Journal of Ecology 1999, **87**, 244–257

Genet structure and determinants of clonal structure in a temperate deciduous woodland herb, *Uvularia perfoliata*

HIROSHI KUDOH*†§, HIROYUKI SHIBAIKE†¶, HIDEKI TAKASU‡, DENNIS F. WHIGHAM* and SHOICHI KAWANO*†

**Smithsonian Environmental Research Center, Edgewater, MD 21037, USA; †Department of Botany, Graduate School of Science, Kyoto University, Kyoto 606–8502, Japan; and ‡Department of Biology, College of Education, Wakayama University, Wakayama 640–8441, Japan*

Summary

1 We used isozyme variation to examine the genet structure of *Uvularia perfoliata* patches in gap and closed canopy habitats in a temperate deciduous forest in Maryland, USA.

2 A large patch in a gap habitat was composed of a small number of widely spread genets with many ramets, and a large number of genets with more restricted distribution and few ramets. Genets with many ramets were patchily distributed at a metre scale. Analysis of genet structure on a scale of square centimetres, however, revealed that the genets were highly intermingled with no clear boundaries between them. The presence at both scales of sampling of many genets with unique multilocus genotypes indicated continuing genet recruitment within the population.

3 In the closed canopy habitat, the patches examined were each composed of a single unique multilocus genotype, suggesting that each had developed by asexual propagation following the establishment of a single genet.

4 The clonal structure of *U. perfoliata* patches in both gap and closed canopy habitats therefore appears to depend on recruitment patterns of genets. Populations in closed canopy habitats are characterized by a 'waiting' strategy, in which asexual ramet production maintains populations until genet recruitment by seed production can occur under the more optimal conditions associated with canopy gaps. Extended sampling suggests that the genetic diversity of *U. perfoliata* populations is primarily controlled by the disturbance regime of the forest canopy.

Keywords: clonal diversity, gap, genet structure, genetic variation, ramet, spatial structure, *Uvularia perfoliata*

Journal of Ecology (1999) **87**, 244–257

Introduction

In clonal plant species, a single genetic individual (genet) can be composed of many potentially independent subunits (ramets), and genets have the potential of increasing in size indefinitely by producing new independent ramets (Cook 1983; Kawano 1984, 1985; Watkinson & White 1985). Populations of some clonal species have been reported to be dominated by one or only a few genets (Harberd 1967; Oinonen 1967; Kemperman & Barnes 1976; Mitton & Grant 1980; Suzuki 1987). Most available evidence, however, indicates that populations of clonal species are multi-clonal (reviewed in Ellstrand & Roose 1987; Hamrick & Godt 1989; Pleasants & Wendel 1989) and thus leads to questions related to their genet structure (Cook 1979). A number of authors have investigated the genet and ramet structure of natural populations of clonal plants (Wu *et al.* 1975; Gray *et al.* 1979; Silander 1979; Mitton & Grant 1980; Reinartz & Popp 1987; Maddox *et al.* 1989; Lokker *et al.* 1994; Falińska 1995) but little is known about the processes that determine this structure (but see Falińska 1995).

Correspondence: Dr Hiroshi Kudoh/Dr Shoichi Kawano, Department of Botany, Graduate School of Science, Kyoto University, Kyoto 606–8502, Japan.

§Present address: Department of Biology, Graduate School of Science, Tokyo Metropolitan University, Hachioji, Tokyo 192–0397, Japan.

¶Present address: Laboratory of Vegetation Conservation, National Institute of Agro-environmental Sciences, 311 Kannondai, Tsukuba, Ibaraki 305–8604, Japan.

The number of genets and the number of ramets of each genet in a population depend on the demographic balance between the number of ramets produced by clonal growth, ramet mortality and the number of seedlings recruited (Silvertown & Lovett-Doust 1993; Watkinson & Powell 1993; McLellan *et al.* 1997). In temperate deciduous forests, many summergreen clonal species can persist in the low-light conditions of the understorey as a result of clonal growth and/or asexual reproduction: sexual reproduction is infrequent and is often localized to gap habitats where availability of resources (e.g. light, water, nutrients) increases (Wijesinghe & Whigham 1997). Therefore, the number and relative sizes of genets of clonal species are expected to be influenced by the distribution, size, frequency of occurrence and closure rates of canopy gaps, as these are the primary focal points of disturbance in temperate deciduous forests. The long-term turnover rates of canopy should therefore influence both the size and genetic structure of populations of herbaceous species in temperate deciduous forests.

In this paper we report the results of a study on the spatial structure of ramet/genet patches of *Uvularia perfoliata* L. (Liliaceae), a temperate deciduous woodland perennial. Seed-producing individuals are mainly found under gaps in the forest canopy (Wijesinghe & Whigham 1997). Most individuals in patches under closed canopy conditions are non-flowering and produce 0–2 ramets per year. The ramets develop at the tips of elongated stolons, which become disconnected from the parent ramet at the end of the growing season (Whigham 1974; Wijesinghe & Whigham 1997). Such asexually produced ramets do not have an aerial shoot during the year they are formed and their initial growth thus depends entirely on resources supplied by the parent ramet. Seed production is typically low, even in gap habitats. Whigham (1974) found that the number of seeds produced ranged from zero to 12 seeds m^{-2} in an area where patches averaged 241 aerial shoots m^{-2}. To evaluate the relationships between asexual and sexual reproduction on the clonal structure of *U. perfoliata*, we examined the spatial genet structure of distinct patches in gap and non-gap habitats by mapping the locations of multilocus genotypes using isozyme variation.

The study was based on a set of predictions related to the spatial genet structure expected for *U. perfoliata* patches in gap and non-gap habitats.

Non-flowering patches of ramets under closed canopy conditions

If seedling recruitment is restricted to gap habitats, such patches should be the result of clonal expansion of genets that became established during a 'past' gap event: several seedlings are likely to colonize a single gap and the patches are therefore likely to contain multiple genets. Alternatively, if seeds dispersed from a gap habitat can establish in a non-gap habitat, uniclonal patches are more probably developed by asexual propagation of a single founder genet.

Boundaries and the size distribution of genets in dense patches of shoots in gap habitats

If between-genet interactions act to prevent genets from expanding into adjacent areas that are already occupied by pre-existing genets, patches in gap habitats would be composed of a limited number of genets and boundaries between genets would be distinct or narrow. Furthermore, recruitment of new genets into dense patches would be minimal, and the patch would consist of a small number of genets each with many ramets. However, if between-genet interactions are weak or absent, we would predict that genets would become intermingled as a result of asexual propagation. We would expect the patches to consist of many individuals of the small numbers of genets that became established before or soon after the gap formed and a few ramets of each of the many genets that became established later.

The scale of genet patches in gap habitats

If under gap conditions, genets expand very little by asexual propagation and therefore persist only in the areas where seedlings had initially become established. We would expect to find a high degree of genetic diversity at a small scale (e.g. centimetres). If genets expand continuously by asexual propagation, we would expect to find that genet structure would be organized at a larger scale (e.g. metres).

Methods

PLANTS

Uvularia perfoliata is a temperate deciduous woodland perennial distributed in eastern North America. The life history of *U. perfoliata* was described by Whigham (1974). Almost all individuals reproduce asexually by stolons that begin to form at about the same time that aerial shoots appear above the ground in spring. Flowering also occurs during the period of shoot expansion and the species is predominantly an outbreeder. Seeds, which have elaiosomes and are ant dispersed, are shed in late summer or early autumn. In the autumn, aerial shoots senesce and stolons, which connect mother plants with daughter ramets, decompose, and thus separation of the new ramets formed at the tip of stolons occurs at the end of each season.

SAMPLING

The study was conducted at the Smithsonian Environmental Research Center, Edgewater, MD, USA (38°54′N, 76°33′W). Two rectangular sampling sites were established, one in 1994 and a second,

encompassing a further three plots, in 1995. The 1994 plot (16 × 10 m) (Fig. 1a) was located in an area that had previously been a canopy gap but that had been reverting to a closed condition during the previous 10 years and is hereafter referred to as the gap site. The 22 × 6-m plot established in 1995 (Fig. 1b) was located in an area in which no canopy gaps had formed in the previous 20 years and is hereafter referred to as the closed canopy site. Sampling procedures used at the two sites were designed to account for large differences in shoot densities. In the gap site, the 16 × 10-m plot was divided into 160 1-m^{-2} subplots (Fig. 1a). Shoot densities and the number of flowering shoots were scored for all subplots. Plants were sampled for enzyme electrophoresis at two different spatial scales. First, we collected leaves of the shoots nearest to all intersections of the 1 × 1-m grid, unless there were no shoots within 30 cm of the intersection (the Gap-Grid sample). Genotypes of the 129 sampled leaves were determined by isozyme electrophoresis procedures as described below. The results of this survey were used to select two additional 1 × 1-m subplots (Gap-Ql and Gap-Q2; Fig. 1a) to investigate between-genet boundaries and genet structure at a smaller (centimetre) scale. Both subplots were areas of possible between-genet boundaries (see the Results) and had shoot densities >200 shoots m^{-2}. All shoots in the two subplots ($n = 232 + 289$ for Q1 + Q2, respectively) were mapped and measured for shoot height, number of leaves, and the maximum length and width of the largest leaf, and leaves of all aerial shoots were collected for electrophoresis. Shoot height and number of leaves are good indicators of ramet size, and are highly correlated with shoot biomass (Whigham 1974).

