Gene diversity and demographic turnover in central and peripheral populations of the perennial herb Gypsophila fastigiata

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Published in:
Oikos

DOI:
10.1034/j.1600-0706.2002.11907.x

2002

OIKOS 99: 489–498, 2002

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Lönn, M. and Prentice, H. C. 2002. Gene diversity and demographic turnover in central and peripheral populations of the perennial herb *Gypsophila fastigiata*. – Oikos 99: 489–498.

Within-population gene diversity (H_S) was estimated (using allozyme markers) for 16 populations of the perennial, outcrossing plant, *Gypsophila fastigiata*, on the Baltic island of Öland. The populations were characterized by data on extent, density, life-stages, and habitat diversity. Populations were classed as central or peripheral in relation to the distribution of ''alvar'' (habitats with shallow, calcareous soils on limestone bedrock) on southern Öland. Three minimal adequate models were used to explain H_S and the proportions of juveniles and dead adults. In the first model, H_S was significantly lower in peripheral populations and there were no significant additional effects of other explanatory variables. The lower diversity in peripheral populations can be explained by a combination of genetic drift (in populations that vary in size in response to habitat fragmentation) and lower levels of interpopulation gene flow than in central populations. In the two life-stage models, peripheral populations had significantly larger proportions of both juveniles and dead adults – indicating a greater demographic turnover than in the central populations. There were also significant effects of H_S and species diversity on the proportion of juveniles. The central or peripheral position of populations is the strongest predictor of both within-population gene diversity and life-stage dynamics in Öland *G. fastigiata*.

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The population dynamics and genetic structure of fragmented population systems are of central interest within both evolutionary biology and conservation biology (Gilpin 1991, Young et al. 1996, Tew et al. 1997). Habitat loss, as a consequence of, for example, climatic change or anthropogenic disturbance, may lead to the fragmentation of species' regional or local distributions. Progressive habitat loss is accompanied by decreasing population sizes and by increasing spatial disjunction and genetic isolation of the separate populations.

Population genetic theory predicts that random genetic drift will lead to the loss of genetic variation in small populations (Barrett and Kohn 1991). Genetic drift in small and isolated populations is expected to lead to a decrease in gene diversity (Nei et al. 1975) and also, in particular, to a reduction of allelic-richness within populations (Rich et al. 1979) and to an increase in the between-population component of diversity (Brakefield 1989).

Several studies of plant populations have revealed a relationship between population size and levels of allelic-richness or the structuring of genetic diversity (Billington 1991, van Treuren et al. 1991, Dolan 1994, Prentice and Andersson 1997, Luijten et al. 2000), whereas other studies have failed to reveal a relationship between population size and genetic variation (e.g. Godt et al. 1995). However, it is unrealistic to expect a general association between present day population size and levels of genetic variation (Lönn and Prentice 1995). Presently large populations may previously have been considerably smaller or have had a history of population bottlenecks or founder events (Dolan 1994,

Accepted 14 June 2002

ISSN 0030-1299

Lönn et al. 1995). Presently small populations may only recently have been fragmented and may, particularly in long-lived perennial plants, show a genetic structure that is a result of demographic inertia (Michaud et al. 1995). In general, levels of genetic diversity in present-day populations may be more likely to reflect the populations' accumulated history of size fluctuations than to be related to present population size (Linhart and Premoli 1994).

Genetic variation that has been lost from small populations may be replenished, over time, by gene flow from surrounding populations (Travis 1990, Richards 2000, Cruzan 2001). Disjunct populations that occupy a peripheral position in relation to the main concentration of populations are likely to show a greater degree of genetic isolation than more central populations. Central populations are open to gene flow from all directions from other surrounding populations, whereas gene flow into peripheral populations may be directionally-constrained. Because central populations are likely to have a greater chance of regaining genetic variation lost during episodes of small size, we can predict that habitat fragmentation will have a greater cumulative impact on levels of gene diversity in peripheral than in central populations (Prober and Brown 1994).

Population marginality on a large geographic scale may reflect species' overall physiological and adaptive limits, with distributional boundaries being determined by climatic factors (Sykes et al. 1996). Population marginality may also be associated with edaphic or climatic limits on a local scale (Jones and Gliddon 1999). If geographic marginality is associated with ecological marginality, peripheral populations will be those that are most likely to become fragmented as a consequence of climatic or environmental change. Several studies support the prediction that ecologically marginal populations are characterized by demographic variability (Nantel and Gagnon 1999) and increased demographic turnover (Grant and Antonovics 1978, Johansson 1993).

A general susceptibility to population disjunction, high demographic turnover and genetic bottlenecks, coupled with reduced possibilities for incoming gene flow is expected to make peripheral populations particularly prone to the cumulative loss of genetic variation over time.

