THE ECOLOGY AND GENETICS OF FITNESS IN FOREST PLANTS II MICROSPATIAL HETEROGENEITY OF THE EDAPHIC ENVIRONMENT

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SUMMARY

(1) Variation in the physical environment was investigated at scales of 0.1–50 m in the understorey of a southern Quebec forest, dominated by Acer saccharum and Fagus grandifolia, to determine if the spatial heterogeneity necessary to account for the evolution and maintenance of genetically diverse populations of forest plants exists in this old-growth forest.

(2) To characterize spatial heterogeneity in the understorey during the late summer, soil pH and the availability of K⁺ and NO₃⁻ ions in the soil solution were measured. The sampling programme involved analysis of replicate soil cores at 555 points dispersed throughout a 50-m × 50-m area in a valley previously undisturbed by human activities.

(3) A geostatistical analysis demonstrated that all three edaphic measures are predictably similar up to about 2 m, but that at greater distances the predictability is negligible and the environmental regime is best estimated by the mean for the site as a whole.

(4) These results demonstrate environmental variance at scales relevant to seed dispersal and genetic neighbourhood size in understorey herbs such as Impatiens. This spatial pattern of environmental variation may contribute to the maintenance of genetic variation through selection of locally adapted subpopulations.

INTRODUCTION

Impatiens species provide a useful illustration of how the regional distribution, abundance and performance of forest herbs vary with the physical and chemical environment (Lechowicz, Schoen & Bell 1988) and with biotic factors (Schoen et al. 1986). The survival, growth and fecundity of Impatiens plants native at a given site are superior to individuals transplanted from other nearby sites (Schoen et al. 1986). These results suggest that Impatiens may adapt to environmental variation at scales of 100–1000 m. Other studies have revealed evidence for local adaptation at much finer spatial scales within single populations of Impatiens. Schemske (1984) identified genetically differentiated subpopulations of Impatiens pallida separated by 30–50 m within a forest in Illinois. Such differentiation has been found in populations of Impatiens capensis separated by only 10 m in a forest in Rhode Island (Argyres & Schmitt 1991), and shown to have adaptive advantage (Schmitt & Gamble 1990).
Stewart & Schoen (1987) demonstrated differences in selection among twenty-five 1-m\(^2\) plots of *I. pallida*, in many cases separated by only a few metres, within a 2500-m\(^2\) area of forest understorey in southern Quebec, similar results were found for both *I. pallida* and *I. capensis* growing in the same vicinity (Brassard & Schoen 1990) These differences in selection at sites separated by only a few metres coincide with the existence of patches of similar genotypes, or neighbourhoods, in both species at the scale of 2–4 m (Schoen & Latta 1989) These results, which are consistent with those in other herbaceous species (Levin 1988), suggest that fine-scale environmental variance and spatially structured genetic diversity normally occur in natural populations of forest herbs

One possible explanation for the maintenance of fine-scale genetic structure within plant populations is that relative fitness changes from site to site within the area occupied by the population (Levene 1953) If neighbourhoods in natural populations of *Impatiens* are maintained by such fine-scale adaptation, then the local environment must also vary at the same fine scale In other words, the fine-scale distribution of patches of genetically similar *Impatiens* should essentially map the fine-scale pattern of environmental factors important to *Impatiens* survival and fecundity The effect of such fine-scale selection will be reinforced if gene flow is sufficiently limited so that locally adapted genotypes are not broken up or dispersed This is likely to be the case in *Impatiens* given the importance of clonal genetics in its reproductive biology (Schmitt & Ehrhardt 1990, Schmitt & Gamble 1990), seed dispersal over only 1–2 m (Schmitt, Ehrhardt & Swartz 1985), and pollen transport over only a few metres (Dube 1988) In a previous paper (Bell & Lechowicz 1991) we reported the results of bioassays with *Arabidopsis thaliana* and *Hordeum vulgare* in soil cores taken from a forest understorey habitat of *I. pallida* that demonstrate the existence of the necessary fine-scale environmental heterogeneity Our purpose in this paper is to complement these bioassays with an investigation of selected soil chemical variables at scales from 0.1 to 50 m in the understorey of an old-growth northern hardwood forest where we have investigated the ecology and genetics of *I. pallida* In this vicinity edaphic factors are important correlates of the distribution, abundance and performance of *Impatiens* species at the scale of the local landscape (Lechowicz, Schoen & Bell 1988) Only if there is predictable similarity over the short distances typical of neighbourhood sizes in *Impatiens* can the genetic substructure of populations be accounted for by selection of specialization to local edaphic conditions

