Seasonal Changes in Sexual Allocation within Flowers of *Chelidonium majus* (Papaveraceae)

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애기똥풀(양귀비과) 꽃에서 일어나는 성적자원 분배의 계절적 변화

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ABSTRACT

Seasonal variation in size and number of floral structures was examined in two Massachusetts populations of *Chelidonium majus,* a self-compatible herb. All floral characters except for anther number per flower declined significantly during a 3 week period. However, temporal patterns were not identical among characters or between populations. The results indicate that floral characters varied in conjunction with flower diameter, but that the pattern of changes in floral characters in response to environments may not be easy to predict. Principal Components Analysis was conducted to identify the functional relationship among floral characters. The first three components appeared to represent components of attractiveness, male function, and female function, respectively, perhaps reflecting the functional distinction of floral characters. Based on this pattern, the relative allocation to sexual structures within flowers was examined; male allocation was relatively greater than female allocation early or in the middle of the flowering season, depending upon populations. Temporal variation in relative allocation within flowers was not independent of seed yield components; different combinations of the size and number of floral characters were correlated with different seed yield components, for example, either seed size or number per fruit, during a season. In particular, allocations to attractive and male structures were highly correlated with mean seed weight only earlier in the season. These results provide some evidence that flowering phenology is an important component to be considered in the study of sexual allocation.

INTRODUCTION

Resource allocation to both sexual functions in hermaphroditic plants may not be sym-

metrical. This may be due to differential marginal return of male and female allocations (Charnov, 1982). The exact pattern of marginal return through both sexual functions may change depending upon the selfing rate and attractiveness (Charlesworth and Charlesworth, 1987; Lloyd, 1987). However, sexual allocation patterns potentially affecting reproductive success of plants may also vary during a season. Although it is already known that insect visitation rate (Thomson, 1982), pollen flow distance (Handel and Le Vie Mishkin, 1984; Palmer *et al*., 1988), allele frequencies in the pollen pool (Sampson *et al*., 1990), pollen or ovule production (McKone, 1989; Thomson *et al*., 1989), and flower characters (Huether, 1969; Stace and Fripp, 1977; Kang and Primack, 1991) change during the course of a growing season, flowering phenology during a season has rarely been considered in the study of sexual allocation.

It is likely that variations in ovule and pollen production, which represent the primary characters of sexual allocation, are correlated with flower diameter, which is a major component of attractiveness. In other words, if flower or petal size varies seasonally, one may suspect that there will be correlated variation in the sexual structures within flowers.

However, it is difficult to make predictions on the amount and direction of concomitant changes in size and number of floral structures within flowers because the developmental and genetic correlations among floral characters are still uncertain (e.g., Primack, 1987; Stanton and Preston, 1988) and partly because there are few empirical data that can be used for generating hypotheses or theories (Thomson, 1989).

In general, sexual allocation models (e.g., Geber and Charnov, 1986; Charlesworth and Charlesworth, 1987) assume two separate reproductive processes such as flowering and seed maturation. However, flowering and seed maturation occur continuously and simultaneously within and among inflorescences in many plants during a season. Thus, in light of a serial adjustment theory of maternal investment (Lloyd, 1980) and modular reproduction in plants (Stanton and Galloway, 1990), both resource allocations before and after fertilization are subject to adjustment within plants. Despite its ecological as well as evolutionary importance, seasonal variation in floral characters and subsequent changes in seed size and number have not been seriously considered in most allocation studies.

Previous study in the common, self-compatible herb, *Chelidonium majus,* showed that flower diameter and seed yield components (seed size and number per fruit) declined significantly toward the end of a growing season (Kang and Primack, 1991). In this study, the same species was used to investigate seasonal variation in floral structures representing sexual allocation and the relationship between such seasonal variation and seed yield components. A comprehensive analysis was undertaken to examine seasonal patterns of sexual allocation. First, temporal variation in size and number of various floral characters was described. Second, because determining the unique role of each floral structure to male and female functions is extremely difficult (Goldman and Willson, 1986), a statistical method was used to infer the functional relationship of floral

characters. Only the allocation to the sexual structures within flowers was considered as primary sexual allocation in this paper. And finally, the relationship between floral characters and seed yield components was explored to examine the temporal pattern in the relationship among allocations within flowers (attractiveness, male and female allocations) and fitness components.

