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Genetic Variation among Insular Populations of *Gekko hokouensis* (Reptilia: Squamata) near the Northeastern Borders of the Oriental and Palearctic Zoogeographic Regions in the Northern Ryukyus, Japan

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ABSTRACT—Genetic variation among populations of *Gekko hokouensis* from Kyushu, and the northern and the central Ryukyus was investigated by use of protein electrophoresis. From the results, the following three groups were recognized: Group 1, consisting of populations of Kyushu except for that of the southwestern part; Group 2, consisting of populations of southwestern Kyushu and the northern Ryukyus; and Group 3, consisting of populations of the central Ryukyus. Samples belonging to the Group 2 were genetically fairly uniform with low heterozygosities despite their wide geographic range that includes both the Oriental and Palearctic faunal regions delimited by the Tokara Tectonic Straits. This strongly suggests that the populations composing this group have spread over their current range recently. In addition, the degree of genetic differentiation between populations of the Groups 1 and 2 and the Group 3 (Nei's D: 0.083–0.175, $\bar{x} = 0.129$) was too small to have resulted merely from the deduced geographic isolation between the northern Ryukyu–Kyushu region and the central Ryukyus. This suggests that at least one of those two groups has colonized after formation of the Tokara Tectonic Straits by secondary dispersals. Circumstantial evidences seem to favor the secondary dispersals of the northern Ryukyu–Kyushu groups. Data for geographic patterns of allelic distributions and of intrapopulational genetic variabilities suggest that the southern Kyushu populations have received gene flow also from an extralimital source as well.

INTRODUCTION

A number of authors have argued that two major terrestrial faunal regions, the Palearctic and Oriental regions, have their borders in the northern Ryukyus (e.g., Kuroda, 1931; Inger, 1950; Hikida *et al.*, 1989, 1992; Nishida, 1990). Recent geological studies of the northern Ryukyus strongly suggested that the Tokara Tectonic Straits, located between Kodakarajima and Akusekijima within the Tokara Group, has persisted since its initial formation at the early Pliocene, and that the faunal discontinuity observed in the northern Ryukyus reflects the effect of this long standing strait as a significant barrier against dispersals of terrestrial organisms (Kizaki and Oshiro, 1980; Kato, 1989).

Gekko hokouensis Pope, 1928, has an exceptionally broad distributional range among terrestrial vertebrates occurring in the East Asian islands. This gecko, while being widely distributed in the southwestern side of the Tokara Tectonic Straits (i.e., the central and the southern Ryukyus, Taiwan and southeastern China), also occurs in a part of the northeastern side of the strait, from the northern Tokara Islands to southern Kyushu (Zhou *et al.*, 1982; Ota, 1986, 1989; Shibata, 1989, 1990; Hikida *et al.*, 1992). Such distribution seems to be inconsistent with the geographic faunal pattern mentioned above, and suggests that the species has an extraordinary zoogeographical history among terrestrial animals occurring in and around the northern Ryukyus. In spite of such apparent biogeographical interests, no studies have been made on the intraspecific geographic variation of *G. hokouensis* in this region. In this study, we survey the genetic variation among populations of *G. hokouensis* from Kyushu, and the northern and the central Ryukyus by means of protein electrophoresis, and discuss historical implications of the geographic pattern revealed.

MATERIALS AND METHODS

Specimens were collected from four localities in Kyushu, seven localities in the northern Ryukyus and 13 localities in the central Ryukyus (Fig. 1 and Table 1). A preliminary electrophoretic survey has revealed existence of an undescribed cryptic species which occurs sympatrically with *Gekko hokouensis* in some localities. That species very closely resembles *G. hokouensis* (sensu stricto) morphologically, but clearly differs from the latter on the basis of allelic