**METHODS**

The study site is the 'Yellow Valley quadrat' of Bell & Lechowicz (1991), a 50-m \(\times\) 50-m quadrat in the understorey of an old-growth forest at Mont St Hilaire, Quebec (45°31' N, 73°8' W), that has never been cultivated, grazed, cut or otherwise disturbed by human activity The canopy, which is dominated by *Acer saccharum* Marsh and *Fagus grandifolia* Ehrh., was opened up by a glaze-ice storm in December 1983 (Melancon & Lechowicz 1987) At the time of the sampling in early August 1989 a dense population of *Impatiens pallida* Nutt., dominating the understorey vegetation, was in full flower The sampled area includes both the wall of the valley and the gently sloping valley floor, which is drained by an intermittent stream

Two soil cores from the rooting zone of *Impatiens* at each of 555 sampling sites were collected in this Yellow Valley quadrat, each core was 3 cm in diameter and
5 cm deep and both replicate cores came from within a 0.1-m x 0.1-m area. These replicate soil cores were collected in a variety of sampling designs: (i) thirty sets at the 10-m grid intersections in the 50-m x 50-m quadrat, (ii) 100 sets at random points throughout the quadrat, (iii) two sets of 100 at the 1-m grid intersection in two random 10-m x 10-m areas within the quadrat, (iv) one set of 100 at the 0.1-m grid intersections in a random 1-m x 1-m area within the quadrat, and (v) a series of five random sets nested at 10 m, 1 m and 0.1 m within the quadrat. This number and distribution of sample points provides well-resolved estimates of the spatial structure in this type of forest (Fortin, Drapeau & Legendre 1989).

The logistic demands of sampling intensively at a fine spatial scale limited our survey to the more mobile ions amenable to biologically meaningful assay by ion-electrode methods (Bailey 1976, Koryta 1982). Potassium was chosen because it is correlated with the performance of Impatiens (Lechowicz, Schoen & Bell 1988), and nitrate because it is an important nitrogen source for forest herbs and is known to be correlated with site productivity in deciduous forests (Zak, Host & Pregitzer 1989). Soil pH was also measured as a general indicator of soil quality known to be correlated with the distribution and abundance of forest herbs (Ellenberg 1988, Falkengren-Gerup 1989a, b, Palmer & Dixon 1990).

The fresh soil cores were brought to the Mont St Hilaire Research Centre and processed within 12 h of collection. The soil was sieved to remove coarse debris and pebbles and 10 g (fresh weight) was incubated in 50 ml of distilled water for 3 h at room temperature. The solution was filtered through Whatman number 4 paper, immediately frozen, and stored at −20°C until analysed. This simple aqueous extraction combined with filtration gives better estimates of NO₃⁻ than alternative extractions intended to remove ions interfering with the subsequent ion-electrode analysis (Pedrazzini, Castelli & Nannipieri 1979). The protocol was chosen to mimic the flush of nutrients into the soil solution following saturation of the soil by a summer rainstorm. The data are point-in-time samples at peak flowering of Impatiens, not estimates of the resource availability over a seasonal or annual cycle that would usually be considered in ecosystem-level studies (Binkley & Hart 1989). It is, however, the nutrients available in the soil solution that are actually taken up by forest herbs at a point in time (Mengel 1985, Robinson 1986).

After thawing to room temperature, each filtered sample was assayed for pH using a temperature-compensated meter (Fisher Accumet, Model 950, Ottawa, Ontario). The sample was split into two parts for analysis of NO₃⁻ and K⁺ using ion electrodes (Orion 93−07 NO₃⁻ electrode and 93−19 K⁺ electrode, Boston, MA, U S A). To minimize the possible effects of other ions in the soil solution, the electrode signals before and after addition of known NO₃⁻ or K⁺ standards were used to calculate the ion concentration in the solution (cf Bailey 1976, Koryta 1982). Following the recommendations for the Orion electrode manuals, we also added 1 ml of 5-M NaCl or 2-M (NH₄)₂SO₄ (for NO₃⁻ or K⁺, respectively) to the solutions to adjust ionic strength to the optimum for performance of the electrodes. The units are mg l⁻¹ K⁺ or NO₃⁻ in the soil solution. Electrode performance was checked daily and electrodes replaced as necessary (Bailey et al. 1988). Preliminary experiments confirmed that the protocol gave consistent readings across the range of surface soils sampled in the Yellow Valley quadrat.