MATERIALS AND METHODS

Chelidonium majus (Papaveraceae) is a common perennial herb that occurs in damp, disturbed habitats of the northeastern U. S. A. Following germination in the fall, a leafy rosette is formed. Aerial shoots with leaves are produced in the spring. Several umbels are borne on each aerial shoot, with usually four to five flowers per umbel. The total number of flowers produced during a season ranges from about 30 to 200 per plant. Flowering continues from early May until late June. Yellow, showy flowers usually last two days, and are visited mostly by small bees. The flowers contain 1 ovary with $15-90$ ovules in two layers, and $13-27$ anthers. Fruit set (the percent of flowers developing into fruits) is almost 100% in nature, in contrast to seed set per fruit (the percent of ovules developing into seeds), which ranges from 13 to 100% (Kang, unpublished). Fruits mature about three‚ weeks after anthesis, and contain small, arillate seeds (range of seed number per fruit is $1-73$).

The two study populations (Natick and Newton) were located in eastern Massachusetts, and separated by about 16 km. The Natick population was located along a forest trail in secondary forest in the Broadmoor Audubon Sanctuary, and consisted of three patches of plants separated by about 100 m. The Newton population occurred inside a young scrubby woods in the Newton-Webster Conservation Area. These two populations of *C. majus* differed in plant size (plant height and lateral spread) and density of plants; the plants in the Newton population were smaller, and more widely and evenly distributed. A total of 80 plants in Natick and 86 plants in Newton were selected at random for study.

Flowers were sampled from every tagged individual in each of the populations, by collecting one flower in each of the three successive weeks from mid May to early June, 1988. Concurrently, another flower within the same inflorescence was marked, and collected as a fruit about 3 weeks later. Thus, for each plant three flowers and three fruits were collected. In most cases, the first and second flowers in an inflorescence were sampled as flower and fruit, respectively. Since pollen grains were dispersed shortly after anthesis occurred, only recently opened flowers with undehisced anthers were collected. The flower diameter was measured at each sampling time in the field using calipers. The petal length was also measured for 30 randomly chosen flowers to determine the relationship between flower diameter and petal size. These two characters were highly correlated ($r = 0.78$, $P < 0.001$). Sampled flowers were preserved in FAA for later measurement.

The pistil length, ovule number per flower, filament length, anther length, and anther number per flower were determined under a dissecting microscope with an ocular micrometer (therefore, sexual allocation indicates relative allocation, not absolute allocation, throughout this paper). The total volume of anthers per flower was estimated by multiplying the volume of an anther ($(4/3) \times \pi \times$ (anther length$/2$)3) by the number of anthers per flower. It is assumed that this formula may well reflect the relative volume of anthers, although anthers may not be exactly sphere-shaped. Seed number and weight per fruit were determined for air-dried fruits. Seed set was calculated by dividing the number of seeds per fruit by the number of ovules per flower. Seed set was arcsine-transformed prior to analysis.

Analysis

To examine whether floral characters changed through time, a 2-way ANOVA was conducted for each character with population and sampling time as main factors (SAS G LM procedure, 1982). The significance level for the overall model of 2-way ANOVA on each character was adjusted by dividing alpha $= 0.05$ by 3 because such a model in fact represented three tests on the effects of population, sampling time, and 2-way interaction. When the interaction between population and sampling time was significant, a 1-way ANOVA with sampling time as a factor was performed for each character in each population. Following ANOVA, if necessary, the means of each character were tested to find which means were significantly different from each other (pairwise t-test) after a Bonferonni correction.

Principal Components Analysis (PCA) was performed on the measured floral characters to infer the functional relationship among floral characters. Anther number per flower was not included in the PCA because this variable was incorporated in total anther volume per flower. Based on this PCA pattern, the primary female character (ovule number per flower) and male characters (anther size and total anther volume) were inferred. These primary sexual characters were standardized for subsequent analyses, in order to avoid the problem that arises from utilizing different measurement units. For a comparison of temporal allocation to sexual structures during a season, new variables representing femaleness (SFEMALE) and maleness (SMALE) were generated. SFEMALE was the standardized ovule number per flower; the difference between the number of ovules per flower and the population mean was divided by the standard deviation. SMALE was a cluster variable which combined standardized values of anther size and total anther volume per flower. The standardization was performed on the combined data set of the two populations. RMALE (relative sexual allocation) was generated by subtracting SMALE from SFEMALE, i.e., RMALE $=$ SFEMALE$-$SMALE. In other words, RMALE is equal to 0 in the case of equivalent sexual allocation, and is negative in the case of higher male allocation. The 2-way ANOVA was performed to examine the effects of population and sampling time on RMALE. The type III sums of squares were used to test a signifi-

cance of main factor(s) in all of ANOVAs in this study. Sampling time is a fixed effect, while population is a random effect.