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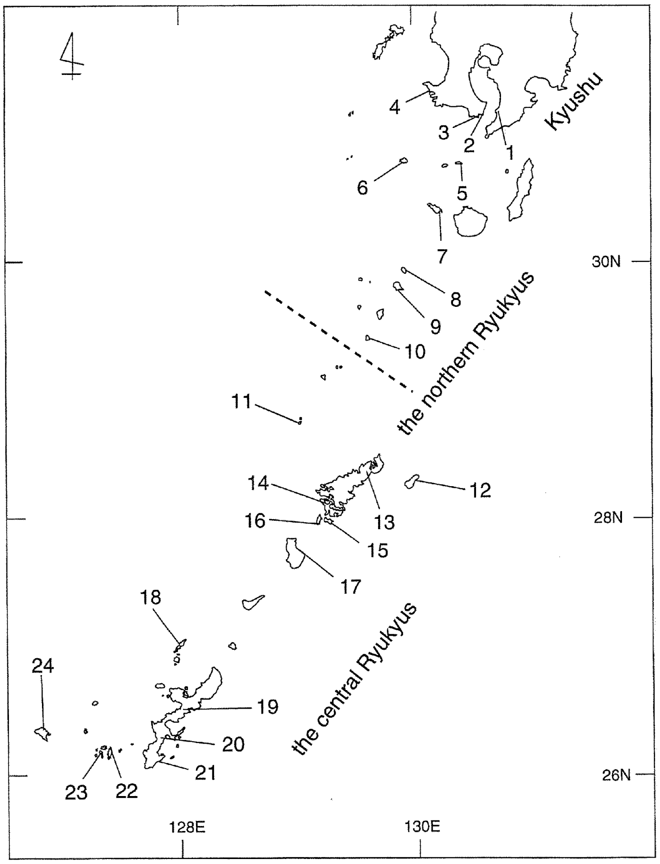

Fig. 1. A map of the central Ryukyu–southern Kyushu region, showing sampling localities of *Gekko hokouensis*. A dotted line shows the location of the Tokara Tectonic Straits.

Table 1. Names and sizes of samples used in the present study. The sample numbers correspond to those in Fig. 1.

Samples	(N)
Kyushu	
1. Nejime	20
2. Yamagawa	20
3. Ibusuki	14
4. Bonotsu	20
Osumi Group (the northern Ryukyus)	
5. Takeshima	20
6. Kuroshima	20
7. Kuchierabujima	20
Tokara Group (the northern Ryukyus)	
8. Kuchinoshima	20
9. Nakanoshima	20
10. Akusekijima	20
11. Yokoatejima	12
Amami Group (the central Ryukyus)	
12. Kikaijima	20
13. Amamioshima	15
14. Kakeromajima	19
15. Ukejima	20
16. Yorojima	20
17. Tokunoshima	20
Okinawa Group (the central Ryukyus)	
18. Iheyajima	14
19. Nago (Okinawajima)	20
20. Nakagusuku (Okinawajima)	22
21. Chinen (Okinawajima)	20
22. Tokashikijima	20
23. Akajima	18
24. Kumejima	20

compositions at no less than eight presumptive loci (Toda, unpublished data). Therefore, we discriminated data for this species using such electrophoretic markers and excluded them from the present analysis. Consequently 454 specimens of *G. hokouensis* were used for population genetic analyses.

Liver and muscles were removed from each specimen, and their extracts, obtained following Toda *et al.* (1997), were subjected to electrophoretic assay. Voucher specimens were deposited in the collection of the Department of Zoology, Kyoto University (KUZ: see Appendix for further details).

Thirty-two presumptive loci were examined (Table 2). Enzyme nomenclature and E.C. number follow Murphy *et al.* (1996). Notations of presumptive loci were made by use of abbreviations of enzymes. In case that more than one loci control an enzyme system, we added numerical supplementaries, with “1” representing the most anodal locus. We made notations for loci encoding peptidase with hyphenated abbreviations of the substrates. Electromorphs were designated alphabetically in order of their anodal mobility.

In order to estimate overall genetic differentiations among samples, we calculated Nei’s (1978) unbiased genetic distance for all pairwise comparisons of samples on the basis of their allele frequencies. The coefficient was subjected to two kinds of clustering methods, the UPGMA algorithm (Sneath and Sokal, 1973) and the Neighbor-joining (NJ) procedure (Saitou and Nei, 1987). The UPGMA phenogram is expected to depict the accurate phylogeny when the assumption of equal rates of molecular evolution among branches is met (Nei, 1987), whereas the NJ analysis does not need such presumption. The NJ dendrogram was rooted at the mid-point of the longest pathway.

To evaluate magnitude of genetic differentiation within each geographical unit that are recognized from the distance analyses (see below), Wright’s (1965) F-statistics (F_{ST}) was calculated. We also calculated expected heterozygosities (*He*: Nei, 1978) for each sample to estimate intrapopulational genetic variability. NJ analysis was made by using N. Saitou’s original computer program. The other computations were made by using BYOSIS-1 computer program (Swofford and Selander, 1981).

