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Genetic Variation in a Dwarfed Lineage, the Mexican Salamander Genus *Thorius* (Amphibia: Plethodontidae): Taxonomic, Ecologic and Evolutionary Implications

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Patterns of genetic relationship and taxonomic identity are examined in *Thorius*, an enigmatic genus of minute neotropical salamanders that represent the smallest tailed tetrapods. Data comprise an electrophoretic analysis of 16 protein loci in 69 population samples from 55 localities throughout the range of the genus in southern México. Eight of the nine described species are genetically distinct: Nei's D commonly exceeds 0.9. There is no biochemical evidence of more than a single species at Zoquitlán, Pue., type locality of *T. schmidti* and *T. maxillabrochus*. Genetic differentiation between disjunct subspecies of *T. pennatulus* is less than the mean pairwise value between described species, but exceeds that observed between some sympatric species pairs. Seven additional as yet undescribed species are identified, based either on their sympatric occurrence with, or great genetic differentiation from, described species. Most species initially identified by electrophoresis are morphologically distinct. Electrophoretic data fail to resolve the taxonomic affinity of a few remaining populations, although they do provide clearcut and distinguishable hypotheses of relationship that can be tested using other sources, such as morphology.

The 15 species constitute three faunal units: Veracruz and eastern Puebla; Sierra de Juárez, Oaxaca; southern and western Oaxaca, and Guerrero. Local endemism is observed within each region, in which sympatry involving two or three species is common. Species frequently demonstrate narrow elevational zonation and distinct habitat and microhabitat preference (e.g., arboreal vs terrestrial). The distribution of four species on Cerro Pelón, Oax., is examined in detail.

RADIATION of the lungless salamanders (Plethodontidae) in the New World tropics rivals—in the extent and nature of morphological diversity—that of any vertebrate taxon of comparable rank. Comprising at least 11 genera (Wake and Elias, 1983), neotropical plethodontid salamanders display a remarkable array of structural and functional configuration, which in many instances is associated with changes in locomotion, feeding mechanics and ecology (Alberch, 1981; Lombard and Wake, 1977; Wake, 1966). But perhaps the most intriguing trend in the group is phylogenetic size reduction, or dwarfism. Reduced adult body size has evolved in several genera, which each contain at least one species whose adult body size may approach that of the smallest urodeles. By far the most extreme example is the Mexican genus *Thorius*, a small-size "specialist" in which adult body size may be as little as 13 mm SL (Standard Length: snout to posterior end of vent), making these salamanders among the smallest of all tailed tetrapods.

The morphology of *Thorius* is highly derived and unique among plethodontid salamanders (Hanken, 1980, 1982; Wake, 1966); indeed, because of the several novel attributes that belie its affinities with the Plethodontidae, at one time *Thorius* was placed in its own (i.e., monotypic) family, Thoriidae (Cope, 1869). Ironically, the morphology of *Thorius*, though readily distinguishing it from all other plethodontid genera, superficially appears highly conservative within the genus. This retarded past attempts to resolve even alpha-taxonomic relationships using morphological evidence and, as a result, specialists familiar with the genus agree that, although nine species are formally described, taxonomic identities remain poorly known (Wake and Lynch, 1976). (In fact, although external morphology is relatively invariant, internal morphology, particularly osteology, is highly variable, including many characters that distinguish species. But because of substantial intraspecific variation, in most cases it has not been possible to reliably infer, a priori, taxo-

nomic boundaries and relationships from morphology.)

Many recent taxonomic studies have benefitted greatly from the advent of biochemical procedures that may be used to quantify molecular differentiation, and thus affinity, among populations (Avise, 1974; Wake, 1981; Wilson et al., 1977). This is particularly true for vertebrate groups that previously have defied systematic analysis by standard morphological criteria. Molecular differentiation often is large and may be used, by considering patterns of genetic relationship, to tease apart a complex of cryptic species and delineate taxonomic boundaries. Plethodontid genera in which this approach has been used effectively include *Batrachoseps* (Yanev, 1978, 1980), *Desmognathus* (Tilley et al., 1978; Tilley and Schwerdtferger, 1981; Tilley, 1981); *Plethodon* (Duncan and Highton, 1979; Highton, 1979; Larson and Highton, 1978) and *Pseudoeurycea* (Lynch et al., 1977). In the hope of gaining the necessary insight into species boundaries which morphology alone had failed to provide, I performed an electrophoretic analysis of protein variation and differentiation among populations of *Thorius* from throughout its range in southern Mexico. I used allozyme evidence mainly in two ways: 1) identification of sympatric, often cryptic, species, and 2) cluster analysis of populations based on observed levels of differentiation. Consideration of the pattern of local differentiation in a single species, *T. macdougalli*, aided interpretation of the patterns seen among more distant allopatric populations and the cluster analyses. The results provide a hypothesis of genetic relationship and taxonomic identity that may be used, together with morphology, to resolve taxonomic uncertainty and define the systematics of this previously intractable group.

Systematics of *Thorius*

Characters that have been used to distinguish the nine species of *Thorius* involve often slight differences in external morphology (e.g., head and nostril shape, foot and digit proportions, degree of interdigital webbing, trunk and tail shape), external coloration and adult body size. Unfortunately, many of these characters, while of potential use for distinguishing species, are susceptible to preservation artifact; such artifacts have not been considered in previous species descriptions based on preserved material. Furthermore, both sexual and seasonal variation—known to affect most external characters (Freeman, 1977)—have not been taken into account when evaluating species differences. Osteological characters, with the exception of the presence/absence of maxillary teeth and the relative size of premaxillary teeth in males, have been ignored. As a result, available species descriptions often are of little use in establishing the identity of specimens from any but a type locality, and are sometimes unable to provide unequivocal identification of additional specimens collected at type localities from which more than one species has been described. A brief summary of each species is provided below. More detailed descriptions, and additional references, are provided in Freeman (1977) and Hanken (1980).

Thorius pennatulus, type species for the genus, was described by Cope (1869) based on seven specimens from "Orivaza," Ver. This terrestrial species is known from several localities in Veracruz in the vicinity (mostly to the southeast) of Volcán Orizaba at elevations of 800–1,200 m—elevations lower than those of any other species. Shannon and Werler (1955a) described a disjunct population from Volcán San Martín in the isolated Tuxtla Range of extreme southeastern Veracruz as *T. p. narismagnus*. Both subspecies have the distinctive circular nostril which is not seen in any other species of *Thorius*; they are distinguished by the slightly larger nostril in *T. p. narismagnus* and slight differences in external coloration.

Thorius pulmonaris and *T. narisovalis*, two terrestrial species from the montane forest of Cerro San Felipe, Oax., were described by Taylor (1939). These species were distinguished by nostril dimensions (larger and more elongate in *T. pulmonaris*) and different elevational distributions (about 2,000 m for *T. pulmonaris* vs 2,600–3,000 m for *T. narisovalis*). Taylor also suggested slight differences in microhabitats favored by the two species: *T. narisovalis* was typically found under the bark of fallen logs, whereas *T. pulmonaris* was found invariably in leaf litter.

Thorius dubitus and *T. troglodytes* were described by Taylor (1941) from the mountains above Acultzingo, Ver. Prominent characters used to differentiate these species were adult body size (greater in *T. troglodytes*) and external proportions. These two terrestrial species also differed slightly in elevational distribution and microhabitat preference: *T. dubitus* occurred at slightly higher elevations and was collected from

under moss and other plants; *T. troglodytes* was found under rocks. Shannon and Werler (1955b), however, later collected both species at the same elevation (3,050 m) near the type locality.

Thorius minutissimus and *T. macdougalli* were described by Taylor (1949) from separate, distant Oaxacan localities. *Thorius minutissimus,* from Santo Tomás Tecpan in the Sierra Madre del Sur of southern Oaxaca near the Isthmus of Tehuantepec, resembles *T. narisovalis* in nostril morphology, but differs in its (smaller) adult body size, in which it resembles *T. pennatulus.* Subtle aspects of dorsal head coloration, and the supposed frequent absence of the fifth digit from the hind foot, were additional characters believed unique to this species. Similarly, Taylor allied *T. macdougalli,* from Cerro de Humo (Cerro Pelón) in the Sierra de Juárez of north-central Oaxaca, to *T. pulmonaris* based on the shape of the external naris, but justified its specific designation because of its smaller adult body size, relatively short tail, rounded digit tips, and dorsal coloration.

Thorius schmidti and *T. maxillabrochus* were described by Gehlbach (1959) from a remote locality near Zoquitlán in extreme southern Puebla. All 12 specimens in both type series have maxillary teeth which are not seen in any other described species; Gehlbach further differentiated the Zoquitlán material on the basis of nostril shape, body size, relative tail length and foot proportions. All specimens were found under fallen logs.

At least six additional populations of *Thorius* from widely scattered localities do not closely match the description of any nominate form. Each population has, at various times, been suspected of representing an undescribed species, but morphological analysis alone has not allowed unambiguous delineation of species boundaries. As long as 40 years ago, Taylor (1941:107) suggested that the "high mountain specimens" collected near Xometla on Volcán Orizaba represented an undescribed species. More recently, both R. W. McDiarmid (pers. comm.) and Freeman (1977) distinguished two populations, which differ in ventral coloration and nostril shape, sympatric at montane localities in pine-oak forest on Volcán Orizaba. At Puerto del Aire, Ver., the type locality of *T. dubitus* and *T. troglodytes,* two terrestrial species, specimens collected in arboreal microhabitats, such as bromeliads, are distinguished by limb and feet proportions and external coloration (McDiarmid, pers. comm.). In the Sierra de Juárez, Oax., from which only a single species, *T. macdougalli,* is described, the presence of two additional species, differing in adult body size and external coloration, has been suspected (Freeman, 1977; D. B. Wake, pers. comm.). Lastly, populations collected in cloud forest localities west of the Rio Atoyác in the western Sierra Madre del Sur of Guerrero differ in adult body size and proportions from *T. minutissimus,* from the eastern Sierra Madre del Sur, as well as other, more distant species (Freeman, 1977; Wake and Lynch, 1976).

Materials and Methods

Salamanders were collected during four field trips in the summer and winter of 1976 and 1978. The 69 population samples from 55 localities include animals from all but one type locality (Fig. 1; Table 1). I visited Santo Tomás Tecpan, Oax., the type locality of *T. minutissimus,* but found no salamanders; a sample from south of Sola de Vega, Oax.—west of Santo Tomás Tecpan in the southern Sierra Madre del Sur—was used instead as a reference population for this species. Sample sizes ranged from 1 to 30 specimens; at least 20 specimens were analyzed when available; nearly 800 animals were analyzed overall. Exact locality data were presented elsewhere (Hanken, 1980) and are available upon request.