The temporal relationship between seed traits and floral characters was analyzed using a canonical correlation analysis. In the canonical correlation analysis, the constants in the linear combinations of dependent variables and those of independent variables were estimated so as to maximize the eigen values (Marascuilo and Levin, 1983). If the linear combinations of dependent variables are indicated by Y1, Y2, etc., and those of independent variables are indicated by X1, X2, etc, the maximization of eigen value corresponds to the maximization of the correlation between pairs of covariates, e.g., X1 and Y1. The individual seed weight, seed number, and seed set per fruit were the dependent variables, and floral characters (flower diameter, ovule number per ovary, anther length, anther number and total anther volume per flower) were the independent variables. However, the canonical correlation analysis does not require a clear distinction between dependent and independent variables. All of the reproductive characters were standardized before an analysis for each sampling time in each population. When more than one pair of canonical covariates was significant, a type 1 error decision was made following Marascuilo and Levin (1983); the successive tests in the sequential procedure became more conservative. For example, in order to retain two pairs of covariates, the significance of the second pair of covariates was tested at alpha = 0.0167 level, and that of the first pair was tested at alpha = 0.0083 level.

RESULTS

Temporal variation of floral characters

All floral characters (except for anther number per flower) declined significantly in mean size as well as mean numbers toward the end of a season in both populations (Table 1). The pistil length declined over time, but in different amounts between sampling intervals in two populations (interaction in a 2-way ANOVA; $F_{2,474} = 4.96$, $P<0.01$). The mean pistil length was significantly shorter at successively later weeks (the Natick population) or was shorter at week 3 than at weeks 1 and 2 (the Newton population) (Table 1). Since the 2-way interaction between population and sampling time for the number of ovules per flower was barely not significant after adjustment of the alpha level ($F_{2,474} = 3.56$, $P = 0.029$), variation in ovule number was examined separately in each population. The number of ovules declined significantly only at the first interval (the Natick population) or at both intervals (the Newton population) (Table 1). This corresponded to a 13% and 24% reduction of ovule number in the Natick and Newton population, respectively, for a 3 week period.

The pattern of temporal decline in filament length differed between populations (2-way

Table 1. Temporal variation in size and number of floral characters over three sampling dates in *Chelidonium majus.* Sample size is 79 to 80 plants in Natick and 85 to 86 plants in Newton. Means and standard deviations are based on measured floral characters. Within each site and colum, different letters indicate significant differendes among means at $P<0.05$. The analysis was done separately for each population because 2−way interactions between population and sampling time were significant for several characters

	Flower diam (mm)[a]		Pistil len (mm)		Ovule no. /flower		Filament len (mm)		Anther len (mm)		Anther no. /flower		Total anther volume (mm³)	
	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD
Natick														
Wk 1	30.3ª(3.5)		9.31ª(1.08)		59.7ª(9.6)		6.78ª(0.80)		2.05ª(0.17)		19.1ª(2.6)		88.25ª(23.72)	
Wk 2	27.4ᵇ(3.0)		8.86ᵇ(1.09)		55.3ᵇ(9.9)		6.23ᵇ(0.80)		1.95ᵇ(0.17)		19.9ª(2.4)		79.46ᵇ(22.47)	
Wk 3	23.6ᶜ(2.5)		7.82ᶜ(1.01)		52.1ᵇ(8.8)		5.73ᵇ(0.70)		1.75ᶜ(0.20)		19.3ª(2.2)		55.97ᶜ(19.86)	
Newton														
Wk 1	27.8ª(2.8)		7.84ª(0.58)		54.9ª(9.1)		5.87ª(0.50)		1.92ª(0.14)		16.6ª(1.9)		62.63ª(15.49)	
Wk 2	24.9ᵇ(2.6)		7.75ª(0.73)		48.3ᵇ(7.6)		5.52ᵇ(0.53)		1.80ᵇ(0.14)		18.8ᵇ(1.8)		58.65ª (13.70)	
Wk 3	23.0ᶜ(2.7)		6.98ᵇ(0.85)		42.0ᶜ(6.9)		5.36ᵇ(0.58)		1.61ᶜ(0.14)		19.3ᵇ(2.5)		42.99ᵇ(13.08)	

a. Date from Kang and Primack(1991).

interaction; $F_{2,475} = 7.01$, $P<0.001$). The filament length declined significantly at both intervals (the Natick population) or declined only at the first interval (the Newton population) (Table 1). The temporal decline in anther length did not vary between populations ($F_{2,475} = 0.12$, $P\gg0.05$); mean anther length declined at each interval in both populations (Table 1). Therefore, the reduction in mean anther length during a season was similar between populations (15% and 16% in the Natick and Newton population, respectively).

