

Among-population variation and correlations in sexually dimorphic traits of *Silene latifolia*

L. F. DELPH,* F. N. KNAPCZYK* & D. R. TAYLOR†

*Department of Biology, Indiana University, Bloomington, IN, USA

†Department of Biology, University of Virginia, Charlottesville, VA, USA

Keywords:

common-garden experiment;
dioecious;
resource allocation;
sexual dimorphism;
sexual selection;
Silene latifolia.

Abstract

The degree of sexual dimorphism in a trait may be determined directly by disruptive selection, as well as by correlations with other traits under selection. We grew seeds from nine populations of the dioecious plant *Silene latifolia* in a common-garden experiment to determine whether phenotypic variation and correlations existed for floral, leaf and resource allocation traits, and whether this variation had a genetic component. We also determined the traits which were sexually dimorphic, the degree of dimorphism, and whether it varied among populations. Seven traits exhibited among-population variation and sexual dimorphism. Variation in the degree of dimorphism occurred only for two traits, suggesting that dimorphism may be evolving more slowly than trait means. Males had more, smaller flowers, shorter leaves, and allocated less of their total biomass to stems and more to leaves than females. Flower production was the most sexually dimorphic trait and was correlated with all measured traits. Most traits exhibited significant correlations between the sexes. The pattern of correlations and the degree of sexual dimorphism among traits lead us to suggest that intrasexual selection for an exaggerated floral display in males has indirectly led to sexual dimorphism in a host of other traits.

Introduction

Sexual dimorphism is an emergent characteristic of many plant species that exhibit gender dimorphism (Geber *et al.*, 1999). Dimorphism in primary sexual organs is a necessary consequence of gender dimorphism; but less obviously, dimorphism in traits that are not directly related to physical reproduction is also pervasive. Species can be sexually dimorphic in size, colour, and longevity of vegetative and reproductive structures, in resource acquisition and allocation, and in interactions with other community members (see reviews by Delph *et al.*, 1996; Ågren *et al.*, 1999; Dawson & Geber, 1999; Delph, 1999; Eckhart, 1999). This study focuses on sexual dimorphism in a variety of traits in the dioecious, flowering plant *Silene latifolia*.

Correspondence: Lynda F. Delph, Department of Biology, Indiana University, Bloomington, IN 47405, USA.
Tel.: 812-855-1831; fax: 812-8556705;
e-mail: ldelp@bio.indiana.edu

Sexual dimorphism is thought to arise because the sexes have different reproductive roles, and consequently, are differentially selected to perform these roles (Darwin, 1871). Both natural and sexual selection can lead to sexual dimorphism. Natural selection could cause dimorphism to evolve in homologous, nondimorphic traits if viability and/or growth are maximized at different values for males and females (e.g. Lloyd & Webb, 1977; Wallace & Rundel, 1979; Meagher, 1984). Sexual selection as a mechanism for the evolution of sexual dimorphism was originally invoked by Darwin (1871), and was described as having intrasexual and intersexual components. With intrasexual selection, selected individuals are those that acquire the most mates or matings. Intersexual selection for dimorphism arises if one sex, usually females, can directly choose their mates from a pool of potential mates.

The extent of sexual dimorphism is constrained by the amount of genetic variation in sexually dimorphic traits in a population that natural and sexual selection can act

upon. Between-sex genetic correlations for homologous traits can also limit, or at least slow, their evolutionary trajectory towards dimorphism (Lande, 1980). In addition, if sexual dimorphism occurs in more than one trait, the degree to which it is expressed can be constrained by correlations among traits within a single sex (Lande, 1980; Meagher, 1994). Furthermore, if exaggeration of a trait in one sex enhances its fitness via mating success but also reduces fitness via viability or growth in the other sex, an increase in the degree of sexual dimorphism would be constrained. Hence, the degree of sexual dimorphism, as well as the average phenotype for a trait, will reflect a balance between what is optimal for males and females, the genetic variance-covariance structure, and what values are optimal under natural vs. sexual selection (Lande, 1980).

Determining the relative influence of correlations, natural selection, and sexual selection is a current focus of studies on the evolution of sexual dimorphism in plants (e.g. Bond & Maze, 1999; Davis, 2001). Here, we report the results of a common-garden experiment looking at the phenotypic variation and correlations among populations of *S. latifolia*. We grew seeds from diverse populations in a single environment to eliminate differences that may be caused by environmental differences among the populations (e.g. Kohorn, 1995). Hence, barring strong maternal effects, any among-population differences observed could be attributed to a genetic component.

Our objectives were (1) to determine whether sexual dimorphism evolves as rapidly as trait means, and (2) to investigate why the magnitude of sexual dimorphism varies across traits. We considered the hypothesis that disruptive selection may lead to sexual dimorphism in one trait, with dimorphism evolving in other traits simply by virtue of the fact that they are genetically correlated with the one under direct selection. For example, sexual selection might favour differences between the sexes in characters such as the number and size of flowers. If true, then we would predict a large degree of sexual dimorphism of traits such as flower number, and the magnitude of sexual dimorphism in other traits would be correlated with the magnitude of their genetic correlation with the trait under selection.