Populations of many European plants that require open habitats are dependent on traditionally managed pastures or meadows. Such habitats were previously widespread but have declined dramatically over the last century (Bernes 1994). The remaining semi-natural grasslands are becoming increasingly fragmented and populations of many grassland species are disjunct and small (van Treuren et al. 1991, Fischer and Stöcklin 1997, Rosquist and Prentice 2001). The relationships between population size/fragmentation and measures of genetic diversity or different aspects of reproductive success or individual fitness have been examined in a number of studies of declining grassland plants in Europe (van Treuren et al. 1993, Kéry et al. 2000). However, few studies have attempted to assess the relative importance of habitat, demographic and genetic characteristics as predictors of population dynamics in declining plant species (but see Ouborg and Van Treuren 1995 and Menges and Dolan 1998).

On the Baltic island of Öland, there are still more-or-less extensive areas of grassland and dwarf shrub vegetation that are traditionally maintained and which contain a high diversity of plant species that depend on open habitats. One such species is *Gypsophila fastigiata*, a relict from the open habitats that surrounded the Scandinavian ice sheet at the end of the last glaciation (Berglund 1966, Prentice 1992).

In the present study of *G. fastigiata* on Öland, we investigate whether the central or peripheral positioning of populations is a more important determinant of levels of within-population gene diversity or life-stage structure than the present-day extent or density of populations. We use minimal adequate models to explore associations between gene diversity, life-stage structure, habitat diversity, population density, population extent and population position (whether populations are peripherally or centrally located).

Material and methods

The species

Gypsophila fastigiata L. is a long-lived, perennial herb with cushion-like rosettes and a robust, central taproot. On Öland, *G. fastigiata* is confined to open "alvar" communities on fissured, outcropping limestone bedrock overlain by thin, calcareous soils (Bengtsson et al. 1988). These true alvar habitats, with soils that are derived from in situ weathering of the limestone bedrock, are widely, but patchily, distributed on Öland. The Great Alvar on southern Öland represents the most extensive area of alvar habitats. Within this area, the sparsely vegetated, true alvar habitats are interspersed with areas of deeper, base-poor morainic soils that support species-rich grasslands that are kept free from scrub encroachment by grazing (Bengtsson et al. 1988). *Gypsophila fastigiata* is absent from the closed grasslands on the deep, morainic soils.

The species is self-compatible, but predominantly outcrossing in the wild (outcrossing rate $>99\%$, H.C. Prentice and M. Lönn, unpubl.) and produces up to 200 inflorescences per individual. The nectar-rich flowers are visited by an exceptionally wide range of insects – from ants and beetles to syrphids, solitary bees and small butterflies and moths (H.C. Prentice and L.A. Nilsson, unpubl. obs. 1987). The seeds lack specialized adaptations for dispersal and are predominantly grav-

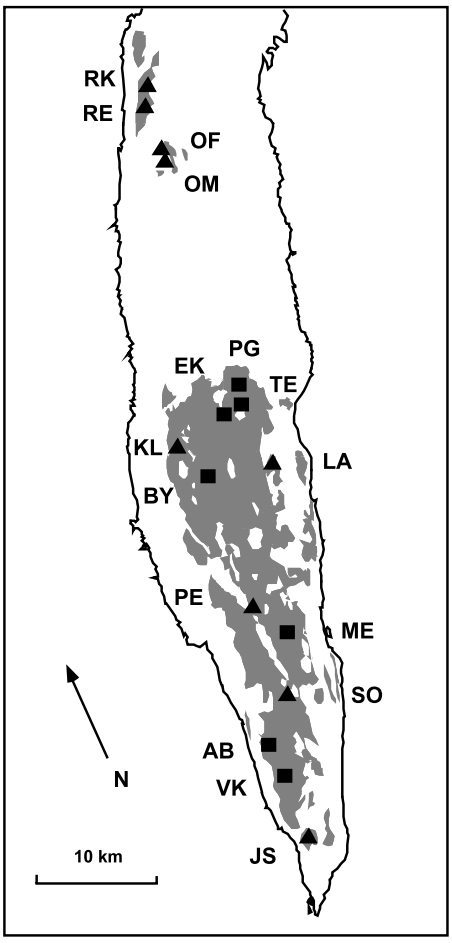

Fig. 1. Locations of the 16 studied populations of *Gypsophila fastigiata* on the Baltic island of Öland. Peripheral populations are indicated by triangles and central populations are indicated by squares. The shaded areas show the distribution of the alvar grasslands (after Königsson 1968) that are the main habitats for *G. fastigiata*. Site details are given in Table 1.

ity-dispersed within a radius of 20 cm from the mother plant (Bengtsson 2000). Laboratory germinability tests suggest that *G. fastigiata* may potentially have a moderately long-lived (ca 5 yr) seed bank (Bengtsson 2000).