Most specimens were frozen whole within 24 hours of collection and stored in liquid nitrogen (−196 C) until return to the laboratory. There they were transferred to a freezer (−76 C) for up to several months, whereupon the specimens were thawed and samples of liver, heart, gut, body wall and tail were removed for analysis. Salamanders that were not frozen in the field were returned to the laboratory alive where they were anesthetized in 30% chloretone before dissection. Carcasses were individually tagged, preserved in 10% neutral-buffered formalin, and retained as voucher specimens; they are deposited in the permanent collection of The University of California Museum of Vertebrate Zoology (the list of specimens is presented in Appendix 1). For each animal, tissue samples were pooled and proteins assayed using standard horizontal starch gel electrophoresis and histochemical staining procedures (Ayala et al., 1972; Harris and Hopkinson, 1976; Selander et al., 1971) (Table 2).

Allele frequencies were used to compute stan-

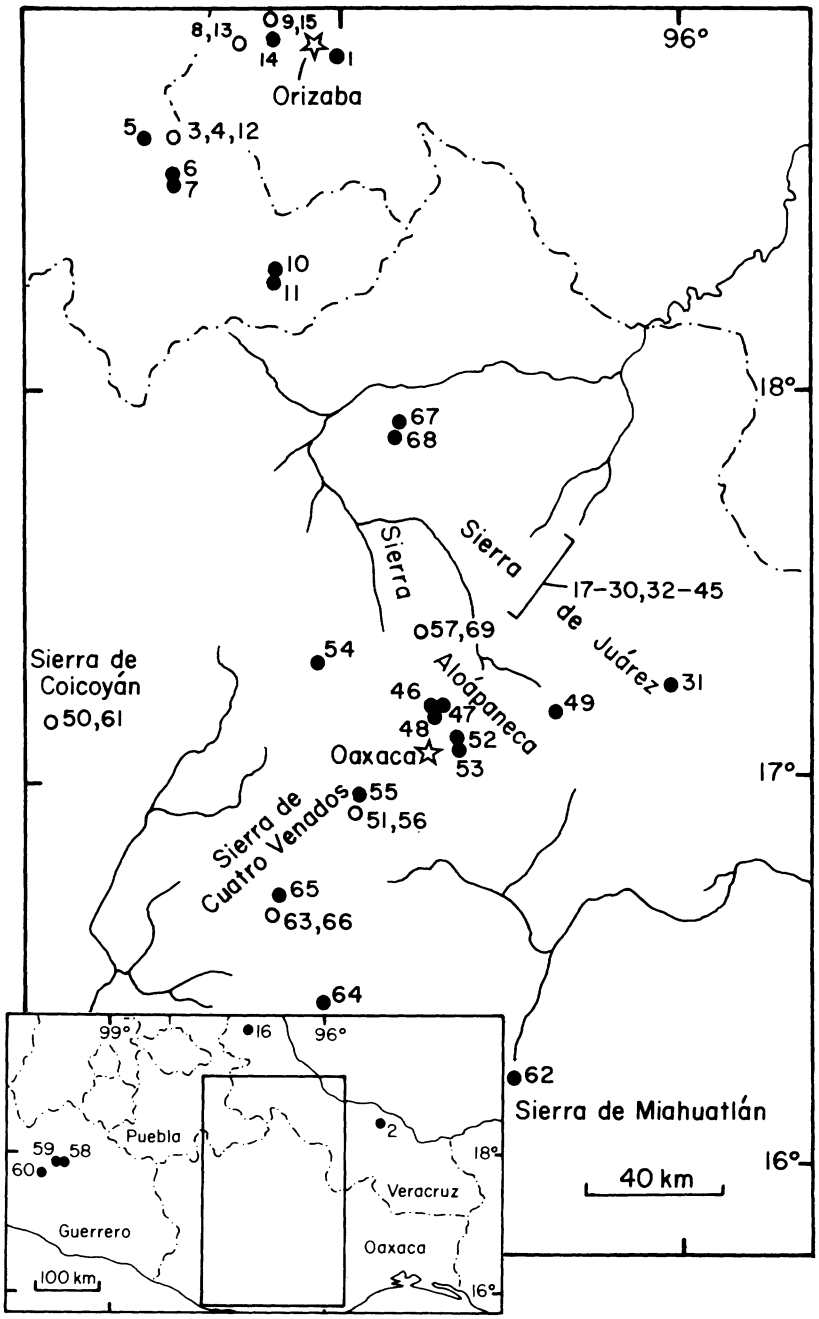

Fig. 1. Collection localities of *Thorius* in southern México. The enlarged map includes the rectangular area outlined on the inset. Closed circles identify those localities at which only one species occurs; open circles identify sympatric localities. Populations are numbered as in Table 1. Populations 17–30 and 32–45 from the vicinity of Cerro Pelón include five sympatric localities not indicated on map. Outlying localities from Guerrero (58–60) and Veracruz (2, 16) are indicated on the inset.

TABLE 1. COLLECTION LOCALITIES. Asterisk denotes populations sympatric with one (*) or two (**) additional species. Type localities are underlined, except for population 64 which is the reference sample for *T. minutissimus*.

Population	Species	Locality	Latitude	Longitude	Elevation (m)	N
1	*T. p. pennatulus*	Ver.: Cerro Chicahuaxtla, Cuautlapam	18°45′	97°00′	980–1,150	16
2	*T. p. narismagnus*	Ver.: Volcán San Martin	18°30′	95°10′	890–1,200	8
3**	*T. dubitus*	Ver.: 3.2 km S Puerto del Aire	18°30′	97°20′	3,280	18
4**	*T. troglodytes*	Ver.: 3.2 km S Puerto del Aire	18°30′	97°20′	3,280	16
5	*T. troglodytes*	Pue.: 1.1 km W Puerto del Aire	18°30′	97°20′	—	1
6	*T. troglodytes*	Pue.: 10 km E San Felipe	18°25′	97°20′	2,790	30
7	*T. troglodytes*	Pue.: 12 km E San Felipe	18°25′	97°20′	2,660	25
8*	*T. schmidti*	Ver.: Volcán Orizaba: El Berro	19°00′	97°15′	2,550	22
9*	*T. schmidti*	Ver.: Volcán Orizaba: Xometla	19°00′	97°15′	2,625	17
10	*T. schmidti*	Pue.: 9 km W Zoquitlán	18°20′	97°05′	2,470	19
11	*T. schmidti*	Pue.: 10 km W Zoquitlán	18°20′	97°05′	2,760	6
12**	*T.* sp. A	Ver.: 3.2 km S Puerto del Aire	18°30′	97°20′	2,500	7
13*	*T.* sp. B	Ver.: Volcán Orizaba: El Berro	19°00′	97°15′	2,500	8
14	*T.* sp. B	Ver.: Volcán Orizaba: Texmola	19°00′	97°15′	2,640	27
15*	*T.* sp. B	Ver.: Volcán Orizaba: Xometla	19°00′	97°15′	2,625	23
16	*T.* sp. B	Ver.: Las Vigas	19°40′	97°05′	2,525	20
17	uncertain	Oax.: Cerro Pelón: 10.3 km N Guelatao	17°15′	96°25′	2,230	1
18	*T. macdougalli*	Oax.: Cerro Pelón: 18 km N Guelatao	17°20′	96°25′	2,790	20
19	*T. macdougalli*	Oax.: Cerro Pelón: 20.6 km N Guelatao	17°20′	96°25′	2.760	1
20	*T. macdougalli*	Oax.: Cerro Pelón: 22.6 km N Guelatao	17°20′	96°25′	2,875	2
21	*T. macdougalli*	Oax.: Cerro Pelón: 23.1 km N Guelatao	17°20′	96°25′	2,850	8
22*	*T. macdougalli*	Oax.: Cerro Pelón: 25 km N Guelatao	17°25′	96°25′	2,850	18
23*	*T. macdougalli*	Oax.: Cerro Pelón: 26 km N Guelatao	17°25′	96°25′	2,885	20
24*	*T. macdougalli*	Oax.: Cerro Pelón: 30.5 km N Guelatao	17°25′	96°25′	2,950	20
25	*T. macdougalli*	Oax.: Cerro Pelón: 37 km N Guelatao	17°30′	96°25′	2,905	1
26	*T. macdougalli*	Oax.: Cerro Pelón: 37.6 km N Guelatao	17°30′	96°25′	2,905	1
27	*T. macdougalli*	Oax.: Cerro Pelón: 42 km N Guelatao (Machín)	17°30′	96°25′	2,725	18
28	*T. macdougalli*	Oax.: Cerro Pelón: 46 km N Guelatao	17°35′	96°25′	2,820	2
29	*T. macdougalli*	Oax.: Cerro Pelón: 50.2 km N Guelatao	17°35′	96°25′	2,955	11
30**	*T. macdougalli*	Oax.: Cerro Pelón: 51 km N Guelatao	17°35′	96°25′	2,930	17
31	uncertain	Oax.: Totontepec	17°10′	96°00′	2,600	9
32*	*T.* sp. C	Oax.: Cerro Pelón: 25 km N Guelatao	17°25′	96°25′	2,850	5
33*	*T.* sp. C	Oax.: Cerro Pelón: 26 km N Guelatao	17°25′	96°25′	2,885	1
34*	*T.* sp. C	Oax.: Cerro Pelón: 30.5 km N Guelatao	17°25′	96°25′	2,950	5
35	*T.* sp. C	Oax.: Cerro Pelón: 46.9 km N Guelatao	17°35′	96°25′	2,890	3
36**	*T.* sp. C	Oax.: Cerro Pelón: 51 km N Guelatao	17°35′	96°25′	2,930	10
37	*T.* sp. D	Oax.: Cerro Pelón: 54 km N Guelatao	17°35′	96°25′	2,475	9
38*	*T.* sp. D	Oax.: Cerro Pelón: 52 km N Guelatao	17°35′	96°25′	2,755	11
39**	*T.* sp. D	Oax.: Cerro Pelón: 51 km N Guelatao	17°35′	96°25′	2,930	4
40*	*T.* sp. E	Oax.: Cerro Pelón: 52 km N Guelatao	17°35′	96°25′	2,755	8
41	*T.* sp. E	Oax.: Cerro Pelón: 56 km N Guelatao	17°40′	96°25′	2,335	8
42	*T.* sp. E	Oax.: Cerro Pelón: 57.3 km N Guelatao	17°40′	96°25′	2,270	1
43	*T.* sp. E	Oax.: Cerro Pelón: 65 km N Guelatao	17°40′	96°25′	2,170	3
44	*T.* sp. E	Oax.: Cerro Pelón: 65.3 km N Guelatao	17°40′	96°25′	—	1
45	uncertain	Oax.: Cerro Pelón: 81 km N Guelatao	17°45′	96°20′	1,530	1
46	*T. nariscovalis*	Oax.: Cerro San Felipe: 4 km W La Cumbre	17°10′	96°45′	2,850	20
47	*T. nariscovalis*	Oax.: Cerro San Felipe: 9 km W La Cumbre	17°10′	96°40′	3,080	19
48	*T. nariscovalis*	Oax.: Cerro San Felipe: 15 km W La Cumbre	17°10′	96°40′	3,185	16
49	*T. nariscovalis*	Oax.: 4 km NE Cuajimoloyas	17°05′	96°30′	3,170	20
50*	*T. nariscovalis*	Oax.: 29.5 km NE Tlaxiaco	17°15′	97°40′	3,080	1
51*	*T. nariscovalis*	Oax.: 25 km W Zaachila	16°55′	97°00′	2,780	16

TABLE 1. Continued.

