

**BOTTLENECK-INDUCED DISSOLUTION OF
SELF-INCOMPATIBILITY AND BREEDING SYSTEM
CONSEQUENCES IN *ASTER FURCATUS*
(ASTERACEAE)¹**

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Aster furcatus is a rare species with extremely limited genetic variation at isozyme loci. We utilized crossing experiments and seed set data obtained from natural populations to verify that there is also little allelic variation at the self-incompatibility (S) locus. Seed set in several populations was limited by a low number of S-alleles. Associated with a low number of S-alleles in populations was the dissolution of the incompatibility system, manifest by individual variation in self-compatibility, and by complex dominance relationships among S-alleles. Plant self-compatibility was correlated with mean number of ovules per inflorescence. Thus, self-compatibility appeared to be under partial environmental influence. Computer simulations revealed that the shapes of seed set distribution curves of modeled self-incompatible plant populations depend on the number of incompatibility alleles in the populations. By varying the number of S-alleles in modeled populations, we generated seed set distribution curves similar to those of natural populations. Genetic bottlenecks reduce the number of S-alleles and the proportion of compatible matings in populations of multiallelic self-incompatible species. Self-compatible genotypes are at a selective advantage in populations that lack a sufficient number of S-alleles to produce compatible crosses. *Aster furcatus* appears to be evolving self-compatibility as a result of bottleneck-induced losses of S-alleles.

The evolution of self-incompatibility (SI) and obligate outcrossing is considered to be an essential factor in the successful rise of angiosperms (Stebbins, 1950, 1957; De Nettancourt, 1977; Beach and Kress, 1980). Despite the increased heterosis expected as a result of outcrossing enforced by SI systems, the loss of self-incompatibility has occurred repeatedly in the flowering plants (Stebbins, 1950). Stable, self-compatible species exist throughout the angiosperms, and are believed to be derived from self-incompatible progenitors (Stebbins, 1957). In a historically outcrossing species, the progeny of self-pollinations would be expected to suffer the effects of severe inbreeding depression (Jain, 1976; Schemske, 1983). As inbreeding continues, it is expected that deleterious alleles will be purged from the population and the effects of inbreeding depression will diminish as particularly fit inbred strains are selected (Wright, 1969; Lande and Schemske, 1985).

In the majority of cases, evidence for the evolution of self-compatibility is post facto, supported mainly by the existence of derived, self-compatible species. Few studies have demonstrated the active dissolution of an SI system (e.g., Barrett, Morgan, and Husband, 1989). However, others have reported observations such as intraspecific polymorphism in levels of self-compatibility in normally incompatible genera (e.g., Berry and Calvo, 1989) that

seem to indicate active SI dissolution. Genetic models of self-fertilization and inbreeding depression have found predominant selfing and predominant outcrossing to be alternative stable states (Lande and Schemske, 1985). Plants with intermediate levels of selfing are thought to be in a state of flux (Schemske and Lande, 1985; Waller, 1986; but see Aide, 1986).

Uyenoyama (1986) grouped hypotheses to explain the evolution of selfing into negative and positive factors. The primary negative hypothesis is the reproductive assurance hypothesis (Jain, 1976), which suggests that selfing arises because of an unreliable supply of potential mates. Positive hypotheses include automatic selection for selfing genes because of a doubling of parent offspring relatedness and local adaptation allowed by reduced pollen flow (Antonovics, 1968).

We are interested in the evolutionary impact of population bottlenecks on species with multiallelic self-incompatibility as a special case of the evolution of self-compatibility to ensure reproduction. Bottlenecks cause losses of allelic variation at the SI locus. Because self-incompatibility systems require large numbers of incompatibility (S) alleles to maintain high levels of cross-compatibility, bottlenecks are expected to seriously reduce the potential for sexual reproduction in the bottlenecked population (Les, Reinartz, and Esselman, 1991). In such instances, we predict strong selection for the dissolution of SI and an increased production of seed by selfing. Generally, inbreeding coefficients are expected to be relatively high in small populations that are likely to experience biparental inbreeding regardless of the breeding system (Solbrig, 1976; Uyenoyama, 1986).

