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Spatial genetic structure in three populations of *Hemerocallis hakuunensis* (Liliaceae)

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Abstract. Ninety-nine, 75, and 71 individuals were mapped and leaf samples were collected from three populations (10 × 15-m plot [MIM], 40 × 50-m plot [KOS], and 15 × 20-m plot [NOD]) of *Hemerocallis hakuunensis* to determine if spatial genetic structure existed in the three populations in terms of their ecological characteristics. A substantial spatial genetic structuring was found in MIM, whereas a weak structure was observed in NOD and KOS. Moran's *I* values were significantly different from the expected value (-0.010) in 137 (52.7%) of 260 cases in MIM, whereas significant Moran's *I* values were observed in 25 (10.4%) of 240 cases and 55 (20.4%) of 270 cases in NOD and KOS, respectively. The approximate minimum patch width also varied among the three populations: NOD = 3 m, MIM = 5 m, and KOS = 10 m. The differences in patch width may result from the differences in density, colonization history, thinning processes or any of a number of other factors among populations.

Keywords: Allozymes; Density; *Hemerocallis hakuunensis*; Moran's *I*; Spatial autocorrelation.

Introduction

During the past decade, spatial genetic structure has been quantified using spatial autocorrelation statistics using allozymes as genetic markers to understand the evolutionary dynamics of plant populations (e.g., Berg and Hamrick, 1995; Chung and Epperson, 1999; Dewey and Heywood, 1988; Epperson and Clegg, 1986; Perry and Knowles, 1991; Schnabel and Hamrick, 1990). These studies showed that individuals are not likely to be randomly distributed owing to factors such as limited seed and pollen dispersal, isolation in small patches, differential mortality, and microenvironmental selection. In addition to these factors, differences in density, pollinator behaviors, topography, and colonization history could result in a different genetic architecture in plant populations of a species (e.g., Chung et al., 1998; Knowles et al., 1992; Young and Merriam, 1994). All these aspects should be considered in studies of natural plant populations to gain insights into their maintenance mechanisms of genetic diversity.

Hemerocallis hakuunensis Nakai is commonly found in grasslands of mountainous areas, under pine-oak forests on the hillsides, and under disturbed orchards of chestnut trees of the southern, central, and northwestern Korean Peninsula (Chung and Kang, 1994a). Individuals of the species have 5 to 16 flowers on the branched inflorescence. Flowers are orange-yellow and are usually visited by bees, bumblebees, and flies. They start to open before sunrise and remain open until the afternoon (Chung, pers. obs.). The species has no specialized mechanisms for seed dispersal (Chung, pers. obs.).

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In this study, spatial autocorrelation using allozyme markers was conducted in three populations of *H. hakuunensis* to determine whether differences in spatial genetic structure existed in populations under different ecological conditions.

Materials and Methods

In August 1995 and August 1997, 99, 75, and 71 individuals were mapped and leaf samples were collected within a 10 × 15-m plot (Mich'eon-myeon, Chinju-shi, Prov. Gyeongsangnam-do, hereafter referred to as MIM), a 40 × 50-m plot (Sangri-myeon, Kosung-gun, Prov. Gyeongsangnam-do, hereafter referred to as KOS), and a 15 × 20-m plot (Nogodan, Mts. Chiri, Prov. Chollanam-do, hereafter referred to as NOD), respectively. Ecological characteristics of the three populations are summarized in Table 1. Leaf samples were placed in plastic bags wrapped with a wet paper towel and stored on ice to prevent protein denaturation prior to returning to the laboratory, where they were stored at 4°C until protein extraction.

Leaf samples were cut finely, and crushed with a mortar and pestle. A phosphate-polyvinylpyrrolidone extraction buffer (Mitton et al., 1979) was added to the leaf samples to facilitate crushing and to aid enzyme stabilization. The cellular extract was absorbed onto 4 × 6-mm Whatman 3MM chromatography paper wicks, which were then stored at -70°C until needed. Electrophoresis was performed using 11% starch gels. Fourteen putative loci were resolved from eight enzyme systems using three gel/electrode buffer combinations. Two Poulik buffer systems were used: a modification (Haufler, 1985) of Soltis et al. (1983) "system 6" was used to resolve leucine aminopeptidase (LAP), fluorescent esterase (FE) and a modi-