Population	Species	Locality	Latitude	Longitude	Elevation (m)	N
52	*T. pulmonaris*	Oax.: Cerro San Felipe: 3.4 km S La Cumbre	17°05′	96°35′	2,530	3
53	*T. pulmonaris*	Oax.: Cerro San Felipe: 6.8 km S La Cumbre	17°05′	96°35′	2,180	13
54	*T. pulmonaris*	Oax. Tejocotes	17°15′	97°00′	2,350	1
55	*T. pulmonaris*	Oax.: 24 km W Zaachila	16°55′	97°00′	2,750	1
56*	*T. pulmonaris*	Oax.: 25 km W Zaachila	16°55′	97°00′	2,780	3
57*	uncertain	Oax.: 15 km NE San Juan del Estado	17°15′	96°45′	2,550	10
58	*T.* sp. F	Gue.: Carrizal de Bravos	17°25′	99°50′	2,520	20
59	*T.* sp. F	Gue.: Puerto del Gallo	17°25′	100°10′	2,500	20
60	*T.* sp. F	Gue.: 9 km NE Puerto del Gallo	17°25′	100°10′	3,110	10
61*	uncertain	Oax.: 29.5 km NE Tlaxiaco	17°15′	97°40′	3,080	9
62	uncertain	Oax.: Suchixtepec	16°05′	96°30′	2,700	20
63*	uncertain	Oax.: 15.5 km W San Vicente Lachixio	16°40′	97°05′	2,730	18
64	*T. minutissimus*	Oax.: 18.5 km S Sola de Vega	16°20′	97°00′	2,150	20
65	uncertain	Oax.: 13.2 km W San Vicente Lachixio	16°40′	97°05′	2,710	10
66*	uncertain	Oax.: 15.5 km W San Vicente Lachixio	16°40′	97°05′	2,730	5
67	*T.* sp. G	Oax.: 8 km NE Santos Reyes Pápalo	17°50′	96°50′	2,670	20
68	*T.* sp. G	Oax.: 11 km NE Santos Reyes Pápalo	17°50′	96°50′	2,820	20
69*	*T.* sp. G	Oax.: 15 km NE San Juan del Estado	17°15′	96°45′	2,550	9

dard estimates of protein divergence among populations, Nei's D (Nei, 1972; Nei and Roychoudhury, 1974) and Rogers' S (Rogers, 1972). Alternate clustering dendrograms based on these measures were obtained from three different methods: 1) Unweighted pairgroup method with arithmetic averages (UPGMA; Sneath and Sokal, 1973) from the Numerical Taxonomy Package NT-11; 2) Fitch-Margoliash method (Fitch and Margoliash, 1967) with iterative averaging from the Evolves program supplied by W. Fitch; 3) Distance Wagner procedure (Farris, 1972) using the "Cladistic Inference by Parsimony" program provided by J. S. Farris.

RESULTS

Protein variation.—Sixteen protein loci could be reliably scored in all populations (Table 2). Many additional enzymes were included in initial runs, but because of the tiny amount of tissue sample available from a single specimen these loci provided only faint or otherwise poor resolution of banding patterns; they were not used in subsequent runs. Several groups of populations, usually those separated by only a few km or less, showed relatively slight differences in allele frequencies ($D \leq 0.10$ in most instances) and consistently linked together in the initial cluster analyses based on observed levels of genetic differentiation (see below). In such cases, only one population of each group is included in Table 3 which presents allele frequencies and heterozygosity values, for 35 populations from 28 principal localities. Estimates of genetic differentiation (D and S) among these 35 populations, computed from observed allele frequencies at all 16 loci, are given in Table 4. Allele frequencies and estimates of genetic differentiation for the remaining populations are not provided due to space limitation, but are available upon request.

Variability was high. Mean total number of alleles per locus was 10.6 (range 3–19) and each locus was polymorphic in at least one population. (A locus was considered polymorphic if at least two alleles were present, in any frequency, in a given population.) For a given locus, the mean number of alleles per population ranged from a low of 1.06 (Gapdh) to a high of 2.16 (Mpi); as many as eight alleles were identified at one locus (Got-1) in a single population (67). Heterozygosity ($\bar{H}$ determined by direct count of homozygotes and heterozygotes) in samples of five or more specimens ranged from 0.02 in population 62 to 0.21 in population 30 ($\bar{x} = 0.11$; $N = 50$). Overall, D ranged from as little as 0.01, between adjacent populations 37 and 38 on the north face of Cerro Pelón, to as high as 2.07, between Las Vigas (16) and Zaachila (55).

TABLE 2. PROTEIN VARIATION AT 16 ELECTROPHORETIC LOCI IN *Thorius*.

Locus	Buffer[1]	No. alleles in genus	No. alleles per population[2] ($\bar{x}$, range)	Heterozygosity[2,3] ($\bar{x}$, range)
leucine aminopeptidase (Lap)	TC 7.0	8	1.18, 1–2	.03, 0–.50
isocitrate dehydrogenase 1 (Icd-1)	TC 8.0	8	1.16, 1–3	.02, 0–.44
isocitrate dehydrogenase 2 (Icd-2)	TC 8.0	6	1.37, 1–3	.10, 0–.60
glyceraldehyde-phosphate dehydrogenase (Gapdh)	TC 8.0	7	1.06, 1–2	.03, 0–.80
malate dehydrogenase 1 (Mdh-1)	TC 7.0	11	1.35, 1–3	.08, 0–.59
malate dehydrogenase 2 (Mdh-2)	TC 7.0	12	1.29, 1–3	.05, 0–.55
phosphoglucomutase (Pgm)	TC 7.0	11	1.53, 1–3	.12, 0–.70
glucosephosphate isomerase (Gpi)	GPI phosphate	12	1.69, 1–4	.10, 0–.58
lactate dehydrogenase 1 (Ldh-1)	Poulik, TC 7.0	16	1.24, 1–2	.02, 0–.17
lactate dehydrogenase 2 (Ldh-2)	TC 8.0	6	1.16, 1–3	.02, 0–.40
glutamate oxaloacetate transaminase 1 (Got-1)	TVB, TC 7.0 Poulik	15	2.14, 1–8	.26, 0–.85
glutamate oxaloacetate transaminase 2 (Got-2)	Tris HCL	3	1.37, 1–3	.06, 0–.55
α-glycerophosphate transaminase (Gpd)	TC 8.0	19	2.00, 1–5	.20, 0–.73
peptidase (Pep)	GPI phosphate	11	1.82, 1–4	.20, 0–.67
mannose-phosphate isomerase (Mpi)	TC 8.0	19	2.16, 1–5	.24, 0–.69
serum general protein 2 (Pt-2)	TVB, Tris HCL	6	1.16, 1–2	.02, 0–.44

[1] Abbreviations: Tris citrate pH 7.0 (TC 7.0); Tris citrate pH 8.0 (TC 8.0); Tris verzene borate (TVB).
[2] Based on populations of $N \geq 5$.
[3] Determined by direct count of homozygotes and heterozygotes.

Identification of sympatric species.—Extreme deviations of genotypic arrays from Hardy-Weinberg expectations were observed at 12 localities in Oaxaca and Veracruz (Table 5). At each locality there was conspicuous and total absence of heterozygote phenotypes between one or more pairs of alleles at a given locus, such that the specimens could be divided among two, or sometimes three, populations within which Hardy-Weinberg expectations were met, but between which heterozygotes were lacking. Concordant patterns of fixed differences involving as many as 12 loci were found at each locality such that the composition of each population was consistent. Remaining loci often demonstrated additional between-group differences in allele frequencies (Table 3). This evidence strongly suggests the sympatric occurrence, at each locality, of two or three genetically distinct populations with no evidence of hybridization—in other words, sympatric species. Nei's D between sympatric species ranged from 0.31 to 1.27.

Only one of two published reports of sympatry was confirmed by electrophoresis. Two terrestrial species were detected at Puerto del Aire, Ver., (Table 5, locality I, populations 3 and 4), the type locality of *T. dubitus* and *T. troglodytes*, but electrophoresis also established the presence of a third, arboreal species at this locality (population 12, taken in bromeliads). In fact, this latter species is more differentiated genetically from both terrestrial species than the two terrestrial species are from each other. In contrast, analysis of the 25 animals collected from two localities (populations 10 and 11) near Zoquitlán, Pue., the type locality of *T. schmidti* and *T. maxillabrochus*, revealed no biochemical evidence of more than a single species in these samples.

The remaining 11 instances of sympatry either confirm the existence of undescribed species previously suspected by herpetologists (see above), or represent additional instances previously undetected. In Veracruz, the presence of two sympatric species at high elevations on Volcán Orizaba was established at two localities (Table 5, localities II and III). These species are well-differentiated genetically, displaying fixed allelic differences at 10 or 11 of the 16 loci sampled; D between sympatric populations exceeds 1.0 at both localities. Five sympatric localities were identified on Cerro Pelón in the Sierra de Juárez, Oax. A second, larger terres-

TABLE 3. ALLELE FREQUENCIES AND HETEROZYGOSITY ($\bar{H}$) FOR 35 POPULATIONS OF *Thorius*.

	Population																
Locus	1	2	3	4	6	8	11	12	14	16	17	27	30	31	32	36	37
Lap	c	c	e	e	e	c	c	h	b (.02) c (.98)	c	c	e	e	e	c	c	c
Icd-1	e	g	g	c (.69) g (.31)	e	c (.54) g (.46)	e (.17) f (.58) g (.25)	e (.56) g (.44)	g	g	d	d	d	d	d	d	d
Icd-2	c (.92) d (.08)	c	c (.71) e (.29)	c	c	c	c (.92) d (.08)	c	c	c	c	b (.42) c (.58)	b (.62) c (.38)	b (.94) c (.06)	c	c	c
Gapdh	b	b	d	d	d	d	f	e	g	g	a	f	f	c (.11) f (.89)	b	b	g
Mdh-1	c	g	h	e	e	e (.98) h (.02)	e	h	e	e	b	a (.22) e (.78)	a (.27) b (.03) e (.70)	a (.11) e (.89)	i	i	i
Mdh-2	b	b (.25) f (.75)	h	h	d (.05) h (.95)	f	c	d	a (.05) c (.95)	c	e	e (.11) j (.89)	e	c (.06) h (.94)	e	e	e
Pgm	d (.10) e (.31) f (.59)	a (.06) e (.50) g (.44)	e	e	e (.75) f (.25)	e	e	c	e (.03) g (.97)	j	e	c (.03) e (.94) g (.03)	e (.94) g (.06)	e (.94) k (.06)	e	e	e (.78) g (.22)
Gpi	f	f	f	f	f (.98) g (.02)	f	f	f	f (.98) j (.02)	f	f	f	f	f (.94) j (.06)	h	h	h
Ldh-1	e	e	l	l	l	k	i	l	h	h	e	f (.14) l (.86)	h (.06) l (.94)	p	l	j	l
Ldh-2	c	c	b	b	b	d	a	b	c	c	d	a (.03) d (.97)	d	d	d	d	d

TABLE 3. CONTINUED.