We expected some delays in analysing the frozen samples, so an extra set of cores was taken at the 100 random points as a control for any effects of freezing or
storage This set was assayed immediately without the filtrate being frozen and the values compared against the replicates from the matching sample points analysed after various times in frozen storage. The regressions of change in assayed values against storage time were used to adjust of the effects of different lengths of storage on the assayed values of pH, NO₃⁻ and K⁺. After adjustment for the effects of storage, the mean pH or the mean of the natural logarithm of NO₃⁻ or K⁺ based on the two replicate cores at each sampling point were analysed.

First, the data for the 555 irregularly distributed sampling points were used to estimate a regular array of points at a 1-m resolution by bivariate interpolation (SAS procedure G3GRID) and then contoured (SAS procedure GCONTOUR) to illustrate the general nature of spatial patterns in the Yellow Valley quadrat. The same method of interpolation was used for the subset of cores sampled more intensively at resolutions of 1 m and 0.1 m. These contour maps illustrate the spatial pattern of the environment without being contingent on the subsequent geostatistical analysis, as they would be if we had used kriging (Robertson 1987) to make the interpolations.

Secondly, the predictability over distance of the spatial variation within the Yellow Valley quadrat was analysed by geostatistical methods summarized as a set of semivariograms (Trangmar, Yost & Uehara 1985, Webster 1985, Robertson 1987). A variety of other methods for spatial pattern analysis is available (Legendre & Fortin 1989), but the semivariogram provides a straightforward summary of the variance over a short range of distances. The semivariogram is a plot of how the mean square deviation changes as a function of distance within the sampled area. In calculating the semivariograms, the pattern of environmental variation was assumed to be isotropic (cf. Robertson 1987), both visual inspection of the site and preliminary anisotropic analyses to detect up- or cross-valley gradients suggested that this isotropic analysis was the most appropriate.

**RESULTS**

The soil environment of this old-growth forest shows a high degree of spatial complexity and variation (Fig 1). Some of the extreme values recorded can be related to conditions observed at the site. For example, the weakly developed drainage system on the valley floor tends to have higher levels of K⁺. Some areas especially rich in NO₃⁻ occur at the sites of tip-up mounds, canopy gaps caused by the 1983 ice storm, or seasonal seeps in the valley wall. On the other hand, there is a pronounced pattern in resource availability that could not be related to topography or drainage. Furthermore, the complexity of this soil environment occurs at all spatial scales (Fig 2) — uniformity does not appear to exist even at the scale of 1 m². All three edaphic variables varied substantially at the 0.1-m scale within 1 m².

The geostatistical analyses summarize more formally this complex spatial pattern (Fig 3). Although soil pH has lower overall variability than either NO₃⁻ or K⁺, none of the three soil measures is very predictable at distances >2 m, the greatest and most consistent changes in semivariance occur within 2 m. Beyond 2 m, NO₃⁻ is weakly predictable up to about 20 m. Potassium shows additional weak predictability at distances out to perhaps 35 m, but this modest trend is marked by discontinuities at about 7 and 12 m. Beyond a few metres, the soil environment is best predicted by the mean for the site as whole. Any progeny dispersed more than a few metres from
Fig. 1. Spatial patterns of variation in late summer 1989 for pH, ln K⁺ concentration and ln NO₃⁻ concentration in the soils of a 50-m × 50-m grid in the understorey of an old-growth Fagus-Acer forest in southern Quebec.
Fig. 2. Spatial patterns of soil K⁺ at increasingly finer-scale sampling resolution: (a) at the scale of 1 m in a 10-m × 10-m grid and (b) at the scale of 0.1 m in a 1-m × 1-m grid. Both sampled grids are in turn located in the 50-m × 50-m grid (cf. Fig. 1b), the 100-m² grid centred at (16 east, 6 north) and the 1-m² grid at (41.5 east, 42.5 north).

their parent plants have little likelihood of growing in an environment consistently similar to that of either of their parents.

