arid and rocky with a sparse, unique vegetation and a maritime climate. Other important features are the virtual absence of predators, a highly modified insect fauna, and the occasional overwhelming presence of sea bird nesting colonies (not in the Adriatic, however). This distinct set of conditions affects only a tiny frac-

tion at most of large island and mainland lizards.

The radically different ecology of small islands eventually results in predictable evolutionary changes in behavior, size, color, scales and other characters (Mertens, 1934; Soulé, 1966). Evolutionary rates seem to be particularly high for the small island *Lacerta*. This is more easily demonstrable for color and size than for allozymes. Thus Jabuka, Kamik, and Brusnik may have achieved melanism and relatively large body size independently, but even if they form a single stock, Mali Barjak is tending in the same direction with a population that is distinctly larger, darker and less patterned than on the parent island of Vis.

What we postulate, then, for these Adriatic *Lacerta* is differential rates of loss of alleles, the rate being proportional to the ecological distinctness of the island compared to the mainland. The populations are apparently not old enough to have accumulated new electrophoretic variation through mutation.

The reasons we do not invoke genetic drift to explain the correlation of \bar{H} and A' are (1) the allele frequency data do not support a drift effect except on very tiny islands, none of which are included in analyses III, IV and V, and (2) genetic drift is not a satisfactory explanation for the intermediate \bar{H} values on intermediate sized islands such as Biševo and Svetac; these islands have lizard populations, perhaps 10^4 to 10^5 , too large to be subject to much drift.

A confounding problem in this study is the correlation between the two variables A' and M ($r = 0.91$). This is a geographic coincidence, a consequence of the absence of large, distant islands in the Adriatic. Hence, the importance of M in analysis III could easily be an artifact.

Another possible explanation for the importance of M is that it is an estimate of island age; i.e., the deeper the channel, the longer the island in question has been cut off from a larger island or the mainland.

This, of course, assumes the validity of the commonly accepted figure of 100 to 150 meters for the eustatic sea level rise following the last Pleistocene glacial period. As stated above, Jabuka is the only island included in this study that probably has not been connected to the Yugoslav coast during the last glaciation. Only Jabuka, and perhaps Svetac and its fringing islands, could have populations isolated for more than about 15,000 years.

It is, of course, possible that all of the island populations stem from post-Pleistocene, over-water colonizations, since persistence of the island during glaciation does not mean persistence of these heliothermic lizards. There are other reasons, however, for rejecting this hypothesis. First, island populations such as Vis and Biševo have higher levels of electrophoretic variation than would be expected if these populations were founded by rafting propagules. Second, the virtual electrophoretic identity of alleles and allele frequencies of these two populations (as well as others) strongly implies a pre-existing connection in the Pleistocene; the most recent time such a connection could have existed was during the last glaciation.

If, in fact, M values are fair estimates of population age (time since isolation), why should there have been a steady attrition of variation with time? It is explicit in the hypothesis discussed above, that the level of variation in an island population is a function of the length of time selection has been affecting a diminution of allelic diversity. The longer this time, the less variation is to be expected. Indeed, M is negatively correlated with \bar{H} .

In conclusion, we have proposed two explanations for the major result of the multiple regression analyses. The first is that the loss of allelic diversity is a function of the ecological distinctness of the island. The second is that the loss of variation is a function of the age of the population. Because the two relevant independent variables, A' and M , which we believe to be correlated with ecological distinctness and

time, respectively, are so highly correlated with each other, we cannot distinguish between these two factors or allocate to each the fraction of the variance that it might explain. What we can say is that these are the two variables that would be expected to be important given the general validity of the time-divergence hypothesis.

Some technical points of comparison between the *Uta* and *Lacerta* studies remain to be noted. In the *Uta* study we considered that the variables that best represented time were (1) phenetic distance from a mainland population (Q), and, perhaps (2) D^2 , a measure of isolation. Finally, M in this study and F or F' in the *Uta* study, although they are all channel depth variables, are not strictly comparable. All of the islands in the *Lacerta* study are strictly continental (connected to the mainland in the recent past), whereas many of the islands in the *Uta* study are oceanic (never with a mainland connection). There is no *a priori* reason for supposing a correlation between channel depth and age for oceanic islands.