fication (electrode buffer of pH 8.6) of Haufler (1985) resolved diaphorase (DIA), alcohol dehydrogenase (ADH), and β-galactosidase (β-GAL). A morpholine citrate buffer system, a modification (Chung and Kang, 1994b) of that of Clayton and Tretiak (1972), was used to resolve malate dehydrogenase (MDH), phosphoglucoisomerase (PGI), and 6-phosphogluconate dehydrogenase (6PGD). Stain recipes were taken from Soltis et al. (1983), except for the DIA and β-Gal, which were taken from Cheliak and Pitel (1984). The genetic basis of enzyme banding patterns was inferred from observed segregation patterns in light of typical subunit structure and subcellular compartmentalization (Weeden and Wendel, 1989). Putative loci were designated sequentially, with the most anodally migrating isozyme designated '1', the next '2', etc. Likewise, alleles were designated sequentially with the most anodally migrating allele designated 'a'.

Lap-2, *Dia-2*, and *Pgd-2* were not included in this study due to faint or inconsistent staining. *Pgi-1* and *β-Gal* were monomorphic in all individuals. Moran's *I*, a spatial autocorrelation index, is a spatially weighted product-moment correlation coefficient in which neighboring individuals are compared in terms of their deviation from the mean of all observations. For spatial autocorrelation analysis, the genotypic data were coded so that allele frequency values of 1.0, 0.5, or 0.0 were assigned to individuals being homozygous for a given allele, heterozygous for that allele or genotypes with no copies of that allele, respectively, for each polymorphic locus (Sokal and Oden, 1978). Only one allele was considered at a dialellic locus, because the second allele contributes identical information. For a locus having more than two alleles, all alleles at that locus, regardless of their frequencies, were used for the spatial analysis. However, alleles that were presented by less than five copies (frequencies <2.6%, <3.3%, and <3.5% in MIM, KOS, and NOD, respectively) were excluded as non-informative for spatial analysis. Every possible pair of individuals was considered as a join (a connection between two individuals) and was assigned to one of ten distance classes. Because measures of small-scale autocorrelation more accurately represent the spatial structure, the first of ten distance classes was designed based on an estimate of the average distance which separates nearest neighbor individuals (see Tables 1-3). Moran's *I* values (Sokal and Oden, 1978) were calculated for each of ten distance classes by interpopulational distance classes. Each *I* value was used to test significant deviations from the expected values, $E(I) = -1/(N-1)$ (Cliff and Ord, 1981). A significant positive value of Moran's *I* indicates that the neighboring individuals have more alleles in common than would be expected by chance, whereas a significant negative value suggests that distant individuals have fewer alleles in common. Overall significance of each correlogram, and hence the presence of spatial structure, for each allele was tested using Bonferroni's criterion (Sakai and Oden, 1983). All calculations and statistical analyses were performed using the SAAP program (ver. 4.3) written by D. Wartenberg.

Results

According to the criteria above, 26 (MIM and KOS) and 24 alleles (NOD) were used for spatial autocorrelation analysis. The spatial autocorrelation coefficients, Moran's *I*, for the three populations are presented in Tables 2 through 4. For population MIM, Moran's *I* values were significantly different from the expected value (-0.010) in 137 (52.7%) of 260 cases, and the overall correlogram was significant for 19 (73%) of 26 alleles (Table 2). For distance classes 1 to 5 (0 < 5 m), 68 statistically significant positive values were observed and eight significant negative values were detected. This indicates that individuals are genetically similar when separated by 5 m or less. For population KOS, somewhat weaker spatial genetic structure was observed (Table 3). Moran's *I* values were significantly different from the expected value (-0.013) in 55 (20.4%) of 270 cases, and the overall correlogram was significant for 10 (37%) of 27 alleles (Table 3). For distance classes 1 and 2 (0 < 9 m), 15 significantly positive values were found, but no significant negative value was observed within the distance classes. For population NOD, a considerably weak spatial genetic structuring was found (Table 4). Moran's *I* values were significantly different from the expected value (-0.014) in 25 (10.4%) of 240 cases, and the overall correlogram was significant for only four (16.7%) of 24 alleles (Table 4). For distance classes 1 and 2 (0 < 4 m), only three significant positive values were ob-

Table 1. Ecological characteristics of *Hemerocallis hakuunensis* examined.