Three isolated patches were found within the closed canopy site, and 103 shoots were mapped, measured and sampled for electrophoresis as in Gap Q1 and Q2 (Fig. 1b). The three patches are hereafter referred to as Clo-1, Clo-2 and Clo-3 (Fig. 1b): other than the

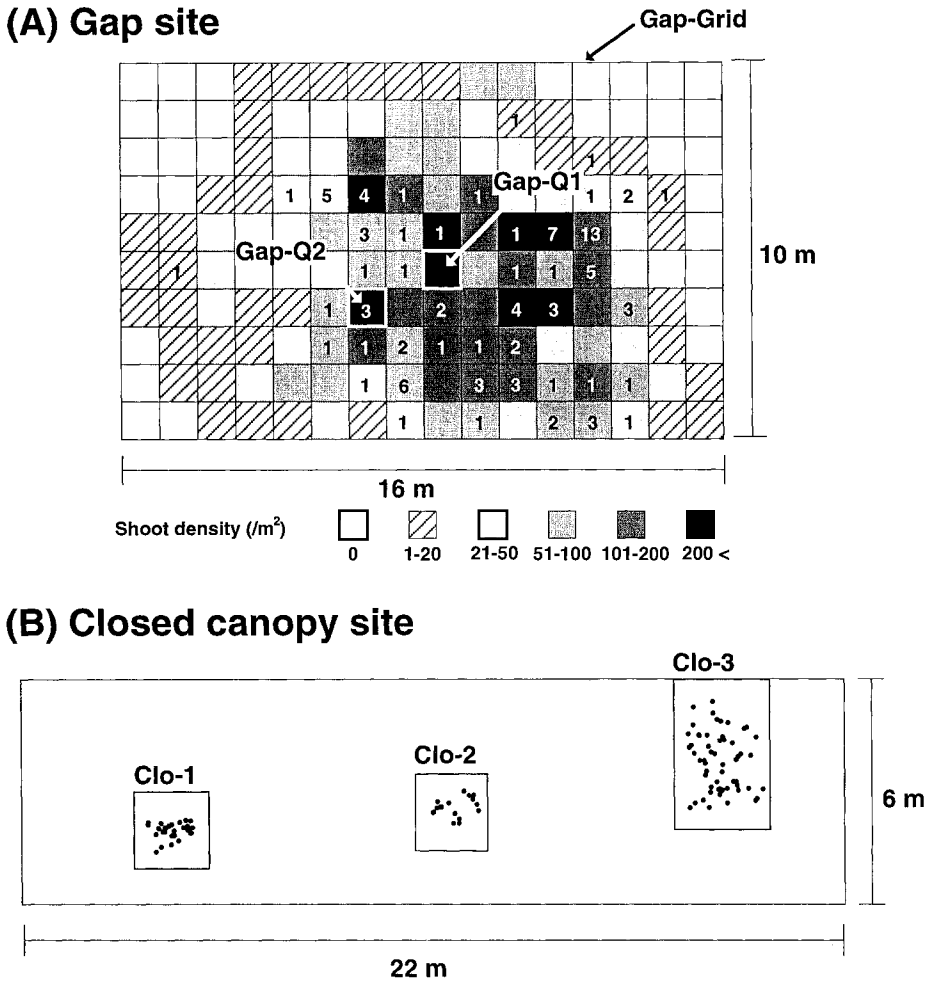

Fig. 1 Two sampling sites used in this study: (a) gap site and (b) closed canopy site. In (a) shoot density is shown by shading according to the scale provided. Numbers within subplots indicate the densities of flowering shoots: no number indicates that no flowering shoots were present. The locations of the two 1 × 1 m subplots, Gap-Q1 and Gap-Q2, are also shown. In (b) the locations of all shoots were mapped in three patches within the closed canopy site; no flowering shoots were present.

sampled shoots, the nearest individual of *U. perfoliata* was more than 10 m from the edge of the site.

ELECTROPHORESIS

Collected leaves were stored at 4 °C prior to electrophoresis. Approximately 80 mg of leaf tissue was homogenized in 1.2 ml of extraction buffer (Shiraishi 1988). The extracts were loaded on polyacrylamide vertical slab gels (Davis 1964; Orstein 1964) after refining by centrifugation (15 000 r.p.m., *c.* 20 000 **g** for 45 min at 1 °C). Electrophoresis was carried out at 4 °C, 11 mA cm^{-2} for approximately 180 min. Enzymes were stained following the protocols of Shiraishi (1988).

We initially used 180 randomly selected leaf samples together with additional samples from adjacent populations to test 22 enzyme systems. Consistently clear and polymorphic band patterns were detected for six enzyme systems, i.e. phosphogluconate dehydrogenase (6PGDH), phosphoglucomutase (PGM), diaphorase (DIA), esterase (EST), asparate aminotransferase (AAT), and acid phosphatase (ACP). All samples were then analysed for these six enzymes, and the genotypes of seven putative loci, i.e. 6*Pgdh*-1, 6*Pgdh*-2, *Pgm*, *Dia*, *Est*, *Aat*-3 and *App*, were scored for all plants (Fig. 2a). *Aat*-2 was polymorphic in the initial screening analysis, but was monomorphic in the population analysed in this study (Fig. 2a). In 6PGDH, although one of the three loci, 6*Pgdh*-3, was monomorphic, gene duplication led to polymorphy in the inter-locus heterodimers formed between the two loci, 6*Pgdh*-1 and 6*Pgdh*-3 (Fig. 2b) (Nishino & Morita 1994; S. Kawano, T. Ohkawa, K. Kitamura, H. Takasu & D. H. Whigham, unpublished observations). The number of alleles and allele frequencies of the seven polymorphic loci are shown in Table 1. There was no detectable evidence for linkage among loci.

ANALYSIS

Differences between sites (gap and closed canopy sites) in ramet traits (shoot height, number of leaves, and the maximum leaf length and width) were tested using nested analysis of variance (ANOVA) (Sokal & Rohlf 1981). The subplots (Gap-Ql, Gap-Q2, Clo-1, Clo-2 and Clo-3) were nested in the sites, and the site effect was tested against between-subplots variances ($P < 0.05$, 0.01, 0.001). Multiple comparisons of means between subplots within sites were made by Tukey's test ($P < 0.01$) (Zar 1984).

All sampled shoots were sorted by multilocus genotype based on the seven putative polymorphic loci (Table 1). Each distinct multilocus genotype detected was assumed to be a distinct genet. We used two indices as measures of genet diversity. The first measure was G/N, where G is the number of genets and N is the number of ramets sampled (Pleasants & Wendel 1989). G/N is the probability that the next ramet sampled will be a different genotype. The second measure of genet diversity is the Simpson's index corrected for finite sample size, $D = 1 - \Sigma\{[n_i(n_i - 1)]/[N(N - 1)]\}$, where n_i is the number of ramets of the ith genet (Simpson 1949; Pielou 1969). D increases as the number of genets increases and as the inequality of genet size decreases. The characteristics of D and its use in studies of clonal variation are discussed in detail by Peet (1974) and Parker (1979). Genet size was defined as number of ramets per genet in the samples, and the inequality of genet size was calculated by the Gini coefficient, a ratio of the average deviation of ramets/genet from the mean to the mean ramets/genet (Weiner & Solbrig 1984).

To investigate spatial genet structure, we compared the distribution of different multilocus genotypes in the Gap-Grid, Gap-Q1 and Gap-Q2 plots using spatial autocorrelation analyses (Sokal & Oden 1978a,b; Heywood 1991). Moran's I was calculated using the spatial analysis program written by Duncan (1990). Distance intervals used to calculate Moran's I were 1 m for the Gap-Grid and 0.1 m for the Gap-Q1 and Gap-Q2 subplots. Separate analyses were performed for the five most abundant genets, and observed Is were tested under the null hypothesis that the spatial distribution of each genet was independent ($P = 0.05$). Distribution data for plants in the closed canopy site were not analysed because each patch contained only one genotype.