RESULTS

Of the 32 presumptive loci examined, 23 were polymorphic, of which eight (*Acoh-1, Aat-1, Est, Fbp, Hbdh, Pgm-1, Pgm-2* and *Pgdh*) showed high heterogeneity in allele frequencies among samples (Table 3). Based on the pattern of shared predominant alleles in these highly variable loci, the samples were divided into three groups—Group 1 consisting of samples from southern Kyushu (samples 1–3: Fig. 1, Table 1), Group 2 consisting of samples from southwestern Kyushu and the northern Ryukyus (samples 4–11), and Group 3 consisting of samples from the central Ryukyus (samples 12–24). Group 1 samples exhibited predominant alleles at three (*Aat-1, Fbp* and *Hbdh*) and five presumptive loci (*Acoh-1, Aat-1, Est, Fbp* and *Pgdh*) that are different from those in samples of the Groups 2 and 3, respectively. Group 2 samples differed from the Group 3 samples in predominant alleles at four loci (*Aat-1, Est, Hbdh* and *Pgdh*). However, in the particularly highly

Table 2. Enzymes, loci, tissues and buffer systems used in the analysis of allozyme variations among populations of *Gekko hokouensis* from Kyushu, and the northern and the central Ryukyus

Enzyme	E. C. number	Locus	Tissue	Buffer system
1. Aconitate hydratase	4.2.1.3	*Acoh-1*	L	CAPM6, TC8
2. Aconitate hydratase	4.2.1.3	*Acoh-2*	L	CAPM6
3. Adenosine deaminase	3.5.4.4	*Ada*	L	TC7, TC8
4. Aspartate aminotransferase	2.6.1.1	*Aat-1*	L	CAPM6
5. Aspartate aminotransferase	2.6.1.1	*Aat-2*	L	CAPM6, TC7
6. Adenylate kinase	2.7.4.3	*Ak*	M	CAPM6
7. Creatine kinase	2.7.3.2	*Ck*	M	CAPM6
8. Esterase	3.1.1.–	*Est*	L	CAPM6, TC7
9. Fructose-biphosphatase	3.1.3.11	*Fbp*	L	TBE8.7
10. Fumarate hydratase	4.2.1.2	*Fumh*	L	TC7
11. General protein	———	*Gp*	M	CAPM6
12. Glucose-6-phosphate isomerase	5.3.1.9	*Gpi*	M	CAPM6
13. Glycerol-3-phosphate dehydrogenase	1.1.1.8	*G3pdh*	L	TC8
14. Guanine deaminase	3.5.4.3	*Gda*	L	TBE8.7
15. 3-Hydroxybutyrate dehydrogenase	1.1.1.30	*Hbdh*	L	TC7
16. L-Iditol dehydrogenase	1.1.1.14	*Iddh*	L	CAPM6, TC7
17. Isocitrate dehydrogenase	1.1.1.42	*Idh-1*	L	CAPM6, TC7
18. Isocitrate dehydrogenase	1.1.1.42	*Idh-2*	L	CAPM6, TC7
19. L-Lactate dehydrogenase	1.1.1.27	*Ldh-1*	L	TC7
20. L-Lactate dehydrogenase	1.1.1.27	*Ldh-2*	L	TC7
21. Malate dehydrogenase	1.1.1.37	*Mdh-1*	L,M	TC8, CAPM6
22. Malate dehydrogenase	1.1.1.37	*Mdh-2*	L,M	TC8, CAPM6
23. Malate dehydrogenase (NADP$^+$)	1.1.1.40	*Mdhp*	M	CAPM6
24. Mannose-6-phosphate isomerase	5.3.1.8	*Mpi*	M	CAPM6
25. Dipeptidase(leucyl-glycine)	3.4.–.–	*Pep-lg*	L	CAPM6, TC7
26. Peptidase(leucyl-glycyl-glycine)	3.4.–.–	*Pep-lgg*	L	TBE8.7
27. Dipeptidase(leucyl-proline)	3.4.–.–	*Pep-lp*	L	TBE8.7
28. Phosphoglucomutase	5.4.2.2	*Pgm-1*	L	TC8
29. Phosphoglucomutase	5.4.2.2	*Pgm-2*	L	TC8
30. Phosphogluconate dehydrogenase	1.1.1.44	*Pgdh*	L	CAPM6
31. Purine-nucleoside phosphorylase	2.4.2.1	*Pnp*	L	TC8
32. Superoxide dismutase	1.15.1.1	*Sod*	L	TBE8.7

Tissue: L = liver, M = muscle.
Buffer: CAPM6 = Citrate-aminopropylmorphorine, pH 6.0 (Clayton and Tretiak, 1972); TC7 = Tris-citrate, pH 7.0 (Show and Prasad, 1970); TC8 = Tris-citrate, pH 8.0 (Clayton and Tretiak, 1972); TBE8.7 = Tris-borate-EDTA, pH 8.7 (Boyer *et al.*, 1963).

polymorphic locus, *Pgm-2*, the predominant alleles varied even within each group.

