Comparison of quantitative genetic variation and allozyme diversity within and between populations of *Scabiosa canescens* and *S. columbaria*

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We quantified allozyme diversity and quantitative genetic variation within and between six populations of the locally rare *Scabiosa canescens* and the more common and widespread *S. columbaria* (Dipsacaceae). For the within-population component, we quantified allozyme diversity as the expected heterozygosity (H_e), followed by a sib analysis to estimate the heritability (h^2) for eight vegetative and floral characters. There was no consistent association between h^2 and H_e , or between h^2 and current population size. Regarding the between-population component, we compared the overall level of population differentiation for the allozymes (F_{ST}), assumed to be selectively neutral, with the corresponding estimates for the quantitative traits (Q_{ST}) to determine if the marker genes and the polygenic characters have been influenced by the same evolutionary force(s). Differences between estimates of F_{ST} and Q_{ST} for *S. canescens* were too small to be declared as significant, indicating that neutral phenotypic evolution has led to the observed differentiation in this species. Our data indicate higher levels of phenotypic differentiation in *S. columbaria* with values of Q_{ST} exceeding the allozymic F_{ST} for all traits. This finding indicates a great potential for *S. columbaria* to respond to local selection pressures, perhaps explaining why this species has a broader ecological amplitude than *S. canescens*.

Keywords: allozymes, genetic diversity, population differentiation, quantitative genetics, rarity, *Scabiosa*.

Introduction

Heritable variation is necessary for adaptation and long-term survival of populations in a changing environment (Falconer, 1989) and has become a major topic in conservation biology (Schemske *et al.*, 1994), even though demographic data are considered more useful for monitoring and managing populations in the short term (Lande, 1988). Genetic studies of rare and declining species usually focus on allozymes or molecular marker genes, whereas few attempts have been made to assess patterns and amounts of heritable variation in ecologically important characters controlled by a large number of loci (Widén & Andersson, 1993; Schemske *et al.*, 1994; Storfer, 1996).

This focus on marker genes has several limitations if the aim is to provide information on adaptation and long-term survival of populations (Lynch, 1996). First, the presumably high mutation rate of quantitative characters (Falconer, 1989) suggests that the recovery time after a bottleneck will be shorter for polygenic variation than for single-locus polymorphisms (Lande & Barrowclough, 1987). Secondly, the expected loss of additive genetic variance caused by genetic drift does not follow the same pattern as the reduction in single-locus heterozygosity when there are high levels of nonadditive genetic variance in the character (Goodnight, 1988; Willis & Orr, 1993). Thirdly, the effect of small population size on genetic variation is expected to differ for monogenic and polygenic characters as a result of selection acting differently on these types of character (Foley, 1992). These differences suggest that conservation

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strategies based solely on the variation at marker gene loci may be misleading and that quantitative genetic analyses therefore serve as an important complement in studies of rare and threatened species (Lande & Barrowclough, 1987; Lynch, 1996; Storfer, 1996).

Genetic variation not only contributes to differences between individuals within populations, but also accounts for differences between populations. As for adaptive traits with a polygenic basis, the proportion of variation within a population attributable to the additive effects of genes can be quantified as the heritability (Falconer, 1989). Regarding the between-population component, it is desirable to quantify the level of population subdivision of quantitative characters in a form that allows direct comparison with the presumably neutral marker genes. For example, to examine whether single-locus and polygenic characters are influenced by the same evolutionary force(s), it may be useful to compare estimates of the population structure based on Wright's (1951) F -statistics (Spitze, 1993; Podolsky & Holtsford, 1995; Yang *et al.*, 1996). In this approach, diversifying selection may be invoked when the level of population differentiation in a quantitative character (hereafter referred to as Q_{ST} following Spitze, 1993) is significantly higher than the corresponding parameter for the marker genes (F_{ST}), whereas unifying selection may be invoked when Q_{ST} is significantly lower than F_{ST} . If the estimates of Q_{ST} and F_{ST} are similar, the hypothesis that the population structure of the quantitative trait is caused by random drift cannot be rejected (Spitze, 1993; Podolsky & Holtsford, 1995; Yang *et al.*, 1996).