Results

STAGE AND SIZE CLASS STRUCTURE OF POPULATIONS

Uvularia perfoliata shoots formed a continuous cover of varying densities over most of the gap site, and this large patch included approximately 8500 shoots, of which 102 were flowering shoots (Fig. 1a). Shoot density was highest in the centre of the grid but much lower near the edges (Fig. 1a). Eight subplots near the centre of grid had >200 shoots, and 19 subplots had between 101 and 200 shoots (Fig. 1a). The density of flowering shoots averaged 0.8 m^{-2} for the 129 subplots with *U. perfoliata* shoots. The majority of subplots contained none or only one flowering shoot, but five plots contained five or more (Fig. 1a). The relative frequency of flowering shoots was low (0.012) compared to total ramet number. In the closed canopy site, 103 shoots were distributed in three distinct patches that were separated by 5–6 m (Fig. 1b), and the number of shoots per patch were 29, 17 and 57 for Clo-1, Clo-2 and Clo-3, respectively, with no flowering shoots.

Results of nested ANOVAs indicated that for subplots where plant architecture was measured shoot height, number of leaves per shoot, and the maximum leaf length and width differed significantly between

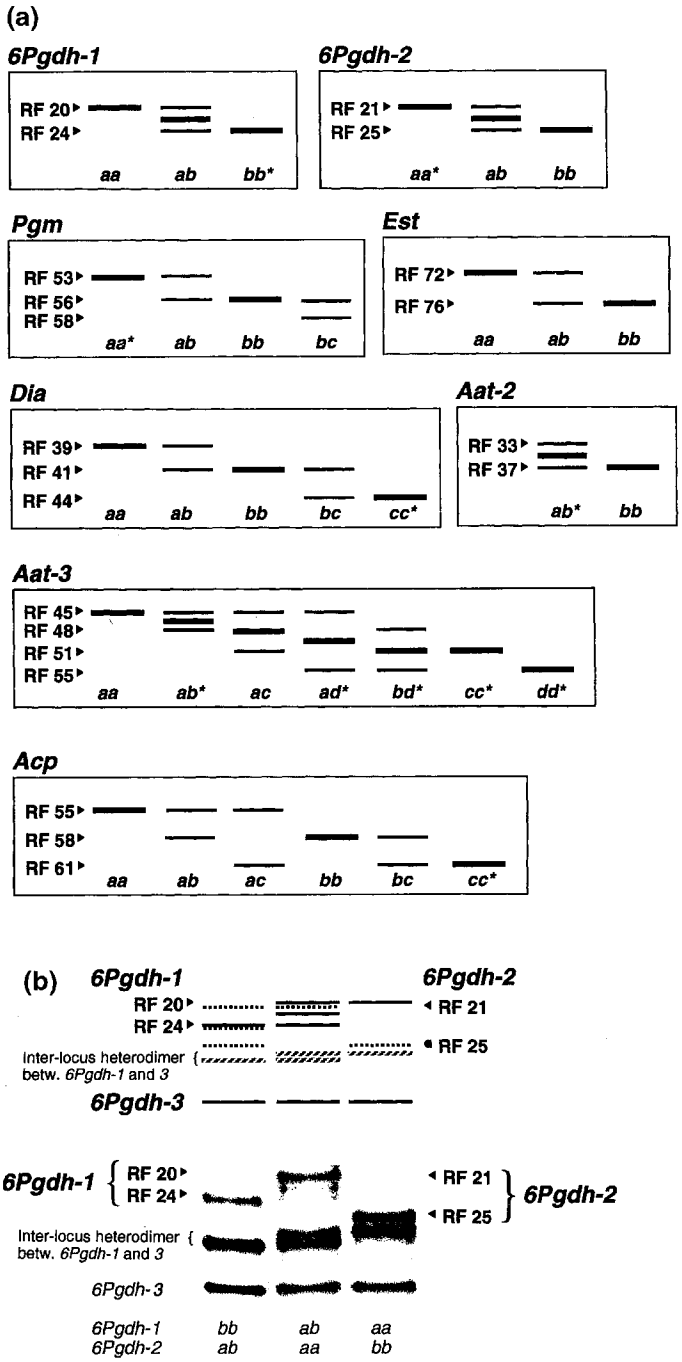

Fig. 2 (a) Zymograms illustrating genotypes of eight putative polymorphic loci of *U. perfoliata* found using samples from the study site and adjacent patches. Asterisks indicate that the genotypes were not found in the patches sampled in this study. *Aat*-2 was thus polymorphic in the initial screening analysis but monomorphic in the patch that was sampled. Rf values indicate migration distance of bands relative to the front (Rf = 100). Putative genotypes are indicated by letters that code alphabetically, with the most slowly migrating allozyme designated *a*. (b) Genotypes of three loci of 6PGDH and the presence of an inter-locus heterodimer between 6*Pgdh*-1 *and* 6*Pgdh*-3 due to gene duplication. 6*Pgdh*-3 was found to be monomorphic.

subplots within sites, but there were no significant differences (at $P < 0.05$) between sites (Table 2). Mean values of all four traits were significantly greater in the Gap-Q2 subplot than in Q1 (Table 2). In the closed canopy site, significant differences among the three patches were detected for shoot height and number of leaves per shoot (Table 2).

GENET DIVERSITY AND GENET SIZE DISTRIBUTION

Using polymorphism at seven putative loci in six enzyme systems (Fig. 2), we detected 41 multilocus genotypes in the 753 ramets sampled (Tables 1 and 3). Thirty-eight multilocus genotypes were identified in

Table 1 Forty-one multilocus genotypes based on the variation of the seven putative loci. Putative genotypes for each locus correspond to the zymograms in Fig. 2(a,b). The number of alleles and allele frequencies are also listed

Multilocus genotype	6*Pgdh*-1	6*Pgdh*-2	*Dia*	*Pgm*	*Est*	*Aat*-3	*Acp*
A	*ab*	*bb*	*bb*	*bb*	*aa*	*aa*	*ab*
B	*aa*	*bb*	*bb*	*ab*	*ab*	*aa*	*ab*
C	*aa*	*bb*	*bb*	*ab*	*ab*	*ac*	*ab*
D	*aa*	*bb*	*bb*	*bb*	*aa*	*aa*	*bc*
E	*aa*	*bb*	*bc*	*bb*	*bb*	*aa*	*ab*
F	*aa*	*bb*	*bc*	*bb*	*ab*	*aa*	*ac*
G	*aa*	*bb*	*bc*	*ab*	*ab*	*aa*	*ac*
H	*aa*	*bb*	*bc*	*ab*	*bb*	*aa*	*bc*
I	*ab*	*bb*	*bb*	*ab*	*ab*	*aa*	*ab*
J	*aa*	*bb*	*bb*	*ab*	*ab*	*aa*	*bb*
K	*aa*	*ab*	*bb*	*ab*	*aa*	*aa*	*ac*
L	*aa*	*bb*	*ab*	*bb*	*ab*	*ac*	*ab*
M	*aa*	*bb*	*bb*	*ab*	*aa*	*aa*	*ab*
N	*aa*	*bb*	*bb*	*ab*	*aa*	*aa*	*bb*
O	*aa*	*bb*	*bb*	*bb*	*aa*	*aa*	*aa*
P	*aa*	*bb*	*bb*	*bb*	*aa*	*aa*	*ab*
Q	*aa*	*bb*	*bb*	*bb*	*bb*	*ac*	*ab*
R	*aa*	*bb*	*bc*	*bb*	*ab*	*aa*	*ab*
S	*aa*	*bb*	*bc*	*bb*	*bb*	*ac*	*ab*
T	*ab*	*bb*	*bb*	*bc*	*aa*	*aa*	*ab*
U	*ab*	*bb*	*bb*	*bb*	*ab*	*ac*	*bc*
V	*ab*	*bb*	*bb*	*ab*	*ab*	*aa*	*ac*
W	*aa*	*bb*	*bb*	*bb*	*aa*	*aa*	*bb*
X	*aa*	*bb*	*bb*	*bb*	*aa*	*ac*	*bc*
Y	*aa*	*bb*	*bc*	*bb*	*aa*	*aa*	*bc*
Z	*aa*	*bb*	*bb*	*ab*	*ab*	*aa*	*bc*
a	*aa*	*bb*	*bb*	*bb*	*ab*	*ac*	*bc*
b	*aa*	*bb*	*bc*	*bb*	*aa*	*aa*	*ab*
c	*aa*	*bb*	*ab*	*ab*	*aa*	*aa*	*bc*
d	*aa*	*ab*	*bb*	*ab*	*ab*	*aa*	*bc*
e	*aa*	*bb*	*bb*	*ab*	*bb*	*ac*	*ab*
f	*aa*	*bb*	*bc*	*ab*	*bb*	*aa*	*aa*
g	*aa*	*bb*	*bc*	*bb*	*bb*	*aa*	*bc*
h	*aa*	*bb*	*aa*	*ab*	*ab*	*ac*	*ab*
i	*aa*	*bb*	*ab*	*ab*	*ab*	*ac*	*ab*
j	*aa*	*bb*	*bb*	*ab*	*ab*	*ac*	*bc*
k	*aa*	*bb*	*bc*	*ab*	*aa*	*aa*	*bc*
l	*aa*	*bb*	*bc*	*ab*	*bb*	*aa*	*ab*
m	*aa*	*bb*	*bb*	*bb*	*bb*	*aa*	*ab*
n	*aa*	*bb*	*bb*	*bb*	*ab*	*aa*	*ab*
o	*aa*	*bb*	*bc*	*bb*	*bb*	*ac*	*bb*
Alleles*	2	2	3	3	2	2 (4)	3
Allele frequencie†							
a	0.94	0.02	0.05	0.24	0.55	0.85	0.33
b	0.06	0.98	0.79	0.73	0.45		0.47
c			0.16	0.01		0.15	0.20