Relatively large genetic distance values were observed between samples from the central Ryukyus (i.e., Group 3) and the remainder (Groups 1 and 2), though these values considerably varied [e.g., D = 0.083–0.175 ($\bar{x}$ = 0.129)]. The D values between samples of the Groups 1 and 2 were mostly less than the above values, but were still at the noticeable level, ranging from 0.059 to 0.085 ($\bar{x}$ = 0.070). Within-group D values were substantially larger in Group 3 [D = 0.000–0.062 ($\bar{x}$ = 0.025)] than in Groups 1 and 2 [D = 0.005–0.017 ($\bar{x}$ = 0.010) and 0.000–0.018 ($\bar{x}$ =0.005), respectively].

The expected heterozygosity (*He*) for each sample examined is presented in the bottom of Table 3. *He* values were highly variable among samples, ranging from 0.012 to 0.102. Samples of the Group 1 showed relatively high values of *He* (0.078–0.100, $\bar{x}$ = 0.093), whereas the values in the Group 2 were distinctly lower (0.023–0.049, $\bar{x}$ =0.040). *He* values in samples of the Group 3 were largely intermediate between those in the Groups 1 and 2 samples (0.012–0.102, $\bar{x}$ = 0.053).

The F_{ST} calculated for all samples examined was 0.575. We recalculated F_{ST} for each of the three sample groups (i.e., Groups 1–3) to evaluate genetic differentiation within each of these groups. The F_{ST} value for the Group 3 (0.305) was much larger than those for the Groups 1 and 2 (0.079 and 0.126, respectively).

Results of NJ and UPGMA clusterings are shown in Figs. 2 and 3, respectively. In both phenograms, three clusters corresponding to above mentioned groups (i.e., Groups 1–3) were recognized, with the primary bifurcation located between the Group 3 and the remainder. These phenograms slightly differ from each other in the inner topology of two large clusters corresponding to the Groups 2 and 3. Such difference may result from small genetic differentiations and/or substantial heterogeneity in the rate of molecular change among populations within each group.

Table 3. Allele frequencies at 23 polymorphic loci in 24 samples of *Gekko hokouensis*, together with a measure of intrapopulational those in Fig. 1.

	Kyushu				the northern Ryukyus							
Locus	1	2	3	4	5	6	7	8	9	10	11	12
Acoh-1												
a												.075
b	.950	.700	.786	.737	.725	.800	.775	.725	.625	.650	.722	.050
c	.050	.300	.214	.263	.275	.200	.225	.275	.375	.350	.278	.875
d												
e												
Aat-1												
a												
b	.300	.100	.107	1.000	1.000	1.000	1.000	1.000	.950	1.000	1.000	1.000
c	.700	.900	.893						.050			
Ck												
a												
b	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
Est												
a												1.000
b										.150		
c	.975	1.000	1.000	1.000	.975	1.000	1.000	1.000	1.000	.850	1.000	
d	.025				.025							
Fbp												
a	.607	.844	.714				.200		.050			.125
b	.393	.156	.286	1.000	1.000	1.000	.800	1.000	.950	1.000	1.000	.875
Gda												
a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
b												
G3pdh												
a												
b	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
c												
d												
Gpi												
a	.025											.300
b	.975	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.700
Hbdh												
a	.222	.500	.464	.900	.974	1.000	1.000	1.000	1.000	.921	1.000	.026
b	.778	.500	.536	.100	.026					.079		.974
Iddh												
a	.639	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
b	.361											
Idh-1												
a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
b												
Idh-2												
a	1.000	1.000	1.000	1.000	.775	.975	.875	1.000	.650	.600	1.000	1.000
b					.225	.025	.125		.350	.400		
Ldh-2												
a												
b	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
Mdh-1												
a	1.000	1.000	1.000	1.000	1.000	1.000	.975	1.000	1.000	1.000	1.000	1.000
b							.025					
Mdh-2												
a												
b									.025			
c	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.975	1.000	1.000	1.000
Pep-lg												
a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
b												
Pep-lgg												
a												
b	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
Pep-lp												
a		.025	.036									.025
b	.975	.725	.964	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.975
c	.025	.250										
Pgm-1												
a		.025										
b												
c	1.000	.975	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
Pgm-2												
a	.350	.125	.321								.056	.100
b	.125	.200	.321	.725	.475	.375	.925	.525	.889	.975	.833	.050
c				.225	.450	.625	.075	.475	.083	.025	.111	
d	.175	.400	.071									.850
e	.350	.275	.250	.050	.075				.028			
f			.036									
Pgdh												
a	.100	.075		.150	.025	.175	.025		.025			.975
b	.900	.925	1.000	.850	.975	.825	.975	1.000	.975	1.000	1.000	.025
c												
Pnp												
a	.075	.350	.107	.175				.125	.050	.025		
b	.925	.650	.893	.825	1.000	1.000	1.000	.875	.950	.975	1.000	1.000
Sod												
a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
b												
He	.096	.100	.078	.049	.047	.036	.036	.036	.048	.046	.023	.041
	(.033)	(.035)	(.032)	(.021)	(.024)	(.020)	(.016)	(.021)	(.021)	(.022)	(.016)	(.018)