In the present investigation of *Scabiosa canescens* and *S. columbaria*, we quantified levels of variation between and within populations to determine whether the heritability of quantitative characters is correlated with allozyme diversity and population size, and whether allozymes and polygenic traits have been influenced by the same evolutionary force(s). Comparison of the locally rare *S. canescens* and the more common *S. columbaria* also provided an opportunity to assess the relationship between rarity and quantitative genetic variation.

Materials and methods

Scabiosa columbaria L. and *S. canescens* W. & K. (Dipsacaceae) are diploid ($2n = 16$), perennial plants with basal leaf rosettes, a variable number of branched stems with a determinate flowering pattern and blue, bilabiate, protandrous, self-compatible and

outcrossing flowers arranged in dense, terminal inflorescences (hereafter 'heads') visited by syrphids, butterflies and bees (Van Treuren *et al.*, 1994). Fertilized flowers develop into one-seeded, indehiscent fruits ('seeds') enclosed in umbrella-shaped epicalyces crowned by spines.

Scabiosa columbaria occurs in various grassland habitats in large parts of Europe and is locally common in southern Sweden with ≈ 100 localities in Skåne, the southernmost region. *Scabiosa canescens* has a more eastern distribution in Europe and occupies a much narrower range of habitats: the 25 localities in Sweden represent steppe-like sites with sandy soils in Skåne (pers. obs.).

In 1993, one of us (P.W.) selected six different-sized populations of each species in Skåne. Population size was determined by direct counts if the number of individuals was below 250, whereas the sizes of larger populations were estimated by multiplying the number of plants in a 10×10 m square by the approximate number of squares that could be fitted by eye to the population. The populations used in the present investigation were separated from other populations by more than 1 km and showed a more than 100-fold difference in population size (Table 1). Most of these populations have been relatively stable between 1993 and 1997 (unpubl. data), suggesting that differences in population size could have persisted for a long time.

One flower head with ripe seeds and one leaf rosette were collected from each of 25 randomly

Table 1 Information on the populations of *Scabiosa columbaria* and *S. canescens* used in the present investigation

Species/Location	Abb.	Coord.	<i>N</i>
<i>S. columbaria</i>			
Smygehuk	Sm	1C7i21941	55
Bjärred	Bj	2C6e20740	59
Klagshamn	Kl	2C1e34201	236
Billebjär	Bi	2C5i20938	600
Grödbby	Gr	3E3d44146	1500
Margaretevall	Ma	1D8 h21233	5000
<i>S. canescens</i>			
Hällestad	Hä	2C5j20046	25
Veberöd	Ve	2D3b34008	100
Degeberga	De	2D8i10528	138
Vitemölla	Vi	2D5j21241	2000
Yngsjö	Yn	2E9a11620	4000
Åhus	Åh	2E9a43538	5000

Abb., abbreviation; Coord., RUBIN coordinates; *N*, the estimated number of flowering individuals in 1993.

selected plants in each population. The seeds were stored in separate paper bags at room temperature for later use in the quantitative genetic study (see below), and the leaf rosettes were wrapped in moist paper and placed in a portable cooling bag for transportation to a refrigerator in the laboratory (4°C). Extraction was performed by grinding $\approx 1 \text{ cm}^2$ of a leaf in 120 μL of extraction buffer (2.4% Tris, 2% polyvinylpyrrolidone, 0.1% EDTA, 0.2% MgCl_2 , 1% sodium metabisulphite and 0.2% di-mercaptoethanol at pH 7.5). The tissue extracts were absorbed onto eight filter paper wicks per individual and stored at -80°C until used.