E) Biogeographical correlates of heterozygosity: Lacerta sicula.—As stated above, we cannot apply the same type of analysis to *L. sicula* because there are too few populations. But we can discuss our observations in the context of our working hypotheses relative to *L. melisellensis*.

It was our original belief based on biogeographic data that most *L. sicula* populations, in contrast to the *L. melisellensis* populations, were founded by over-water colonizations. If this is correct, it follows that the *sicula* populations never had very much electrophoretic variation. The results support our hypothesis. Instead of a positive correlation between A' and \bar{H} as in *melisellensis*, we observe an inverse relationship between area and \bar{H} ; thus Sušac is the largest, but the least heterozygous of the three island populations and Palagruža is the smallest and most heterozygous.

Next we note an inverse relationship between distance and \bar{H} . Palagruža is

closest to the Italian mainland and is most heterozygous; it is approximately on the 100 meter contour and may have been connected to the Italian mainland. Sušac and Kopašte are virtually equidistant from Palagruža (or the mainland) and are less heterozygous. Finally we note that Sušac and Kopašte are surrounded by deep water (> 130 m) and were probably not connected to each other, nor to Palagruža nor the mainland.

SUMMARY

Twenty populations of *Lacerta sicula*, *L. melisellensis*, and *L. oxycephala* from the coast and islands of the Adriatic were studied electrophoretically. Genetic similarity measured by the Rogers coefficient (Rogers, 1972) ranges from 0.84–1.0 among populations of *melisellensis*, and from 0.88–0.99 among populations of *sicula*. Interspecific values range from 0.61–0.74 in this pair, and both have values of about 0.43 compared to *oxycephala*. There is circumstantial evidence for hybridization and introgression in one population.

Among *Lacerta*, amounts of genetic variation are greater in (1) mainland populations than in putatively relict island populations, and (2) on large islands than on small islands. Some fringing islands that are separated by a short linear distance and shallow channel depth from the parent island show a "small island effect," i.e., a precipitous drop in variation compared with the mother island but only if these fringing islands are tiny ($\approx \leq 0.01$ sq. km). Genetic drift is implicated. Larger fringing islands do not show an area effect and are genetically virtually identical to the parent island.

An analysis of allele frequencies in populations on different islands suggests that most of the polymorphisms are being selected, but that some (e.g., esterases) have a pattern consistent with the neutral-drift hypothesis.

Stepwise multiple regression was used to analyze the geographic correlates of mean heterozygosity. The results, like those of a

previous study of island lizards, are consistent with the predictions of the time-divergence theory of variation, but do not allow firm conclusions. The theory predicts that the smaller (more ecologically distinct) and the older an island, the lower the heterozygosity in its population. It is argued that directional selection is the main force eroding genetic variation; the smaller an island, the more directional selection will prevail over stabilizing selection; the older an island, the longer such selection has operated. Very small islands, where genetic drift appears to reduce variability, were excluded from most of the regression studies.

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APPENDIX

Genetic Distance of the Lacertid Lizards

Several estimates have been proposed to examine genetic similarity (or distance) between populations. The Rogers Coefficient (Rogers, 1972) that we used in the body of our paper was chosen, in part, out of convenience (the program was readily available to us) and, in part, to provide direct comparison with other works that have used this measure. Nei (1972) provided another estimate of genetic distance which is probably superior, because it estimates codon differences per locus, and may well be linearly related to divergence time. Dr. Nei's laboratory, in the person of Dr. A. Roychoudhury, kindly consented to compute genetic distance from our gene frequency data. Their program can only calculate pairwise comparisons for 15 or fewer populations, so we cannot compare all 20 populations in the same analysis. Therefore, with the consent of the editors, we present the Nei estimates of genetic distance as an appendix. We compare distances between populations *within* species, then pool the within species gene frequency data to make between species comparisons. We emphasize that none of the conclusions (or speculations) from the body of the paper are altered, merely refined.

Appendix Tables 1a and 1b present standard genetic distance (and standard error of standard genetic distance) between populations of *Lacerta melisellensis* and *L. sicula*, respectively. Table 2 presents the standard genetic distance between the three species of *Lacerta*.