genetic variabilities, the expected heterozygosities (*He*: standard errors in parentheses). Sample numbers correspond to

the central Ryukyus											
13	14	15	16	17	18	19	20	21	22	23	24
.133						.025	.023				
.033		.025	.050		.036	.300	.182	.200		.056	.050
.833	1.000	.975	.950	1.000	.893	.650	.795	.800	.350	.556	.875
					.071	.025			.350	.111	.075
									.300	.278	
.033					.143		.023				
.967	1.000	1.000	1.000	1.000	.857	1.000	.977	1.000	1.000	1.000	1.000
						.150					
1.000	1.000	1.000	1.000	1.000	1.000	.850	1.000	1.000	1.000	1.000	1.000
.967	.974	1.000	.875	.850	1.000	.974	.977	.950	.600	.500	1.000
	.026		.125	.125		.026	.023	.050	.375	.500	
.033				.025					.025		
.063	.050				.071						
.938	.950	1.000	1.000	1.000	.929	1.000	1.000	1.000	1.000	1.000	1.000
.867	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
.133											
							.045				
1.000	1.000	.975	1.000	.975	1.000	.975	.955	1.000	.975	1.000	.975
				.025		.025					
		.025							.025		.025
.100									.025		
.900	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.975	1.000	1.000
	.029		.029		.346			.100	.231	.028	.528
1.000	.971	1.000	.971	1.000	.654	1.000	1.000	.900	.769	.972	.472
.857	1.000	1.000	.850	1.000	.964	.925	1.000	1.000	1.000	1.000	.929
.143			.150		.036	.075					.071
1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.972	1.000
										.028	
1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.972	1.000
										.028	
								.125			
1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.875	1.000	1.000	1.000
1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	.825	1.000	1.000
									.175		
										.028	
						.050	.023				
1.000	1.000	1.000	1.000	1.000	1.000	.950	.977	1.000	1.000	.972	1.000
1.000	1.000	1.000	1.000	1.000	1.000	.950	1.000	1.000	1.000	1.000	1.000
						.050					
	.053			.250							
1.000	.947	1.000	1.000	.750	1.000	1.000	1.000	1.000	1.000	1.000	1.000
.267	.028		.050	.050	.042		.023		.025	.028	
.733	.972	1.000	.950	.950	.958	1.000	.977	1.000	.975	.944	.825
										.028	.175
.033	.026		.025						.675	.583	1.000
			.050								
.967	.974	1.000	.925	1.000	1.000	1.000	1.000	1.000	.325	.417	
.067	.053	.050			.107	.300	.452	.500	.579	.528	.289
.033	.026	.075	.026			.025	.238	.056	.105	.139	.026
			.026								
.800	.895	.850	.842	1.000		.475	.024	.222			.211
.100	.026	.025	.105		.893	.200	.286	.222	.316	.306	.474
										.028	
.767	.974	1.000	.950	1.000	.929	.925	1.000	.500	.925	.917	.925
.233	.026		.050		.071	.075		.500	.075	.083	.050
											.025
						.050			.050		
1.000	1.000	1.000	1.000	1.000	1.000	.950	1.000	1.000	.950	1.000	1.000
1.000	1.000	1.000	1.000	.975	.964	.950	1.000	1.000	1.000	1.000	1.000
				.025	.036	.050					
.076 (.023)	.021 (.008)	.012 (.009)	.040 (.014)	.027 (.015)	.051 (.018)	.069 (.026)	.040 (.023)	.064 (.028)	.102 (.034)	.086 (.033)	.064 (.028)