Population code	Sample size	Elevation (m)	Plot area (m^2)	Population size	Density	Habitat
MIM	97	340	150	Several thousands	0.64	Recently established, 3% hillslopes, understory in chestnut tree orchard, *Hemerocallis hakuunensis* is dominant.
KOS	75	240	2,000	<100	0.04	Undisturbed, 15% hillsides, understory of dense pine-oak forests.
NOD	71	1,350	300	Several thousands	0.24	Undisturbed, open grasslands with *Hosta capitata* (Liliaceae) and other herbs, *H. hakuunensis* is abundant.

Table 2. Spatial autocorrelation coefficients (Moran's *I*) over ten distance classes for 26 alleles in population MIM of *Hemerocallis hakuunensis*. The expected E(*I*) = -0.010.

Allele	Distance class (upper distance bound, m)										P^1	Allele frequency
	1(1)	2(2)	3(3)	4(4)	5(5)	6(6)	7(9)	8(10)	9(12)	10(14)		
Adh^{d}	0.06	0.03	-0.04	-0.02	-0.08*	-0.04	0.03*	-0.02	-0.03	0.07	0.230	0.076
Adh^{e}	0.12*	0.22**	0.14**	0.05	0.06*	-0.10*	-0.15**	-0.04	-0.04	0.11	0.000	0.308
Adh^{g}	0.21**	0.03	-0.08*	0.17**	0.01	-0.06	-0.04	-0.02	-0.14**	-0.54*	0.000	0.611
Dia-1^{c}	0.27**	-0.04	-0.06	0.07*	-0.09*	-0.02	0.01	0.01	-0.14**	-0.33	0.000	0.330
Dia-1^{e}	0.15**	0.03	-0.04	-0.04	-0.01	0.03	0.02	-0.11*	-0.25**	0.81**	0.000	0.595
Dia-1^{g}	0.23**	0.17**	0.15**	0.17**	0.03	-0.06	-0.13**	-0.19**	-0.24**	0.36*	0.000	0.040
Fe-1^{e}	0.00	0.05	0.01	0.00	-0.09*	0.01	0.01	-0.01	-0.15**	-0.05	0.027	0.084
Fe-1^{g}	0.80**	0.64**	0.46**	0.30**	0.18**	-0.14**	-0.32**	-0.62**	-0.76**	0.20	0.000	0.738
Fe-1^{i}	0.13*	0.12**	-0.07	-0.13**	-0.12**	0.01	0.06**	-0.06	-0.08	-0.01	0.007	0.121
Fe-2^{c}	0.66**	0.55**	0.35**	0.27**	0.11**	-0.23**	-0.32**	-0.28**	-0.35**	0.14	0.000	0.676
Fe-2^{e}	0.41**	0.28**	0.29**	0.25**	0.08**	-0.09*	-0.24**	-0.21**	-0.34**	0.23	0.000	0.152
Fe-2^{g}	-0.02	-0.00	0.03	-0.05	-0.00	-0.03	-0.01	-0.05	0.03	0.26	1.000	0.121
Lap-1^{c}	0.80**	0.68**	0.42**	0.25**	0.18**	-0.15**	-0.39**	-0.57**	-0.43**	0.26	0.000	0.954
Mdh^{d}	0.65**	0.51**	0.46**	0.24**	0.18**	-0.19**	-0.28**	-0.43**	-0.70**	-0.17	0.000	0.919
Pgd-1^{c}	0.16**	0.09*	-0.02	-0.02	-0.00	-0.08*	-0.04	0.03	-0.00	0.11	0.047	0.801
Pgd-1^{f}	0.49**	0.54**	0.21**	0.15**	0.14**	-0.11**	-0.22**	-0.39**	-0.48**	0.10	0.000	0.066
Pgd-1^{g}	0.04	-0.02	-0.04	-0.02	0.03	-0.06	-0.00	0.07	-0.10*	0.17	0.428	0.122
Pgi-2^{d}	0.00	-0.00	-0.03	-0.02	0.05	-0.03	0.00	-0.12*	-0.01	0.11	0.204	0.052
Pgi-2^{g}	0.67**	0.42**	0.37**	0.27**	0.01	-0.11**	-0.13**	-0.61**	-0.84**	-0.16	0.000	0.805
Pgi-2^{i}	0.61**	0.42**	0.27**	0.13**	0.00	-0.10**	-0.15**	-0.36**	-0.56**	0.24	0.000	0.052
Pgi-2^{k}	0.06	0.04	-0.06	0.07*	-0.01	-0.07	-0.01	-0.04	-0.04	-0.12	0.347	0.038
Pgi-3^{a}	0.05	0.05	0.01	-0.07	-0.02	0.02	-0.03	-0.00	-0.04	0.04	0.761	0.081
Pgi-3^{c}	0.10*	0.10*	-0.08*	-0.03	-0.09*	0.01	0.04**	-0.07	-0.16**	0.17	0.017	0.237
Pgi-3^{e}	0.49**	0.33**	0.18**	0.02	0.14**	-0.08*	-0.24**	-0.17**	-0.10*	-0.03	0.000	0.581
Pgi-3^{g}	0.52**	0.30**	0.20**	0.09*	0.12**	-0.08*	-0.20**	-0.22**	-0.35**	-0.29	0.000	0.066
Pgi-3^{i}	0.03	0.07*	-0.07	-0.08*	-0.01	0.00	-0.03	0.03	0.09*	-0.49*	0.106	0.035
Average	0.30	0.22	0.13	0.08	0.04	-0.07	-0.11	-0.17	-0.25	0.03		