* Number of alleles found from the samples in this study. For *Aat*-3, two additional alleles were found in adjacent patches, thus the total number of alleles is listed in parentheses.
† Allele frequencies were calculated on a genet basis using all genets found in this study.

the gap site, with 20 in the Gap-Grid sample, and 14 and 17 in the smaller-scale sampling of Gap-Q1 and Gap-Q2, respectively (Table 3). Each patch in the closed canopy site was composed of a single genotype unique to the patch (Table 3). The low genetic diversity within the closed canopy site resulted in a low G/N value (0.03) compared to the gap site (Table 3). Within the gap site, the probability of finding a new genet was greatest in the Gap-Grid ($G/N = 0.16$) compared to within the two smaller Gap-Q1 and Gap-Q2 subplots (Table 3). Clonal diversity showed a similar pattern to G/N, with D-values of 0.82, 0.72, 0.68 and 0.59 for Gap-Grid, Gap-Q1, Gap-Q2 and the closed canopy site, respectively (Table 3).

The Gap-Grid sample and the Gap-Q1 and Gap-Q2 subplots were all made up of a few genets with many ramets and many genets with a few ramets. The largest genet in the Gap-Q1 subplot was Genotype D which accounted for 46.1% of the shoots in this subplot and the three most abundant genotypes in sub-

Table 2 Means and standard deviations of shoot height, number of leaves, leaf length and leaf width of ramets sampled from the Q1 and Q2 subplots in the gap site and three patches (Clo-1, -2, and -3) in the closed canopy site. *F*-values of one-way nested ANOVAs are also listed (NS = non-significant at $P = 0.05$; * $P < 0.01$; ** $P < 0.001$). Different letters indicate significant differences ($P < 0.01$) between subplots and patches within the gap and closed canopy sites, respectively

	Gap site			Closed canopy site				Nested ANOVA	
	Q1 ($n = 232$)	Q2 ($n = 289$)	Total ($n = 521$)	Clo-1 ($n = 29$)	Clo-2 ($n = 17$)	Clo-3 ($n = 57$)	Total ($n = 103$)	Sites	Subplots (nested in sites)
Shoot height (cm)	11.3 ± 4.2 b	13.5 ± 5.3 a	12.5 ± 4.9	11.3 ± 3.2 b	11.1 ± 4.0 b	15.3 ± 5.0 a	13.5 ± 4.6	0.09 NS	15.7**
Number of leaves	3.9 ± 0.9 b	4.2 ± 1.0 a	4.1 ± 1.0	3.7 ± 1.0 b	3.9 ± 0.9 ab	4.5 ± 0.9 a	4.2 ± 1.0	0.23 NS	9.39**
Leaf length (cm)	3.8 ± 1.2 b	4.3 ± 1.3 a	4.1 ± 1.3	4.2 ± 1.2 a	4.0 ± 1.2 a	4.5 ± 1.1 a	4.3 ± 1.2	2.19 NS	7.59**
Leaf width (cm)	1.6 ± 0.5 b	1.8 ± 0.5 a	1.7 ± 0.5	1.7 ± 0.4 a	1.7 ± 0.5 a	1.7 ± 0.5 a	1.7 ± 0.4	0.0004 NS	4.79*

plot Gap-Q1 together accounted for 79.7% of shoots (Table 3). In the Gap-Q2 subplot, C was the most abundant genotype (52.6% of the total) and 80.3% of the ramets belonged to three genets (Table 3). While we only sampled a small percentage of the total number of shoots present in the Gap-Grid, the size distribution of the genotypes was similar to the pattern for the subplots (Fig. 3). Only five of the 20 genotypes identified represented more than 5% of the sampled shoots, and 15 genotypes had less than five shoots (Table 3). The pattern of asymmetry in the genet size distribution at the gap site was also indicated by high values for Gini coefficients (0.71, 0.78 and 0.80 for Gap-Grid, Gap-Q1 and Gap-Q2, respectively) compared to the patches at the closed canopy site (Gini = 0.19) (Table 3). The three most abundant genotypes in the Gap-Q1 and Gap-Q2 subplots were all also found in the Gap-Grid (Table 3). Eight genotypes were unique to Gap-Q1 and 10 genotypes were unique to Gap-Q2 (Table 3): all contained few ramets and most only had a single ramet (Table 3).

SPATIAL DISTRIBUTION OF GENETS AND BETWEEN-GENET BOUNDARIES

A patchy distribution of genets was detected at a metre scale in the Gap-Grid (Fig. 4). For example, genotype A was spread over an area of at least 30 m^2 (Fig. 4). Spatial autocorrelation analyses indicated that the probability of finding ramets of the same genet was significantly higher than expected within 3–5 m and lower at distances greater than 5 m for genotypes A, B and C (Fig. 6). This pattern was not clear for genotypes D and E (Fig. 6), which were less abundant and spatially intermingled with each other and with other adjacent genets (Fig. 4).

In contrast to the patchy pattern at the meter scale, there was no pattern in the Gap-Q1 and Gap-Q2 subplots where genotypes were intermingled without any distinct between-genet boundaries (Fig. 5). Spatial autocorrelation analyses indicated that spatial dependency of ramet locations of a genet was absent or very weak at a spatial scale of less than 1 m (Fig. 7). Although several genets showed significantly positive Moran's *I* within a distance of 30 cm (Fig. 7), the values were much less than those calculated for Gap-Grid samples at the metre scale (Fig. 6). In the closed canopy site, each patch had a distinct genet, i.e. m in Clo-1, n in Clo-2 and o in Clo-3 (Fig. 1b and Table 3).

Discussion

CLONAL DIVERSITY

Clonal diversities measured by *D* were higher in the gap habitat (range 0.68–0.82) than under the closed canopy (0.59) (Table 3). The values obtained in the gap site are slightly higher than the mean *D* (0.62) reported in a review of studies of multi-clonal popu-

Table 3 Number of ramets for each multilocus genotype found in the samples from the gap and closed-canopy sites. Percentages of the total number of ramets for each sample are shown in parentheses. Total number of ramets, number of genotypes, *G*/*N*, *D* and Gini coefficients are also listed. Alphabetical letters indicating multilocus genotypes correspond to those in Table 1 and Figs 4 and 5

	Gap site			Closed canopy site		
Multilocus genotype	Grid	Q1	Q2	Clo-1	Clo-2	Clo-3
A	44 (34.1)	1 (0.4)	2 (0.7)			
B	26 (20.2)	2 (0.9)	2 (0.7)			
C	15 (11.6)		152 (52.6)			
D	12 (9.3)	107 (46.1)	54 (18.7)			
E	7 (5.4)	33 (14.2)	26 (9.0)			
F	4 (3.1)					
G	3 (2.3)					
H	3 (2.3)					
I	3 (2.3)					
J	2 (1.5)	45 (19.4)				
K	1 (0.8)					
L	1 (0.8)					
M	1 (0.8)					
N	1 (0.8)					
O	1 (0.8)	18 (7.8)	5 (1.7)			
P	1 (0.8)		3 (1.0)			
Q	1 (0.8)					
R	1 (0.8)					
S	1 (0.8)					
T	1 (0.8)					
U		19 (8.1)				
V		1 (0.4)				
W		1 (0.4)				
X		1 (0.4)				
Y		1 (0.4)				
Z		1 (0.4)				
a		1 (0.4)				
b		1 (0.4)				
c			16 (5.5)			
d			12 (4.2)			
e			7 (2.4)			
f			3 (1.0)			
g			2 (0.7)			
h			1 (0.3)			
i			1 (0.3)			
j			1 (0.3)			
k			1 (0.3)			
l			1 (0.3)			
m				29 (100)		
n					17 (100)	
o						57 (100)
Total	129	232	289	29	17	57
Genotypes	20	14	17	1	1	1
G/*N*	0.16	0.06	0.06	0.03*		
D	0.82	0.72	0.68	0.59*		
Gini coefficient	0.71	0.78	0.80	0.19*		

* Clo-1, 2, and 3 were pooled for calculation of *G*/*N*, *D* and Gini coefficient for the closed canopy site.