Demographic studies of *G. fastigiata* from six populations on Öland showed high levels of mortality in the pre-reproductive stages and relatively low adult mortality (Bengtsson 2000). Individuals that were present in six Öland populations in a census in 1985 had half-lives that ranged from 1.5 to 3.2 years during the period 1985-1992 and from 2.0 to 4.2 years for the period 1985–1998 (Bengtsson 2000). However, the survivorship of reproductive adults was high (66–98%) (Bengtsson 2000). The dynamics of *G. fastigiata* populations on Öland are strongly influenced by cycles of drought and there has been an overall decline in the numbers of individuals in the six populations that have been censused since 1985 (Bengtsson 2000).

Sites and sampling

Populations of *G. fastigiata* were sampled from sixteen sites in alvar habitats on Öland (Fig. 1, Table 1). The sampling sites and grids are those that were used in an earlier study of *G. fastigiata* (Prentice 1992). In the present study, we include additional habitat, population and demographic data that were not included in the earlier study. All the demographic and habitat data were collected in 1984, at the same time as the seed samples were collected for genetic analysis (Prentice 1992). Within each population, data were collected using a grid with a 7 m mesh-size and a maximum extent of 49×49 m. If a grid cell contained *G. fastigiata* individuals, the individual nearest to the centre of the cell was sampled, giving a maximum of 49 *G. fastigiata* individuals per grid. We scored the numbers

Table 1. Sampling sites for *Gypsophila fastigiata* populations on Öland. All the sites contained open steppe-like alvar vegetation on thin, base-rich soils over limestone bedrock (see Bengtsson et al. (1988) for detailed habitat descriptions).

Site code	Location and site details	
RK	Räpplinge Alvar:	1 km NNW of Greby. Disturbed edge of limestone quarry
RE		200 m NNW of Greby gryn
OM	Karums Alvar:	500 m NE of Odens flisor. Heavily disturbed, cattle-grazed
OF		500 m ENE of Odens flisor. Heavily-disturbed, cattle-grazed
PG	Stora Alvaret:	Prästgropen, 1.6 km SW of Skarpa Alby. Lightly cattle-grazed
TE		1 km N of Ekelunda. Lightly cattle-grazed
EK		Ekelunda, 100 m NNW of Ekelundamossen. Lightly cattle-grazed
KL		Kleva Alvar, 400m N of Kleva stenbrott
LA		Lillalvaret, 500 m SW of Ödmanshorvan. Disturbed and cattle-grazed
BY		Bårby Alvar, 1.3 km SSW of Klovenhall. Heavily sheep-grazed
PE		Kastlösa, Penåsa. Sporadically cattle-grazed
ME		1.3 km N of Mellby. Cattle-grazed
SO		Solberga, 600 m SW of Möckelbrunnen. Ungrazed
AB		Albrunna, 700 m ESE of Hålkärr. Cattle-grazed
VK		Ventlinge, 900 m ENE of church. Cattle-grazed
JS		Jutas stubbe, 800 m ESE of Parboäng. Sheep and cattle-grazed

of *G. fastigiata* individuals belonging to different life-stage categories in a 1×1 m quadrat centred on the sampled individual. We recorded vascular plant species presence/absence within a 40 cm diameter circular plot centred on the sampled *G. fastigiata* individual. We also characterized the extent and the geographic position (peripheral or central) of the populations.

Life-stage categories

The numbers of juveniles, live-adult and dead-adult individuals were scored in the 1×1 m quadrats. Juvenile individuals were those that had formed small leaf rosettes but had not reached 2 cm in diameter. Adults were larger than 2 cm in diameter, and the category adult included the categories of sub-adult, reproductive and ''resting'' adult used by Bengtsson (2000). Dead adults were those that had died the previous summer (1983) during a severe drought (Bengtsson 2000). ''JUVENILES'' and ''DEAD'' are variables that give the number of juvenile and dead-adult individuals respectively. JUVENILES is given as a proportion of the total number of juveniles plus live-adults per population. DEAD is given as a proportion of the total number of live plus dead individuals per population.

Population characteristics

''EXTENT''. The spatial extent of each population was ranked subjectively on a scale of 1 to 16, with the rank of 1 being assigned to the most spatially restricted populations (Table 2). Populations with a score of 1 occupied an area less than 42×42 m and the population with a score of 16 occupied an area of more than 1000×1000 m.