[1]Overall correlogram significance (Bonferroni approximation). * = $P < 0.05$; ** = $P < 0.01$.

Table 3. Spatial autocorrelation coefficients (Moran's *I*) over ten distance classes for 27 alleles in population KOS of *Hemerocallis hakuunensis*. The expected E(*I*) = -0.013.

Allele	Distance class (upper distance bound, m)										P^1	Allele frequency
	1(1)	2(2)	3(3)	4(4)	5(5)	6(6)	7(9)	8(10)	9(12)	10(14)		
Adh^{b}	0.14**	-0.05	-0.01	-0.05	0.07	-0.14**	-0.29**	0.03	-0.00	-0.03	0.003	0.247
Adh^{d}	0.15**	0.00	0.01	-0.03	0.03	-0.13**	-0.33**	-0.22**	-0.05	0.08*	0.001	0.700
Adh^{e}	0.44**	0.06	0.09**	0.09*	0.06	0.10**	-0.04	-0.63**	-0.58**	-0.35**	0.000	0.053
Dia-1^{a}	0.08**	0.05	-0.11**	-0.07	-0.05	0.01	-0.01	0.07	-0.11*	0.01	0.037	0.047
Dia-1^{b}	0.16**	-0.08	-0.13**	0.05	0.01	-0.04	-0.07	-0.09	-0.10	0.10*	0.001	0.437
Dia-1^{d}	0.12**	-0.08	-0.05	0.02	0.03	-0.11*	-0.15	-0.06	-0.02	0.06	0.018	0.520
Fe-1^{b}	0.03	-0.02	-0.01	-0.06	-0.03	-0.04	-0.03	0.04	-0.05	0.00	1.000	0.887
Fe-1^{c}	0.03	-0.01	-0.05	-0.08	-0.00	-0.00	-0.02	0.02	-0.04	0.01	1.000	0.107
Fe-2^{a}	0.09**	-0.02	-0.05	-0.11	-0.03	-0.01	-0.02	0.08	-0.11*	0.01	0.098	0.840
Fe-2^{b}	0.03	-0.01	-0.05	-0.08	-0.00	-0.00	-0.02	0.02	-0.04	0.01	1.000	0.107
Fe-2^{c}	0.12**	-0.04	-0.02	-0.08	-0.06	-0.03	-0.01	0.12*	-0.16**	-0.00	0.013	0.053
Lap-1^{a}	-0.02	-0.01	-0.03	0.00	-0.00	-0.04	0.01	-0.05	0.01	0.01	1.000	0.213
Lap-1^{b}	-0.05	0.07*	-0.02	-0.05	-0.04	-0.03	0.11	-0.07	0.02	-0.01	0.255	0.707
Lap-1^{c}	-0.04	0.05	-0.00	-0.05	-0.09*	-0.03	0.15*	-0.04	0.01	-0.02	0.187	0.080
Mdh^{a}	0.04	0.00	0.00	-0.02	-0.06	-0.04	-0.15*	-0.08	0.11**	-0.07	0.060	0.040
Mdh^{b}	0.09*	0.05	-0.01	0.04	-0.09	-0.17**	-0.25**	-0.03	0.06	-0.01	0.003	0.913
Mdh^{c}	-0.01	0.01	-0.02	0.09*	-0.09	-0.06	-0.10	0.01	-0.05	0.04	0.216	0.040
Pgd-1^{b}	0.23**	0.13**	-0.05	-0.19**	-0.35**	-0.04	0.30**	-0.34**	0.15**	-0.03	0.000	0.733
Pgd-1^{c}	0.20**	0.09**	-0.04	-0.09	-0.39**	-0.10*	0.24**	-0.11	0.05	0.02	0.000	0.227
Pgd-1^{d}	0.02	0.01	-0.01	-0.02	0.03	0.04	0.03	-0.13*	-0.07	-0.08	0.173	0.040
Pgi-2^{b}	-0.00	0.09*	-0.09*	0.12**	-0.07	-0.09*	0.03	-0.06	-0.03	0.02	0.100	0.040
Pgi-2^{e}	-0.02	-0.05	-0.02	-0.02	-0.01	0.04	-0.08	0.04	-0.06	0.02	1.000	0.840
Pgi-2^{h}	-0.04	-0.03	0.07*	-0.06	-0.06	0.01	-0.15	-0.01	-0.03	0.02	0.212	0.067
Pgi-3^{c}	-0.02	-0.00	0.02	0.02	-0.02	0.05	-0.06	-0.10	-0.06	-0.05	0.812	0.460
Pgi-3^{e}	-0.02	-0.03	-0.07	-0.01	0.08*	0.01	-0.09	-0.00	0.03	-0.03	0.444	0.180
Pgi-3^{f}	-0.01	-0.05	0.01	0.06	-0.03	-0.02	-0.07	-0.07	0.02	-0.01	1.000	0.087
Pgi-3^{g}	-0.01	-0.02	-0.06	-0.04	0.03	0.02	0.03	-0.07	0.06	-0.03	0.979	0.207
Average	0.07	0.00	-0.03	-0.02	-0.04	-0.03	-0.04	-0.06	-0.04	-0.02		