lations (Ellstrand & Roose 1987). High clonal diversity and inequality of genet size in the gap habitat may be explained by the low ratio of recruitment from sexual reproduction compared with asexual ramets (a value of 0.054 was reported for *U. perfoliata* in a North Carolina forest; Whigham 1974). Watkinson & Powell (1993) predicted similar results using a computer simulation to analyse the processes that determine the number and relative size of genets in *Ranunculus repens*. They found that *G*/*N* and the inequality of genet size were determined by the ratio of seedling to ramet recruits rather than by ramet mortality, rate of clonal growth or seedling number. Although the model developed for *R. repens* is not directly applicable to this study, their simulations showed that a very low ratio of seedling to ramet recruits is enough to maintain approximately 20 genets m^{-2} in a population with approximately 200 ramets m^{-2} (Wat-

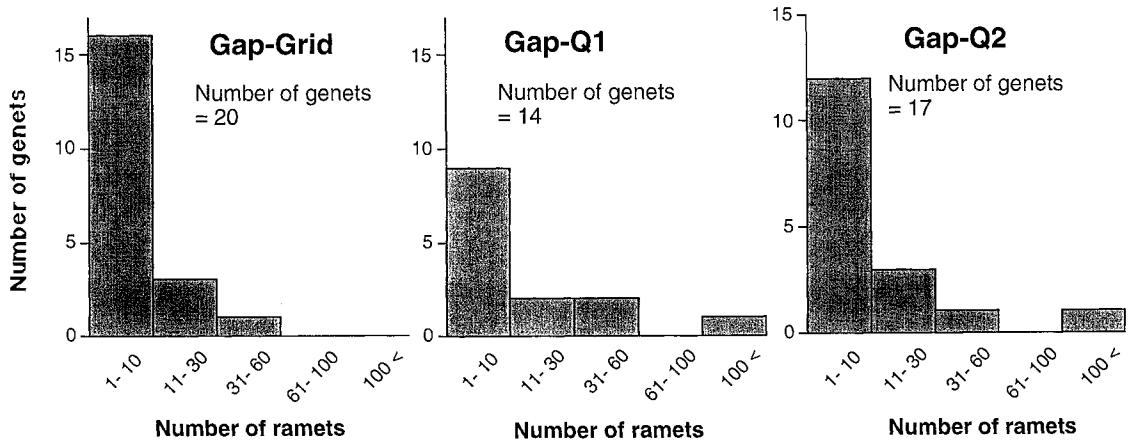

Fig. 3 Genet size structure in the gap site. Genet size was based on the number of ramets within each sample.

kinson & Powell 1993), values close to those observed in this study.

SPATIAL GENET STRUCTURE IN GAP AND NON-GAP HABITATS

Flowering individuals only occurred in the gap site, suggesting that seed production, the source of new genets, is localized in canopy gap habitats. The large number of genets in the gap site with only a few ramets (Table 3) indicates that seedling recruitment occurs in gaps. Results of isozyme analyses of three patches in the closed canopy habitat (Table 3), however, indicate that seedling recruitment also occurs in closed canopy habitats (our first prediction). Seed production of *U. perfoliata* mainly occurs under gaps, and it therefore appears that seed dispersal away from such sites is critical for genet recruitment under closed canopy conditions. Seeds of *U. perfoliata* are dispersed by ants, which are attracted by elaiosomes attached to the seeds. Seeds are removed by ants very effectively, 98% of the fresh seeds that were placed on litter, and 56% of the seeds placed beneath the litter, were removed by ants within 3 days (Whigham 1974). We do not know how far ants disperse *U. perfoliata* seeds, but we have observed ant dispersal distances of greater than 2 m (D.F. Whigham, unpublished data). A very similar situation, seedling recruitment both in gap and closed canopy habitats, has been reported in *Disporum smilacinum*, a temperate woodland herb with a similar life history to that of *U. perfoliata* (Kawano 1984; M. Nakagawa & S. Kawano, unpublished observations).

Inferences about the distribution of genets (our second prediction) depend on the scale of sampling. Although sampling at a metre scale showed clearly defined patches for the most abundant genets (A, B and C) (Fig. 4), thus supporting the first alternative, results at a centimetre scale indicated that genets were intermingled with one another, even though a few genets contributed the majority of the ramets in the Gap-Q1 and Q2 subplots. At both scales, most of the genets contained between 1 and 10 ramets (Fig. 3). The large patches of genets A, B, and C suggested by the Gap-Grid do not therefore represent a continuous cover of ramets of a small number of genets but intermingled patches that include a large number of genets with few individuals (i.e. the second alternative of prediction 2). The data collected in this study have been used in a simulation study to determine the probability of identifying individual clones (Harada *et al.* 1997). The simulation study clearly showed that the design that is used to sample a patch influences the probability of correctly identifying the genetic structure of patches in plant populations (Barkham & Hance 1982; Epperson & Allard 1989; Kitamura *et al.* 1997a, b; Kawano & Kitamura 1997).

The lack of distinct boundaries between genets at a centimetre scale indicates that between-genet interactions are minimal in *U. perfoliata*. Our findings contrast with patterns found for *Xanthoxylum americanum*, *Comus racemosa* and *Rhus glabra*, where genet patches had distinct boundaries and genets with only a few ramets were absent (Reinartz & Popp 1987). Distinct genet patches may also develop because of differential responses of genets to a patchy environment (Wu & Antonovics 1976; Burdon 1980). Spatial heterogeneity of microenvironments does not, however, appear to be important for *U. perfoliata*, because we found many intermingled genets at a very small (centimetre) scale. We infer that the presence of a few distinct genet patches at the metre scale (Fig. 4) simply reflects the fact that genets A, B, and C colonized the area earlier than the other genets and have spread further asexually (see our third prediction).

In gap environments, *U. perfoliata* plants typically produce two daughter ramets that establish at approximately 20 cm from the parent (Whigham 1974; Wijesinghe & Whigham 1997). An *U. perfoliata* genet could, thus, expand to cover the area observed in this study over a 5–10-year period. Although we do not know the exact year that the canopy gap was

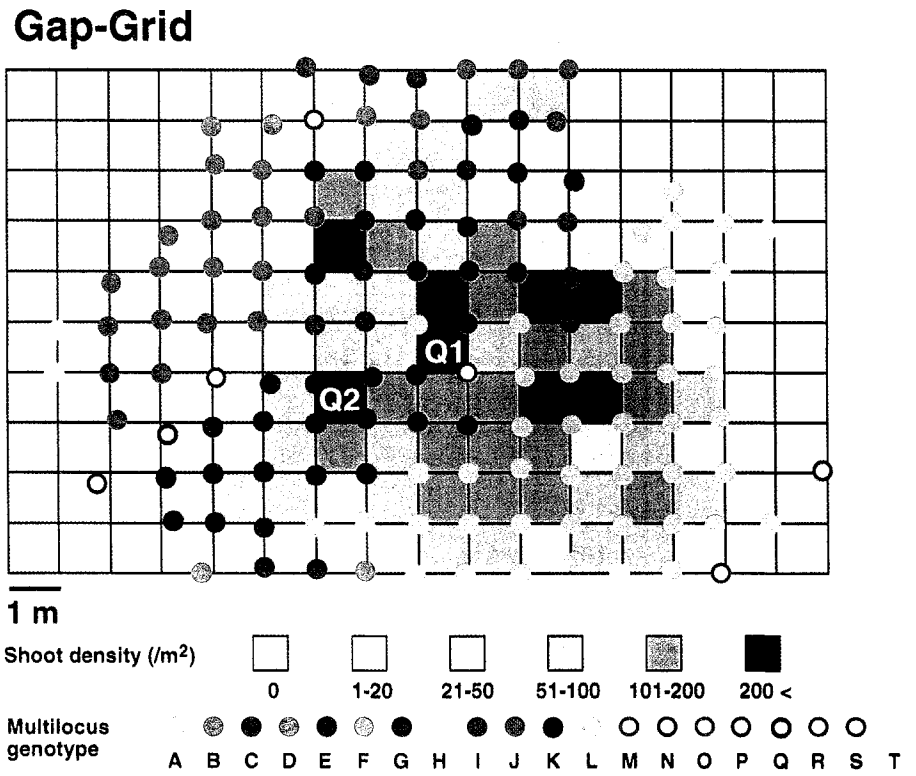

Fig. 4 Spatial genet structure at a 1 × 1-m scale (Gap-Grid) in the gap site. The genetic identities of the shoots at the intersections of the grid are colour-coded to specific multilocus genotypes listed in Tables 1 and 3. Shoot densities within 1 × 1-m subplots are also shown.