Electrophoresis was carried out on 12% starch gels (Reppin PSG 1000) using two buffer systems. A morpholine-citrate buffer with pH 6.1 (Wendel & Weeden, 1989) was used for the following enzymes: aconitase (EC 4.2.1.3, ACO), phosphoglucanate dehydrogenase (EC 1.1.1.44, PGD), phosphoglucisomerase (EC 5.3.1.9, PGI) and shikimate dehydrogenase (EC 1.1.1.25, SKD), and a LiOH-borate buffer with pH 8.3 (Ashton & Braden, 1964) was used for aspartate aminotransferase (EC 2.6.1.1, AAT), phosphoglucisomerase (EC 5.3.1.9, PGI) and triose phosphate isomerase (EC 5.3.1.1, TPI). Hence, both buffer systems were used for the PGI system. Staining schedules were according to Wendel & Weeden (1989). Twelve other enzyme systems were also tested, but the banding patterns of these systems were not interpretable in terms of alleles at Mendelian loci.

Deviations from Hardy–Weinberg expectation and linkage equilibrium were tested for each population using the exact tests in the GENEPOP package (Raymond & Rousset, 1995), and the allelic diversity at polymorphic loci was quantified as the mean expected heterozygosity (H_e) with BIOSYS-1 (Swofford & Selander, 1989). To assess levels of population divergence, the global deficit of heterozygotes (F_{IT}) was partitioned into two components: heterozygote deficit within populations (F_{IS}) and heterozygote deficit between populations (F_{ST} , Wright, 1951). These F -statistics, with 95% confidence intervals (CI) of each estimate, were quantified with FSTAT (Goudet, 1995) according to the variance component approach by Weir & Cockerham (1984).

In May 1994, 20 seeds per plant were sown in large pots with a mixture of standard soil ($\approx 90\%$), sand (10%) and lime (1%). Some families were represented by fewer than 20 seeds, but only one family per species had to be excluded because of low seed-set. The resulting seedlings were raised under standard growth conditions and fertilized once a week. After 75 days, five randomly chosen seedlings

per family were planted in separate pots and randomized in an unheated greenhouse. For some populations, it was necessary to compensate for low germination by increasing the number of seedlings from families with a high germination rate. The plants were overwintered outdoors to enhance flowering in 1995.

The following traits were scored on each individual: leaf number, length and area of the longest leaf, first flowering date, height of the main shoot, maximum plant height, number of heads and flower size. The leaf traits were measured 75 days after sowing (1994), whereas other characters were measured in the first flowering season (1995). Leaf size characters were quantified with an image analysis system (IMAGEGRABBER, OPTILAB) on a Macintosh computer. Flower size, measured under a dissecting microscope, refers to the average distance from the base of the corolla to the tips of the left and right side lobes on the lower lip (based on one marginal flower from the terminal head on each plant). Phenotypic data were obtained for a maximum of 619 and 751 plants of *S. canescens* and *S. columbaria*, respectively.

A variance component analysis with population and family (nested within population) as random effects was conducted for each trait and species with the REML directive in GENSTAT 5 (1995). The (average) level of genetic variance within populations (V_G) was estimated as twice the between-family variance component under the assumptions that maternal effects were weak, that offspring within families were related as full-sibs and that most of the genetic variance was additive. The between-population component (V_{pop}) was assumed to quantify the variance attributable to heritable differences between populations.

Estimates of V_{pop} and V_G were used to quantify the level of population differentiation (Q_{ST}) for each of the continuous characters (Wright, 1951; see also Spitze, 1993; Podolsky & Holtsford, 1995). Provided that V_{pop} and V_G have a strong additive component and that populations are in Hardy–Weinberg equilibrium ($F_{IS} = 0$), it can be shown that $Q_{ST} = V_{\text{pop}} / (2V_G + V_{\text{pop}})$. This parameter is analogous to F_{ST} and therefore provides an opportunity to compare population divergence in phenotypic characters and in single-locus allele frequencies. To obtain approximate 95% CI of Q_{ST} , we used the VFUNION option in GENSTAT 5, a procedure based on the delta method (Kendall & Stuart, 1969). We also employed the REML directive and the VFUNION procedure in GENSTAT 5 to estimate the heritability (h^2) and the 95% CI of h^2 for each character and population,

calculated as twice the proportion of the variance attributable to family differences and based on the same assumptions as the estimation of V_G (see above).