We previously documented the scarcity of isozymic genetic variability in *Aster furcatus*, a threatened species with sporophytic self-incompatibility (Les, Reinartz, and Esselman, 1991). Because preliminary indications were

¹ Received for publication 29 April 1993; revision accepted 19 October 1993.

The authors thank S. Krauskopf, R. Tucker-Adams, B. Leffin, L. Leitner, and L. Nelson for assistance with various aspects of the study; and J. Karron and K. Holsinger for helpful comments on earlier drafts of this manuscript. This work was partly supported by a grant from the Wisconsin Department of Transportation.

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that self-incompatibility was retained in the species, the lack of genetic variation was attributed to bottlenecks and drift. At that time we advocated a more intensive study of the breeding system of *A. furcatus* to ascertain the extent of self-compatibility in the species. Here we report the results of studies that test several hypotheses: 1) *A. furcatus* populations carry low numbers of S-alleles relative to other self-incompatible plants; 2) There is a positive relationship between the number of S-alleles and seed set in natural populations of the species; and 3) The self-incompatibility system of *A. furcatus* is breaking down due to bottleneck-induced losses of S-alleles.

MATERIALS AND METHODS

Reciprocal crossability—We carried out extensive crossing studies in one population (“Sheboygan Falls”) with numerous individuals, high levels of seed set, and for which we had prior evidence of multiple genets (Les, Reinartz, and Esselman, 1991). There were 13 spatially separated patches of *Aster furcatus* ramets in the field population. We assumed confinement of genets to single patches since it was unlikely that vegetative reproduction could result in the occurrence of a genet in more than one of the widely spaced patches. We electrophoresed leaves collected from several ramets in each patch and determined their genotype at the variable triphosphate isomerase (TPI) locus (Les, Reinartz, and Esselman, 1991). A minimum number of distinct genets in the population was determined by selecting one ramet of each unique TPI genotype in each patch (a maximum of three genets per patch since the TPI locus had two alleles and three genotypes). By this method, we identified a minimum of 24 different genets in the population.

From three to eight connected ramets of each of the 24 genets were transplanted to an experimental area at the University of Wisconsin-Milwaukee Field Station. The 24 genets were spaced widely (20-m minimum distance between genets) to preclude the possibility of wind transfer of pollen, and surrounded by wire cages supporting 1-mm fiberglass mesh screening, which completely enclosed the plants and excluded insect pollinators. We attempted to make all reciprocal crosses and self-pollinations between the 24 genets using three to nine heads for each cross. Two genets (2A-1 and 11-1) could not be used as pollen recipients in the crosses because an insufficient number of heads were in flower when the other plants were at anthesis. Twenty other crosses were not made or not harvested due to either the unavailability of pollen donor heads or loss of the pollinated heads before harvest. Altogether, 510 pollinations of multiple heads were performed (Table 2). Pollinations made and harvested numbered: 22 self pollinations, 432 reciprocal cross pollinations, and 56 nonreciprocal crosses. Entire heads were pollinated by gently rubbing a removed pollen donor head at anthesis on a female recipient head with receptive stigmas. This is the standard method for hand pollinating the Asteraceae. Some self pollen was undoubtedly transferred to stigmas at the same time that cross pollen was applied by this method.

All pollinations were made between 25 July and 26 August 1990. Heads were collected when seeds were ripe, just prior to shattering (between 1 September and 22 Oc-

tober). All dates of pollinations and seed ripening were recorded. We stored the heads in paper bags until we could count the developed achenes. Developed achenes were easily identified by a plump, three-dimensional cross section, whereas unfilled seeds were flattened. Number of filled achenes was recorded for each head. For each genet we determined the mean number of florets per head by counting the number of floret scars on a random sample of receptacles after the achenes had been removed. Seed set was expressed as number of developed achenes per floret according to the following formula: achenes per floret = number of developed achenes in all heads ÷ (number of heads × mean number of florets per head for the genet). We used Bateman's (1947) method to obtain a rough estimate of the number of S-alleles in a sample of plants using the total number of pairs of plants and the number that had at least one S-allele in common ($4 \div \text{number of S-alleles} = \text{number of plants sharing an S-allele} \div \text{total number of crosses}$).