''POSITION''. The categories ''peripheral'' and ''central'' are defined in terms of the distribution of the true alvar habitats (thin, calcareous soils over fissured limestone bedrock) to which *G. fastigiata* is confined on southern Öland. Although POSITION is defined in relation to the distribution of alvar habitats, the term is a descriptor of the position of the local population in relation to the geographic distribution of the species and does not include an assessment of ecological marginality. The map in Fig. 1 shows the overall distribution of alvar habitats on southern Öland. The areas of alvar are broken up by more-or-less extensive ridges of deep, base-poor morainic soils. The white patches that interrupt the main area of alvar in Fig. 1 represent the largest of the moraine ridges on the Great Alvar. These ridges were used for arable cultivation during periods of high agricultural activity in the past (Königsson 1968). The populations KL, PE and SO (Fig. 1) are surrounded on at least two sides by areas of deep morainic soils and are classed as ''peripheral'', despite the fact that they have relatively central geographic locations within the Great Alvar. In contrast, all the ''central'' populations, including the geographically marginal AB, are surrounded in all directions by at least 1 km of true alvar habitats on limestone bedrock overlain with shallow, calcareous soils. These shallow alvar soils are impossible to cultivate.

The density of *G. fastigiata* individuals within populations was characterized on two spatial scales. ''NQUADRATS'' is the number of sampled (i.e. containing at least one fruiting adult individual) quadrats within a 49×49 m population grid and is an approximate measure of the large-scale density/evenness of *G.*

Table 2. Data from sixteen populations of *Gypsophila fastigiata* on the Baltic island of Öland. Populations are listed from north to south and codes are explained in Table 1. The population parameters and measurements of *G. fastigiata* density, habitat diversity and life stages are described in the materials and methods. Extent is a rank scale from 1 to 16. Peripheral populations are indicated by 'P' and central populations by 'C'. The H_S (within population gene diversities) values are means over four loci.

Pop	Population parameters		*Gypsophila* density		Habitat diversity		Life stages		Gene diversity
	EXTENT	POSITION	NQUADRATS	NADULTS	INVJACC	SHANNON	JUVENILES	DEAD	H_S
RK	1	P	6	1.83	0.635	3.22	0.000	0.000	0.414
RE	1	P	27	1.52	0.540	3.10	0.226	0.000	0.441
OM	1	P	9	1.56	0.531	3.03	0.222	0.000	0.392
OF	1	P	9	2.00	0.685	3.44	0.100	0.053	0.361
PG	11	C	33	1.85	0.600	3.67	0.032	0.062	0.409
TE	11	C	26	2.00	0.595	3.59	0.088	0.000	0.418
EK	11	C	21	2.43	0.733	3.80	0.105	0.056	0.405
KL	8	P	8	1.00	0.605	2.93	0.111	0.200	0.360
LA	5	P	27	2.11	0.618	3.70	0.260	0.050	0.325
BY	9	C	24	3.12	0.680	3.68	0.461	0.014	0.495
PE	7	P	28	1.29	0.606	3.36	0.333	0.250	0.414
ME	16	C	35	2.74	0.576	3.39	0.010	0.040	0.458
SO	10	P	7	1.43	0.622	2.91	0.333	0.111	0.346
AB	14	C	45	2.44	0.631	3.39	0.018	0.009	0.417
VK	15	C	44	1.84	0.590	3.38	0.100	0.000	0.471
JS	6	P	7	1.29	0.418	3.10	0.100	0.000	0.359

fastigiata individuals within populations. "NADULTS" is the mean number of adult individuals per 1×1 m quadrat (i.e. fine-scale density).

Habitat diversity was characterized in two ways. "INVJACC" is the mean (for each population) of the between-plot values for the inverse of the Jaccard similarity coefficient (based on vascular plant species presence/absence in the 40 cm diameter plots). INVJACC is a measure of the large-scale vegetation heterogeneity within a population. The inverse Jaccard coefficients (Jaccard 1901) were calculated using the program CLUS, written by Lajos Hajdu. Overall vascular plant species diversity for each population was characterized by "SHANNON" (the Shannon-Weaver index calculated, using natural logarithms, from species presence/absence in the 40 cm diameter plots within each grid).

Gene diversity within and between populations

Allozyme data are taken from Prentice (1992) and the relative allele frequencies in the 16 populations are shown in Appendix 1. The data are based on one polymorphic locus for each of the enzyme systems; aspartate aminotransferase (AAT, EC 2.6.1.1), esterase (EST, 3.1.1.-), isocitrate dehydrogenase (IDH, EC 1.1.1.42) and phosphoglucose isomerase (PGI, EC 5.3.1.9) with a total of 13 alleles.

The gene diversity statistic H (Nei 1973), corrected for sample size (Nei 1978), was estimated for each population (H_S) and for the total material (H_T). The between-population component of the total diversity was calculated using G_{ST} (Nei 1973) ($G_{ST} = (H_T - \overline{H}_S)/H_T$), where $\overline{H}_S$ is the mean over populations of H_S, weighted for sample size. Mean over loci values of G_{ST} were obtained using the approach of Chakraborty et al. (1982). Jackknifed estimates of mean G_{ST} and standard errors were obtained by a jackknifing procedure where pseudovalues were calculated by the sequential removal of one population at a time (Lönn and Prentice 1995). The difference between the jackknifed mean G_{ST} values for the peripheral and central groups of populations was tested for significance by an approximate t-test.