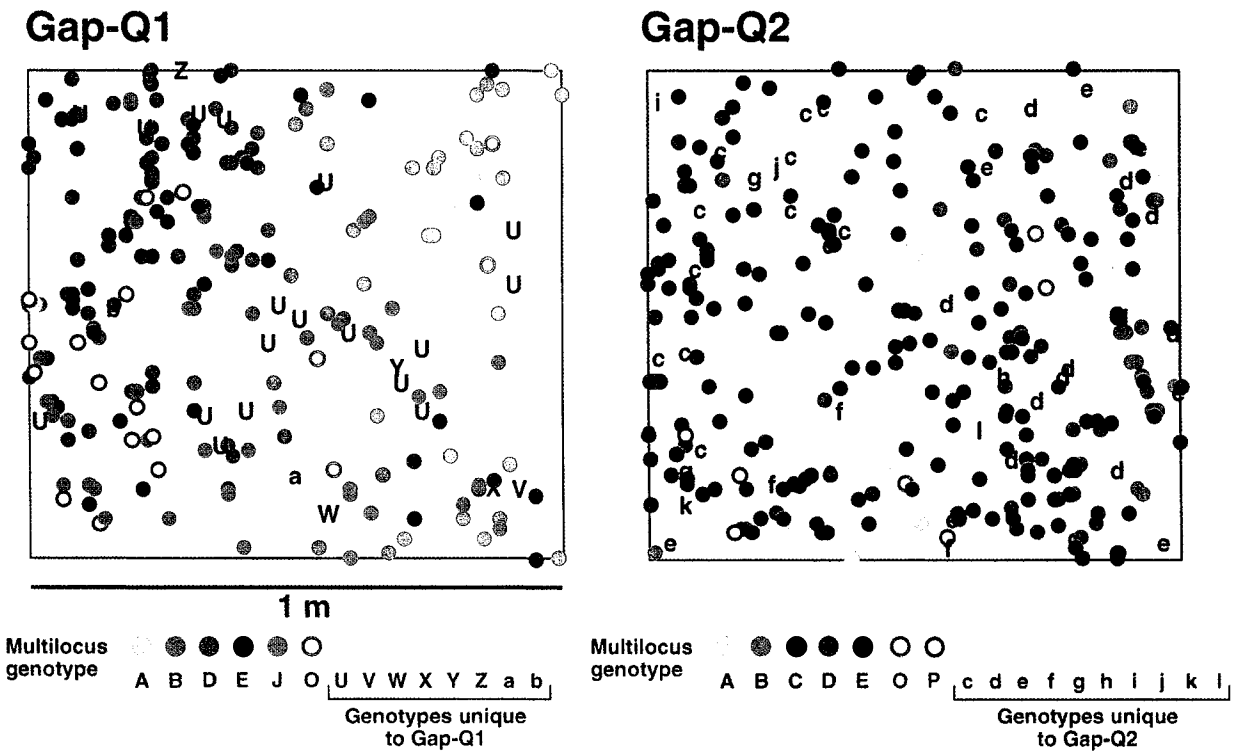

Fig. 5 Spatial genet structure at a centimetre scale (Gap-Q1 and Gap-Q2) in the gap site. The multilocus genotypes that were found in the 1 × 1-m grid (Fig. 4) are shown by coloured symbols. Multilocus genotypes that were unique to the subplots are shown by letters (as defined in Tables 1 and 3).

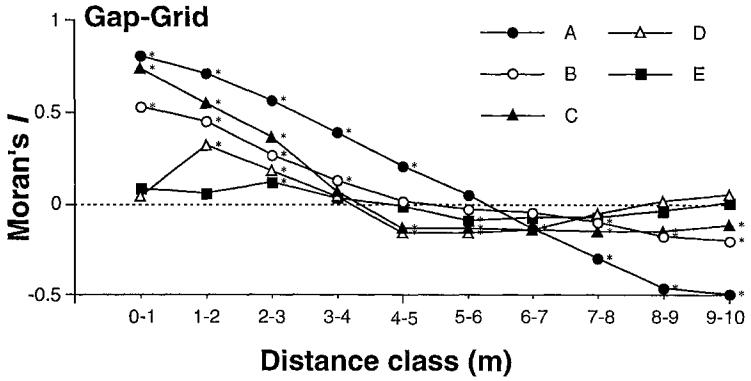

Fig. 6 Spatial autocorrelations measured by Moran's *I* at 10 different distance intervals for the five largest genets in the 1 × 1-m Gap-Grid. Separate analyses were performed for the most abundant five genets. Asterisks indicate significant deviation (at $P = 0.05$) of observed *I* from the random spatial distribution of genets.

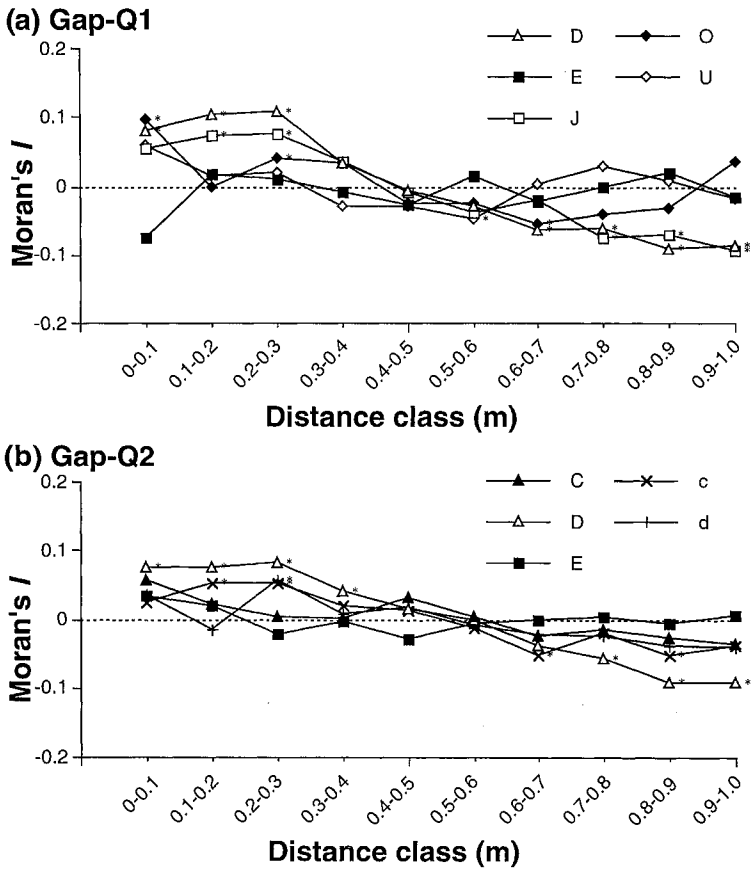

Fig. 7 Spatial autocorrelations measured by Moran's *I* at 10 different distance intervals for the five largest genets in the Gap-Q1 (a) and Gap-Q2 (b) subplots. Separate analyses were performed for the most abundant five genets for each subplot. Asterisks indicate significant deviation (at $P = 0.05$) of observed *I* from the random spatial distribution of genets.

formed, it was first observed in 1977, at which time the tree that had formed the gap was already in an advanced stage of decomposition (D. F. Whigham, personal observation). This indicates that the gap was 20–25 years old by the time of our study. Whigham (1974) reported that it takes several years for seedlings to start asexual reproduction, and this together with the amount of space covered by genets such as A, B and C (Fig. 3) leads us to believe that these genets may have become established under closed canopy conditions and been maintained by clonal propagation until the time of gap creation, when other genets were recruited. Observations at the closed canopy site clearly demonstrate that under low light conditions *U. perfoliata* can form distinct patches of ramets from a single genet. For example, genotype o in Clo-3 covered an approximately 6-m^2 area with 57 ramets (Fig. 1b and Table 3).

The genet structure observed in the gap habitat thus suggests that between-genet interaction is absent

or weak enough to allow many genets to become established. Other authors have found similarly low competitive abilities of different genets within clonal species (Rumbaugh 1970; Solangaarachchi & Harper 1989), and genet mortality is very low following seedling establishment (Ernst 1979; Solbrig *et al.* 1980; Barkham & Hance 1982; Hartnett & Bazzaz 1985; Inghe & Tamm 1985; Pitelka *et al.* 1985). Others (de Kroon *et al.* 1992; Hara & Srutek 1995) have also found evidence, based on studies of shoot growth and mortality, that competition between genets of clonal species is weak and symmetric. However, the relationships between inter-genet competition and the genet structure of patches of clonal species remain largely unknown. Because inter-genet competition depends on interference between ramets at the boundary between genets (Bülow-Olsen *et al.* 1984), studies on the boundary structure of clonal species are needed. There is some limited evidence that suggests that clonal species with long-lived connections and a high degree of physiological integration between ramets have patchily distributed genets with distinct boundaries (Reinartz & Popp 1987; Falińska 1995). Other clonal species, such as *U. perfoliata*, with short-term connections between ramets, appear to have intermingled genets without clear boundaries (Lokker *et al.* 1994; M. Nakagawa & S. Kawano, unpublished observations).