We measured nine floral, leaf, and resource allocation traits and determined whether populations differed in phenotype. Furthermore, we determined whether or not there were phenotypic correlations among traits at the among-population, within-sex and between-sex levels. We also determined the extent of sexual dimorphism in these traits and whether sexual dimorphism varies to the same degree as phenotypic means among populations. Lastly, we investigated whether the degree of sexual dimorphism in a trait was related to its correlation with the most strongly dimorphic trait, flower number. We found that many traits exhibit sexual dimorphism, and although there is among-population variation in phenotype,

variation in the degree of sexual dimorphism was less prevalent. Sexual dimorphism in flower production was pervasive, and was correlated with all of the other sexually dimorphic traits in a pattern that suggests there has been disruptive selection on flower number, with dimorphism in other traits evolving as a consequence of between-trait correlations.

Materials and methods

Study system

Silene latifolia (Caryophyllaceae) is a dioecious, weedy, short-lived perennial. It is native to eastern Europe and the Mediterranean region (Baker, 1948), and was introduced and has spread within North America (McNeill, 1978). Its flowers have white petals that open for the first time in the late afternoon and are primarily pollinated at night by moths (Shykoff & Bucheli, 1995). Gender is determined by sex chromosomes, with males being the heterogametic sex (Warmke, 1946).

A common-garden experiment was set up using seeds from nine wild populations (Table 1), and a variety of traits were measured. Seven European populations from Douglas Taylor's *S. latifolia* collection (which can be viewed online at wsrv.clas.virginia.edu/~drt3b/home.html) were included. In addition, seeds collected (in 2000) from two Virginia populations (Giles County) were included. Eight seeds from four to nine families from each of the nine populations (for a total of 440 seeds) were planted in 196-celled trays in November 2000 and were thereafter maintained in a greenhouse at Indiana University (see Table 1 for number of families used per population). Up to four seedlings from each family were transplanted into 4-in clay pots in December 2000. Plants were grown in a 2 : 1 mixture of Metromix and sterilized soil, under a 16 : 8 h light : dark cycle. Plants were fertilized once a month with half-strength all purpose 20:20:20 (J.R. Peters Inc., Allentown, PA, USA), and were watered daily as needed. The position of each plant within the greenhouse was altered weekly to reduce position effects.

Variation in floral, leaf, and resource allocation traits

We choose which traits to measure, based on previous reports of sexual dimorphism in *S. latifolia* (Lawrence, 1963; van Nigtevecht, 1966; Gross & Soule, 1981; Meagher, 1992; Delph & Meagher, 1995). We measured flower production, calyx width, length, petal-limb length (the portion of the petal above the fused calyx), leaf length, specific leaf area and percentage of total mass allocated to reproduction, leaves and stems. These traits are unlikely to be affected by maternal effects, as seed size does not influence any growth factors after the first week following germination and has no effect on floral

Table 1 Source populations and number of plants used in our study.

Population*	Origin	No. of families	No. of individuals harvested	No. of individuals used to determine SLA
1	Virginia (A) (Giles County)	6	11	6
2	Virginia (B) (Giles County)	6	21	11
3 – CRC	Portugal (Cabo de Roca)	5	15	11
4 – REC	France (between Paris and Alsace)	7	10	11
5 – WIN	France (Alsace)	5	18	10
6 – ROS	Italy (Tuscany)	9	25	15
7 – SVL	Austria (St Valentin)	4	12	7
8 – ZAG	Croatia (Zagreb)	5	18	9
9 – BDA	Hungary (Budapest)	7	26	14

*The three-letter designations were taken from D. Taylor's *Silene* collection web site (wsrv.clas.virginia.edu/~drt3b/home.html).

characters (L. Delph and T. Meagher, unpublished data). Flower production refers to the number of flowers produced during the first 30 days of flowering for each plant. All flowers produced during this 30-day period were collected as they abscised, and were used for calculating the total biomass of each individual (see below). Calyx and petal measures were taken on the third, fourth, and fifth flower to open on each plant using digital calipers. Data from these three flowers per plant were averaged for statistical analyses. Leaf length was always measured for one of the two leaves at the second node down from the node with the first flower to open; this measurement was always taken on the same day the third flower was measured. The leaf area (using a LiCor leaf-area meter) and dry mass of a rosette leaf were determined to calculate specific leaf area (SLA = area/mass); larger SLA values indicate thinner leaves. This value was obtained for a subset of plants in May 2001 (see Table 1).

After each plant had flowered for 30 days, the plants were harvested and their tissues were divided into leaf, stem and reproductive parts (including all remaining flowers and buds, as well as the flowers collected during the first 30 days of flowering). Plant parts were weighed after being dried in a 60 °C oven. Biomass allocation to reproduction, leaves, and stems was determined by dividing the mass of each by the total mass of the plant. Tests of all three sets of parts would not allow for independence, as allocation to any two determines allocation to the third. We therefore chose to analyse allocation only to leaves and stems. This was based on the fact that our unpollinated females would be allocating many fewer resources to reproduction than would pollinated females (Delph & Meagher, 1995), making allocation to reproduction the least relevant of the three allocation measures. Our nonpollinated females would have made more flowers than if we had pollinated them (Delph & Meagher, 1995), but flower and leaf size traits are unlikely to have been affected (L. Delph and J. Gehring, unpublished data).