Statistical methods

The main analyses were carried out with the help of generalized linear models – allowing the simultaneous evaluation of variables and avoiding repeated tests on the same data. (McCullagh and Nelder 1989). Generalized linear models also allow the choice of error distributions that are appropriate for different types of response variable. Generalized linear models were used for analyses of deviance (McCullagh and Nelder 1989). We constructed minimal adequate models by the stepwise model simplification procedure of Crawley (1993). All terms of interest were included in a maximal model. The individual terms were then removed and replaced in the maximal model, one by one. The term that caused the smallest change in deviance on removal from the model was deleted from the model. This procedure was repeated until only significant terms remained in the model, which then represented the minimal adequate model. Significance testing was carried out by referring F-ratios to an F-distribution. The competition between explanatory variables for a position in a minimal adequate model can be seen as an objective means of identifying the variables that have the greatest explanatory power.

Separate models were constructed for the response variables H_S, JUVENILES and DEAD. The models were constructed to allow biologically plausible conclusions. For example, we felt that it was unrealistic to attempt to explain H_S (within-population gene diversity) in terms of the numbers of juvenile individuals present in a particular year, whereas it is reasonable to attempt to explain H_S in terms of descriptors of population position or habitat diversity.

Because H_S is also equivalent to the expected proportion of heterozygous individuals under Hardy-Weinberg equilibrium, it was treated as a pseudo-binomial observation and analyzed in a generalized linear model using a binomial error distribution but without expecting a dispersion factor (mean residual error deviance) of one (McCullagh and Nelder 1989, p. 328). The response variables JUVENILES and DEAD are both counts out of a total possible count and were analyzed in generalized linear models using a binomial error distribution with expectation of a unit dispersion factor. The models for JUVENILES and DEAD both had a dispersion factor larger than one. Deviance changes were divided by the dispersion factor to correct for this overdispersion and the resulting F-ratio was referred to an F-distribution for significance testing. The analyses were carried out using the program package GENSTAT 5, release 3 (Lawes Agricultural Trust 1995).

Results

Differences in population and habitat variables between peripheral and central populations

The mean (over the four loci) gene diversities and the proportions of juveniles and dead individuals in each population are given in Table 2, together with the estimates of population extent, individual density, habitat diversity and the position (peripheral or central) of the populations.

Central populations had significantly higher values than peripheral populations for the variable EXTENT ($P < 0.001$, Table 3) and had a higher density of *G. fastigiata* individuals, both within the sampling grid (NQUADRATS) and within quadrats (NADULTS)

Table 3. Mean values (S.E. in parentheses) of population, life-stage, habitat diversity and gene diversity descriptors, for central and peripheral groups of *Gypsophila fastigiata* populations. The values for the separate populations are given in Table 2. Pairs of variables that show a significant positive correlation (Spearman rank correlation test, $P < 0.05$) are indicated by shared lower case letters (a-c). Significant differences between means in peripheral and central populations (Mann-Whitney U tests) are indicated by * = $P < 0.05$ and *** = $P < 0.001$. § The variables JUVENILES, DEAD and H_S are used as response variables in the generalized linear models (Tables 4–6) and are not included in the non-parametric tests.

	Central populations			Peripheral populations	
Population size					
EXTENT a	12.43	(2.57)	***	4.44	(3.54)
Gypsophila density					
NQUADRATS a, b	32.57	(9.50)	*	14.11	(9.99)
NADULTS b, c	2.34	(0.48)	*	1.56	(0.36)
Habitat diversity					
INVJACC	0.63	(0.06)		0.60	(0.05)
SHANNON c	3.56	(0.26)	*	3.20	(0.26)
Life stages					
JUVENILES §	0.12	(0.16)	§	0.19	(0.12)
DEAD §	0.03	(0.03)	§	0.07	(0.09)
Gene diversity					
H_S §	0.44	(0.04)	§	0.38	(0.04)

($P < 0.05$; Table 3). SHANNON (species diversity) is higher in central than in peripheral populations ($P < 0.05$; Table 3).

Correlations between population and habitat variables

Population size (EXTENT) is positively correlated with the large-scale density of *G. fastigiata* individuals (NQUADRATS) within the sampling grids ($P < 0.05$; Table 3). Restricted populations are characterized by a discontinuous internal distribution of *G. fastigiata* individuals whereas *G. fastigiata* is more evenly distributed within extensive populations. The number of quadrats containing individuals of *G. fastigiata* (NQUADRATS) ranged from 6 (population RK) to 45 (population AB) (Table 2). NQUADRATS is also positively correlated with the fine-scale density (NADULTS) of *G. fastigiata* individuals ($P < 0.05$; Table 3). The fine-scale density of adult individuals ranged from 1.00 m^{-2} (population KL) to 3.12 m^{-2} (population BY) (Table 2).