THE WAITING STRATEGY OF *U. PERFOLIATA*

Genet recruitment under closed canopy conditions may be important in the long-term maintenance of genet diversity in *U. perfoliata* populations. Data from the three closed canopy patches that were sampled suggest that genets can become established under closed canopy conditions and that they can form distinct patches with many ramets. In another study of *U. perfoliata*, Wijesinghe & Whigham (1997) showed that ramets in gap habitats produce significantly more and larger daughter ramets, and in this study we found that ramets in gap habitats became large enough to reproduce sexually. The average rate of gap formation in the canopy of temperate deciduous forests in eastern North America, estimated at approximately 1% of total land area per year (Runkle 1982; Runkle & Yetter 1987), suggests that most of the understorey will be in a closed canopy condition at any moment in time and that perennial understorey clonal and non-clonal herbs will most often persist in a vegetative state. Asexual propagation would thus enhance genet survival under suboptimal conditions (Barkham & Hance 1982; Hartnett & Bazzaz 1985; Inghe & Tamm 1985; Pitelka et al. 1985; Kelly 1995). Because the 'waiting' stage under suboptimal conditions is a suppressed phase unfavourable for sexual reproduction, the cost of maintenance of inter-ramet connections is most likely to be a factor selecting for short-term connections between parents and ramets (Kelly 1995), a life-history feature referred to as 'pseudo-annual' (Kawano 1985; cf. Salisbury 1942). The pseudo-annual strategy is shared by several herbaceous species [e.g. *Medeola virginiana* (Bell 1974; Cook 1979, 1983), *Allium monanthum* (Kawano & Nagai 1975), *Adoxa moschatellina*, *Disporum sessile* (Kawano 1985; Hori *et al.* 1992, 1995) and *D. smilacinum* (Kawano 1985)] of temperate deciduous forests (Kawano 1970, 1975).

We suggest that the life-history strategy of *U. perfoliata* can be referred to as a 'waiting strategy', which is characterized by a combination of predominantly vegetative spread under suboptimal conditions in closed canopy sites, and effective genet and ramet recruitment under optimal conditions in canopy gaps. Although for any one given time and place the existence of a genet of *U. perfoliata* under a gap is unpredictable, for the forest as a whole gap formation occurs often enough for the 'waiting strategy' to be of evolutionary significance. To evaluate further the results of this study, we have recently expanded our sampling of *U. perfoliata* to another area of the same forest and analysed the genetic structure of a further 20 *U. perfoliata* patches of varying size that had a wide range of number of ramets (10–1600 per patch; S. Kawano, T. Ohkawa, K. Kitamura, H. Takasu & D. H. Whigham, unpublished data). The 11 smallest patches had between 10 and 244 ramets per patch and were composed of only one or two genets, whereas the three largest patches had 672, 774 and 1602 ramets and contained multiple genotypes. These additional data support our conclusion that large patches, such as found in our gap site, are multi-clonal and that genet diversity increases during gap events which may occur at the same site. The presence of numerous patches with one or two genets also supports the conclusion that seeds are dispersed away from gap habitats and that patches can develop under closed canopy conditions by clonal propagation. An alternative hypothesis, however, is that the patches with one or only a few genotypes that are found under closed canopy conditions are remnant patches that resulted from the break-up of large multi-clonal patches that had developed beneath canopy gaps. While analysis of patterns such as those described in this paper provide insight into the development of patterns of genetic diversity and the distribution of genets in clonal species such as *U. perfoliata*, validation of these findings will require either long-term monitoring of populations or experimental manipulations of genets in gap and non-gap habitats.

Acknowledgements

We thank Tatsuyoshi Morita, Keiko Kitamura, Jay O'Neill, and Yumi Kudoh for assistance in field and laboratory work, and also Yoh Iwasa and Yuko Harada for discussion during the course of the present study. Thanks are also due to Chris Peterson for pro-

viding computer software used in the spatial analysis and Don Weller for consultation in the spatial analysis. We also thank to the two anonymous referees for their helpful comments. This study was supported by a Smithsonian Regents fellowship to S. Kawano (corresponding author), Grants-in-Aid no. 01041053 and no. 05401090 for Overseas Research from the Ministry of Education, Science and Culture of Japan (S. Kawano), a Smithsonian Postdoctoral fellowship and JSPS Postdoctoral Fellowship for Research Abroad (H. Kudoh), and the Smithsonian Environmental Sciences Program (D. F. Whigham), for which we extend our gratitude.

References

Barkham, J.P. & Hance, C.E. (1982) Population dynamics of the wild daffodil (*Narcissus pseudonarcissus*). III. Implications of a computer model of 1000 years population change. *Journal of Ecology*, **70**, 323–344.

Bell, A.D. (1974) Rhizome organization in relation to vegetative spread in *Medeola virginiana*. *Journal of the Arnold Arboretum*, **55**, 458–468.

Bülow-Olsen, A., Sackville Hamilton, N.R. & Hutchings, M.J. (1984) A study of growth form in genets of *Trifolium repens* L. as affected by intra- and interplant contacts. *Oecologia*, **61**, 383–387.

Burdon, J.J. (1980) Intra-specific diversity in a natural population of *Trifolium repens*. *Journal of Ecology*, **68**, 717–735.

Cook, R.E. (1979) Asexual reproduction: a further consideration. *American Naturalist*, **113**, 769–772.

Cook, R.E. (1983) Clonal plant populations. *American Scientist*, **71**, 244–253.

Davis, B.J. (1964) Disk electrophoresis. II. Method and application to human serum proteins. *Annals of the New York Academy of Science*, **121**, 404–427.

De Kroon, H., Hara, T. & Kwant, R. (1992) Size hierarchies of shoots and clones in clonal herb monocultures: do clonal and non-clonal plants compete differently? *Oikos*, **63**, 410–419.

Duncan, R. (1990) *Spatial Analysis Programmes*. School of Forestry, University of Canterbury, Christchurch, New Zealand.

Ellstrand, N.C. & Roose, M.L. (1987) Patterns of genotypic diversity in clonal plant species. *American Journal of Botany*, **74**, 123–131.

Epperson, B.K. & Allard, R.W. (1989) Spatial autocorrelation analysis of the distribution of genotypes within populations of lodgepole pine. *Genetics*, **121**, 369–377.

Ernst, W.H.O. (1979) Population biology of *Allium ursinum* in Northern Germany. *Journal of Ecology*, **67**, 347–362.

Falińska, K. (1995) Genetic disintegration in *Filipendula ulumaria*: consequences for population dynamics and vegetation succession. *Journal of Ecology*, **83**, 9–21.

Gray, A.J., Parsell, R.J. & Scott, R. (1979) The genetic structure of plant populations in relation to the development of salt marshes. *Ecological Processes in Coastal Environments* (eds R.L. Jefferies & A.J. Davy), pp. 43–64. Blackwell Scientific Publications, Oxford, UK.

Hamrick, J.L. & Godt, M.J.W. (1989) Allozyme diversity in plant species. *Plant Population Genetics, Breeding and Genetic Resources* (eds A.H.D. Brown, M.T. Clegg, A.L. Kahler & B.S. Weir), pp. 43–63. Sinauer, Sunderland, MA.

Hara, T. & Šrůtek, M. (1995) Shoot growth and mortality patterns of *Urtica dioica*, a clonal forb. *Annals of Botany*, **76**, 235–243.

Harada, Y., Kawano, S. & Iwasa, Y. (1997) Probability of clonal identity: inferring the relative success of sexual vs. clonal reproduction from spatial genetic patterns. *Journal of Ecology*, **85**, 591–600.

Harberd, D.J. (1967) Observations on natural clones in *Holcus mollis*. *New Phytologist*, **66**, 401–408.

Hartnett, D.C. & Bazzaz, F.A. (1985) The genet and ramet population dynamics of *Solidago canadensis* in an abandoned field. *Journal of Ecology*, **73**, 407–413.

Heywood, J.S. (1991) Spatial analysis of genetic variation in plant populations. *Annual Review of Ecology and Systematics*, **22**, 335–355.

Hori, Y., Yokoi, T. & Yokoi, Y. (1992) Size-structures and reproductive characteristics in diploid and triploid populations of *Disporum sessile D.* Don (Liliaceae). *Plant Species Biology*, **7**, 77–85.

Hori, Y., Yokoi, T. & Yokoi, Y. (1995) Effects of light intensity on the size structure and establishment of diploids and triploids of *Disporum sessile*. *Plant Species Biology*, **10**, 11–16.

Inghe, O. & Tamm, C.O. (1985) Survival and flowering of perennial herbs. IV. The behaviour of *Heptica nobilis* and *Sanicula europaea* on permanent plots during 1943–1981. *Oikos*, **45**, 400–420.

Kawano, S. (1970) Species problems viewed from productive and reproductive biology. I. Ecological life histories of some representative members associated with temperate deciduous forests in Japan. *Journal of the College of Liberal Arts, Toyama University, Japan*, **3**, 181–213.