The temporal variation in anther number per flower differed between populations (2-way interaction; $F_{2,475} = 12.30$, $P<0.001$). It also occurred almost in an opposite direction of other floral characters. In Natick, the number of anthers was somewhat higher in the middle of the season (week 2) rather than the earlier week (week 1), although it was not statistically significant (Table 1). In the Newton population, mean number of anthers increased significantly later in the season (weeks 2 and 3). A temporal change in mean total anther volume per flower differed between populations (2-way interaction; $F_{2,475} = 4.76$, $P<0.01$). Mean total anther volume per flower declined significantly at both intervals in the Natick population, while it declined significantly only after the second interval in the Newton population (Table 1). Overall, mean total anther volume declined about 37% (Natick population) and 31% (Newton population) toward the end of a season. The larger flowers in the Natick population tended to have larger floral characters and higher amounts of total anther volume than the smaller flowers in the Newton population across sampling times. The only exception was the number of anthers per flower which was higher in the Newton population later in the season.

Temporal variation in relative sexual allocation

When PCA was conducted on floral characters, a simple pattern emerged from the first three components after a varimax rotation (Table 2). Three criteria were satisfied by the extraction of three components; (1) when rotated, each component was distinguished by a unique group of characters that possessed extremely high loading scores, (2) the eigen values of these three components were greater than 1, and more than 91% of the total variance was accounted for by these components, and (3) the separation into three distinct components appeared to be biologically meaningful because it generated three clusters of characters, i.e., characters responsible for attractiveness, male function, and female function.

The first component accounted for 36% to 41% of the total variance in the two populations (Table 2). The second and third components explained the remaining variance. The first component appeared to be a function of characters such as flower diameter, filament length, and pistil length because the variance in these characters was associated mostly with the first component. Anther length and total anther volume per flower were the major, almost equally important, elements of the second component. For example, the second component accounted for at least 72.2% (the Natick population) and 71.4% (the Newton population) of variance in these characters. The third component accounted for the considerable amount of variance due mainly to the extremely high loading score of ovule number per flower. Therefore, the first component seemed to represent a general size component. The second component undoubtedly represented a male component, and the third represented a female component. Since the pattern of character clusters was quite similar in two populations, only characters highly loaded on the second

Table 2. Principal Components Analysis (PCA) for floral characters in two population. Loading scores were obtained after varimax rotation

Characters	Component 1		Component 2		Component 3	
	Natick	Newton	Natick	Newton	Natick	Newton
Flower diamter	0.816	0.710	0.381	0.337	0.270	0.500
Pistil length	0.800	0.745	0.363	0.320	0.364	0.481
Filament length	0.898	0.906	0.298	0.287	0.081	0.106
Anther length	0.404	0.351	0.850	0.845	0.268	0.328
Total anther volume / flower	0.361	0.303	0.878	0.877	0.244	0.303
Ovule number / flower	0.227	0.267	0.274	0.418	0.929	0.836
Eigen value	2.458	2.163	1.935	1.957	1.206	1.391
Varance(%)	40.96	36.11	32.26	32.61	20.11	23.19

and third components were standardized, and used as primary sexual characters through-
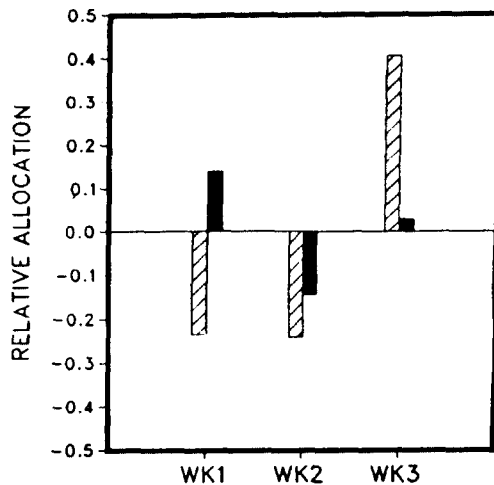out this paper (SMALE and SFEMALE, respectively; see Analysis).

Fig. 1. Temporal variation in relative sexual allo-
cation in the Natick(dashed bar) and
Newton(soild bar) population. The value of
zero for relative allocation indicates
equivalent allocation to female and male
structures, since relative allocation was
calculated by subtracting male allocation
from female allocation.

The temporal change in RMALE differed between populations (2-way interaction; $F_{2,474}$ = 9.73, P<0.001), although sampling time exerted significant effects on the relative allocation in both populations (1-way ANOVA: $F_{2,226}$ = 12.92, P<0.001 in the Natick population: $F_{2,248}$ = 4.28, P<0.05 in the Newton population). In the Natick population, RMALE was significantly lower at weeks 1 and 2 than at week 3 (P<0.001 for both pairwise comparisions), suggesting the higher primary male allocation earlier in the season (Fig. 1). In the Newton population, RMALE was significantly lower at week 2 than at week 1 (P<0.01).