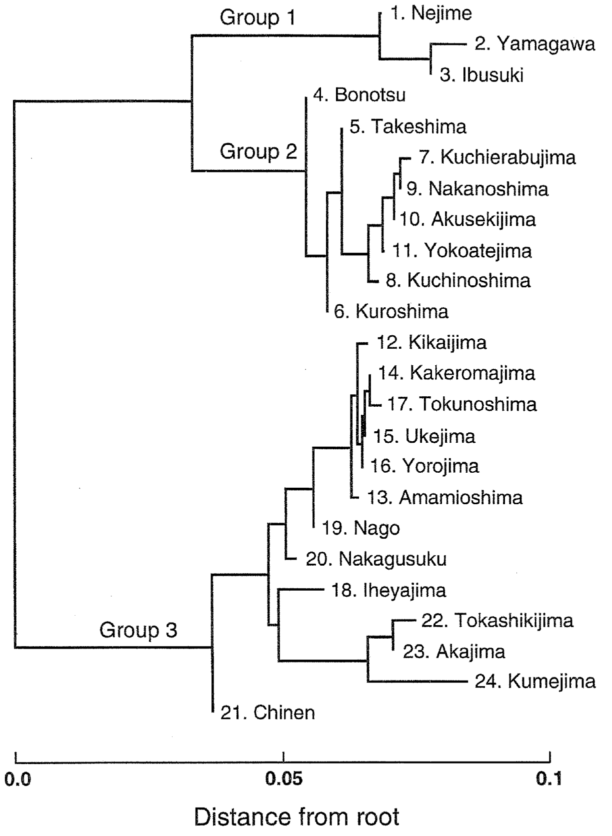

Fig. 2. Neighbor-Joining phenogram among populations of *Gekko hokouensis* from Kyushu, and the northern and the central Ryukyus based on Nei's (1978) unbiased genetic distance. Minus values in branch lengths are expressed as zero.

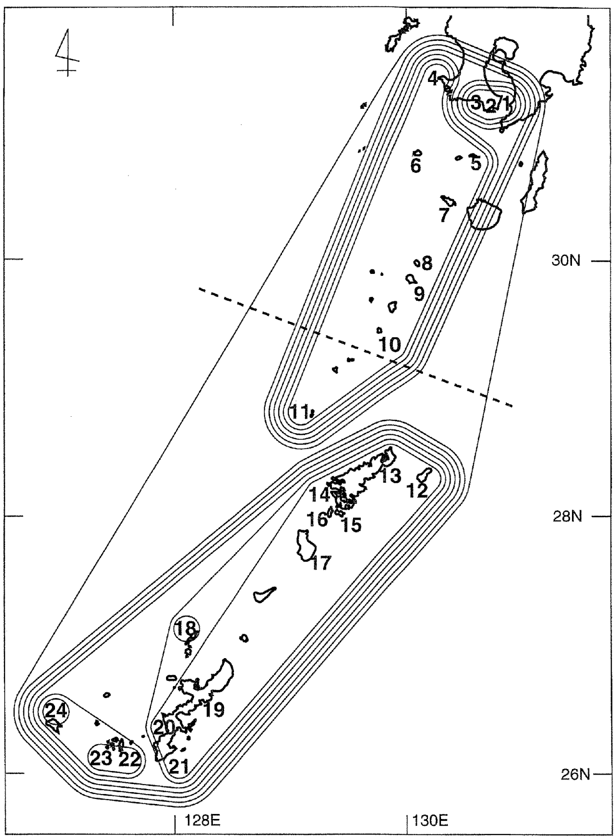

Fig. 3. Genetic relationship among populations of *Gekko hokouensis* from Kyushu, and the northern and the central Ryukyus based on contours of Nei's (1978) unbiased genetic distance values derived from the UPGMA algorithm. Each contour line is drawn at an interval of D=0.02. A dotted line shows the location of the Tokara Tectonic Straits.

DISCUSSION

Relationship of Yokoatejima and other northern Ryukyu and southwestern Kyushu populations

Present results indicate that the most prominent genetic differentiation in *Gekko hokouensis* exists between populations of the central Ryukyus and those of the northern Ryukyus and Kyushu, with the primary boundary being located between Amamioshima and Yokoatejima (Figs. 2 and 3). Superficially, this appears to coincide with the pattern of divergence of many other terrestrial vertebrate lineages occurring in the central Ryukyu–southern Kyushu region (Nishida, 1990). However, detailed investigation reveals a discrepancy between the geographic pattern of genetic variation in *G. hokouensis* and the deduced paleogeography in this region (Kizaki and Oshiro, 1980; Kato, 1989). Hikida *et al.* (1992), in a review of distributions and variations of amphibians and reptiles in the northern Ryukyus, demonstrated the presence of the primary herpetofaunal gap exactly between Akusekijima and Kodakarajima, where the Tokara Tectonic Straits is located (Fig. 1). Present results, however, indicate that the population of *G. hokouensis* on Yokoatejima (sample 11), an island located southwest to the strait, is genetically much closer to those of islands northeast to the strait (D = 0.001–0.008) than to those of the central Ryukyu islands (D = 0.092–0.147) (also see Figs. 2 and 3). This indicates that the Yokoatejima population has not been genetically isolated from populations of other northern Ryukyu islands until recently, despite the presumed long geographical isolation between Yokoatejima and the latter.