Locus	Population																
	1	2	3	4	6	8	11	12	14	16	17	27	30	31	32	36	37
Got-1	j	i (.87) j (.13)	i	h (.31) i (.63) m (.06)	h (.15) i (.67) m (.18)	i	i	a (.69) i (.31)	f (.18) h (.55) j (.27)	j (.60) m (.40)	m	j (.92) m (.08)	g (.06) j (.73) m (.21)	b (.89) e (.11)	c (.70) g (.10) i (.20)	g (.21) i (.79)	j
Got-2	b	b	b	b	a (.03) b (.97)	b	b	b	b	a (.04) b (.96)	b	b	a (.15) b (.85)	a (.06) b (.94)	b	b	b
Gpd	d	d	c (.04) d (.96)	d (.19) g (.75) l (.06)	d (.03) g (.50) h (.17) k (.10) l (.20)	d	d (.25) f (.75)	d	c (.60) d (.37) i (.03)	d	h	h (.61) l (.33) p (.06)	h (.41) m (.59)	d (.11) h (.78) r (.11)	m	m	m
Pep	g (.31) h (.69)	i	c (.25) h (.68) k (.07)	c (.25) f (.59) j (.16)	c (.37) f (.60) j (.03)	j	h (.92) j (.08)	c (.31) g (.69)	c	d	d	c (.06) d (.47) g (.47)	c (.73) e (.27)	c (.06) d (.88) g (.06)	e	c	e
Mpi	a (.50) d (.04) i (.46)	i	o	i	i (.97) n (.03)	i (.05) l (.95)	h (.67) m (.33)	a	c (.04) f (.83) k (.13)	k	j	h (.08) j (.70) n (.22)	h (.67) j (.12) q (.21)	d (.11) j (.61) o (.28)	e (.70) i (.30)	i	i
Pt-2	c	c	c	e	e	e	e	c	d	d	c	c	c	e	f	f	c (.17) d (.83)
$\tilde{H}$[1]	.12	.07	.06	.13	.15	.04	.13	.09	.09	.03	—	.19	.21	.14	.08	.03	.05

[1] Determined by direct count of homozygotes and heterozygotes for populations of $N \geq 5$.

TABLE 3. (CONTINUED.)

Locus	Population																	
	39	41	45	46	50	51	52	54	55	57	59	61	62	63	64	66	67	69
Lap	c	e (.69)	c	c (.95)	c	c	f	f	f	b (.45) c (.55)	e	c	d (.03) g (.97)	f	g	g	a (.03) b (.97)	c
Icd-1	d	d	d	d	d	d (.97) b (.03)	b (.83) d (.17)	d	a (.50) d (.50)	d	d	d	b	d	d (.38) f (.62)	a (.05) d (.95)	b (.02) d (.90) e (.08)	b (.06) d (.94)
Icd-2	c	b (.50) c (.50)	c	c	c	c (.97) d (.03)	c	c	c	c	c (.97) d (.03)	b (.28) c (.72)	c	c	b (.03) c (.97)	b (.40) c (.30) f (.30)	c	a (.75) c (.25)
Gapdh	g	f	f	b	b	b	c	c	c	f	b (.40) f (.60)	f	f	f	b	b	b	b
Mdh-1	i	e	e	j	i	j	b (.17) e (.83)	f	c	b	e	e	e	e (.87) j (.13)	e	b (.11) e (.89)	b	b (.89) d (.11)
Mdh-2	e	e (.06) j (.94)	g	c (.15) h (.85)	h	h	e (.67) g (.33)	e	k	b (.80) e (.20)	e	e	e	e	e	e	e (.97) l (.03)	g
Pgm	e	e	e	b (.03) e (.97)	e	e (.97) f (.03)	c (.50) e (.50)	e (.50) f (.50)	e	e (.35) f (.65)	e	e	e	e	e	c (.31) e (.69)	e (.90) f (.10)	e
Gpi	h	c (.69) d (.06) h (.25)	c	e (.03) g (.70) j (.27)	k	i	f	f	f	f	a (.05) f (.62) k (.33)	f (.83) k (.17)	c (.08) f (.92)	b (.02) f (.98)	d (.10) f (.90)	f	g (.97) i (.03)	g
Ldh-1	l	l	l	q	e (.50) i (.50)	d (.06) g (.94)	d	d	d	l	l	i (.78) o (.22)	e	i	a (.03) e (.97)	m	k	d (.06) l (.94)
Ldh-2	d	d	b	d	d	d (.97) f (.03)	d	d	d	d	a (.03) d (.97)	d	d	d	d	d	a (.03) d (.97)	a (.06) d (.94)
Got-1	g (.13) j (.87)	j	g	c (.03) g (.94) n (.03)	g	e (.25) g (.75)	l	l	l	l (.90) n (.10)	l (.48) n (.52)	j (.11) l (.89)	l	l	n	l (.31) n (.69)	e (.12) f (.20) h (.17) i (.35) j (.05) k (.08) l (.03)	l

TABLE 3. (CONTINUED.)

Locus	Population																	
	39	41	45	46	50	51	52	54	55	57	59	61	62	63	64	66	67	69
Got-2	b	a (.19) b (.81)	b	a (.03) b (.97)	b	b	b	b	b	b	a (.03) b (.94) c (.03)	b	a (.03) b (.97)	b	b (.77) c (.23)	b	b	b
Gpd	m	h (.43) j (.31) m (.25)	g	d (.74) f (.20) h (.03) k (.03)	d	d (.84) h (.16)	h (.17) n (.83)	n	n	h	h	c (.17) h (.83)	h	h (.43) n (.57)	g	h (.95) l (.05)	n	n (.81) q (.19)
Pep	c (.30) d (.70)	c (.31) e (.69)	e	e	e	b (.06) e (.94)	d	b	b	d (.10) e (.90)	b (.10) c (.87) e (.03)	b (.22) c (.50) e (.28)	c	b	c	c (.45) e (.55)	f	b (.56) d (.38) e (.06)
Mpi	i	b (.13) f (.06) i (.81)	g	e (.08) i (.69) l (.08) m (.10) p (.05)	m	i (.44) m (.53) r (.03)	e (.83) j (.17)	j	f	e (.05) i (.90) l (.05)	c (.03) f (.97)	f	f	f	f	f (.85) j (.15)	i	i (.81) s (.19)
Pt-2	d	c	c	b (.03) e (.97)	e	e	c	c	c	c	e	c (.72) e (.28)	c (.97) e (.03)	c	c	c	c	c (.06) d (.94)
$\bar{H}^1$	.06	.13	—	.12	—	.11	.08	—	—	.13	.13	.14	.02	.05	.08	.18	.08	.14

TABLE 4. COEFFICIENTS OF NEI'S D (ABOVE THE DIAGONAL) AND ROGERS' S (BELOW THE DIAGONAL) AMONG 35 POPULATIONS OF *Thorius*. Comparisons between populations of described species from type localities are underlined.

	1	2	3	4	6	8	11	12	14	16	17	27	30	31	32	36	37
1	—	.39	1.00	1.30	1.09	1.04	1.01	.85	1.02	1.00	.89	1.02	1.17	1.78	1.22	1.16	1.14
2	.65	—	.75	.96	1.07	.68	.92	.95	.82	.92	.86	1.20	1.28	1.73	1.10	.90	1.14
3	.30	.46	—	.39	.45	.75	.93	.56	1.30	1.42	1.19	.81	.81	1.02	1.35	1.41	1.43
4	.35	.38	.64	—	.07	.55	.75	.94	1.10	1.24	1.31	.78	.81	.72	1.19	1.14	1.13
6	.34	.34	.64	.93	—	.67	.77	.87	1.16	1.30	1.30	.78	.81	.74	1.25	1.26	1.17
8	.35	.51	.47	.58	.51	—	.58	1.20	1.01	1.05	.96	.99	1.06	.91	1.07	.98	1.17
11	.37	.38	.39	.47	.46	.56	—	1.37	.85	.89	1.12	.93	.94	.89	1.27	1.16	1.40
12	.43	.39	.57	.39	.42	.30	.25	—	1.36	1.49	1.34	1.05	1.14	1.91	1.58	1.78	1.54
14	.36	.44	.27	.33	.31	.37	.43	.26	—	.26	1.34	1.30	1.23	1.56	1.59	1.32	1.01
16	.36	.40	.24	.29	.27	.35	.41	.23	.77	—	1.08	1.17	1.40	1.36	1.64	1.66	1.06
17	.41	.42	.30	.27	.27	.38	.33	.26	.26	.34	—	.53	.59	.73	.80	.82	.81
27	.37	.32	.45	.46	.46	.37	.40	.35	.27	.31	.59	—	.17	.38	.94	1.12	.81
30	.31	.28	.45	.45	.45	.35	.39	.32	.29	.25	.55	.84	—	.48	.69	.78	.61
31	.17	.18	.36	.49	.48	.40	.41	.15	.21	.26	.48	.69	.62	—	1.30	1.30	1.37
32	.30	.33	.26	.30	.20	.34	.28	.21	.20	.19	.45	.39	.50	.27	—	.21	.23
36	.31	.41	.24	.32	.31	.38	.31	.17	.27	.19	.44	.33	.46	.27	.81	—	.36
37	.32	.32	.24	.32	.31	.31	.25	.21	.36	.35	.44	.45	.54	.25	.79	.70	—
39	.31	.31	.25	.35	.33	.33	.26	.21	.37	.40	.49	.47	.54	.32	.74	.73	.94
41	.30	.28	.36	.44	.44	.32	.32	.26	.23	.21	.42	.83	.79	.54	.48	.41	.58
45	.28	.29	.37	.45	.42	.32	.39	.33	.26	.25	.38	.50	.50	.32	.46	.33	.45
46	.37	.40	.29	.40	.38	.46	.38	.19	.22	.21	.40	.36	.34	.42	.57	.53	.50
50	.38	.40	.31	.35	.32	.45	.40	.20	.19	.41	.32	.32	.41	.59	.53	.51	.46
51	.37	.39	.30	.39	.36	.46	.37	.19	.21	.20	.40	.33	.33	.43	.55	.50	.48
52	.29	.30	.29	.31	.30	.36	.30	.32	.27	.33	.53	.49	.47	.40	.34	.29	.30
54	.29	.28	.28	.24	.24	.29	.23	.27	.20	.19	.54	.48	.48	.35	.36	.42	.36
55	.36	.30	.26	.20	.19	.26	.21	.20	.19	.13	.41	.38	.38	.30	.23	.23	.22
57	.43	.40	.35	.37	.39	.34	.34	.35	.25	.24	.61	.62	.62	.43	.50	.40	.55
59	.22	.24	.38	.55	.56	.44	.43	.27	.37	.24	.50	.67	.75	.63	.50	.48	.46
61	.33	.34	.30	.36	.36	.47	.54	.24	.43	.32	.62	.65	.71	.57	.49	.50	.49
62	.34	.35	.31	.35	.35	.38	.39	.28	.38	.25	.56	.56	.63	.44	.32	.38	.32
63	.29	.30	.31	.33	.32	.38	.46	.27	.32	.25	.54	.61	.64	.47	.40	.39	.39
64	.39	40	.31	.39	.37	.37	.35	.27	.38	.24	.52	.44	.56	.35	.41	.47	.33
66	.35	.35	.31	.33	.33	.36	.28	.36	.24	.52	.55	.55	.62	.45	.41	.45	.34
67	.40	.46	.33	.40	.39	.42	.29	.28	.22	.20	.57	.47	.51	.33	.48	.53	.45
69	.35	.39	.29	.36	.34	.36	.31	.23	.30	.31	.57	.46	.51	.37	.58	.54	.58

trial species is sympatric with *T. macdougalli* at three localities south of the crest in the vicinity of Llano de las Flores (Table 5, localities IV–VI). These two species are then joined by a third terrestrial species at locality VII, just below and north of the crest. This latter species, in turn, is sympatric at locality VIII with a fourth, arboreal and terrestrial species which is endemic to the cloud forest at intermediate elevations on the north slope (see below). Previously unreported instances of sympatry, each with two species, were detected at four Oaxacan localities (Table 5, localities IX–XII); Nei's D between sympatric populations ranged from 0.46 to 1.27.