Statistical analyses

All analyses were performed using SPSS 10.0 for Macintosh. We performed two-way mixed-model ANOVAS to test for the effect of sex and population for each trait. The interaction term was removed from the model if it was not significant at $P > 0.20$, and the model was then rerun with only the main effects. When the effect of sex was significant, we ran one-way ANOVAS combining sex and population into one categorical variable, to determine the populations which exhibited a significant difference between the sexes. When the main effect of population was significant, we used an *a posteriori* test (Tukey) to determine which populations differed significantly from each other. The ln of flower number and leaf length were used in analyses of these traits in order to obtain equal variances; we graphed the nontransformed values in Figs 1 and 2. We calculated two-tailed tests of Pearson's correlations to investigate correlations among traits at three levels: across populations, within the sexual morphs and between the sexual morphs across populations. A one-tailed test of the Pearson's correlation was calculated to find the correlation between the percentage of sexual dimorphism in the traits and the correlation of each trait with flower number, as we predicted *a priori* that this relationship would be positive. The percentage of sexual dimorphism was calculated as the difference between the female and male trait mean, divided by the male mean, multiplied by 100. This was calculated separately for each population and then averaged across populations to get the mean percentage sexual dimorphism for each trait.

Results

Floral traits

Males produced approximately 3.5 times as many flowers as unpollinated females over a 30-day period [157 ± 11

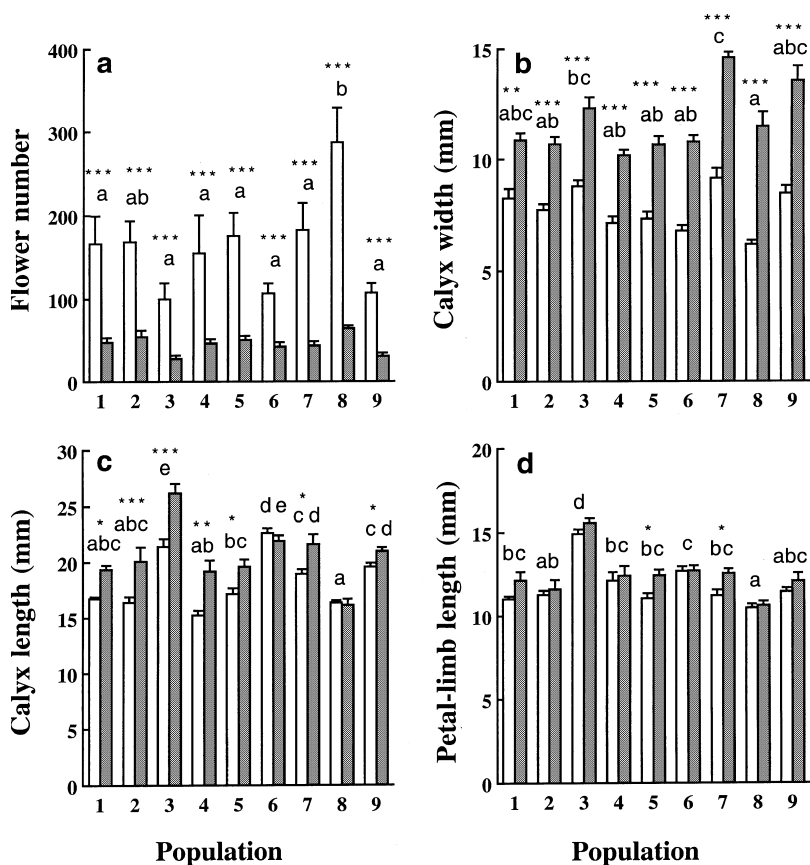


Fig. 1 Means (± 1 SE) for floral traits by population and sex (males – open bars, females – dark bars). Populations with different superscripts are significantly different from each other. Asterisks indicate a significant difference between the sexes (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$).

flowers (mean ± 1 SE), $n = 79$ vs. 45 ± 2 , $n = 80$, respectively; sexual dimorphism = 71%). The sex \times population interaction was not significant ($P = 0.69$) and was therefore removed from the ANOVA model. The sex of the plant had a significant effect on flower production (Table 2), with males producing significantly more flowers than females in all nine study populations (Fig. 1a). In addition, the populations differed significantly in flower production (Table 2), with individuals from the Croatian population (Pop 8) differing significantly from seven of the other eight populations. Flower production by both males and females were greatest for individuals from Croatia and lowest in individuals from Portugal (Pop 3). Males from Croatia produced 2.9 times more flowers, on average, than males from Portugal, and females produced 2.3 times more flowers.

Calyx width was also found to be sexually dimorphic (sexual dimorphism = 51%; Table 2). On average, the calyces of flowers on females were 1.5 times wider than those on males (11.6 ± 0.2 mm vs. 7.7 ± 0.1 mm, $n = 83$ and 84, respectively). Just as with flower number, calyx width differed significantly between the sexes in all nine populations (Fig. 1b). Population also had a significant effect on calyx width (Table 2), with flowers from the Austrian population (Pop 7) having significantly wider calyces than those of five other populations. In addition,

the population with the greatest flower production (Croatia – Pop 8) produced the narrowest flowers, and this difference was significant in comparison with two other populations (Portugal – Pop 3 and Austria – Pop 7). There was a significant sex \times population interaction for calyx width (Table 2), indicating that the degree of sexual dimorphism differed among populations for this trait. The Austrian population (Pop 7) exhibited both the largest means for calyx width and the largest difference between the sexes.