A sample from southwestern Kyushu (i.e., Bonotsu sample) is genetically much closer to those from the northern Ryukyus including Yokoatejima (D = 0.001–0.008) than to the other three Kyushu samples (D = 0.060–0.076) (also see Figs. 2 and 3). Apparently it is also difficult to explain this pattern of geographic variation on the vicariance ground alone, since there are no paleogeographical evidence that supports existence of recent exclusive land connection of the northern Ryukyu islands and southwestern Kyushu. This result strongly suggests that the southwestern Kyushu population has been very recently isolated from the northern Ryukyu populations genetically.

Level of genetic differentiation among the northern Ryukyu populations including Yokoatejima and Bonotsu populations (i.e., Group 2) is fairly low (D < 0.019). Although the maximum geographic distance between the Group 2 populations (318 km, between Yokoatejima and Bonotsu populations) is nearly as large as that between the central Ryukyu populations (395 km, between Kumejima and Kikaijima populations), the F_{ST} value for the former (0.126) is much smaller than that for the latter (0.305). Moreover, the Group 2 populations collectively show low intrapopulational genetic variabilities (Table 3). From these results, as well as from the geographic pattern of their distribution that are not concordant with the deduced paleogeography (see above), it is highly likely that populations of the Group 2 have recently spread from a source population through overseas dispersals by rafting or artificial transportation.

This does not necessarily imply that the source population of the whole of the Group 2 is located outside of the current range of this group. Nevertheless, considering the relatively small genetic distance between the Groups 2 and 1 (Figs. 2 and 3), we can reasonably postulate that these groups had relatively recently derived from a common source population.

Relationship between population groups and its zoogeographical implications

As mentioned at the beginning part of this paper, the current distribution of *Gekko hokouensis,* which includes both southwestern and northeastern sides of the Tokara Tectonic Straits, is unusual among those of non-volant terrestrial vertebrates occurring in East Asian islands. To explain this, one may argue that the ancestral populations of the Groups 1 and 2 and those of the Group 3 have long persisted in the northern Ryukyu–Kyushu region and the central Ryukyus, respectively, being involve in an *in situ* vicariant divergence through the formation of the Tokara Tectonic Straits. If this is the case, initial divergence between the Groups 1 and 2, and the Group 3 should have synchronized with those in other animal clades that should have also been subjected to this vicariance. Several authors reported comparable data of Nei's D between populations of terrestrial vertebrates on both sides of the Tokara Tectonic Straits. Although these include values from both intra- and interspecific comparisons, most of them are considered to reflect the vicariant event at the strait (Nishida, 1990; Toda *et al.*, 1997). These values are: D = 0.18 for *Oryzias latipes* (Sakaizumi *et al.*, 1983), D = 0.19 for *Plecoglossus altivelis* (Nishida, 1985), D = 0.52 for *Rhacophorus* (Nishioka *et al.*, 1987), D = 0.36 for *Cynops* (Hayashi and Matsui, 1988), and D = 0.31–0.60 for *Eumeces* (Kato *et al.*, 1994). Of these, the first two are for fishes that are amphidromous or with a high sea-water tolerance. Relatively small D values for these fishes are therefore considered to reflect the delay of timing of initial divergence by the vicariance (Nishida, 1990). Thus, D values between populations of strict terrestrial vertebrates, all amphibians and reptiles, on both sides of the Tokara Tectonic Straits range from 0.31 to 0.60. Compared to these, D values between the populations of *Gekko hokouensis* in the central Ryukyus and the remaining regions (0.083–0.175, $\bar{x}$ = 0.129) are considerably smaller. As such, our results do not support hypothesis of *in situ* divergence between the central Ryukyu and the remaining populations of *G. hokouensis* examined here, but suggest a relatively recent colonization of at least one of them by secondary dispersals.