Kawano, S. (1975) The productive and reproductive biology of flowering plants. II. The concept of life history strategy in plants. *Journal of the College of Liberal Arts, Toyama University, Japan*, **8**, 51–86.

Kawano, S. (1984) Population biology and demographic genetics of some liliaceous species. *Korean Journal of Plant Taxonomy*, **14**, 43–57.

Kawano, S. (1985) Life history characteristics of temperate woodland plants in Japan. *The Population Structure of Vegetation* (ed. J. White), pp. 515–pp. 549. Dr W. Junk Publishers, Dordrecht, the Netherlands.

Kawano, S. & Kitamura, K. (1997) Demographic genetics of the Japanese beech, *Fagus crenala*, in the Ogawa Forest Preserve, Ibaraki, Central Honshu, Japan. III. Population dynamics and genetic substructuring within a metapopulation. *Plant Species Biology*, **2**, 157–177.

Kawano, S. & Nagai, Y. (1975) The productive and reproductive biology of flowering plants. I. Life history strategies of three *Allium* species in Japan. *The Botanical Magazine, Tokyo*, **88**, 281–318.

Kelly, C.K. (1995) Thoughts on clonal integration: facing the evolutionary context. *Evolutionary Ecology*, **9**, 575–585.

Kemperman, J.A. & Barnes, B.V. (1976) Clone size in American aspens. *Canadian Journal of Botany*, **54**, 2603–2607.

Kitamura, K., Shimada, K., Nakashima, K. & Kawano, S. (1997a) Demographic genetics of the Japanese beech, *Fagus crenala*, in the Ogawa Forest Preserve, Ibaraki, Central Honshu, Japan. I. Spatial genetic substructuring in local populations. *Plant Species Biology*, **12**, 107–135.

Kitamura, K., Shimada, K., Nakashima, K. & Kawano, S. (1997b) Demographic genetics of the Japanese beech, *Fagus crenala*, in the Ogawa Forest Preserve, Ibaraki, Central Honshu, Japan. II. Genetic substructuring among classes in local populations. *Plant Species Biology*, **12**, 137–155.

Lokker, C., Susko, D., Lovett-Doust, L. & Lovett-Doust, J. (1994) Population genetic structure of *Vallisneria americana*, a dioecious clonal macrophyte. *American Journal of Botany*, **81**, 1004–1012.

McLellan, A.J., Prati, D., Kaltz, O., Schmid, B. (1997) Structure and analysis of phenotypic and genetic variation in clonal plants. *The Ecology and Evolution of Clonal Plants* (eds H. de Kroon & J. van Groenendael), pp. 185–pp. 210. Backhuys Publishers, Leiden, the Netherlands.

Maddox, G.D., Cook, R.E., Wimberger, P.H. & Gardescu, S. (1989) Clonal structure in four *Solidago allissima* (Asteraceae) populations: rhizome connections within genotypes. *American Journal of Botany*, **76**, 318–326.

Mitton, J.B. & Grant, M.C. (1980) Observations on the ecology and evolution of quaking aspen, *Populus tremuloides*, in the Colorado front range. *American Journal of Botany*, **67**, 202–209.

Nishino, T. & Morita, T. (1994) 6-phosphogluconate dehydrogenase (6PGD) gene duplication in *Kalimeris* (Asteraceae). *Plant Species Biology*, **9**, 91–97.

Oinonen, E. (1967) The correlation between the size of Finnish bracken [*Pteridium aquilinum* (L.) Kuhn] clones and certain periods of site history. *Acta Forestalia Fennica*, **83**, 1–51.

Orstein, I. (1964) Disk electrophoresis. I. Background and theory. *Annals of the New York Academy of Science*, **121**, 321–349.

Parker, E.D. Jr (1979) Ecological implications of clonal diversity in parthenogenetic morphospecies. *American Zoologist*, **19**, 753–762.

Peet, R.X. (1974) The measurement of species diversity. *Annual Review of Ecology and Systematics*, **5**, 285–307.

Pielou, E.C. (1969) *An Introduction to Mathematical Ecology*. WileyInterscience, New York, NY.

Pitelka, L.F., Hansen, S.B. & Ashmun, J.W. (1985) Population biology of *Clintonia borealis*. I. Ramet and patchy dynamics. *Journal of Ecology*, **73**, 169–183.

Pleasants, J.M. & Wendel, J.F. (1989) Genetic diversity in a clonal narrow endemic, *Erythronium propullans*, and in its widespread progenitor, *Erythronium albidum*. *American Journal of Botany*, **76**, 1136–1151.

Reinartz, J.A. & Popp, J.W. (1987) Structure of clones of northern prickly ash (*Xanthoxylum americanum*). *American Journal of Botany*, **74**, 415–428.

Rumbaugh, M.D. (1970) A clonal competition experiment with alfalfa, *Medicago sativa* L. *Agronomy Journal*, **62**, 51–55.

Runkle, J.R. (1982) Patterns of disturbance in some old-growth mesic forest of eastern North America. *Ecology*, **63**, 1533–1546.

Runkle, J.R. & Yetter, T.C. (1987) Treefalls revisited: gap dynamics in the southern Appalachians. *Ecology*, **68**, 417–424.

Salisbury, E.J. (1942) *The Reproductive Capacity of Plants*. Bell & Sons Ltd, London, UK.

Shiraishi, S. (1988) Inheritance of isozyme variations in Japanese black pine, *Pinus thunbergii* Parl. *Silvae Genetica*, **37**, 93–100.

Silander, J.A. Jr (1979) Microevolution and clone structure in *Spartina patens*. *Science*, **203**, 658–660.

Silvertown, J. & Lovett-Doust, J. (1993) *Introduction to Plant Population Biology*, 3rd edn. Blackwell Scientific Publications, Oxford, UK.

Simpson, E.H. (1949) Measurement of diversity. *Nature*, **163**, 688.

Sokal, R.R. & Rohlf, F.J. (1981) *Biometry*, 2nd edn. W. H. Freeman & Co., San Francisco, CA.

Sokal, R.R. & Oden, N.L. (1978a) Spatial autocorrelation in biology. I. Methodology. *Biological Journal of the Linnaean Society*, **10**, 199–228.

Sokal, R.R. & Oden, N.L. (1978a) Spatial autocorrelation in biology. II. Some biological implications and four applications of evolutionary and ecological interest. *Biological Journal of the Linnaean Society*, **10**, 229–249.

Solangaarachchi, S.M., Harper, J.L. (1989) The growth and asymmetry of neighbouring plants of white clover (*Trifolium repens* L.). *Oecologia*, **78**, 208–213.

Solbrig, O.T., Newells, S.J. & Kincaid, D.T. (1980) The population biology of the genus *Viola*. I. The demography of *Viola soraria*. *Journal of Ecology*, **68**, 521–546.

Suzuki, W. (1987) Comparative ecology of *Rubus* species (Rosaceae). I. Ecological distribution and life history characteristics of three species, *R. palmutus* var. *coptophyllus*, *R. microphyllus* and *R. crataegilblius*. *Plant Species Biology*, **2**, 85–100.

Watkinson, A.R. & Powell, J.C. (1993) Seedling recruitment and the maintenance of clonal diversity in plant populations – a computer simulation of *Ranunculus repens*. *Journal of Ecology*, **81**, 707–717.

Watkinson, A.R. & White, J.H. (1985) Some life-history consequences of modular construction in plants. *Philosophical Transactions of the Royal Society of London, Series B*, **313**, 31–51.

Weiner, J. & Solbrig, O.T. (1984) The meaning and measurement of size hierarchies in plant populations. *Oecologia*, **61**, 334–336.

Whigham, D.F. (1974) An ecological life history study of *Uvularia perfoliata* L. *American Midland Naturalist*, **91**, 343–359.

Whigham, D.F., O'Neill, J. & Cipollini, M. (1993) The role of tree gaps in maintaining the population structure of a woodland herb: *Cynoglossum virginianum* L. *Plant Species Biology*, **8**, 107–115.

Wijesinghe, D.X. & Whigham, D.F. (1997) Costs of producing clonal offspring and the effects of plant size on population dynamics of the woodland herb *Uvularia perfoliata* (Liliaceae). *Journal of Ecology*, **85**, 907–919.

Wu, L. & Antonovics, J. (1976) Experimental ecological genetics of *Plantago*. II. Lead tolerance in *Plantago lanceolata* and *Cynodon dactylon* from a roadside. *Ecology*, **57**, 205–208.

Wu, L., Bradshaw, A.D. & Thurman, D.A. (1975) The potential for evolution of heavy metal tolerance in plants. III. The rapid evolution of copper tolerance in *Agrostis stolonifera*. *Heredity*, **34**, 165–187.

Zar, J.H. (1984) *Biostatistical Analysis*, 2nd, edn. Prentice-Hall, Englewood Cliffs, NJ.

Received 27 November 1997
revision accepted 4 September 1998