Large-scale vegetation heterogeneity (INVJACC) ranged from 0.42 (population JS) to 0.73 (population EK). Species diversity (SHANNON) ranged from 2.91 (population SO) to 3.80 (population EK) (Table 2) and SHANNON is positively correlated with NADULTS ($P < 0.05$; Table 3).

The proportion of juveniles (JUVENILES) ranged from 0.00 (population RK) to 0.46 (population BY) and the proportion of dead individuals (DEAD) ranged from 0.00 (populations RK, RE, OM, TE, VK, JS) to 0.25 (PE) (Table 2).

Gene diversity

The gene diversity, H_S, ranged from 0.325 (population LA) to 0.495 (population BY) (Table 2).

The between-population component of gene diversity (mean over 4 loci) is significantly greater in the peripheral group of populations ($G_{ST} = 0.068$) than in the central group of populations ($G_{ST} = 0.021$; $t = 3.78$; d.f. = 14; $P < 0.01$).

Minimal adequate models

The minimal adequate model explaining gene diversity within populations (H_S) is shown in Table 4. Gene diversity is significantly associated ($P < 0.01$) with population position and predictions from the model show that H_S is higher in central than in peripheral populations. There are no additional significant associations with extent, individual density or habitat diversity.

Table 5 shows a minimal adequate model explaining the proportion of juveniles in the populations. The response variable JUVENILES (number of juvenile individuals as a proportion of the total number of

Table 4. A minimal adequate model explaining gene diversity (H_S) (mean over four allozyme loci) in 16 populations of *Gypsophila fastigiata*. A binomial error distribution is assumed for the response variable H_S. The values for the explanatory and response variables are given in Table 2. Deleted terms (in order of deletion): INVJACC, NADULTS, NQUADRATS, SHANNON and EXTENT, starting with a maximal model. The predicted values and standard errors for H_S, for the two categories of POSITION, are calculated from the minimal adequate model.

Explanatory variable	df	F-ratio
POSITION	1	11.22**
Residual	14	
(** = $P < 0.01$)		

Predicted values of H_S for the two categories of POSITION:

	H_S	S.E.
Peripheral	0.379	0.012
Central	0.440	0.014

Table 5. A minimal adequate model explaining the proportion of juveniles out of the total number of juveniles plus live-adults per population (JUVENILES) in 16 *Gypsophila fastigiata* populations. A binomial error distribution is assumed for the response variable JUVENILES. The values for the explanatory variables and the response variable are listed in Table 2. Deleted terms (in order of deletion): NADULTS, INVJACC, EXTENT and NQUADRATS, starting with a maximal model. Predicted values and standard errors for JUVENILES (the probability of finding a juvenile among the total number of juveniles and adults in a site) for the two categories of POSITION are calculated from the minimal adequate model when H_S and SHANNON are kept at their mean values (Table 3) for peripheral and central populations respectively. The regression coefficients and approximate standard errors are given for the continuous variables H_S and SHANNON.

Explanatory variable	df	F-ratio	Regression coefficient	S.E.
POSITION	1	18.94***		
H_S	1	15.24**	0.22	0.06
SHANNON	1	9.74**	3.52	1.19
Residual	12			

(** = $P<0.01$ and *** = $P<0.001$)

Predicted values of JUVENILES for the two categories of POSITION:

	JUVENILES	S.E.
Peripheral	0.147	0.049
Central	0.098	0.029

juveniles plus live adults) is significantly associated with population position ($P < 0.001$), and the predictions from the model show that there is a higher proportion of juveniles in the peripheral populations. After removal of the effect population position, the proportion of juveniles within populations is also positively associated with both gene diversity (H_S) ($P < 0.01$) and species diversity (SHANNON) ($P < 0.01$).

A minimal adequate model explaining the proportion of dead adult individuals (number of dead adult individuals as a proportion of the total number of live and dead adult individuals) is shown in Table 6. The response variable DEAD is significantly associated with population position ($P < 0.05$), with a higher proportion of dead individuals in peripheral populations.

Discussion

In the outcrossing perennial plant *G. fastigiata*, the central or peripheral positioning of the populations in southern Öland is a more important determinant of levels of within population gene diversity than the present size (i.e. extent or density) of the populations.