Seasonal relationship between floral allocation and seed yield traits

The canonical correlation analysis showed that the set of predictor variables (floral characters) had significant impacts on the set of dependent variables. Also, the overall temporal pattern was similar between populations. In both populations, two pairs of canonical variates were significant at weeks 1 and 2, while a single pair was significant at week 3 (Tables 3a, b). The correlations between the first canonical variate (Y1) and dependent variables consistently loaded most heavily on seed number per fruit. The first canonical variate (X1) for the predictor set was most highly correlated with ovule number per flower throughout the season. Its correlation coefficient was always greater than 0. 97, indicating that there was little change in terms of contribution of ovule number to X1. On the other hand, flower diameter and some male characters such as anther size were also correlated considerably with X1 (Tables 3a, b).

The second pair of canonical coveriates explained a significant amount of variance (close to or more than 20%) in the system. The second pairs of canonical variates were

Table 3a. Correlation coefficients between the significant canonical variates and the original vaiables measured in the Natick population. Floral characters and seed yield traits were the independent and dependent variables, respectively. Since the standardized canonical coefficients(pattern matrix) and correlation coefficients (structure matrix) showed a similar pattern in terms of loadings with coveriates, the latter was reported

Characters	Wk 1				Wk 2				Wk 3	
	X1	Y1	X2	Y2	X1	Y1	X2	Y2	X1	Y1
Flower diameter	-0.427		0.739		-0.514		-0.733		-0.524	
Anther length	-0.560		0.533		-0.496		-0.099		-0.461	
Total anther volume /flower	-0.487		0.791		-0.466		0.064		-0.429	
Anther number /flower	-0.037		0.696		-0.164		0.115		-0.004	
Ovule number /flower	-0.998		0.021		-0.993		0.085		-0.997	
Seed number /fruit		-0.506		0.277		-0.465		-0.372		-0.414
Mean seed weight		-0.060		0.969		-0.424		-0.865		0.056
Seed set /fruit		0.347		0.265		0.357		-0.452		0.030
Eigen value		6.026		0.331		7.290		0.252		4.087
Canonical correlation		0.926		0.499		0.938		0.449		0.896
Variance		85.77		24.90		87.94		20.16		80.34

Table 3b. Correlation coefficients between the significant canonical variates and the original variables measured in the Newton population

Characters	Wk 1				Wk 2				Wk 3	
	X1	Y1	X2	Y2	X1	Y1	X2	Y2	X1	Y1
Flower diameter	-0.448		-0.785		-0.582		0.516		-0.607	
Anther length	-0.543		-0.588		-0.503		0.756		-0.539	
Total anther volume /flower	-0.622		-0.551		-0.482		0.803		-0.583	
Anther number /flower	-0.245		-0.044		0.066		0.090		-0.362	
Ovule number /flower	-0.998		-0.037		-0.993		0.085		-0.979	
Seed number /fruit		-0.580		-0.486		-0.503		0.862		-0.466
Mean seed weight		-0.044		-0.780		0.114		-0.121		0.002
Seed set /fruit		0.484		-0.533		0.336		0.936		0.113
Eigen value		25.957		0.351		11.320		0.238		5.405
Canonical correlation		0.981		0.509		0.959		0.437		0.919
Variance		96.29		29.96		91.88		19.24		84.39

defined with somewhat different variables in the two populations. In the Natick population, Y2 was highly correlated with mean seed weight at weeks 1 and 2 (correlation coefficients > 0.86). In the Newton population, Y2 was highly correlated with mean seed weight at week 1, but with seed set and also with seed number per fruit at week 2. In contrast to X1, X2 was clearly defined by a high correlation with flower diameter and/or male characters in two populations.

Consequently, throughout the season, a highly significant canonical correlation of the first pair of covariates (X1 and Y1) existed due to the strong ties between ovule number per flower and seed number per fruit (Fig. 2). However, the contribution of seed number per fruit to Y1 decreased as the season proceeded, as shown by the lowered correlation coefficients from week 1 to week 3 (Tables 3a, b). The second canonical correlation appeared to represent the relationship between nonfemale characters and seed yield components, especially seed weight. In 3 out of 4 cases, Y2 was mainly represented by mean seed weight. Even at week 2 in the Natick population, the standardized canonical coefficients of male characters (in the pattern matrix) were extremely high (-2.014 and 2.574 for anther size and total anther volume per flower, respectively). This great differ-
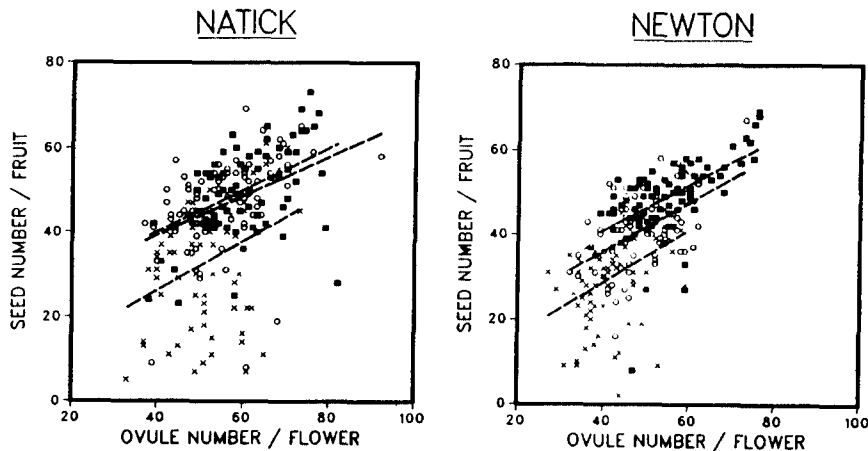