Associations between population size, allozyme diversity and heritability were estimated by computing the Pearson correlation coefficient for each of 1000 bootstrap samples (Efron & Tibshirani, 1986) with the BOOTSTRAP procedure in GENSTAT 5. The BOOTSTRAP procedure was also employed to test for differences between species in the mean heritability for each trait and for the mean heritability over traits (averaged over populations). Finally, we utilized Mantel's (1967) matrix permutation method with 2000 randomizations to test for associations between pairwise interpopulation distances based on allozymes (F_{ST} values) and quantitative characters (Q_{ST} values).

Results

Allozymes

Electrophoretic analyses of *S. columbaria* revealed five polymorphic loci (*AAT-1:2*, *ACO-2:3*, *PGI-2:3*, *PGD-2:2* and *TPI-1:2*; the first number denotes the locus and the second refers to the number of alleles found at this locus). The banding pattern of SKD could not be interpreted in terms of Mendelian loci and had to be excluded from all analyses, whereas the interpretation of *PGI-2* and *PGD-2* agreed with allozyme data from Van Treuren & Bijlsma (1992). Only two variable loci (*AAT-1:3*, *SKD-1:3*) were found in *S. canescens*; the *PGD* and *TPI* loci turned out to be monomorphic and the banding patterns of *PGI* and *ACO* were impossible to interpret in terms of alleles. The two species appear to have similar sets of alleles at the *AAT*, *PGD* and *TPI* loci.

There was no significant deviation from Hardy-Weinberg equilibrium ($P > 0.05$) in any of the populations of *S. canescens*, and only two out of 30 comparisons (six populations $\times$ five loci) revealed a significant deviation from random mating ($P < 0.05$) in *S. columbaria*. There was no evidence for nonrandom associations of alleles between pairs of loci in *S. canescens* ($P > 0.05$) and only three out of 60 pairwise combinations of loci were found to be in significant linkage disequilibrium ($P < 0.05$) in *S. columbaria*.

The F_{ST} -values for individual loci in *S. columbaria* ranged from 0.09 (*PGI-2*) to 0.15 (*TPI-1*), with a mean F_{ST} (0.123) significantly greater than 0 (CI 0.101–0.142). Although the number of polymorphic loci in *S. canescens* was too small for calculation of

CI (Goudet, 1995), the mean F_{ST} (0.164) differed significantly from 0 according to a permutation test ($P < 0.001$; F_{STAT} , Goudet, 1995). As for the within-population variation, the expected mean heterozygosity (H_e) ranged from 0.18 to 0.45 for *S. columbaria* and from 0.04 to 0.39 for *S. canescens* (Table 2a,b). These data are not directly comparable for the two species, as they are based on different sets of loci. There was no significant correlation between H_e and population size ($P > 0.05$) in either of the species.

Quantitative characters

Only four plants of *S. columbaria* and 11 plants of *S. canescens* died during the winter, and nearly all surviving plants flowered in their first season (100 and 98% in *S. columbaria* and *S. canescens*, respectively). A distribution-fit-test (DISTRIBUTION directive in GENSTAT 5) showed that most of the characters were approximately normally distributed, the only exceptions being leaf number, head number and first flowering date, which had to be square root transformed to reach normality.