Population position was the only significant factor in a minimal adequate model for gene diversity (Table 4). Peripheral populations had significantly lower gene diversity than central populations. Peripheral populations were also, on average, smaller than central ones (Table 3), but population size had no additional effect on gene diversity after the effect of population position was accounted for in the explanatory model. Central populations were denser and had a greater diversity of vegetation types than peripheral populations (Table 3). However, the population density and vegetation diversity variables did not add to the explanatory power of the model for gene diversity (Table 4).

Populations with a small effective size are vulnerable to the random loss of gene diversity through genetic drift (Nei et al. 1975, Caballero 1994), and peripheral populations may be generally more subject to spatial isolation and genetic bottlenecks than are central populations (Brewer et al. 1990). The fact that the peripheral populations of *G. fastigiata* on Öland show a higher between-population component of gene diversity (G_{ST}) than the central populations also supports the idea that the peripheral populations have been more subject to random changes in gene frequencies (Brakefield 1989) than the central populations. Peripheral populations are expected to have had a history of size-fluctuation and restricted interpopulation gene flow (Ellstrand and Elam 1993). Peripheral populations are also likely to have been colonized or recolonized more recently than central populations. The process of colonization has been shown to result in a high between-population

Table 6. A minimal adequate model explaining the proportion of dead adults out of all live plus dead individuals (DEAD) in 16 *Gypsophila fastigiata* populations. A binomial error distribution is assumed for the response variable DEAD. The explanatory variables and the response variable are listed in Table 2. Deleted terms (in order of deletion): INVJACC, H_S, NQUADRATS, NADULTS, SHANNON and EXTENT), starting with a maximal model. Predicted values and standard errors of DEAD (the probability of dying for an adult individual) for the two categories of POSITION are calculated from the minimal adequate model.

Explanatory variable	df	F-ratio
POSITION	1	4.66*
Residual	14	

(* = $P<0.05$)

Predicted values of DEAD for the two categories of POSITION:

	DEAD	S.E.
Peripheral	0.087	0.032
Central	0.024	0.012

component of gene diversity in *Silene dioica* (Wade and McCauley 1988, Giles and Goudet 1997).

Genetic variation that has been lost as a result of episodes of small population size may be restored by gene flow from other populations, and the probability of "genetic rescue" will be dependent on levels of incoming gene flow (Richards 2000). Central populations are expected to have access to gene flow from surrounding populations in several directions. In contrast, peripheral populations are, by definition, not surrounded by other populations and incoming gene flow is less likely to be multidirectional. We suggest that the lower gene diversity in peripheral *G. fastigiata* populations may reflect both a history of repeated episodes of small population size and a relatively lower potential for replenishing lost diversity through incoming gene flow.

We designated populations as being central or peripheral on the basis of the distribution of alvar habitats. However, a spatially peripheral position may also be associated with ecological marginality. Several studies have shown that ecologically marginal populations are characterized by high demographic turnover (Grant and Antonovics 1978, Johansson 1993). The two demographic characteristics (the proportions of juveniles and dead adults) that we recorded in the present study were influenced by population position. Peripheral populations contained more juveniles and more dead adult individuals (Table 5 and 6), indicating a faster turnover of individuals than in the central populations. The fact that the demographic characters were scored the year after a severe drought (Bengtsson 2000), suggests that peripheral populations may be more sensitive to climatic fluctuations. The density of adult *G. fastigiata* individuals in peripheral populations was significantly lower than in central populations on both of the density scales that we used in the present study (Table 3), and the fact that high adult mortality in the peripheral populations followed a drought-year also suggests that the peripheral locations may be ecologically marginal for *G. fastigiata*.

As well as being ecologically marginal, peripheral populations may also be particularly vulnerable to anthropogenically-driven changes in habitat quality and extent. Pollen analytical evidence shows that the extent of the open alvar grasslands on Öland has varied substantially during historical times (Königsson 1968). There have been many periods with a higher agricultural intensity than at the present day in the investigated area. Peripheral *G. fastigiata* habitats on Öland are often located on the outer rim of the alvar grasslands, adjacent to farmland or separated from each other by grassland areas, on deep morainic soils, that have been used for arable cultivation during periods when the human population on Öland was denser that at present (Königsson 1968).

When the effects of population position and habitat diversity are taken into account, there is still a positive effect of within-population gene diversity on the proportion of juveniles within populations (Table 5). The offspring from a genetically diverse population may be expected to show high levels of individual heterozygosity, and it is possible that more heterozygous individuals are intrinsically more fit (Burdon et al. 1983, Turelli and Ginzburg 1983). Positive relationships between the mean observed heterozygosity within populations and fitness-related characters in offspring were found in *Gentiana pneumonanthe* by Oostermeijer et al. (1994).