There are a few species of amphibians and reptiles that, while occurring in the central Ryukyus, are distributed on some islands northeast to the Tokara Tectonic Straits as well (Hikida *et al.*, 1992; Kato *et al.*, 1994). These species are considered to belong to the Oriental faunal elements, since their ranges overwhelmingly spread over the southwestern side of the strait. Occurrence of these species in the northeastern side of the strait is, therefore, considered to be the consequence of secondary dispersals. Kato *et al.* (1994) provided a strong support for such overseas dispersal of an Oriental species by demonstrating the poor genetic differentiation between populations of the Okinawan five-striped skink, *Eumeces marginatus*, on both sides of the strait. By contrast, no representatives of the Palearctic faunal elements occurring in the northeastern side of the Tokara Tectonic Straits seem to have extended their range to the southwestern side of the strait after the formation of the strait (Hikida *et al.*, 1992). Ota *et al.* (1995) argued that the prevalence of the presumed recent northeastward dispersals across the Tokara Tectonic Straits in amphibians and reptiles is attributable to the direction of the prevailing sea current there (i.e., grossly from southwest to northeast). *Gekko hokouensis* is considered to be an Oriental species whose distribution is remarkably wide in the southwestern side (i.e., ranging from the central Ryukyus to southeastern China), but is quite narrow in the northeastern side of the strait (i.e., the northern Ryukyus and southern Kyushu: Zhou *et al.*, 1982; Shibata, 1989, 1990; Ota, 1986, 1989; Hikida *et al.*, 1992). Taking these into considerations, we can safely conclude that the northern Ryukyu and Kyushu populations have colonized to their current ranges most probably by secondary dispersals after formation of the strait.

However, this hypothesis leaves difficulty in explaining data for the level of intrapopulational genetic variabilities and for the distributions of several alleles in the southern Kyushu populations. If the southern Kyushu populations, as well as the northern Ryukyu populations, have originated from the central Ryukyu populations by secondary dispersals, genetic variability of the former should be lower than that of the latter as a consequence of operation of the founder effect (Wright, 1931). However, the southern Kyushu populations actually show a relatively high level of heterozygosity (Table 3). Moreover, the southern Kyushu populations possess several alleles that are not observed in the central Ryukyu populations (e.g., *Aat-1*: c, *Est*: d). To explain these, it seems necessary to postulate the occurrence of gene flows into the southern Kyushu populations from extralimital populations. Further extensive electrophoretic surveys of *G. hokouensis* from wider ranges are needed to verify this hypothesis.

The geographic pattern of genetic variation in *G. hokouensis* revealed by this study provides a counterexample for general zoogeographic view on the Ryukyu–Kyushu region. This gecko seems to have dispersed across the Tokara Tectonic Straits at which dispersals of many other terrestrial vertebrate lineages must have been interrupted. Gibbons

(1985) argued that successful overseas dispersals by particular groups of reptiles and amphibians over the others may be facilitated by their specialized eco-physiological traits, such as the strong tolerance of adults and/or eggs for dehydrated condition, and the high ability of flotation (also see Hedges, 1996). For gekkonid species, he suspected that the viability of their hard-shelled eggs in the salt water is an important trait for their overseas dispersals. However, direct supportive data have been obtained from only a few species (Brown and Alcala, 1957). It would therefore be interesting to assess Gibbons' (1985) idea by comparing the degree of sea-water tolerance between eggs of *G. hokouensis* and other northern–central Ryukyu reptiles.

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Appendix

Catalogue number of specimens examined in this study. Nejime KUZ 31192–197, 32253–256, 33393–396, 33494–499; Yamagawa 30492–495, 30594–599, 31198–201, 33653–658; Ibusuki 32245–250, 32331–336, 33659–660; Bonotsu 33285–288, 33363–372, 33555–560; Takeshima 32991–994, 33321–326, 33328–335, 33663–664; Kuroshima 33289–292, 33317–320, 33460–467, 33561–564; Kuchierabujima 32339–346, 33385–392, 33456–459; Kuchinoshima 33293–296, 33301–304, 33520–525, 33687–692; Nakanoshima 33305–310, 33373–380, 33582–587; Akusekijima 33311–316, 33510–519, 33526–529; Yokoatejima 21628–631, 30510–513, 33637–640; Kikaijima 30423–432, 30453,454, 30484–491; Amamioshima 33259–264, 33530–533, 33620–624; Kakeromajima 34127–134, 34178–183, 34247–251; Ukejima 34101–108, 34143–146, 34239–246; Yorojima 34109–116, 34135–140, 34164–169; Tokunoshima 34117–122, 34170–175, 34226–229, 34252–255; Iheyajima 30573–574, 32447, 32453, 32456–459, 33534–539; Nago 32269–275, 32323–330, 33336–340; Nakagusuku 30421–422, 30570–571, 31172–175, 31202–203, 32309–310, 32347–348, 33631–636, 34141–142; Chinen 31176–185, 31238–242, 32257–261; Tokashikijima 31204–215, 32484–487, 33352–355; Akajima 34154–163, 34231–238; Kumejima 30416–420, 30471, 30473–476, 30514–521, 30600–601.