Fig. 2. Relationship between ovules per flower and seeds per fruit. ■, ○, and x indicate each sampling time(week 1, week 2 and week 3, respectively). Regression ovules on seeds with sampling time as a dummy variable showed that all lines were statistically perallel($F_{2,223} = 0.20$, $F_{2,245} = 0.13$, both P≪0.05 in Natick and Newton populations, respectively), but intercepts were not coincident($F_{4,223} = 14.37$, $F_{4,245} = 16.07$, both P<0.001 in Natick and Newton populations, respectively). The order of intercepts corresponds to successive sampling weeks in both populations. Adjusted mean number of seeds were significantly different between both weeks 1 and 2 and week 3(P<0.01 for both pairs of comparison) in the Natick population, between weeks 1 and 2 (P<0.05), and between both weeks 1 and 2 and week 3(P<0.01 for both pair) in the Newton population.

ence between the standardized canonical coefficients and correlation coefficients occurred due to the strong correlation among flower size and male characters.

DICUSSION

Seasonal variation of floral characters

The size and number of all floral characters (except for anther number per flower) decreased significantly toward the end of a season. This temporal variation occurred in conjunction with a reduction in flower diameter. These characters, however, did not decline in an identical pattern between two sampling intervals nor between populations. For example, only flower diameter and anther length declined at both intervals in both populations; the other characters examined declined over time but with different magnitudes between sampling times in two populations. This result indicates that floral characters including flower diameter are affected by seasonal change, but that the sensitivity of certain floral characters may be population-specific. Furthermore, concurrent temporal decline in most floral characters suggests that they are phenotypically positively correlated with flower diameter and with each other.

Only the number of anthers per flower tended to increase (the Natick population) or significantly increased (the Newton population) toward the end of the season. This unique temporal pattern of the number of anthers contributed greatly to temporal changes in total anther volume per flower which also involved variation in anther length. In the Natick population where about 19 anthers were produced per flower during a season, a drastic decline in anther size over time was reflected in corresponding patterns of reduction in mean total anther volume per flower. On the other hand, in the Newton population where flowers produced significantly higher number of anthers later in the season, mean total anther volume per flower declined significantly only after the second interval. If the amount of pollen grains per anther was proportional to the anther size, as in *Raphanus sativus* (Stanton and Preston, 1986), *Erythronium grandiflorum* (Harder and Thomson, 1989), and *Bromus tectorum* (McKone, 1989), the temporal decrease in anther size will result in successively lower production of male gametes toward the end of a season. However, the temporal decline of pollen production per anther was compensated by an opposite change in anther number per flower, which was conspicuous particularly in the Newton population.

A temporal increase in anther number per flower may function to buffer the reduction in size of individual anthers, thereby minimizing the reduction in pollen production per flower during a season. The number of anthers per flower may also work as an attractive unit in *C. majus,* and higher anther number may become more important in attracting pollinators toward the end of a season, especially since flower size declined significantly at

each sampling interval. This has not been tested even though there is evidence that pollinators prefer flowers with anthers to flowers without anthers (Coleman and Coleman, 1982; Solomon, 1987). The ambiguous role of anthers or pollen was also pointed out in *Aralia hispida* (Thomson *et al.*, 1989).

However, regardless of its effect, variability of floral characters itself may not have a selective value. Significant temporal changes in such stamen characters as anther size, the number of anthers, and the total anther volume suggests that most variation occurs within plants, not among plants, reflecting little potential of these characters to respond to natural selection. Alternatively, it may imply that genetic components for these characters have undergone strong directional selection. Yet significant differences in pollen productivity were shown among individuals of a *Lobelia* species (Devlin, 1988), a *Raphanus* species (Stanton and Preston, 1986), and between outcrossed progeny and selfed progeny in a selfing annual, *Scleranthus annuus* (Svensson, 1988).