Cluster analysis.—All 69 populations were clustered using the UPGMA method for both D and S. Generally, most populations fall into discrete clusters whose composition is the same for both distance measures; only the dendrogram of D is presented (Fig. 2).

Populations from Veracruz and Puebla form at least six distinct groups: a) the two subspecies of *T. pennatulus* (1 and 2); b) *T. troglodytes* (4)

TABLE 4. (CONTINUED.)

39	41	45	46	50	51	52	54	55	57	59	61	62	63	64	66	67	69
1.18	1.21	1.28	1.00	.96	1.01	1.24	1.23	1.02	.84	1.50	1.10	1.10	1.26	.92	1.06	.91	1.04
1.19	1.28	1.23	.91	.93	.94	1.22	1.27	1.22	.91	1.44	1.08	1.05	1.21	.89	1.05	.78	.94
1.39	1.01	1.00	1.23	1.19	1.19	1.24	1.29	1.34	1.06	.98	1.22	1.16	1.17	1.18	1.17	1.10	1.24
1.07	.81	.80	.91	1.05	.94	1.18	1.44	1.60	.99	1.02	1.06	1.11	.94	.94	1.12	.92	1.03
1.11	.83	.87	.98	1.13	1.01	1.20	1.44	1.69	.95	.58	1.03	1.04	1.14	1.01	1.10	.94	1.08
1.11	1.16	1.15	.78	.79	.79	1.01	1.23	1.35	1.08	.82	.75	.97	.97	.99	1.03	.88	1.03
1.33	1.13	.94	.98	.91	1.01	1.20	1.47	1.58	1.08	.84	.61	.94	.78	1.09	1.13	1.24	1.19
1.58	1.37	1.12	1.67	1.61	1.64	1.16	1.33	1.61	1.06	1.33	1.41	1.29	1.32	1.32	1.28	1.27	1.48
.99	1.49	1.33	1.53	1.60	1.57	1.31	1.61	1.66	1.40	1.00	.84	.96	1.15	.98	1.03	1.53	1.22
.91	1.55	1.38	1.58	1.65	1.63	1.12	1.65	2.07	1.41	1.44	1.15	1.40	1.39	1.44	1.47	1.63	1.17
.71	.88	.98	.93	.89	.92	.64	.62	.89	.49	.69	.48	.58	.62	.66	.66	.56	.56
.76	.18	.70	1.09	1.15	1.11	.71	.73	.96	.47	.40	.44	.59	.50	.76	.61	.71	.78
.62	.24	.70	1.09	1.14	1.12	.77	.73	.98	.48	.29	.34	.47	.45	.59	.48	.68	.68
1.16	.61	1.14	.87	.89	.85	.91	1.05	1.19	.84	.47	.57	.83	.75	1.07	.80	1.12	1.00
.30	.73	.78	.57	.52	.61	1.08	1.02	1.48	.70	.69	.71	1.13	.93	.90	.89	.74	.55
.31	.89	1.11	.65	.64	.70	1.23	.87	1.49	.91	.73	.70	.97	.95	.76	.81	.64	.62
.06	.55	.81	.69	.68	.74	1.19	1.01	1.50	.60	.79	.72	1.15	.94	1.10	1.09	.80	.54
—	.63	.94	.78	.77	.84	1.07	1.03	1.48	.74	.72	.73	1.08	.94	1.03	1.07	.79	.47
.53	—	.50	.86	1.02	.92	.98	1.04	1.15	.45	.45	.48	.62	.61	.78	.66	.81	.86
.39	.61	—	.79	.81	.83	1.13	1.25	1.50	.66	.72	.64	1.08	.82	.93	1.07	1.16	1.04
.46	.42	.46	—	.25	.08	1.33	1.20	1.46	.79	.80	.80	1.32	1.06	1.04	1.05	.80	.74
.36	.36	.45	.78	—	.22	1.39	1.24	1.36	.94	.77	.75	1.24	1.03	1.00	1.10	.96	.86
.43	.40	.44	.93	.80	—	1.35	1.21	1.45	.83	.80	.79	1.31	1.06	1.07	1.05	.86	.78
.35	.38	.32	.27	.25	.26	—	.38	.56	.72	.79	.61	.51	.42	.76	.68	.68	.70
.36	.35	.29	.30	.29	.30	.69	—	.44	.67	.81	.61	.75	.33	.85	.76	.59	.60
.23	.32	.22	.23	.26	.24	.57	.65	—	.95	.97	.70	.69	.43	.80	.80	.87	.76
.48	.64	.52	.45	.39	.44	.49	.51	.39	—	.55	.40	.59	.51	.94	.74	.50	.46
.49	.64	.44	.45	.45	.45	.45	.45	.38	.58	—	.24	.36	.39	.44	.37	.77	.61
.48	.62	.53	.45	.47	.45	.55	.55	.50	.67	.78	—	.24	.15	.47	.34	.69	.54
.34	.54	.38	.27	.29	.27	.60	.47	.50	.56	.70	.78	—	.33	.29	.29	.81	.87
.39	.55	.44	.35	.36	.35	.66	.72	.65	.60	.68	.86	.72	—	.53	.44	.59	.57
.36	.46	.40	.36	.37	.34	.47	.43	.45	.39	.65	.62	.75	.59	—	.21	.64	.83
.34	.52	.34	.35	.33	.35	.51	.47	.45	.48	.69	.71	.75	.65	.81	—	.63	.44
.45	.44	.31	.45	.38	.43	.51	.55	.42	.61	.46	.50	.44	.56	.53	.54	—	.41
.62	.43	.35	.48	.42	.46	.50	.55	.47	.63	.54	.58	.42	.56	.44	.49	.67	—

from Puerto del Aire, the type locality above Acultzingo, and nearby populations (5–7); e) *T. dubitus* (3), also from Puerto del Aire; d) the third, cryptic, arboreal species sympatric at Puerto del Aire (12); e) *T. schmidti* from Zoquitlán (10 and 11) and the first of two species sympatric at montane localities on Volcán Orizaba (8 and 9); f) the second of two species sympatric on Volcán Orizaba (13–15) and Las Vigas (16).

Most populations from Oaxaca and Guerrero also form robust clusters that remain intact regardless of distance measure. *Thorius narisovalis* from the type locality on Cerro San Felipe (46–48), Cuajimoloyas (49), a locality approximately 40 km southeast in the same Sierra Aloápaneca, and two montane populations from the opposite side of the Oaxaca valley, Zaachila (51) and Tlaxiaco (50), represent one distinct cluster; the average pairwise D between each of the two western populations and the four eastern populations is 0.082 (Zaachila) and 0.259 (Tlaxiaco). *Thorius pulmonaris* from moderate elevations on Cerro San Felipe (52 and 53) is well

TABLE 5. LOCALITIES AT WHICH TWO OR THREE SPECIES OF *Thorius* OCCUR IN SYMPATRY.

	Locality	Species	Population (N)	No. loci showing fixed differences	D
I.	Ver.: 3.2 km S Puerto del Aire	*T. dubitus*	3 (18)	Pop. 3 vs 4: 3	0.39
		T. troglodytes	4 (16)	Pop. 3 vs 12: 5	0.56
		T. sp. A	12 (7)	Pop. 4 vs 12: 9	0.94
II.	Ver.: Volcán Orizaba: El Berro	*T.* sp. B	13 (8)	10	1.03
		T. schmidti	8 (22)		
III.	Ver.: Volcán Orizaba: Xometla	*T.* sp. B	15 (23)	11	1.19
		T. schmidti	9 (17)		
IV.	Oax.: Cerro Pelón: 25 km N Guelatao (Llano de las Flores)	*T. macdougalli*	22 (18)	8	1.09
		T. sp. C	32 (5)		
V.	Oax.: Cerro Pelón: 26 km N Guelatao (Llano de las Flores)	*T. macdougalli*	23 (20)	8	1.04
		T sp. C	33 (1)		
VI.	Oax.: Cerro Pelón: 30.5 km N Guelatao	*T. macdougalli*	24 (20)	8	1.03
		T. sp. C	34 (5)		
VII.	Oax.: Cerro Pelón: 51 km N Guelatao	*T. macdougalli*	30 (17)	Pop. 30 vs 36: 8	0.78
		T sp. C	36 (10)	Pop. 30 vs 39: 6	0.62
		T. sp. D	39 (4)	Pop. 36 vs 39: 3	0.31
VIII.	Oax.: Cerro Pelón: 52 km N Guelatao	*T.* sp. D	38 (11)	4	0.64
		T. sp. E	40 (8)		
IX.	Oax.: 15 km NE San Juan del Estado	uncertain	57 (10)	3	0.46
		T. sp. G	69 (9)		
X.	Oax.: 29.5 km NE Tlaxiaco	*T. narisovalis*	50 (1)	6	0.75
		uncertain	61 (9)		
XI.	Oax.: 25 km W Zaachila	*T. narisovalis*	51 (16)	12	1.27
		T. pulmonaris	56 (3)		
XII.	Oax.: 15.5 km W San Vicente Lachixio	uncertain	66 (5)	4	0.44
		uncertain	63 (18)		

differentiated genetically from *T. narisovalis* from higher elevations (mean D between 52–53 and 46–48 equals 1.36), but demonstrates a relatively close affinity with populations north and west of the Oaxaca valley at Tejocotes (54) and Zaachila (55, 56), respectively. A third cluster unites three populations from north of the Oaxaca valley (67–69), while a fourth group includes all three Guerreran populations (58–60).