Population means and heritabilities (h^2) are shown in Table 2(a,b). Populations of *S. canescens* had higher heritabilities than populations of *S. columbaria* for most of the characters: the difference reached significance for leaf area and length ($P < 0.001$) and for the average heritability ($P < 0.05$). Estimates of h^2 varied independently of H_e in both species ($P > 0.05$), the only exception being leaf area in *S. canescens* ($r = 0.78$, $P < 0.05$). Population size was significantly negatively correlated with h^2 for leaf area, leaf length and flower size ($r = -0.65$, -0.67 and -0.67 , respectively; $P < 0.05$ in all cases) in *S. columbaria*, and with h^2 for first flowering date ($r = -0.72$, $P < 0.05$) in *S. canescens*. The only significant positive association with population size involved h^2 for leaf number in *S. canescens* ($r = 0.78$, $P < 0.05$). Estimates of h^2 for other characters showed a nonsignificant relationship with population size ($P > 0.05$).

Estimates of Q_{ST} for *S. columbaria* differed widely between traits (Table 3), but always exceeded the F_{ST} based on allozymes. In the case of flower size and plant stature, the differences were sufficiently large to be declared as significant (nonoverlapping CI). Populations of *S. canescens* showed lower levels of differentiation: none of the Q_{ST} estimates differed significantly from 0 and all but one (flower size) were lower than the allozymic F_{ST} . We found no consistent association between pairwise genetic distances based on F_{ST} and Q_{ST} ($P > 0.05$).

Table 2(a) Population means, heritabilities and expected heterozygosities (H_e) for *Scabiosa columbaria*

Population	Sm	Bj	Kl	Bi	Gr	Ma
Means						
Leaf number	35.2	41.3	41.7	37.4	43.8	52.4
Leaf area (10 ³ mm ²)	17.9	23.1	24.8	27.2	26.5	21.5
Leaf length (10 ³ mm)	1.87	1.70	1.85	2.07	1.76	1.74
Main stem height (mm)	665	756	1060	977	1066	731
Maximum height (mm)	843	896	1347	1214	1306	976
Head number	134	165	226	182	289	211
Flowering date	27.7	35.8	34.2	33.8	40.6	29.1
Flower size (mm)	12.3	12.4	13.0	14.6	11.0	13.9
Heritabilities						
Leaf number	0.108	0.177	0.753*	0.221	0.174	0.414*
Leaf area	0.386*	0.315	0.470*	0	0.184	0.129
Leaf length	0.098	0.154	0.176	0	0.078	0
Main stem height	0.447*	0.109	0.110	0	0.325	0.630*
Maximum height	0.677*	0.082	0.432*	0.147	0.360	0.880*
Head number	0.438*	0.098	0.091	0.405*	0.300	0.338
Flowering date	0.405*	0.198	0.450*	0.222	0.319	0.437*
Flower size	0.526*	0.557*	0.328	0.112	0.499*	0
Average	0.386	0.211	0.351	0.138	0.280	0.354
H_e	0.445	0.221	0.327	0.179	0.422	0.224

Populations have been arranged according to increasing size.

*The 95% CI of the heritability does not overlap zero.

Discussion

Within-population variation

Population genetic theory of finite populations predicts a loss of variation as a result of inbreeding and random genetic drift (Falconer, 1989). Hence, given the small and disjunct populations of many rare, endangered species and the adaptive significance of heritable variation (Lande & Barrowclough, 1987), numerous attempts have been made to estimate the level of genetic variation in populations of rare species (Schemske *et al.*, 1994). Most of these studies have focused on allozymes or molecular marker genes, presumably under the assumption that levels of variation in single-locus characters also provide insights into levels of variation of polygenic characters. The validity of this assumption has been questioned (Lynch, 1996) and the present investigation provides no evidence for a strong association between marker gene diversity and quantitative genetic variation. Admittedly, it is possible that our study lacked the power to detect such relationships, given the large standard errors of most heritability estimates (Falconer, 1989) and the small number of allozyme loci scored, especially for *S. canescens*.