In particular, within-individual variation in anther or petal number per flower is often explained as an indication of developmental disorder because it tends to occur during a senescent phase of plants (Huether, 1969; Ellstrand, 1983; Webb, 1984). Furthermore, the temporal pattern of stamen characters and ovule number per flower appears to be different among species (Bawa and Webb, 1983; more references in Thomson, 1989). For example, the temporal pattern in *C. majus* is quite different from that in a tropical tree, *Muntingia calabura,* where pistil length and anther number in a flower varied in an inverse relationship during a flowering period (Bawa and Webb, 1983). These diverse temporal patterns of variation among floral characters and among species suggest that the genetic and developmental basis of these characters may be quite complex and vary among species. Indeed, Shore and Barrett's (1990) study in *Turnera ulmifolia* revealed that different quantitative genetic models were required to explain variance of different floral structures.

Seasonal variation of sexual allocation

Sexual groupings of floral parts can be distinguished by an experimental manipulation on certain floral parts (e.g., Queller, 1983; Bell, 1985), or by careful observation of pollinator activity (Stanton et al., 1986). Alternatively, functional relationships among floral characters can be inferred using statistical tests such as a Principal Components Analysis (PCA), which has often resulted in interpretable sexual groupings of characters (e.g., Stanton and Preston, 1988). The results of this study using PCA suggest the three functional components of flowers. The "size component" was mainly defined by the three characters of flower diameter, pistil length, and filament length. Anther size and total anther volume per flower were almost equally important elements in the "maleness component". Like the relationship between filament length and anther size, ovule number

per flower behaved differently from the pistil length, the former belonging to the third component. It may be premature to conclude that these patterns of correlations among floral characters imply "selective correlations" (see Armbruster, 1988 ; Stanton and Galloway, 1990).   However, the clear and extremely similar groupings of characters in two populations indicates that the primary sexual structures (anthers and ovules) are to some degree independent of attractive structures and of each other even in this self-compatible species.

Relative sexual allocation varied significantly during a season in both populations, although the pattern of temporal variation differed somewhat between populations. Such variation occurred mainly through a rapid reduction of male allocation, especially total anther volume per flower, rather than female allocation, which was accompanied by a decline in flower diameter.   Flower size (or petal size) and male allocation are highly correlated within as well as among species along a gradient of the degree of selfing (Ornduff, 1969; Schoen, 1982; Wyatt, 1984; Cruden and Lyon, 1985).   However, it is not clear whether the seasonal decline in relative male allocation and flower diameter indicates the higher selfing rate at the end of a season.   Nor are clear causal relationships among floral characters.   For example, while surgical elimination of anthers resulted in reduced corolla size in *Glechoma hederacea* (Plack, 1957), anther number varied quite independently of flower diameter in *C. majus*. In any case, temporal variation in relative sexual allocation demonstrated the different degree of sensitivity of sexual structures within flowers.   The sensitivity of male allocation or sex change has also been noted by others (Solomon, 1985; Thompson, 1987; Devlin, 1988).   Variation in relative allocation to sexual structures within flowers in relation to a seasonal change or a change in flower size should deserve more attention.   This is because the sensitivity of sexual allocation to environmental conditions can drastically change the selective pressure acting on certain allocation patterns.   For example, plastic response of individual floral structures may reduce or buffer the effects of selection on sexual functions.

Relationship between sexual allocation and seed yield components

Allocations to attractive and sexual structures in flowers were correlated with seed size and number per fruit, suggesting that these two reproductive processes are not independent of each other.   The first pair of canonical covariates at each sampling time revealed a relationship between ovule number per flower and seed number per fruit. Thus, the allocation to a primary female structure was consistently a good predictor of seed number per fruit in both populations.   This reflects the parallel slopes of regression of seed number per fruit on ovule number per flower (Fig. 2).   Those persistently high slopes indicate relatively high values of mean seed set.   The self-compatibility of *C. majus* may be largely responsible for high seed set.