Populations from Cerro Pelón in the Sierra de Juárez basically define four discrete clusters: *T. macdougalli* (18–30), although populations north of Machín (28–30) form a distinct subgroup relative to those at Machín and south (18–27); populations 32–36; populations 37–39; populations 40–44. Three populations do not demonstrate close genetic affinity to any other population. Population 17, represented by a single specimen which, based on external coloration and proportions, resembles *T. macdougalli*, was collected from the lower elevational limit of this species on the south slope of Cerro Pelón less than 6 km from the nearest sampled population (18). Yet, it clusters neither with *T. macdougalli* nor with any other population from Cerro Pelón. Likewise population 45, a single specimen taken in a bromeliad at Vista Hermosa, represents the lower elevational record of *Thorius* from the cloud forest on the north slope but shows little genetic affinity to the adjacent populations which it resembles in external morphology (mean D between 45 and 40–44 equals 0.541), and clusters remotely from them. Lastly, the sample from Totontepec (31), a village located in the Sierra de Juárez approximately 50 km southeast of Cerro Pelón, morphologically resembles *T. macdougalli*, but clusters with the main group of *T. macdougalli* from Cerro Pelón (18–30) only after the latter joins the cloud forest species from the north slope.

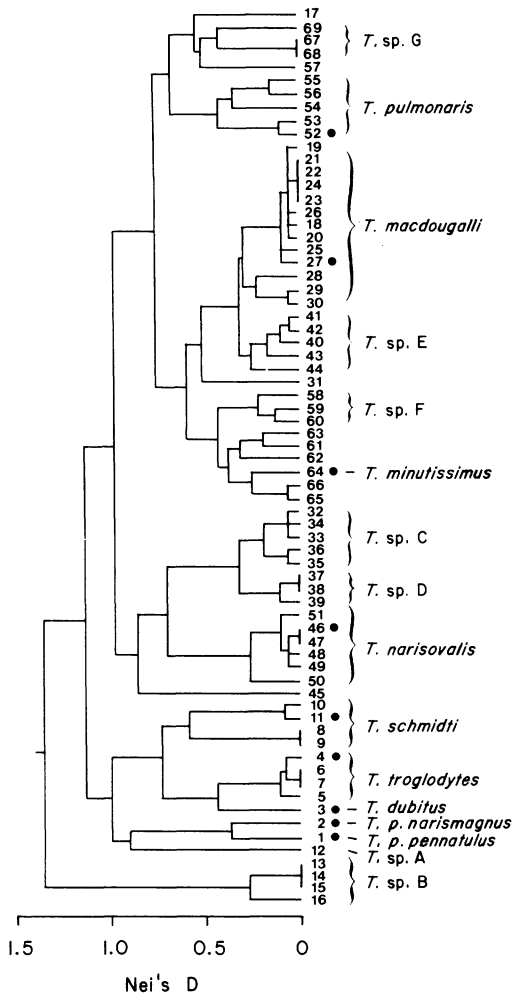

Fig. 2. UPGMA dendrogram based on Nei's D among 69 populations of *Thorius*. Closed circles denote populations from type localities, except population 64 which is the designated reference sample of *T. minutissimus*. Populations are numbered as in Table 1. Suggested species designations are indicated for most populations; the identity of populations 17, 31, 45, 57, 61–63 and 65–66 is uncertain (see text). Cophenetic correlation coefficient equals 0.85.

Six additional Oaxacan populations cluster differently according to which distance measure is used, and thereby present an inconsistent pattern of genetic relationship. Population 57 (San Juan del Estado) demonstrates no consistent affinity to any other population. In the dendrogram of S (not illustrated) it first joins geographically distant population 17 from Cerro Pelón (S = 0.58), whereas in the dendrogram of D (Fig. 2) it clusters with population 17 only after linking with populations 67 and 68 (Santos Reyes Pápalo) and sympatric population 69. The remaining five populations (61–66) comprise a single cluster relative to all other populations in both dendrograms, but the branching sequence among the five populations differs slightly in each: using S, Suchixtepec (62) clusters first with Sola de Vega (64) (Fig. 2).

To examine the effects of different clustering algorithms on the resulting branching sequence, alternate dendrograms were derived using the Fitch-Margoliash and distance Wagner methods based on D (=1 − S) for a "collapsed" matrix of 27 populations (Fig. 3B, C). In this matrix, populations that clustered consistently in the original UPGMA dendrograms of 69 populations were represented instead by one or two populations. For comparison, a UPGMA dendrogram of S was also generated for the smaller matrix (Fig. 3A). Of particular interest was the effect that changing the clustering algorithm would have on the branching sequence of those populations that had either ambiguous or anomalous patterns in the earlier UPGMA dendrograms, viz. 17, 31, 45, 57, 61–66.

All three branching patterns based on the smaller matrix differ from those of the larger matrix and from each other. The Wagner tree is most different (Fig. 3C). *Thorius narisovalis* (46), for example, which clusters with populations 32, 37 and 45 in all other trees (Figs. 2, 3A, B), joins them only after most other populations. And population 57, which joins populations 41 and 27 in the Fitch-Margoliash and UPGMA (smaller matrix) trees, instead first joins population 69. Fitch-Margoliash and UPGMA dendrograms based on the smaller matrix both differentiate populations 64 (*T. minutissimus*) and 66 from 61–63, as does the larger UPGMA dendrogram of D, but they further ally the latter cluster with the Guerreran populations (e.g., 59), which is not seen in either large dendrogram; this pattern is disrupted almost completely in the Wagner tree. The anomalous position of populations 17, 31 and 45 still remains in all of the smaller ones.

Local genetic differentiation in Thorius macdougalli.—Several species each exhibit a pattern of relatively great genetic differentiation among populations within a limited geographic area. In most instances, significant between-locality

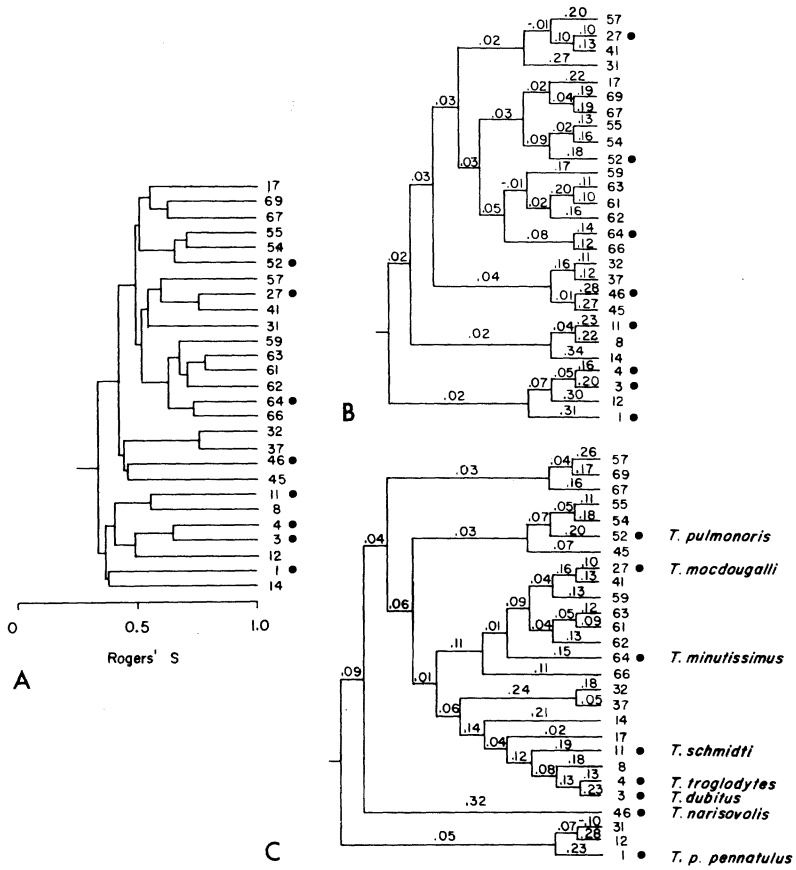

Fig. 3. Three dendrograms based on Rogers' S (A) or D (=1 − S) (B and C) among 27 populations of *Thorius* ("collapsed" matrix). A: UPGMA; cophenetic correlation coefficient equals 0.91. B: Fitch-Margoliash method; % SD = 11.1. C: Distance Wagner procedure; deviation ratio equals 0.37. Closed circles denote populations from type localities, except population 64 which is the designated reference sample of *T. minutissimus*.

difference in allele frequency is limited to a single locus. Fourteen population samples of *T. macdougalli* along a stretch of Mexican Highway 175 that crosses the Sierra de Juárez between Guelatao and Vista Hermosa (Table 1, populations 17–30) provide an opportunity to examine an extreme example of local genetic differentiation, involving several loci, in greater detail than for most species. Sample size at each locality ranged from 1 to 20 ($\bar{x}$ = 11.7).

Allele frequencies of the nine populations with adequate samples (N ≥ 8) reveal high levels of genetic variability. The frequency of polymorphic loci ($\bar{P}$) equals 53.5% (range 37.5–62.5%), and the mean heterozygosity per population ($\bar{H}$) equals 0.13 (range 0.08–0.21). Only one locus, Gapdh, is fixed for the same allele in all populations, although six additional loci—Lap, Icd-1, Pgm, Ldh-2, Got-1 and Pt-2—display only slight variation in allele frequencies. Between-locality variation is greater in Mdh-1, Gpi, Got-2 and Gpd; at each of these loci, all populations share the same common allele, although exact frequencies may differ. Extreme variation is displayed by the five loci remaining: Icd-1, Ldh-1, Mdh-2, Pep and Mpi (Fig. 4). All of these demonstrate a cline in allele frequencies in which the end points, populations 18 and 30, either share no alleles (Mdh-2) or show pronounced differences in the frequency of shared alleles. The steepest portion of the cline occurs between populations 26 and 29 which are sepa-

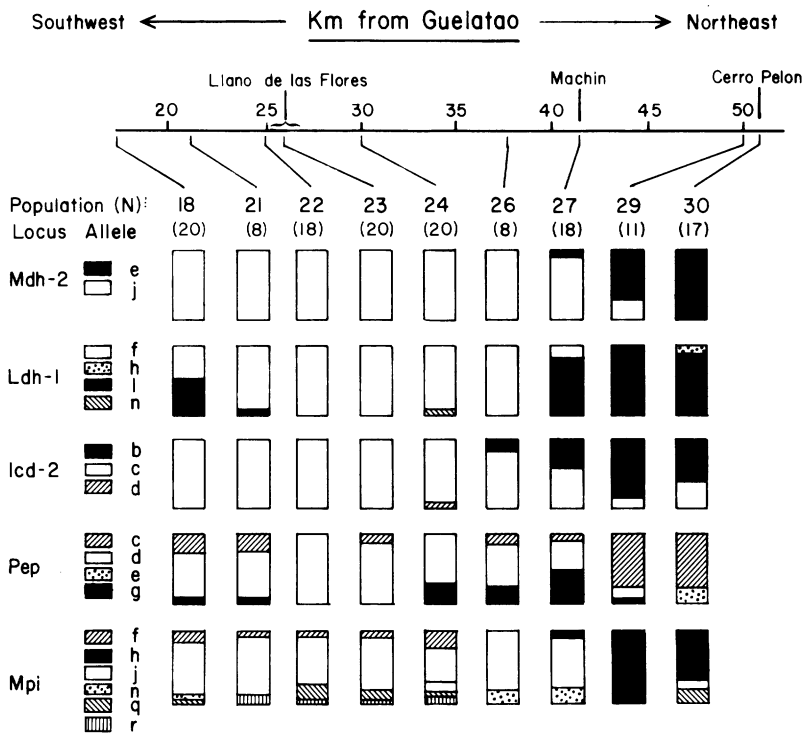

Fig. 4. Allele frequencies at five variable loci in *T. macdougalli*. The nine populations comprise a transect along Mexican Highway 175 between Guelatao and Vista Hermosa in the Sierra de Juárez, Oax. Elevations of all localities range from 2,725 to 2,955 m.

rated by a road distance of only 12.6 km. Consequently, D between populations 26 and 29 is virtually identical to that between populations 18 and 30—0.27 and 0.24, respectively. This break, which occurs near Machín (27) and only a few km southeast of the abrupt transition between cloud forest and the seasonally wet pine-oak-fir forest on the north slope of Cerro Pelón, was detected by the UPGMA cluster analysis (Fig. 2), in which populations 18–27 clustered separately from 28–30 (mean D between 18–27 and 28–30 equals 0.282, range 0.11–0.39). However, distinguishing between these two groups of populations masks the continuous (albeit steep) change of allele frequencies between populations 26 and 29; for example, the mean D between population 27 and 28–30 equals 0.139 (range 0.11–0.17), or less than half the average value between the two groups. This continuous transition of allele frequencies along the transect suggests that, notwithstanding the substantial differentiation, there is at least some gene flow among these populations.