If there is local adaptation of different genotypes to different microhabitats, it is also possible that the wider range of genotypes produced in genetically diverse populations will increase the chance of the "right" genotypes becoming established in the right microhabitats – assuming that gene dispersal distances are greater than the patch size of the microhabitat mosaic (Prentice et al. 1995). Results from earlier studies suggest that there may be fine-scale, local adaptation within Öland populations of *G. fastigiata* (Prentice and Cramer 1990, Lönn et al. 1996).

Changes in agricultural policy and practice have lead to a dramatic and continuing decline in the extent of traditionally managed grassland habitats in northern Europe (Bernes 1994, Fischer and Stöcklin 1997). Many grassland species that occur in fragmented populations are likely to be threatened by a progressive loss of gene diversity as a result of stochastic processes in small populations.

The present study supports the prediction that peripheral populations are likely to be especially vulnerable to the genetic effects of population fragmentation. In Europe, grassland and dwarf shrub communities are characteristically found in areas that are topographically or edaphically unsuitable for arable production. Populations in habitats that are on the interface between productive and economically marginal agricultural areas are likely to have had a history of population fluctuations and local extinctions. Peripherality is likely to be associated with fast demographic turnover, a progressive loss of genetic variability and a reduced likelihood that lost variation will be restored by gene flow from surrounding populations.

Acknowledgements – We would like to thank Karin Bengtsson for help with fieldwork and demographic discussions. The Uppsala University Ecological Research Station on Öland provided an excellent base for the fieldwork. The project was supported by grants (to HCP) from NFR (The Swedish Natural Sciences Research Council) and SJFR (The Swedish Forestry and Agricultural Research Council).

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Appendix 1. Relative allele frequencies (based on the raw data used in Prentice (1992)) in Öland populations of *Gypsophila fastigiata*. *Aat*-3 and *Pgi*-2, 355 individuals; *Idh*, 345 individuals; *Est*-2, 319 individuals. For *Aat*-3 and *Pgi*-2, the numbers of individuals analysed in each population corresponds to NQUADRATS in Table 2, while some individuals lack data from *Idh* and *Est*-2.

Pop	Locus																		
	Aat-3 Allele			*Est*-2 Allele									*Idh* Allele		*Pgi*-2 Allele				
	1	1b	2	1*	1	2*	2	3	4	5*	5	N	1	2	1*	1	2	3	4
RK	0.917	0	0.083	0	0.300	0	0	0.400	0.200	0.100	0	0	0.833	0.167	0	0	0.750	0.250	0
RE	0.889	0	0.111	0	0.313	0	0.042	0.104	0.229	0	0.313	0	0.500	0.500	0	0.019	0.833	0.111	0.037
OM	0.944	0	0.056	0	0.438	0	0	0.063	0.188	0	0.313	0	0.417	0.583	0	0	0.889	0.111	0
OF	1.000	0	0	0	0.333	0	0	0.389	0	0.056	0.222	0	0.722	0.278	0	0	0.833	0.167	0
PG	0.955	0	0.045	0	0.052	0	0.034	0.276	0.397	0	0.241	0	0.742	0.258	0	0.030	0.712	0.258	0
TE	0.981	0	0.019	0	0.021	0	0	0.292	0.292	0.042	0.354	0	0.500	0.500	0	0.058	0.750	0.192	0
EK	0.905	0.024	0.071	0	0	0	0.053	0.263	0.395	0	0.289	0	0.643	0.357	0	0.071	0.857	0.071	0
KL	1.000	0	0	0	0.188	0	0	0.250	0	0	0.188	0.375	0.812	0.188	0	0.125	0.812	0.063	0
LA	0.981	0	0.019	0	0.120	0	0.020	0.620	0.160	0	0.040	0.040	0.889	0.111	0	0.037	0.704	0.241	0.019
BY	0.804	0.022	0.174	0	0.125	0	0.050	0.150	0.175	0	0.350	0.150	0.773	0.227	0	0.091	0.682	0.227	0
PE	0.964	0	0.036	0	0.038	0	0.038	0.423	0.269	0	0.115	0.115	0.714	0.286	0	0.143	0.732	0.125	0
ME	0.824	0	0.176	0	0.102	0.017	0.034	0.288	0.237	0.034	0.254	0.034	0.712	0.288	0.029	0.071	0.829	0.071	0
SO	1.000	0	0	0.100	0	0.100	0	0	0.700	0	0.100	0	0.812	0.188	0	0.063	0.687	0.250	0
AB	0.922	0.011	0.067	0	0.038	0.013	0	0.397	0.436	0	0.115	0	0.605	0.395	0	0.144	0.756	0.100	0
VK	0.932	0	0.068	0.052	0.026	0.026	0.039	0.247	0.234	0.052	0.299	0.026	0.705	0.295	0	0.193	0.636	0.170	0
JS	1.000	0	0	0	0	0	0	0.700	0.100	0	0.200	0	0.500	0.500	0	0.071	0.786	0.143	0