Despite a more than 100-fold difference in population size, there was no tendency for small populations to possess lower levels of quantitative genetic variation than large populations: the highest average heritability was recorded for the smallest population in each species (Sm and Hain Table 2a,b) and the heritability for leaf and flower size was significantly negatively correlated with population size in *S. columbaria*. That small populations can retain high levels of quantitative variation for ecologically important characters has also been demonstrated in other rare plant species, e.g. *Senecio integrifolius* (Widén & Andersson, 1993). It is worth noting that *S. columbaria* and *S. canescens* also show nonsignificant relationships between population size and expected heterozygosity. Leaving aside the large standard errors of H_e , this finding is in accord with data from other allozyme studies (Ellstrand & Elam, 1993), including a recent investigation of *S. columbaria* in The Netherlands (Van Treuren *et al.*, 1991).

The weak effect of population size on both heritability and expected heterozygosity has at least four possible causes. First, the sampling variance of these parameters is large, particularly for small populations (Lynch, 1988). Secondly, occasional long-

Table 2(b) Population means, heritabilities and expected heterozygosities (H_e) for *Scabiosa canescens*

Population	Hä	Ve	De	Vi	Yn	Åh
Means						
Leaf number	33.3	26.7	36.3	26.3	27.1	27.9
Leaf area (10 ³ mm ²)	9.72	10.7	9.22	11.2	9.82	11.6
Leaf length (10 ³ mm)	1.28	1.50	1.25	1.28	1.37	1.36
Main stem height (mm)	564	593	559	525	581	602
Maximum height (mm)	669	769	670	622	687	717
Head number	89.8	97.8	104	70.4	66.5	77.0
Flowering date	44.8	40.3	42.7	43.2	44.4	42.7
Flower size (mm)	9.16	11.1	11.0	10.7	10.8	11.0
Heritabilities						
Leaf number	0.009	0.002	0.500*	0.734*	0.411*	1.010*
Leaf area	0.603*	0.672*	0.409*	0.988*	0.452*	0.583*
Leaf length	0.548*	0.236	0.192	0.546*	0.212	0.570*
Main stem height	0.608*	0.213	0.350	0.408*	0.610*	0.068
Maximum height	0.691*	0	0.395*	0.327	0.778*	0.270
Head number	0.414*	0	0.056	0.014*	0.713*	0.739*
Flowering date	0.557*	1.49*	0.511*	0.418*	0.293	0.098
Flower size	0.470*	0	0.677*	0.403*	0.107	0.176
Average	0.488	0.327	0.386	0.480	0.447	0.439
H_e	0.249	0.363	0.040	0.390	0.225	0.373

Populations have been arranged according to increasing size.

*The 95% CI of the heritability does not overlap zero.

distance gene flow could diminish the effect of genetic drift and the loss of genetic variation in small populations (Ellstrand & Elam, 1993). Thirdly, a subdivided population can maintain higher levels of genetic variation than a panmictic population of the same size (e.g. Goldstein & Holsinger, 1992), a possible explanation of the unusually high heritabilities in the small Sm population, which shows higher levels of spatial structuring than other populations (pers. obs.). Finally, the level of genetic variation

should be more strongly influenced by the long-term population size than by the current number of individuals, perhaps explaining the high heritabilities and the relatively high allozyme diversity in the small Hä population, for which historical evidence indicates a larger population size in the past (L. Knutsson, pers. com.).

Mechanisms that could explain the high heritability in small populations with low allozyme diversity (e.g. Bj, Table 2a) include microsite heterogeneity

Table 3 The level of population differentiation in phenotypic characters of *Scabiosa*, as determined by Q_{ST} and its 95% confidence intervals (CI)

	<i>S. columbaria</i>			<i>S. canescens</i>		
	Q_{ST}	CI		Q_{ST}	CI	
Leaf number	0.154	−0.039	0.346	0.114	−0.043	0.271
Leaf area	0.377	0.039	0.716	0.019	−0.028	0.066
Leaf length	0.526	−0.024	1.076	0.088	−0.044	0.220
Main stem height	0.750	0.494	1.007	0.069	−0.044	0.182
Maximum height	0.570	0.245	0.895	0.104	−0.041	0.250
Head number	0.379	0.050	0.708	0.113	−0.047	0.272
Flowering date	0.380	0.055	0.706	0.041	−0.040	0.122
Flower size	0.485	0.146	0.824	0.217	−0.036	0.469

combined with local selection and/or genotype $\times$ environment interaction (Barton & Turelli, 1989), and the conversion of nonadditive variance into additive variance after a bottleneck (Goodnight, 1988; Willis & Orr, 1993).