On average, the calyx of flowers on males was 0.9 times the length of calyces on flowers of females (18.7 ± 0.31 mm vs. 19.8 ± 0.25 mm, respectively; sexual dimorphism = 12%). The main effects of sex and population were significant, and there was a significant sex \times population interaction (Table 2). Seven of nine populations exhibited significantly shorter calyces on staminate than pistillate flowers (Fig. 1c). Calyx length did not differ significantly in two populations that exhibited the reverse pattern. Flowers from Portugal (Pop 3) had significantly longer calyces than flowers from seven other populations, and those from Croatia (Pop 8) had significantly shorter calyces as compared with five populations.

On an average, mean petal-limb length was 11.8 ± 0.17 mm for flowers on males and 12.5 ± 0.18 mm for

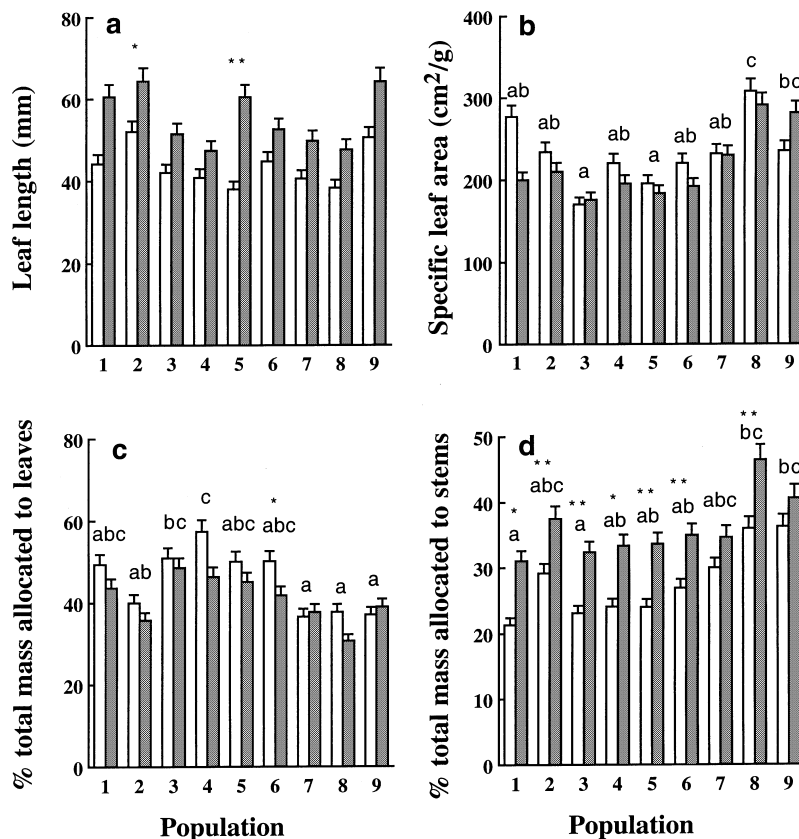


Fig. 2 Means (+1 SE) for leaf and allocation traits by population and sex (males – open bars, females – dark bars). Populations with different superscripts are significantly different from each other. Asterisks indicate a significant difference between the sexes (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$).

flowers on females (sexual dimorphism = 6%). The sex \times population interaction was not significant ($P = 0.54$) and was removed from the model. The effect of sex in the two-way ANOVA was significant (Table 2), and in all nine populations flowers on females had longer petal limbs than those on males; however, the difference was only significant in two populations (Fig. 1d). Significant variation in petal-limb length among populations was found (Table 2). Flowers from Portugal (Pop 3) had longer petal limbs than those of all other populations, and the petal limbs of Croatian individuals (Pop 8) were significantly shorter than those of six other populations (Fig. 1d).

Flower production was significantly negatively correlated with the three floral-size traits, with the strongest correlation between flower production and calyx width (Table 3a). Furthermore, the three flower-size traits were all significantly positively correlated. The directions of the correlations of floral traits within sexes were similar to the direction of the correlations across populations (Table 3b). However, differences did exist in the magnitude of the correlations, with neither sex showing a significant within-sex correlation for calyx width and calyx length. In terms of between-sex correlations, all four floral traits in males were significantly positively correlated with the same traits in females (Table 3c).

Furthermore, flower number was always negatively correlated to the other floral traits, and five out of six of these between-sex correlations were significant (Table 3c).

Leaf traits

On an average, leaves from females were 1.3 times longer than those on males (56 ± 1.9 mm vs. 45 ± 1.3 mm, respectively; sexual dimorphism = 28%). The sex \times population interaction was not significant ($P = 0.86$) and was removed from the model. Sex had a significant effect on leaf length in the two-way ANOVA (Table 2), and leaves on males were significantly shorter than those on females in two populations (Virginia B – Pop 2 and France B – Pop 5; Fig. 2a). None of the populations differed significantly from each other in leaf length for the *a posteriori* test, although the population effect was significant in the two-way ANOVA (Table 2).

Leaves on females were 1.1 times thicker than leaves on males (SLA of 213 ± 7.1 and 231 ± 8.6 cm² g⁻¹, respectively; sexual dimorphism = 6%). The sex \times population interaction was not significant (Table 2). Although males had higher SLA values than females in seven populations, the main effect of sex was not significant in the two-way ANOVA (Table 2). The effect of population was significant (Table 2). The average SLA

Table 2 Results of two-way ANOVAS testing for the effect of sex and population on eight traits. The sex \times population interaction was removed from the model if the *P*-value for the interaction was >0.20 . When the sex \times population interaction was included in the model the error d.f and MS used to test the sex effect is listed directly under the sex-effect line in each case. The population effect was tested using the d.f and MS from the sex \times population interaction when the interaction was included.