**DISCUSSION**

Natural environments vary in space and time, but variation relevant to the establishment and growth of individual plants has rarely been investigated (Bazzaz & Sultan 1987; Wiens 1989). Geographers and landscape ecologists (e.g. Meentemeyer 1989) routinely consider the nature of environmental variation at scales above tens of metres. Soil scientists seek mapping units at scales that minimize variance within the unit (Webster 1985; Trangmar, Yost & Uehara 1985). Ecosystem scientists similarly
focus on reliable estimates of system variables at the scale of watersheds. Zak & Pregitzer's (1990) discussion of sampling for nitrogen in Michigan hardwood forests is a relevant example. Plant community ecologists do often sample variation at scales of a metre or even less, and recently have become interested in spatial pattern at these scales. For example, Robertson et al. (1988) report spatial variation in nitrogen availability in an abandoned field in Michigan at the scale of 1–40 m. More often community ecologists use ordination or gradient analyses (Jongman, Ter Braak & Van Tongeren 1987) that disregard the actual spatial relationships among samples. Other community ecologists have been directly concerned with spatial scale of the environment because of its impact on sampling design (Fortin, Drapeau & Legendre 1989, Palmer & Dixon 1990) or interpretation (Allen 1987) in ordination or gradient analyses. These recent studies, especially those by Palmer (1990), have contributed to our knowledge of the spatial structure of forest environments at scales ranging from centimetres to tens of metres. It is exactly these poorly studied, fine scales, at which plant establishment and growth occur, that are most relevant to evolutionary ecology.

We have shown here that variation in the edaphic environment of understorey herbs in old-growth deciduous forest occurs at the scale of 0.1 m, and that points separated by up to about 2 m are predictably similar. Only two other recent studies have investigated spatial heterogeneity at similarly fine scales in forests: the first concerned variation in soil pH at distances between 0.5 and 4 m in Swedish beech forests (Bringmark 1989), and the second variation in a wide range of soil properties.
at distances from a 0 1 to 50 m in a hardwood forest on the North Carolina piedmont (Palmer 1990) In contrast to our study site, these forests have been previously subjected to some human use and management Bringmark (1989) found that the variance in pH increased with distance, although she recognized plateaux where variance stabilized between 0 1–0 7 m and 1 5–2 5 m Palmer (1990) studied the spatial variation in soil pH, buffered acidity, cation exchange capacity, humic acid, and the availability of P, K, Ca, Mg, Mn and Cu All these diverse soil chemical properties, as well as the soil weight/volume quotient, showed a general increase in the variance of each property over distance His semivariograms show rapid increases for pH up to about 6 m and more gradual increases for available K up to at least 30 m — predictability over greater distances than at our site His other soil variables show good predictability ranging from <1 m for P to at least 40 m for Mn, most variables appear to have reasonable predictability at least up to 5 m Taking these results with our own, it is clear that the necessary environmental heterogeneity exists in both anthropogenically disturbed and more-pristine natural forest environments to drive local adaptation in herbaceous populations at scales up to a few metres

Edaphic heterogeneity at these fine scales in forest understooreys can arise from many sources In the forest understoorey, some heterogeneity is created by local effects of species in the tree canopy Examples include the gradients induced by stemflow at the base of the trunk of trees (Boerner & Koslowsky 1989, Falkengren-Grerup 1989b) or the edaphic mosaic associated with litter deposition and decomposition under different canopy trees (France, Binkley & Valentine 1989, Lodhi & Johnson 1989) Less-predictable, patchily distributed events, such as treefalls that create canopy gaps and tip-up mounds (Pickett & White 1985), influence the understoorey environment on scales up to tens of metres, the patchy activities of fossorial animals may do so at scales below a metre (Andersen 1987) Although many such factors that can contribute to local heterogeneity have been studied, surprisingly little attention has previously been given to the outcome of the sum of factors interacting at a site (Bazzaz & Sultan 1987) — in other words, to the overall pattern of local heterogeneity in the forest understoorey environment The few data available (this paper, Bell & Lechowicz 1991, Palmer 1990) suggest that forest understoorey environments are predictably similar over distances of a few metres This scale of environmental predictability is in accord with the supposition that local selection contributes to the maintenance of spatially structured genetic variation observed in natural populations of forest herbs such as Impatiens (Schemske 1984, Stewart & Schoen 1987, Brassard & Schoen 1990, Argyres & Schmitt 1991)

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