Discussion

Taxonomic implications.—Electrophoretic analysis has established the genetic distinction of all but one of the ten formally described species and subspecies. The mean pairwise D among single populations of all species from their respective type localities (except *T. maxillabrochus*; see below) equals 0.975 (range 0.39–1.30); 15 of the 28 individual comparisons exceed D = 0.9. These values, although high for interspecific comparisons within most vertebrate genera (Ayala, 1975), are not uncommon for plethodontid salamanders, in which interspecific comparisons often exceed D = 1.0 (e.g., *Batrachoseps*—Yanev, 1978; *Bolitoglossa*—Hanken and Wake, 1982; and *Plethodon*—Highton and Larson, 1979). Genetic differentiation between disjunct populations of *T. pennatulus* (1, 2) which are described as separate subspecies is less than that observed among most species—D = 0.39. This value, however, equals that recorded between sympatric *T. dubitus* (3) and *T. troglodytes*

(4), and is greater than that observed between one pair of sympatric species (Table 5, locality VII). Thus, while there is a tendency for increased genetic differentiation with increased taxonomic rank, there is no absolute distinction, in terms of genetic distance value, between interspecific and intersubspecific comparisons.

There was no biochemical evidence of more than a single species in the samples from near Zoquitlán, Pue., the type locality of *T. schmidti* and *T. maxillabrochus*. This supports the suggestion of F. R. Gehlbach (pers. comm.) that there is only a single species at this locality. It is conceivable, however, that a second species, though present, was included in neither sample; therefore, the electrophoretic data do not provide sufficient evidence to resolve this question entirely. Pending morphological analysis of these populations, I have referred the samples from Zoquitlán to *T. schmidti*.

Several populations, singly or in groups, appear to represent additional, undescribed species. For many populations, this interpretation is supported by their sympatric occurrence with other species and the demonstrated maintenance of separate gene pools. This "sympatry test" provides unequivocal and convincing evidence of reproductive isolation; electrophoretic data have been used effectively in this manner to confirm the existence of sympatric species of many plethodontid genera, including *Batrachoseps* (Yanev, 1978), *Desmognathus* (Tilley et al., 1978) and *Plethodon* (Highton, 1979). In some of these studies, sympatric species had been suspected earlier on the basis of morphology, whereas in others, electrophoresis detected cryptic species that previously had eluded detection. Both instances were observed in *Thorius*. For example, one or two additional, undescribed species was suspected at each of three distant localities: Puerto del Aire, Ver.; Volcán Orizaba, Ver.; and Cerro Pelón, Oax. (see above). All of these "suspicions" proved correct (Table 5, localities I, II–III, and IV–VII, respectively). On the other hand, five instances of sympatry, involving two described species and at least three undescribed species, had not been reported (Table 5, localities VIII–XII).

In the absence of the sympatry test, one must infer the taxonomic status of allopatric populations from protein data on the basis of observed levels of differentiation and clustering patterns. However, at present, no satisfactory criterion exists by which to evaluate the taxonomic status of allopatric populations from observed levels of protein differentiation in the absence of significant morphological differentiation (Wake, 1981). The difficulty is particularly acute in *Thorius*, in which many species, for instance *T. macdougalli*, demonstrate the capacity for significant protein differentiation among populations separated by relatively short geographic distance, apparently despite gene flow (also see Slatkin, 1981, for a discussion of protein divergence of local populations of salamanders in the face of gene flow). For this reason, I have chosen not to use a certain minimum D value as a sufficient criterion, by itself, for recognition of any given pair of allopatric populations as belonging to different species. Nevertheless, the protein differentiation of many populations relative to described species is in many cases sufficiently large—sometimes exceeding values observed in pairwise comparisons between populations of any pair of described species—to warrant considering these populations as distinct species. In Veracruz, for example, populations 13–15, one of two species sympatric on Volcán Orizaba, display close affinity only to population 16 (Las Vigas), nearly 100 km to the north (mean pairwise D between 16 and 13–15 equals 0.253). Together, these four populations display only a remote genetic affinity to any other population: the minimum D between any of the four populations and any remaining population equals 0.82 (13 vs 2), whereas most comparisons exceed D = 1.0 and in a single comparison D = 2.07 (16 vs 55).

The following populations deserve consideration as distinct species: *T.* sp. A—12; *T.* sp. B—13–16; *T.* sp. C—32–36; *T.* sp. D—37–39; *T.* sp. E—40–44; *T.* sp. F—58–60; *T.* sp. G—67–69. The mean pairwise D between all but one of these species and each of the described species (except *T. maxillabrochus*) exceeds the minimum D between sympatric species by at least 0.15 units, and usually much more. The single exception is *T.* sp. E, which exhibits relatively little differentiation ($\bar{D}$ = 0.299) from *T. macdougalli* (18–30) with which it is nearly sympatric on Cerro Pelón (see below). Formal descriptions of these species are in preparation and will be presented elsewhere.

In each case that has been examined in detail, once conspecific populations are identified electrophoretically, morphological characters—often including osteological features—have been identified that unambiguously distinguish these species (Hanken, 1980). This is particularly significant in the case of cryptic species that

had not been detected previous to the electrophoretic analysis. For instance, *T.* sp. C and *T.* sp. D, two large terrestrial species that are sympatric on Cerro Pelón (Table 5, locality VII), are virtually indistinguishable based on external morphology, such as adult body size and proportions, although their ventral coloration differs slightly (dark in *T.* sp. C, pale in *T.* sp. D). Moreover, these species are the least differentiated genetically of any sympatric species pair ($\bar{D} = 0.31$). Yet, these species exhibit pronounced differences in skull morphology: the fronto-parietal fontanelle is broad and maxillary teeth are absent in *T.* sp. C, whereas the fontanelle is narrow and maxillary teeth are present in *T.* sp. D. In fact, the primitive cranial osteology of *T.* sp. D readily distinguishes it from all remaining species of *Thorius*, described and undescribed (Hanken, 1980). Thus, after initially sorting populations on the basis of the electrophoretic data, morphological characters that unequivocally distinguish these species were identified which had previously gone unnoticed. In this sense, and with few exceptions (see below), in *Thorius* the problem of evaluating the taxonomic identity of genetically well-differentiated populations that are morphologically indistinguishable may be moot—many presumptive species that were identified on the basis of electrophoretic evidence are distinguishable morphologically. Additional analyses have documented extensive intraspecific variation in osteological features in nearly all species (Hanken, 1980, 1982). In retrospect, this variation can partly explain the great difficulty of previous workers in elucidating taxonomic relationships by morphology alone.

Most remaining populations demonstrate consistent affinity, as revealed by cluster analysis, to either one of the formally described species or one of the seven additional undescribed species. Populations 8 and 9 from Volcán Orizaba consistently cluster first with *T. schmidti* from Zoquitlán (10, 11), yet these two pairs of populations are well differentiated from each other (mean pairwise D between 8–9 and 10–11 equals 0.603). Preliminary analysis (Hanken, 1980) failed to identify morphological features that distinguish these populations. Furthermore, all adults possess maxillary teeth which distinguish *T. schmidti* (including *T. maxillabrochus*) from all other described species. In the absence of any present evidence that would suggest referring these populations to different species (e.g., sympatry of intervening populations), I have referred populations 8 and 9 to *T. schmidti.* Electrophoretic data failed to provide unequivocal evidence of the patterns of genetic relationship of populations 61–63 and 65–66 (southwest Oaxaca); 17, 31 and 45 (Sierra de Juárez); and 57 (San Juan del Estado). For these populations, relationships depicted in the various dendrograms differed according to which clustering algorithm and genetic distance measure were used, and the number of populations included.

Prager and Wilson (1978) advocate the Fitch-Margoliash method as superior to both UPGMA and especially distance Wagner methods in cluster analyses based on electrophoretically-derived estimates of genetic distance. More recently, Farris (1981) discussed properties of both Nei's and Rogers' distance measures that may qualify their use in phylogenetic reconstruction based on electrophoretic data and yield erroneous results. In this study, dendrograms based on D and S for the large matrix yielded almost the same cluster pattern. Using the collapsed matrix of S for 27 populations, Fitch-Margoliash and UPGMA trees also were virtually identical, but differed markedly from the Wagner tree (Fig. 3). Furthermore, in the case of the errant southwest Oaxacan populations, cluster patterns in the former trees (Fig. 3A, B) are congruent with morphological evidence: based on adult body size, populations 65 and 66 resemble *T. minutissimus* (64), whereas populations 61, 62 and 63 resemble the much larger, undescribed species from Guerrero, *T.* sp. F (58–60). None of the dendrograms, however, closely ally populations 17, 31 and 45 to the remaining Sierra de Juárez populations as would be predicted based on morphological similarity or geographical proximity. Populations 17 and 45 each comprise single specimens; their anomalous cluster patterns may be ascribed to sampling error that gives erroneous genetic distances when they are based on a limited number of loci and small samples (see Gorman and Renzi, 1979; Tilley et al., 1978). But population 31 comprises 9 specimens; its remote clustering from the remaining species *T. macdougalli* cannot be as easily ascribed simply to sampling error. Lastly, population 57 demonstrates no consistent genetic affinity to any species. Clearly, electrophoretic evidence has identified the problematic populations and established their lack of close genetic affinity to other populations, but resolution of the taxonomic status of these populations awaits additional information from

other sources, or additional methods for analyzing electrophoretic data.