Source	d.f.	MS	<i>F</i>	<i>P</i>
Flower number				
Sex	1	53.2	299.6	<0.001
Population	8	1.8	9.9	<0.001
Error	149	0.2		
Calyx width				
Sex	1	546.7	124.1	<0.001
Error	8.4	4.4		
Population	8	22.0	4.7	0.021
Sex \times population	8	4.7	3.6	0.001
Error	149	1.3		
Calyx length				
Sex	1	186.8	11.8	0.008
Error	8.3	15.8		
Population	8	104.5	6.2	0.009
Sex \times population	8	16.9	4.6	<0.001
Error	149	3.7		
Petal-limb length				
Sex	1	13.2	10.6	0.001
Population	8	27.9	22.4	<0.001
Error	157	1.2		
Leaf length				
Sex	1	2.0	23.9	<0.001
Population	8	0.2	2.2	0.032
Error	157	0.1		
Specific leaf area				
Sex	1	4387.1	1.8	0.214
Error	8.9	2445.6		
Population	8	14184.8	5.7	0.012
Sex \times population	8	2502.8	1.5	0.183
Error	76	1704.3		
% Total biomass allocated to leaves				
Sex	1	0.07	6.2	0.014
Population	8	0.06	5.6	<0.001
Error	146	0.01		
% Total biomass allocated to stems				
Sex	1	0.24	53.2	<0.001
Population	8	0.04	9.4	<0.001
Error	146	0.01		

of plants from Croatia (Pop 8) stood out as significantly greater than that of seven other populations, with Portugal plants (Pop 3) exhibiting the lowest SLA (Fig. 2b).

Correlation coefficients between leaf traits (leaf length and SLA) and flower number are shown in Table 4a. Flower number was significantly negatively correlated to leaf length and positively correlated to SLA. These correlations were not significant, however, within the sexes (Table 4b). Leaf length and SLA in males were

significantly positively correlated to the same traits in females (Table 4c).

Resource allocation traits

Males allocated $45\% \pm 1.3$ and $28\% \pm 0.8$ of their total biomass to leaves and stems, respectively, whereas females allocated $41\% \pm 1.2$ and $36\% \pm 0.8$ (sexual dimorphism = 9 and 32%, respectively). The sex \times population interactions for the percentage of total mass allocated to leaves or stems were not significant ($P = 0.74$ and 0.90 , respectively) and were removed from the models. Significant sexual dimorphism was found in the percentage of total mass allocated to leaves (Table 2), however, this difference was significant in the *a posteriori* tests only for the Italian population (Pop 6). The effect of population was highly significant (Table 2), with plants from one of the French populations (Pop 4) allocating a significantly greater proportion of total mass to leaves as compared with plants from four other populations (Fig. 2c). Males spent significantly less of their total mass on stems compared with females (Table 2), and this sexual dimorphism was significant in seven of the nine populations (Fig. 2d). Populations also differed significantly in the percentage of total mass allocated to stems (Table 2). Across all populations, plants from Croatia (Pop 8) had the highest percentage allocation of mass to stems and the lowest to leaves.

The percentages of total biomass allocated to either leaves or stems were significantly correlated with flower number, leaf length, and SLA (Table 4a). These correlations were negative for the percentage of total mass allocated to leaves and positive for the percentage of total mass allocated to stems. Furthermore, allocations to leaves and stems were negatively correlated with each other; this was also true of the within-sex correlations for these traits (Table 4b). At the within-sex level for both males and females the percentages of total mass allocated to leaves or stems were, respectively, negatively and positively correlated to leaf length and SLA (Table 4b). Between-sex correlations for the percentage of total mass allocated to leaves and stems were significantly positively correlated, and each was negatively correlated with the other trait in the other sex (Table 4c). We also found that increasing thinness in leaves (i.e. increasing SLA) in either sex was negatively correlated with the percentage of total mass allocated to leaves in the other sex (Table 4c).

Correlation with percentage sexual dimorphism

The most sexually dimorphic trait was flower number (see above). The relationship between the percentage of sexual dimorphism for each trait and the correlation of each trait with flower number was significantly positive ($r = 0.67$, $n = 7$, $P = 0.05$).

Table 3 Correlation coefficients for floral traits (a) across the nine populations (b) within the sexes (male values above the diagonal ($n = 79$ for comparisons with flower number, $n = 83$ for comparisons between floral traits) and female values below the diagonal ($n = 79$ for comparisons with flower number, $n = 84$ for comparisons between floral traits)), and (c) between the sexes (males along the top, females along the bottom of the diagonal). Mean male and female trait values in each population were used to determine the between-sex correlations ($n = 9$).

	Flower number	Calyx width	Calyx length	Limb length
(a) Across populations				
Calyx width	-0.58***			
Calyx length	-0.41***	0.35***		
Limb length	-0.34***	0.31***	0.57***	
(b) Within sexes				
Flower number		-0.27*	-0.41***	-0.35**
Calyx width	-0.27*		0.16	0.27*
Calyx length	-0.28*	0.18		0.58***
Limb length	-0.27*	0.23*		0.52***
(c) Between the sexes				
Flower number	0.88**	-0.68*	-0.68*	-0.72*
Calyx width	-0.10	0.69*	0.38	0.03
Calyx length	-0.80**	0.65	0.75*	0.87**
Limb length	-0.69*	0.54	0.61	0.93***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Table 4 Correlation coefficients for leaf and stem traits (a) across the nine populations (b) between the sexes (male values above the diagonal, female values below the diagonal), and (c) between the sexes (males along the top, females along the bottom of the diagonal). Mean male and female trait values in each population were used to determine the between-sex correlations ($n = 9$).