Note that the greatest within species genetic distance is 0.11 for *L. melisellensis* (Korčula and Kamik) and only 0.06 for *L. sicula* (Trogir and Pod Kopište), but the between species values are much higher (0.4 between *melisellensis* and *sicula*).

Establishing divergence time is very much in the realm of speculation. First, we have to make the major assumption that genetic distance is linearly related to time. And, based upon that premise, which is admittedly questionable, we have to calibrate our clock. Elsewhere (Yang et al., 1974) we have attempted such a "ballpark" estimate, and we shall do so again, emphasizing that we are groping for a realistic estimate, and not pretending that we can provide the exact point in time for divergence. Our estimate was based upon the

APPENDIX TABLE 1a. *Standard genetic distance (and standard error)¹ between populations of Lacerta melisellensis.*

	Kor 1	Las 2	Pdm 3	Vis 4	Gre 5	Mlp 6	Mlb 7	Bis 8	Sve 9	Bru 10	Kam 11	Jab 12
2	.013 .009											
3	.064 .058	.048 .032										
4	.066 .053	.023 .020	.040 .038									
5	.042 .029	.009 .006	.054 .045	.006 .006								
6	.024 .023	.010 .007	.097 .069	.040 .040	.015 .015							
7	.091 .068	.036 .027	.061 .060	.003 .002	.010 .009	.047 .048						
8	.051 .037	.014 .009	.053 .047	.003 .003	.001 .001	.021 .021	.006 .005					
9	.061 .061	.031 .027	.009 .007	.013 .013	.025 .019	.068 .053	.026 .026	.023 .019				
10	.090 .066	.044 .030	.056 .042	.014 .013	.024 .017	.063 .051	.016 .014	.020 .016	.028 .020			
11	.116 .074	.061 .037	.087 .066	.022 .020	.037 .029	.074 .056	.025 .027	.031 .026	.049 .035	.043 .031		
12	.091 .068	.036 .027	.061 .060	.003 .002	.010 .009	.047 .048	.000 .025	.006 .005	.025 .026	.016 .014	.023 .023	

TABLE 1b. *Standard genetic distance (and standard error) between populations of Lacerta sicula.*

	Zad 13	Tro 14	Pes 15	Pala 16	Suš 17	Kop 18	Pdk 19
14	.045 .035						
15	.007 .004	.048 .035					
16	.015 .008	.047 .036	.009 .008				
17	.019 .016	.053 .035	.006 .005	.012 .009			
18	.022 .018	.055 .036	.007 .006	.014 .010	.001 .001		
19	.024 .020	.063 .037	.013 .012	.022 .016	.004 .004	.005 .005	

¹ Standard error appears immediately below standard genetic distance.

concordance between Nei's Genetic Distance and Sarich and Wilson's Immunological Distance (where I. D. 30 = G. D. 1.0) and where I. D. 30 = 18 million years of divergence (see Yang et al., 1974).

This estimate gives a predicted divergence time of about 7 million years between *L. melisellensis* and *L. sicula*, and about 16 million years between *L. oxycephala* and the other species. Since the genus *Lacerta* (*sensu lato*) is known as far back as the Miocene (Romer, 1966), and since Arnold (1973) thinks that *oxycephala* is generically distinct from *melisellensis* and *sicula*, our estimate of 16 million years might not be too high.

At the low end, we have estimated that the populations isolated on islands should probably be dated in the tens of thousands of years, based upon changes in eustatic sea level. When we examine the relationships between Vis and the islands to the west (see Fig. 3), the genetic distances generally lie between 0.01 (180,000 years) and 0.003 (60,000 years). These latter estimates are a bit high, but the genetic distances are so small, and depend so heavily on differences in allele frequencies rather than gene substitutions, that we are not displeased with the order of magnitude provided by the estimates.

APPENDIX TABLE 2. *Standard genetic distance (\pm standard error) of three species of Lacerta.*

	<i>L. melisellensis</i>	<i>L. sicula</i>
<i>L. sicula</i>	0.415 \pm 0.169	
<i>L. oxycephala</i>	0.894 \pm 0.285	0.9478 \pm 0.300

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ADDITIONAL LITERATURE CITED

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