We removed the screening around the individual genets prior to the 1991 growing season, allowing insects to pollinate the plants. All pollination in 1991 was self-pollination (geitonogamy), however, since the isolation of the genets and the large number of ramets and heads per genet restricted pollinator movements to single genets. The distance separating genets precluded the possibility of cross pollinations adding measurably to pollen deposition since we never observed pollinator movement directly from one genet to another. We harvested 20 randomly selected heads from each genet in 1991 to determine the degree of constancy between seasons in the selfing ability of the genets. Number of developed achenes per floret was calculated in the same manner as for the 1990 data.

Plants were not grown in a common garden but were widely spaced through an experimental area that varied considerably in microenvironment, especially soil type and moisture. We were not able to directly evaluate the quality of the environment of each genet or possible genetic components of plant vigor. To explore whether there was a specific effect of plant vigor on florets per head and selfing level, however, we classed the plants into various growth vigor categories. Based on overall size and growth rates of genets and ramets within genets we grouped all 24 genets into five categories: poor, poor to average, average, average to excellent, and excellent.

Seed set in natural populations—We collected seed heads from 13 natural populations of *Aster furcatus* in Wisconsin. An average of ten heads was collected from each of 20 spatially separated ramets in each population. We counted number of florets and number of developed achenes in each head and weighed all of the developed achenes produced by each ramet. Mean number of florets per head, seed set, and weight of developed achenes were calculated for each ramet. We ordered ramets from each population by seed set and examined the distribution of seed set proportions.

In order to interpret the effect of the number of S-alleles in a population on the distribution of seed sets, we constructed a computer simulation model of seed set in populations of sporophytic self-incompatible plants. The model was designed to determine the effect of number of S-alleles in the population on the shape of the seed set

TABLE 1. Mean number of developed achenes per floret in self- and cross-pollinations of 22 genets of *Aster furcatus* from one population. Achenes per floret is calculated for all cross-pollinations and for compatible crosses only.

	Pollen recipient genets																						
	1-1	3-1	4A-1	4A-2	5A-1	5B-1	6A-1	6B-1	7A-1	7A-2	8-1	9A-1	9C-1	9D-2	10A-1	12A-1	12B-1	12B-2	12C-1	12D-3	12D-5	13-1	
Self pollinations																							
1990	0.002	0.040	0	0.010	0.032	0.084	0.036	0.055	0.031	0.005	0.065	0.049	0.002	0.293	0.011	0.033	0.018	0.014	0.087	0.013	0.007	0.014	0.108
1991	0.078	0.045	0.159	—	0.219	0.108	0.059	0.085	0.004	0.025	0.085	0.024	0	0.243	0.012	0.121	0.047	0.055	—	0.084	0.041	0.108	
All cross pollinations																							
1990	0.014	0.150	0.001	0.021	0.048	0.122	0.265	0.120	0.073	0.016	0.199	0.067	0.021	0.203	0.030	0.148	0.067	0.053	0.120	0.259	0.143	0.175	
Compatible cross pollinations																							
1990	—	0.271	—	0.135	0.126	0.172	0.413	0.192	0.339	—	0.253	0.226	0.129	0.270	0.355	0.214	0.192	0.178	0.257	0.288	—	0.264	