	Flower number ($n = 159$)	Leaf length ($n = 159$)	SLA ($n = 87$)	% allocated to leaves ($n = 156$)	% allocated to stems ($n = 156$)
(a) Across populations					
Leaf length	-0.25**				
SLA	0.27*	0.19			
% Total mass allocated to leaves	-0.20*	-0.37***	-0.30**		
% Total mass allocated to stems	0.32**	0.39*	0.28*	-0.82***	
(b) Within the sexes					
Flower number		-0.03	0.33*	-0.43***	0.32**
Leaf length	-0.10		0.25	-0.41***	0.35**
SLA	-0.16	0.26		-0.30	0.38*
% Total mass allocated to leaves	-0.25*	-0.32**	-0.35*		-0.85***
% Total mass allocated to stems	0.19	0.25*	0.36*	-0.84***	
(c) Between the sexes					
Flower number	0.88**	-0.29	0.67*	-0.19	0.19
Leaf length	-0.30	0.70*	-0.05	-0.24	0.04
SLA	0.50	0.14	0.68*	-0.78*	0.93***
% Total mass allocated to leaves	-0.68*	-0.18	-0.74*	0.84**	-0.82**
% Total mass allocated to stems	0.58	0.08	0.60	-0.68*	0.92***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Discussion

Sexual dimorphism

Populations of *S. latifolia* exhibited significant sexual dimorphism in seven of eight floral, leaf, and resource allocation traits. The most extreme sexual dimorphism was found for flower production. Males made many, small flowers in comparison to females (see also Gross & Soule, 1981; Meagher, 1992; Gehring & Linhart, 1993; Delph & Meagher, 1995). In addition to sexual dimorphism in floral traits, individual leaves on males were significantly smaller and the percentage of their total biomass allocated to leaves was higher, as compared with females. Females, in contrast, invested more in stems than did males. There was evidence that the magnitude

of sexual dimorphism for a given trait was affected by its correlation with flower number, with the most sexually dimorphic traits being those that were most correlated with flower number.

Correlations among traits

The traits we found to be sexually dimorphic were generally correlated with each other across populations. For example, populations with plants that produced many flowers relative to other populations made relatively short, thin leaves and small flowers. In addition, they allocated less of their biomass to leaves and more to stems. This pattern of intercorrelations among traits at the phenotypic level concurs with other studies of genetic variation and resource allocation in *S. latifolia*.

For example, calyx width is negatively genetically correlated with flower production in males (Meagher, 1999). Moreover, selection to increase flower production resulted in a significant decrease in calyx width (Meagher, 1994). Similarly, Delph & Meagher (1995) found a phenotypic trade-off between growth and reproduction within both males and females, and Meagher (1992) found a significant negative genetic correlation between the date of first flowering and height. Overall, the correlations seen in our study, combined with those of intrapopulation level studies, suggest that some of the sexual dimorphism may be indirectly caused by selection for dimorphism in flower number, instead of being a consequence of differing optima between the sexes for the whole host of traits. The pattern of correlations suggests that not only do flower number and size tradesoff exist, but that investment in flowers trades off with leaf characters and growth.

Selection for sexual dimorphism

Disruptive selection has been proposed as a mechanism for the evolution of sexual dimorphism from monomorphism (see Geber, 1999, for a review). Meagher (1999) has shown via a quantitative genetic analysis that flowers on males evolved to be smaller in *S. latifolia*, more so than females evolving to be bigger. This conclusion was based on measures of phenotypic means and estimates of genetic variance-covariance matrices, which allowed calculation of selection gradients given different starting scenarios: selection intensity on males appeared to be double that on females (Meagher, 1999). Hence, it seems worthwhile to ask why smaller flowers would be favoured by males but not by females. In both cases the production of smaller flowers would reduce resource expenditure per flower and allow the production of more flowers, as seen from the negative correlations between flower-size traits and flower number that were found in both sexes. In other words, the only way a plant of either sex can make more flowers is to make smaller flowers.

The most sexually dimorphic trait was flower production and the magnitude of sexual dimorphism in other traits was correlated with how correlated these traits were with flower number. These results support the hypothesis that intrasexual selection for greater flower production in males but not in females may have caused sexual dimorphism to evolve in a variety of traits (Bond & Midgley, 1988). Fitness in males might be more affected by the number of flowers produced, if a male's fitness is more mate-limited than that of a female. Males that produce a relatively large number of flowers might be likely to acquire more matings given that pollinators prefer *S. latifolia* plants with large floral displays (Shykoff & Bucheli, 1995). In contrast, producing more flowers may not lead to an equivalent increase in fitness for females for two reasons. First, ovule number per flower might decrease with flower size given that the calyx

houses the ovules and developing seeds. Second, access to mates is unlikely to be the major factor limiting fitness in females given that flower and seed production appears to be greatly impacted by resource availability (Delph & Meagher, 1995; Meagher & Delph, 2001). If this pattern of males being limited more by mates and females more by resources (referred to as Bateman's principle) holds, then selection would favour males with a relatively exaggerated phenotype (Bateman, 1948) – i.e. large flower displays in this particular case. Furthermore, if high flower production were genetically correlated with traits that reduced fitness via natural selection (e.g. smaller leaf size and less growth), then sexual selection for high flower production by males could result in males deviating from the optimum value that natural selection alone would select for (Lande, 1980). The outcome would be sexual dimorphism in flower production, as well as sexual dimorphism in other correlated traits. Similarly, Bond & Maze (1999) demonstrated that although large floral displays increased pollinator attraction in males they also decreased viability in the dioecious shrub *Leucadendron xanthoconus*. They therefore suggested that if the increase in pollinator visits translated into an increase in mating opportunities, then intrasexual selection for sexual dimorphism would be counter-balanced by viability selection.