[1]Overall correlogram significance (Bonferroni approximation). * = $P < 0.05$; ** = $P < 0.01$.

Table 4. Spatial autocorrelation coefficients (Moran's *I*) over ten distance classes for 24 alleles in population NOD of *Hemerocallis hakuunensis*. The expected E(*I*) = -0.014.

Allele	Distance class (upper distance bound, m)										P^1	Allele frequency
	1(1)	2(2)	3(3)	4(4)	5(5)	6(6)	7(9)	8(10)	9(12)	10(14)		
Adh[a]	0.02	0.05	-0.07	-0.18**	0.02	-0.04	0.00	-0.03	0.02	-0.02	0.047	0.082
Adh[b]	0.07*	0.06	-0.06	0.01	0.10**	-0.06	-0.10	-0.15*	-0.16**	-0.27**	0.003	0.253
Adh[d]	0.09*	0.06	-0.09	-0.06	0.05*	0.03	-0.07	-0.13*	-0.13*	-0.22*	0.168	0.664
Dia-1[a]	-0.01	-0.04	-0.06	0.04	0.01	-0.00	-0.11*	0.07	-0.02	0.05	0.360	0.061
Dia-1[b]	-0.01	-0.01	0.05	-0.06	-0.00	-0.05	-0.03	0.02	0.02	0.00	1.000	0.267
Dia-1[d]	0.03	0.01	-0.07	-0.01	-0.02	-0.03	-0.06	0.05	-0.03	0.10	0.578	0.480
Dia-1[e]	0.01	-0.06	-0.08	-0.13*	0.03	0.01	-0.05	0.01	0.01	-0.02	0.458	0.192
Fe-1[b]	0.04	-0.08	-0.07	-0.01	-0.08*	0.04	0.15**	0.03	-0.09	-0.07	0.018	0.897
Fe-2[a]	0.01	-0.08	-0.09	0.05	-0.03	-0.00	0.08	0.03	-0.06	-0.16	0.503	0.767
Fe-2[b]	0.04	-0.08	-0.07	-0.01	-0.08*	0.04	0.15**	0.03	-0.09	-0.07	0.018	0.103
Fe-2[c]	0.03	-0.08	-0.14*	-0.02	0.01	0.03	0.06	-0.01	-0.04	-0.07	0.341	0.130
Lap-1[a]	0.02	-0.01	0.04	-0.06	-0.01	0.03	0.02	-0.05	0.01	-0.06	1.000	0.185
Lap-1[b]	0.02	-0.05	0.01	0.03	-0.04	-0.03	0.09*	-0.13*	0.03	-0.06	0.300	0.610
Lap-1[c]	0.02	-0.02	0.04	0.06	-0.04	-0.02	0.05	-0.05	0.00	0.02	0.960	0.205
Lap-2[a]	0.04	-0.08	-0.01	0.09	-0.01	-0.02	-0.03	0.02	-0.06	0.05	0.544	0.069
Lap-2[b]	0.01	-0.11	-0.02	0.07	-0.00	-0.01	-0.04	-0.06	-0.01	0.08	0.792	0.911
Mdh[b]	0.02	-0.04	0.00	-0.09	0.04*	-0.05	-0.06	0.09*	-0.10	-0.07	0.341	0.720
Pgd-1[b]	0.04	0.08	-0.05	0.01	-0.04	0.02	-0.00	0.04	-0.02	0.03	0.909	0.863