distribution when plants were ordered by decreasing seed set. The model, written in Turbo Pascal, utilized a uniform grid of approximately 100,000 locations, and plants were randomly assigned locations in this grid. We chose a plant density (0.002 plants/grid unit) which could be held constant and which provided a range of seed sets while the number of S-alleles were varied. S-allele genotypes were determined by random assignment. Frequencies of S-alleles were held equal, and only heterozygous plants were modeled. Matings were considered compatible only between plants sharing neither S-allele. Pollen flow was made leptokurtic by modeling the proportion of seeds set due to a compatible pairing as a negative exponential function of the distance between the paired plants. The genotype of a target plant was compared with every other plant in the population, and if the pairing was compatible, the target plant's seed set was incremented as a function of the distance to the pollen donor plant. The proportion of seed set contributed by pollination from a compatible plant one distance unit away in the grid was held constant at 0.90. Plants in the model population were sorted in order of decreasing seed set in order to compare the distribution with that in natural populations.

RESULTS

Breeding system-self-compatibility—We found extensive variation in self-compatibility among the genets of the Sheboygan Falls population. Of the 22 genets for which data on self-compatibility were available for the 1990 experiment, one (9D-2) was fully self-compatible, having a selfing level (0.293 achenes per floret) as high as its level of outcrossed seed set in compatible crosses (0.270) (Table 1). Three others (5B-1, 8-1, 12C-1) were at least partially self-compatible with selfing levels exceeding 6% and between 25% and 50% of their compatible cross pollination seed sets (Table 1). The other 18 genets varied from 0 to 0.055 in their measured selfing ability. In 1991, six of 20 plants were self-compatible (selfing level > 10%; Table 1). In 1990, two of these six plants (5B-1 and 9D-2) were also self-compatible, but four appeared to be self-incompatible. Several plants were entirely self-incompatible but had high rates of seed set when crossed with specific, compatible mates, providing strong evidence for the presence of a functional self-incompatibility system.

Selfing level in both 1990 and 1991 was strongly correlated with the number of florets per head (Fig. 1). In 1990, 55% of the variation in selfing level among plants was accounted for by variation in the number of florets per head; this value reached 71% in our 1991 analysis. The correlation between florets per head and selfing level was highly significant calculated with or without the fully self-compatible plant, 9D-2. Since neither selfing level nor the number of florets per head stayed constant from 1990 to 1991, we transformed both variables to standard normal variates ([observation-mean]/standard deviation). We could then directly compare the difference in the number of florets per head for individual genets between 1990 and 1991 with the difference in the standardized selfing level. Plants that had a greater relative number of florets per head in 1991 than they did in 1990 tended also to have a higher standardized selfing level in 1991 than in 1990 (Fig. 2). An increase or decrease in the