Traits varying in the degree of sexual dimorphism

Of the seven sexually dimorphic traits, only calyx width and length displayed significant among-population variation in the degree of sexual dimorphism. This result is consistent with the hypothesis that phenotypic means will evolve more quickly than the degree of sexual dimorphism (Lande, 1980). Lande (1980) suggested that the evolution of sexual dimorphism is constrained primarily by correlations between the sexes in homologous traits. In our study, there were significant positive between-sex correlations for all of the sexually dimorphic traits. For example, the between-sex correlations for flower number and calyx width were 0.88 and 0.69, respectively. Genetic correlations may be the underlying cause of these phenotypic correlations (see Cheverud, 1988). For example, Meagher (1999) showed for two populations of *S. latifolia* from Virginia that the between-sex genetic correlation for flower number ranged from 0.77 to 0.84 and that for calyx width ranged from 0.82 to 0.90. Moreover, selection to change calyx width in one sex resulted in a correlated response in the other sex (Meagher, 1994). Genetic correlations are overcome when loci develop that modify one trait without modifying correlated traits. However, this process can be slow, and is expected to be orders of magnitude slower than the evolution of the mean phenotype for a sexually dimorphic trait (Lande, 1980). Support for this comes from our result that although only two of the seven sexually dimorphic traits exhibited significant among-population

variation in the degree of sexual dimorphism, all seven exhibited significant among-population variation in their means.

Ecological factors affecting sexually dimorphic traits

By surveying variation in floral, leaf and resource allocation traits among populations in a common-garden experiment, we have shown that there is a genetic basis to phenotypic variation among populations. Given this, we suggest that it would be worthwhile to investigate which factors have contributed to this variation and whether the phenotypic differences among populations are adaptive. Results from previous studies suggest that pollinators, competitors, seed predators, pathogens, and availability of resources are likely to be important factors. For example, having more flowers, and thereby being more attractive to pollinators, might be disadvantageous in the presence of a pollinator-transmitted smut that causes the sterilization of flowers (Alexander & Antonovics, 1988; Thrall & Jarosz, 1994). Further, Lyons *et al.* (1994) found that sexual dimorphism in flower number can vary with the density at which groups of plants are grown. Lastly, flower size or number may influence the level of seed predation by the moth *Hadena rivularis*, which acts as both a pollinator and seed predator (Brantjes, 1976; Goulson & Jerrim, 1997).

Our results suggest that the populations from Portugal and Croatia are remarkably divergent from each other for several traits. Plants from Portugal, for example, exhibited extremely thick leaves and made relatively few flowers, whereas those from Croatia had the thinnest leaves and the most numerous and the smallest flowers. It would be interesting to study the ecology of these populations to see whether ecological factors are responsible for these differences.

In summary, we found that variation in phenotypic means among populations occurs more often than variation in the degree of sexual dimorphism. In addition, the extent of correlations among sexually dimorphic traits suggests that constraints on the independent evolution of some traits may exist, and be a product of selection for dimorphism in the most sexually dimorphic trait, flower number. Our results point the way for future investigations into how intrasexual selection for flower number influences the evolutionary trajectory of a host of other traits in this dioecious species.

Acknowledgments

The experiments and collections were supported by National Science Foundation grants DEB-0075318 and DEB-9876397 (and REU supplements) to LFD and DRT, respectively.