Pgi-2[a]	0.02	0.02	0.07	-0.08	-0.04	0.06	-0.07	0.06	-0.13*	0.02	0.290	0.226
Pgi-2[c]	0.07*	-0.11	0.00	-0.01	-0.06	-0.07	-0.01	-0.02	0.13**	-0.13	0.069	0.048
Pgi-2[d]	0.02	-0.05	0.16**	-0.10	-0.04	0.06	-0.05	0.02	-0.08	-0.07	0.069	0.658
Pgi-2[f]	-0.01	-0.04	-0.09	-0.08	0.02	0.03	-0.05	-0.02	0.05	-0.07	1.000	0.069
Pgi-3[a]	-0.02	-0.08	-0.02	0.08	-0.01	-0.02	0.01	-0.08	-0.02	0.08	0.771	0.315
Pgi-3[c]	-0.03	-0.08	-0.05	0.11*	-0.01	0.03	-0.02	-0.03	-0.01	0.05	0.351	0.678
Average	0.02	-0.03	-0.03	-0.02	-0.01	-0.00	-0.00	-0.01	-0.03	-0.04		

[1]Overall correlogram significance (Bonferroni approximation). * = $P < 0.05$; ** = $P < 0.01$.

served at a scale of 3 m. A relatively higher mean Moran's *I* value in the first distance class was observed in MIM (0.30) than in KOS (0.07) or NOD (0.02).

The distance at which the mean Moran's *I* value first intersects the E(*I*) value may represent the shortest length of an irregularly shaped patch size (Sokal, 1979). The approximate minimum patch width also was varied among the three populations: MIM of 5 m, KOS of 10 m, and NOD of 3 m (Figure 1).

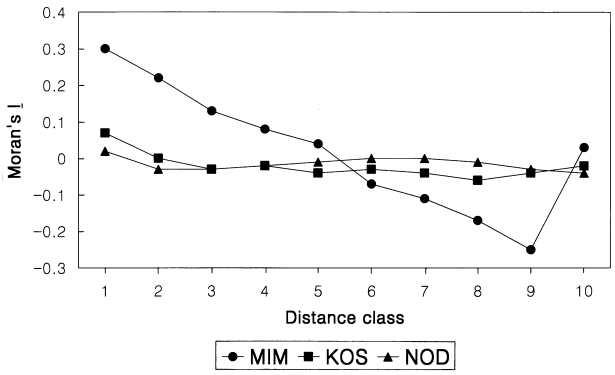

Figure 1. Correlograms for populations MIM, KOS, and NOD using mean values of Moran's *I* and 10 distance intervals. Distance classes in meters as in Tables 2 to 4.

Discussion

The ratio of significant *I* values in populations of *H. hakuunensis* was higher than predicted by a 5% type I error, indicating that genetic structuring within populations existed for the three populations. The mean correlogram of a population of *H. hakuunensis* indicates that the minimum patch width was approximately 3-10 m. The level of genetic relatedness seen among nearby individuals in these sites could easily be explained by localized seed dispersal.