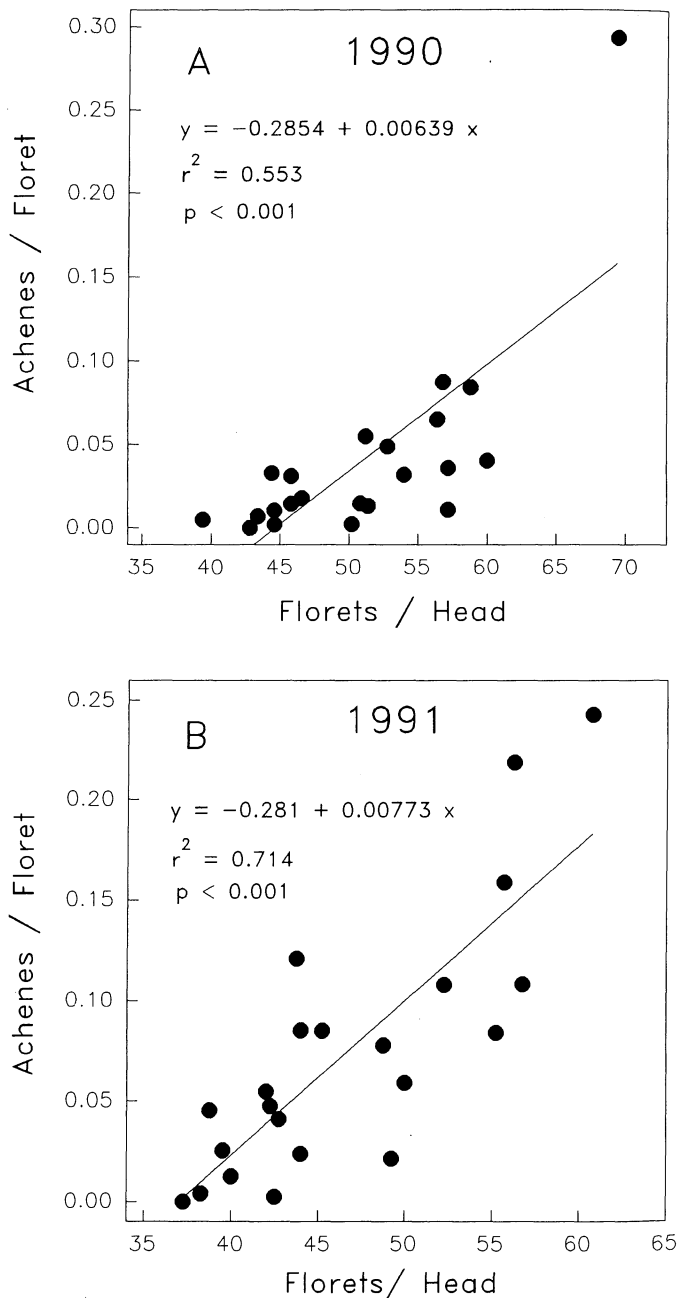


Fig. 1. Relationship of the mean number of developed achenes per floret produced by self-pollinations to the mean number of florets per head in 22 genets of *Aster furcatus* from one Wisconsin population. A. 1990 hand, self-pollinations. B. 1991 insect, self-pollinations.

number of florets per head was concomitant with proportional changes in the selfing level. We found no significant correlation between plant condition classes and either florets per head or selfing level. There was, however, a weak but significant positive correlation between plant condition class and the mean weight of developed achenes produced by the genet ($r^2 = 0.227$; $P = 0.025$).

Cross-compatibility—We examined compatibility in those 19 genets that were self-incompatible or had low self-compatibility in 1990 (Table 2). In this group of 19

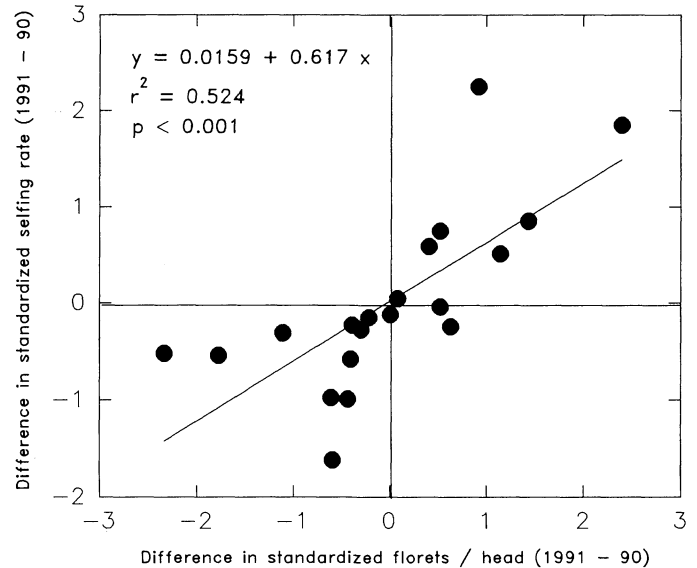


Fig. 2. Relationship between the difference in the standardized scores ($(X_i - \bar{X})/s$) of the number of florets per head from 1990 to 1991 and the difference in standardized scores of the number of developed achenes per self-pollinated floret from 1990 to 1991.

plants, individual genets acting as females (pollen recipients) were compatible with from 0 to 16 pollen donors. Among the 19 genets there are 342 possible reciprocal crosses (self-pollinations excluded). Of the 342 crosses, 93 were found to be compatible and 13 crosses were reciprocally compatible (compatible with either plant used as pollen donor or recipient). We therefore estimated that 158 of 171 pairs of plants (92%) shared at least one S-allele. Bateman's method for estimating number of S-alleles yields an estimate of 4.3 S-alleles (either four or five) in our sample. Because this estimate is likely to be biased upward, we conclude that there are probably as few as four S-alleles in our sample of 19 plants.