References

- Ågren, J., Danell, K., Elmqvist, T., Ericson, L. & Hjalten, J. 1999. Sexual dimorphism and biotic interactions. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 217–266. Springer-Verlag, Berlin.
- Alexander, H.M. & Antonovics, J. 1988. Disease spread and population dynamics of anther-smut infection of *Silene alba* caused by the fungus *Ustilago violacea*. *J. Ecol.* **76**: 91–104.
- Baker, H.G. 1948. Stages in invasion and replacement demonstrated by species of *Melandrium*. *J. Ecol.* **36**: 96–119.
- Bateman, A.J. 1948. Intrasexual selection in *Drosophila*. *Heredity* **2**: 349–369.
- Bond, W.J. & Maze, K.E. 1999. Survival costs and reproductive benefits of floral display in a sexually dimorphic dioecious shrub, *Leucadendron xanthoconus*. *Evol. Ecol.* **13**: 1–18.
- Bond, W.J. & Midgley, J. 1988. Allometry and sexual differences in leaf size. *Am. Nat.* **131**: 901–910.
- Brantjes, N.B.M. 1976. Riddles around the pollination of *Melandrium album* (mill.) Garcke (Caryophyllaceae) during the oviposition by *Hadena bicurvis* Hufn. (Noctuidae, Lepidoptera). *Proc. Koninklijke Nederlandse Akademie Van Wetenschapper, Series C* **79**: 1–12.
- Cheverud, J.M. 1988. A comparison of genetic and phenotypic correlations. *Evolution* **42**: 958–968.
- Darwin, C. 1871. *The Descent of Man, and Selection in Relation to Sex*. Murray, London.
- Davis, S.L. 2001. Phenotypic and genetic correlations among floral traits in two species of *Thalictrum*. *J. Hered.* **92**: 361–366.
- Dawson, T.E. & Geber, M.A. 1999. Dimorphism in physiology and morphology. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 175–216. Springer-Verlag, Berlin.
- Delph, L.F. 1999. Sexual dimorphism in life history. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 148–173. Springer-Verlag, Berlin.
- Delph, L.F., Galloway, L.F. & Stanton, M.L. 1996. Sexual dimorphism in flower size. *Am. Nat.* **148**: 299–320.
- Delph, L.F. & Meagher, T.R. 1995. Sexual dimorphism masks life history trade-offs in the dioecious plant *Silene latifolia*. *Ecology* **76**: 775–785.
- Eckhart, V.M. 1999. Sexual dimorphism in flowers and inflorescences. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 123–148. Springer-Verlag, Berlin.
- Geber, M.A. 1999. Theories of the evolution of sexual dimorphism. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 97–122. Springer-Verlag, Berlin.
- Geber, M.A., Dawson, T.E. & Delph, L.F. 1999. *Gender and Sexual Dimorphism in Flowering Plants*. Springer-Verlag, Berlin.
- Gehring, J.L. & Linhart, Y.B. 1993. Sexual dimorphisms and response to low resources in the dioecious plant *Silene latifolia* (Caryophyllaceae). *Int. J. Plant Sci.* **154**: 152–162.
- Goulson, D. & Jerrim, K. 1997. Maintenance of the species boundary between *Silene dioica* and *S. latifolia*. *Oikos* **79**: 115–126.

- Gross, K.L. & Soule, J.D. 1981. Differences in biomass allocation to reproductive and vegetative structures of male and female plants of a dioecious, perennial herb, *Silene alba* (Miller) Krause. *Am. J. Bot.* **68**: 801–807.
- Kohorn, L.U. 1995. Geographic variation in the occurrence and extent of sexual dimorphism in a dioecious shrub, *Simmondsia chinensis*. *Oikos* **74**: 137–145.
- Lande, R. 1980. Sexual dimorphism, sexual selection, and adaptation in polygenic characters. *Evolution* **34**: 292–305.
- Lawrence, C.W. 1963. Genetic studies on wild populations of *Melandrium*. II. Flowering time and plant weight. *Heredity* **18**: 149–163.
- Lloyd, D.G. & Webb, C.J. 1977. Secondary sex characters in plants. *Bot. Rev.* **43**: 177–216.
- Lyons, E.E., Miller, D. & Meagher, T.R. 1994. Evolutionary dynamics of sex ratio and gender dimorphism in *Silene latifolia*. I. Environmental effects. *J. Hered.* **85**: 196–203.
- McNeill, J. 1978. *Silene alba* and *S. dioica*. North America and the generic delimitation of *Lychnis*, *Melandrium*, and *Silene* (Caryophyllaceae). *Can. J. Bot.* **56**: 297–308.
- Meagher, T.R. 1984. Sexual dimorphism and ecological differentiation of male and female plants. *Ann. Miss. Bot. Gard.* **71**: 254–264.
- Meagher, T.R. 1992. The quantitative genetics of sexual dimorphism in *Silene latifolia* (Caryophyllaceae). I. Genetic variation. *Evolution* **46**: 445–457.
- Meagher, T.R. 1994. The quantitative genetics of sexual dimorphism in *Silene latifolia* (Caryophyllaceae). II. Response to sex-specific selection. *Evolution* **48**: 939–951.
- Meagher, T.R. 1999. The quantitative genetics of sexual dimorphism. In: *Gender and Sexual Dimorphism in Flowering Plants* (M.A. Geber, T.E. Dawson, L.F. Delph, eds), pp. 275–294. Springer-Verlag, Berlin.
- Meagher, T.R. & Delph, L.F. 2001. Individual flower demography, floral phenology, and floral display size in *Silene latifolia*. *Evol. Ecol. Res.* **3**: 845–860.
- van Nigtevecht, G. 1966. Genetic studies in dioecious *Melandrium*. I. Sex-linked and sex-influenced inheritance in *M. album* and *M. dioicum*. *Genetica* **37**: 281–306.
- Shykoff, J.A. & Bucheli, E. 1995. Pollinator visitation patterns, floral rewards and the probability of transmission of *Microbotryum violaceum*, a venereal disease of plants. *J. Ecol.* **83**: 189–198.
- Thrall, P.H. & Jarosz, A.M. 1994. Host-pathogen dynamics in experimental populations of *Silene alba* and *Ustilago violacea*. I. Ecological and genetic determinants of disease spread. *J. Ecol.* **82**: 549–560.
- Wallace, C.S. & Rundel, P.W. 1979. Sexual dimorphism and resource allocation in male and female shrubs of *Simmondsia chinensis*. *Oecologia* **44**: 34–39.
- Warmke, H.E. 1946. Sex determination and sex balance in *Melandrium*. *Am. J. Bot.* **33**: 648–660.

Received 21 January 2002; revised 11 April 2002; accepted 11 June 2002