Surprisingly, the second pair of covariates existed only at earlier weeks and demonstrated a positive relationship of nonfemale characters - flower size and male characters - mainly with seed weight. From the viewpoint of female reproductive success, this relationship may involve the indirect effect of attractiveness on seed yield components through pollinator activity. While pollinators generally prefer larger flowers (e.g., Bell, 1985; Stanton and Preston, 1988; Young and Stanton, 1990a), seasonal changes in pollinator activity or mating patterns often occur (Thomson, 1982; Handel and Le Vie Mishkin, 1984; Pamler *et al*., 1988; Sampson *et al*., 1990). Furthermore, mating patterns affect seed weight; for example, outcrossed seeds tend to be heavier than selfed seed in several species (Galen *et al* ., 1985; Mitchell-Olds and Waller, 1985; Levin and Bulinska-Radomska, 1988). Mean weight of outcrossed seeds of *C. majus* was also slightly heavier than that of selfed seeds in a small sample (Kang, unpublished; 0. 699 mg vs. 0. 647 mg, n = 11 vs. 19 fruits from 8 plants). If reduction of flower size during a season is a rather common phenomenon (Bawa, pers. comm.), previous papers on temporal change of seed size (Wood *et al*., 1977; Cavers and Steel, 1984) and mating patterns may be also related to the temporal modification of floral attractiveness and sexual allocation within flowers. How the male reproductive success is affected by the decline of flower diameter was not directly examined in this study. However, attractiveness can contribute to both sexual functions (Willson and Price, 1977; Charlesworth and Charlesworth, 1987; Campbell, 1989).

In particular, male reproductive success is determined by the pollination mechanisms, not simply by the amount of pollen (Lloyd, 1987). In *C. majus,* the second pair of covariates which were highly loaded by male characters and flower diameter (X1) and by mean seed weight (Y2) existed only at earlier weeks. This may imply the reduced siring ability of male gametes in relation to a decrease in pollen quantity per flower later in the season. The decline in siring ability of pollen can also occur if the pollen quality such as competitive ability deteriorates over time during a season. Young and Stanton's study (1990b) showed that the plants grown in unfavorable conditions were not successful as male parents, although they did not control the quantity of pollen due to small sample size. Both of these results emphasize one of the problems in sexual selection theory which is primarily based on the heritability of sexual characters, for example, competitive ability of male parents and mate choice of female parents (Charlesworth, 1987). Again, however, partly due to the lack of information on causal relationships among floral structures and partly due to the lack of information on maternal and paternal genetic contribution to seeds in *C. majus,* it is difficult to conclude whether correlations between both attractiveness and sexual structures and seed yield components have significance for sexual selection. Nonetheless, these results demonstrate that seasonal variation in floral characters (including flower diameter) can contribute to changes in seed size and number which are major components of reproductive success.

CONCLUSION

Temporal variation in floral characters, including flower diameter, occurred in more complicated patterns than expected. Such temporal variation within flowers was correlated with different seed yield components during a season. Consequently, this may imply that male and female reproductive successes through seeds are affected not only by pollinator activity involved in plant size, density, and reward type and amount, but also by seasonal change in relative allocation relating to flowering phenology. The problems regarding the interpretation and measurement of sexual allocation in flowers have not been completely solved (e.g., Goldman and Willson, 1986). However, the results of this study indicate that flowering phenology is an important variable to be considered in sexual allocation studies, especially because temporal variation in the size and numbers of floral characters may be a common phenomenon in plants. Maternal resource allocation (e.g., Lloyd, 1980) as well as primary sexual allocation may be susceptible to environmental conditions. The sensitivity of various floral characters in response to environmental conditions should be examined in more number of species before one infers the adaptive significance of floral polymorphism because sensitivity of those characters can drastically change the effects of selection acting on certain allocation patterns.

적 요

Massachusetts에 위치한 자가수분하는 초본인 애기똥풀(*Chelidonium majus*: 양귀비과)의 두 집단에서 꽃의 내부구조들의 수와 크기에 있어서의 계절적 변화를 조사하였다. 꽃밥의 수를 제외한 모든 내부구조들이 3주의 조사기간동안 유의하게 감소하였다. 그러나 계절적 양상은 내부구조 사이에서, 집단 사이에서 같지 않았다. 이 결과는 내부기관들이 꽃의 직경과 공변이를 일으키고 있음을 시사하고 또한 내부구조들의 환경에 대한 반응을 예측하기 어렵다는 것을 보여준다. 꽃 직경과 내부기관들의 기능적 관계를 조사하기 위하여 Principal Components Analysis를 수행하였다. 첫 3 components가 각기 매개체(pollinator)의 유인요소(component of attractiveness), 자성요소(male component), 웅성요소(female component)로 해석되었다. 이 결과에 근거하여 꽃의 성적구조에 대한 상대적 자원분배를 조사하였다. 두 집단에서 개화 초기나 중기에는 자성구조에 대한 자원분배의 양이 컸다. 이런 상대적 자원분배의 양상은 종자생산과 무관하지 않았다: 개화기간중 각기 다른 꽃의 형질들의 조합이 종자의 양이나 크기에 상관되었다. 특히 유인구조와 자성구조의 형질들은 단지 개화기의 초기에만 종자의 평균무게와 정적으로 상관되었다. 이 결과들은 개화기가 성적분배 연구에 고려되어야 할 중요한 변인임을 보여주고 있다.

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