Genetic differentiation between populations

Estimates of Q_{ST} for *S. canescens* were similar to, or slightly lower than, the corresponding parameter for the allozyme markers (F_{ST}). Based on the wide overlap between confidence intervals, there is no reason to believe that the phenotypic characters are more strongly influenced by geographically varying selection than by random genetic drift in this species. As for *S. columbaria*, estimates of Q_{ST} always exceeded the allozymic F_{ST} with significantly higher values for traits reflecting plant stature, indicating that diversifying selection has played a major role in shaping large-scale patterns of variation in this species, at least in southernmost parts of Sweden. This scenario is consistent with the weak association between genetic distances based on allozymes (F_{ST}) and quantitative characters (Q_{ST}), and may be particularly relevant for plant height, given the tendency for coastal plants (Sm, Bj and Ma) to be of shorter stature than those representing more mesic inland localities (Kl, Bi and Gr; Table 2a).

Podolsky & Holtsford (1995) assessed patterns and levels of population differentiation in allozymes and a wide range of floral characters in the plant *Clarkia dudleyana* (Onagraceae). Most of the Q_{ST} values were similar to, or higher than, the estimate of F_{ST} based on allozymes, indicating that characters had been influenced by genetic drift or by selection for different optimum phenotypes in different populations. In a study of the conifer *Pinus contorta* ssp. *latifolia*, Yang *et al.* (1996) found the Q_{ST} values for specific gravity, stem diameter, stem height and branch length to be significantly higher than the allozymic F_{ST} , and attributed these differences to diversifying selection, whereas the data for branch angle and branch diameter were consistent with the neutrality hypothesis. A recent study of the rare, endemic plant *Silene diclinis* (P. Waldmann, S. Andersson & H. Prentice, unpubl. obs.) revealed a tendency for some traits to be less differentiated than allozymes, presumably because of unifying selection. When combined with genetic evidence from *Scabiosa* (this study), there is considerable variation in the extent to which marker genes provide a valuable overview of the genetic structure for quantitative characters.

Yang *et al.* (1996) reviewed the basic assumptions underlying comparative studies of population genetic structure. Implicit in the present investigation are the assumptions that the allozymes are selectively neutral and that different allozyme loci provide more or less independent information on single-locus diversity. However, we cannot exclude the possibility that the genetic variances within populations have been overestimated because of nongenetic (maternal) effects, particularly for traits expressed early in the ontogeny, a factor that would lead to a downward bias in Q_{ST} . Inclusion of variance arising from nonadditive gene interactions (dominance, epistasis) could also affect Q_{ST} , but the detection of such effects would require a more complex crossing design. The partitioning of variation into additive and nonadditive genetic components (Falconer, 1989) will be the goal of a future study of *S. canescens*.

Differences between species

Rare species tend to have less allozyme diversity than more common and widespread taxa (Hamrick & Godt, 1996), but there have been few attempts to examine whether rarity is associated with low levels of genetic variation in traits directly related to fitness. For example, Black-Samuelsson & Andersson (1997) demonstrated more divergent genotypes in the broadly adapted *Vicia dumetorum* than in the rare and ecologically restricted *V. pisiformis*. Genetic data from the present investigation not only indicate low levels of population differentiation in the rare *S. canescens*, but also suggest that natural selection may be responsible for the extensive population divergence in *S. columbaria*. One interpretation of these findings is that *S. columbaria* is more responsive to local selection pressures, perhaps explaining why this species has a broader ecological amplitude than *S. canescens*.

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