The number of significant *I* values and the average Moran's *I* values for each distance class indicates that population MIM is more structured than populations KOS and NOD. Factors possibly contributing to the different levels of genetic substructuring include differences in density, topography, colonization history, or any of a number of others (Hamrick and Nason, 1996; J. L. Hamrick, pers. comm.). The area occupied by population MIM was cleared to plant chestnut trees about 20 years ago (J. S. Park, a landowner, pers. comm.). It is highly likely that Population MIM was established by a few seed sources from adjacent areas, and the size of the population has been rapidly increasing due to sufficient resources (i.e., fertilizers) and low interspecific competition. A larger population size of *H. hakuunensis* was observed in an orchard of chestnut trees compared to populations found in an understory of pine forests and forest margins. More limited pollinator flight distance in a dense young population under chestnut trees, coupled with limited seed dispersal may be factors for shaping a strong spatial genetic substructuring in MIM. In contrast, population NOD is located on undisturbed, high elevation, relatively open grasslands in which *H. hakuunensis* is abundant (several thousands individuals are scattered over the grasslands)

with *Hosta capitata* (Liliaceae) and other herbs. In this habitat, the pollinator flight range would be relatively long, resulting in a relatively large neighborhood size. An intermediate degree of genetic structuring was observed in KOS. Only 75 individuals were recorded in a 20 × 50 m area of dense pine forests with other shrubs such as oaks and sumacs. Under such a dense forest, the pollinator flight distance would be restricted. The age of the oldest *Pinus densiflora* was 68 years old in KOS, suggesting no fire had occurred during the past several decades.

Hemerocallis hakuunensis has no specialized mechanisms for seed dispersal (ovate 5 mm long seed), and many seedlings are found near maternal plants in natural populations (Chung, pers. obs.). Since gene movement in seed plants is a sequential (two-step process via pollen then by seed), observed genetic structure may be most strongly influenced by limited seed dispersal compared to pollen movement. Even with long distance pollen flow, limited seed movement can result in patches of halfsibs (Hamrick and Nason, 1996). In other words, the male gamete moves twice, but the female gamete only moves via seed. Thus, family structure can result solely by limited seed dispersal. Considering this argument, the considerably weaker genetic substructuring found in NOD and KOS compared with that of MIM also suggests that the thinning process between the seedling and adult stages does away with most of the genetic structure in populations NOD and KOS, if interspecific competitions with other herbs is strong. The greater density of MIM might indicate less thinning of new recruits, which could explain the higher relatedness values seen in this site.

It may be of interest to compare the present results with a previous study of another outcrossing liliaceous plant which has a similar life history and ecological traits with the *Hemerocallis* species. Maki and Masuda (1994) examined spatial genetic structure using the same computer program as in this study in two outcrossing populations (10 × 20-m and 2 × 20-m areas) of *Chinographis japonica* var. *japonica*. The variety, occurring in the understory of forests in western Japan, is a self-incompatible perennial. As with *H. hakuunensis*, bees visit the flowers and no specialized mechanism of seed dispersal is known (Maki and Masuda, 1994). The pattern of overall correlogram found in *C. japonica* var. *japonica* (Moran's *I* values were significant in 15 [23%] and [27%] of 66 cases in two populations) is similar to those of *H. hakuunensis*, parIn summary, the approximate minimum patch width in the populations of *H. hakuunensis* examined is 3-10 m, indicating that populations are structured on a small scale. This study describes in detail several features of the fine-scale population genetic structure, which may result from the differences in density, colonization history, thinning processes or a number of other factors.

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Hemerocallis hakuunensis (Liliaceae) 三種族群之基因構造的地域分佈

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分別來自 *Hemerocallis hakuunensis* 之三種族群，即［MIM］、［KOS］及［NOD］的 99、75 和 71 個體被定位且收集其葉試樣以推算是否從其生態特性之參數可看出此三族群之基因構造的地域分佈。從 MIM 可發現相當程度之基因構造的地域分佈，但 NOD 及 KOS 者則較弱。在 MIM 之 260 個案例中有 137 個（佔 52.7%）其 Moran's *I* 值和期望值（-0.010）有顯著不同，NOD 240 個案例中之 25 個（佔 10.4%），KOS 270 個案例中之 55 個（佔 20.4%）有顯著之 Moran's *I* 值。粗估之 minimum patch width 在三族群也不同，即：NOD = 3 m，MIM = 5 m 及 KOS = 10 m。此 patch width 之差異可能是密度，棲地繁殖歷史，消減過程或若干其他因子在三族群間之不同所導致。

關鍵詞：異構²¡；密度；*Hemerocallis hakuunensis*；Moran's *I*；地域自相關。