Ecological relationships.—The 15 species of *Thorius* comprise three faunal units that occupy different geographic regions along the southeastern edge of the Mexican plateau: eastern Puebla and Veracruz; Sierra de Juárez in northcentral Oaxaca; southern and western Oaxaca, and Guerrero. The known distribution of each species is contained within a single region, but species within a given region are not necessarily more closely related to one another (as suggested by genetic distance estimates) than to species in different regions. All but two species occur in montane forest above 2,100 m; *T. pennatulus* occurs in lowland forest between 800 and 1,200 m, *T.* sp. E has been taken as low as 1,530 m and as high as 2,475 m. *Thorius* sp. A from the cloud forest at Puerto del Aire, Ver., is exclusively arboreal, and *T.* sp. E is found both on the ground and in arboreal bromeliads; all other species are exclusively terrestrial.

Local endemism is common within regions. For instance, five species are found in montane forest in the vicinity of Volcán Orizaba near the Puebla-Veracruz border (Fig. 1). (*Thorius pennatulus*, a sixth species, also occurs nearby, but at lower elevations.) Both species sympatric at high elevations on Volcán Orizaba are also represented off of the volcano: *T. schmidti* occurs to the south in extreme northern Puebla; *T.* sp. B is found to the north at Las Vigas. Yet neither species occurs at Puerto del Aire, less than 40 km southeast, where three additional species, *T. dubitus*, *T. troglodytes* and *T.* sp. A—each known only from the immediate vicinity of Puerto del Aire—occur. On Cerro Pelón, Oax., four species endemic to the Sierra de Juárez display a complex yet regular pattern of species replacement, involving sympatry of two of three species, that reveals specific elevation and habitat preferences of each species (Fig. 5; Table 5, localities IV–VIII). *Thorius macdougalli*, the most abundant and widespread species, is distributed continuously above 2,230 m in the seasonally-wet pine and pine-oak-fir forests on the southwest face of the mountain, and barely extends into the perennially-wet cloud forest on the northeast, Caribbean face. At elevations of 2,850 m and higher it is joined by *T.* sp. C. These species are sympatric with *T.* sp. D at a single locality which, at 2,930 m, defines both the lower elevational limit of *T. macdougalli* and *T.* sp. C on the northeast face, and the upper

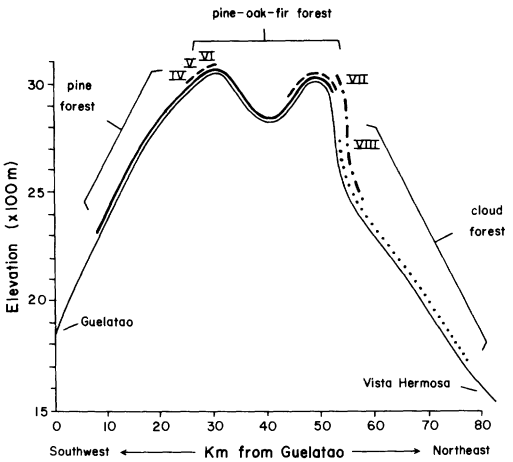

Fig. 5. Schematic cross section of the Sierra de Juárez, Oax., in the vicinity of Cerro Pelón, showing the distribution of four endemic species of *Thorius*: (——) *T. macdougalli*; (– – –) *T.* sp. C; (–·–·–) *T.* sp. D; (·····) *T.* sp. E. Roman numerals identify sympatric localities (Table 3). Distance NE Guelatao along Mexican Highway 175 is indicated on the horizontal axis. The approximate distribution of each of the three major vegetation types is also included.

elevational limit of *T.* sp. D. *Thorius* sp. D is sympatric with *T.* sp. E, the arboreal and terrestrial cloud forest endemic, beginning at 2,755 m, and has been taken as low as 2,475 m. The range of *T.* sp. E extends as low as 1,530 m (Vista Hermosa).

Regional differentiation, local endemism and elevational zonation are typical of other neotropical plethodontid genera, such as *Pseudoeurycea* (Bogert, 1967) and *Bolitoglossa* (Wake and Lynch, 1976), as well as a variety of other tropical vertebrate ectotherms (Bogert and Porter, 1967; Duellman, 1968; Huey, 1978). Moreover, many groups with geographic ranges that overlap that of *Thorius*, particularly those groups limited to moderate to high elevations, show similar patterns of species distribution, geographically and elevationally. For example, in addition to four species of *Thorius*, the list of vertebrates endemic to the Sierra de Juárez in the vicinity of Cerro Pelón includes two species of *Pseudoeurycea* (Regal, 1966; Wake, unpubl.), two species of the frog genus *Hyla* (Caldwell, 1974), one species of the lizard genus *Abronia* (Campbell, 1982), and two snake genera, *Cryophis* (Bogert and Duellman, 1963) and *Exiloboa* (Bogert, 1968), each of which is restricted to a

relatively narrow range of elevation and habitats. Bogert and Porter (1967) stressed the role of habitat and elevational restriction in effecting species diversification in *Abronia*, which is limited to montane pine-oak woodland and cloud forest and which exhibits a pattern of local endemism and regional differentiation that mimics that of *Thorius*. Wake and Lynch (1976) proposed a similar allopatric speciation model in their discussion of the evolution and diversification of neotropical plethodontid salamanders, especially the genera *Bolitoglossa*, *Pseudoeurycea* and *Chiropterotriton* (see also Wake, 1981). They suggested that habitat restriction and narrow elevational zonation—which is associated with narrowly specified thermal preference and limited acclimation ability (Feder, 1978)—promoted, or maintained, the isolation of once continuous populations which often were separated initially as a result of either tectonic events or climatic change. It seems likely that these characteristics—habitat restriction and narrow elevational zonation—which *Thorius* shares with other neotropical salamander genera and many tropical vertebrate ectotherms, have played a similarly large role in promoting species diversification in *Thorius* as they have in these groups, and that they are accountable, at least in part, for both the large number and present distribution of species.

Future studies.—Previous studies of the biology of *Thorius* have been hindered by the confusing systematics of the group and the inability to reliably establish the identity of specimens from most populations. In this study, electrophoretic data unraveled the complex web of taxonomic relationships and established the affinities of most populations. The total of 15 species ranks *Thorius* fourth among the 11 neotropical plethodontid genera behind *Bolitoglossa* (65 spp.), *Pseudoeurycea* (24 spp.), and *Oedipina* (16 spp.) (Wake and Elias, 1983). (Additional analysis may justify recognition of several more species; thus the 15 species may represent only a minimum estimate.) Furthermore, the range of habitat and microhabitat diversity collectively displayed by these species is comparable to or exceeds that of most neotropical genera (Wake and Lynch, 1976). This data has provided the information necessary to initiate studies of patterns of morphological diversification in *Thorius* that relate to phylogenetic size reduction (Hanken, 1980, 1982), and should allow further investigation of other aspects of its biology.

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APPENDIX 1. SPECIMENS EXAMINED (ARRANGED BY POPULATION NUMBER AS IN TABLE 1).*

1: M 2803, M 2834, M 5111–M 5122, M 5143, M 5143a; 2: M 3166–M 3170, M 3171a, M 3171b, M 3171c; 3: M 2945–M 2947, M 2950, M 2955, M 2957, M 2960, M 2963, M 2964, M 5545, M 5547–M 5554; 4: M 2938–M 2942, M 2953, M 2954, M 2956, M 2958, M 2959, M 2961, M 2962, M 5546, M 6006–M 6011; 5: M 6020; 6: M 2908–M 2937; 7: M 2848–M 2872; 8: M 3052, M 3053, M 3055–M 3057, M 3070–M 3080; 9: M 5172–M 5176, M 5179–M 5182, M 5184, M 5185, M 5187, M 5189–M 5191, M 5193, M 5194; 10: M 3909–M 3922, M 3928–M 3932; 11: M 3957–M 3962; 12: M 5537–M 5544; 13: M 3051, M 3054, M 3058–M 3061, M 3068–M 3069; 14: M 2985–M 2999, M 3000–M 3111; 15: M 5155–M 5171, M 5177, M 5178, M 5183, M 5186, M 5188, M 5195; 16: M 3968–M 3977, M 3983–M 3992; 17: M 4889; 18: M 3268–M 3272, M 3278–M 3292; 19: M 4893–M 4896; 20: M 4898–M 4899; 21: M 4974, M 4976–M 4982; 22: M 4950–M 4958, M 4964–M 4972; 23: M 3318–M 3337; 24: M 4901–M 4905, M 4907–M 4915, M 4921–M 4926; 25: M 3438; 26: M 4942–M 4949; 27: M 5007–M 5024; 28: M 5041–M 5042; 29: M 5028–M 5038; 30: M 3462, M 3464–M 3466, M 5060–M 5072; 31: JH 509–JH 514, JH 516–JH 518; 32: M 4959–M 4963; 33: M 3372; 34: M 4916–M 4920; 35: HBS 2085–HBS 2087; 36: M 3461, M 3467, M 5049–M 5051, M 5055–M 5059; 37: M 5077–M 5085; 38: HBS 2198–HBS 2200, HBS 2203–HBS 2206, HBS 2209–HBS 2212; 39: M 5040, M 5052–M 5054; 40: HBS 2220–HBS 2227; 41: M 3249, M 5096–M 5099, HBS 2257–HBS 2259; 42: HBS 2260; 43: M 3234–M 3236; 44: HBS 2266; 45: M 3208; 46: M 3471–M 3474, M 4514–M 4529; 47: M 3492–M 3495, M 4443–M 4457; 48: M 3553–M 3564, M 3569–M 3572; 49: M 3701–M 3705, M 3711–M 3725; 50: JH 388; 51: M 4427, M 4429, JH 488–JH 496, HBS 2046–HBS 2050; 52: HBS 2062–HBS 2064; 53: M 4546–M 4554, HBS 2058–HBS 2061; 54: M 4828; 55: M 4419; 56: M 4426, M 4428, HBS 2051; 57: HBS 2144, HBS 2146, HBS 2150–HBS 2152, HBS 2155–HBS 2158, HBS 2161; 58: M 6045–M 6047, M 6051–M 6062, M 6067–M 6069, M 6071, M 6072; 59: TJP 14595–TJP 14613; 60: TJP 14538, TJP 14540–TJP 14548; 61: JH 387, JH 389–JH 396; 62: M 4705–M 4724; 63: M 4763–M 4765, M 4767–M 4769, M 4771–M 4782; 64: M 4570–M 4583, M 4595, M 4596, M 4606–M 4608; 65: JH 596–JH 605; 66: M 4766, M 4770, JH 566, JH 572, JH 588; 67: M 3799–M 3801, M 3803–M 3812, M 3814, M 3817–M 3822; 68: M 3845–M 3864; 69: M 4824, HBS 2143, HBS 2145, HBS 2149, HBS 2153, HBS 2154, HBS 2169–HBS 2171.

* M = Museum of Vertebrate Zoology Mexican field tag series; JH = J. Hanken field tag series; TJP = T. J. Papenfuss field tag series; HBS = H. B. Shaffer field tag series. All specimens will be deposited in the MVZ permanent collection.