The small proportion of reciprocally compatible crosses suggests the existence of dominance relationships among the S-alleles of this sporophytic incompatibility system. More extensive variation in compatibility occurred among genets acting as pollen recipients (0 to 16 compatible crosses) than among genets acting as pollen donors (two to eight compatible crosses) (Fig. 3). This result suggests that most of the dominance of S-alleles is expressed in the stigma as opposed to in the pollen grain.

Outcrossing level per genet was positively correlated with number of florets per head, but the relationship was much weaker than between selfing level and number of florets per head. With the self-compatible plant (9D-2) removed, $r = 0.543$, $P = 0.011$, and 29% of the variation in outcrossing level was accounted for by the number of flowers per head.

Seed set in natural populations—There were highly significant differences among populations in number of flowers per head, seed set, and weight of developed achenes (Table 3). One population, Roehl Co. Park II, lost seed from heads before they were counted and was therefore excluded from the statistical analysis. Mean weight of developed achenes varied considerably among populations, ranging from about 0.7 mg to 1.3 mg.

TABLE 2. Developed achenes per 100 florets (% seed set) from reciprocal crosses of 19, mostly self-incompatible, genets of *Aster furcatus* from the Sheboygan Falls, Wisconsin population. Compatible crosses (defined as >10% seed set) are boxed; reciprocally compatible crosses are circled; dashes represent crosses that were not made or that were lost before harvest.

Pollen donor	Pollen recipient																		
	12D-3	12A-1	13-1	6A-1	6B-1	3-1	12C-1	12B-1	9A-1	5A-1	7A-1	12B-2	10A-1	9C-1	4A-2	12D-5	7A-2	4A-1	1-1
12D-3	X	26	9	60	1	2	49	5	22	1	2	2	36	0	2	1	3	0	0
12A-1	30	X	7	42	10	30	4	1	5	12	0	3	0	4	3	2	3	0	1
13-1	35	14	X	47	19	28	-	4	30	0	38	8	7	4	1	1	2	0	2
6A-1	47	20	3	X	3	35	-	1	7	17	2	1	1	6	0	2	2	0	0
6B-1	25	22	47	51	X	1	6	42	26	1	4	2	1	2	0	0	0	0	1
3-1	10	2	2	-	29	X	12	5	0	10	0	14	1	1	2	0	0	0	3
12C-1	44	26	3	42	29	9	X	14	0	6	37	3	0	2	0	8	1	0	1
12B-1	30	24	21	4	31	28	29	X	4	5	2	2	0	0	2	1	1	1	1
9A-1	48	18	28	21	11	30	-	22	X	1	1	14	0	4	3	5	1	0	2
5A-1	21	4	12	4	8	3	-	13	2	X	0	-	2	2	0	0	0	0	4
7A-1	4	8	14	3	22	17	-	2	5	10	X	2	2	0	2	0	0	0	6
12B-2	16	32	17	45	1	6	13	0	8	2	0	X	4	2	1	0	1	1	0
10A-1	20	14	33	27	28	6	21	1	22	1	1	26	X	2	1	1	0	0	0
9C-1	22	3	7	4	5	26	4	1	0	2	0	5	0	X	2	1	1	0	0
4A-2	6	20	19	57	12	1	6	12	2	2	0	1	1	0	X	0	1	0	3
12D-5	30	2	-	-	12	-	3	-	0	7	18	8	1	6	1	X	5	0	3
7A-2	30	3	5	5	10	30	6	0	1	7	8	0	0	0	2	0	X	0	0
4A-1	21	2	2	16	5	1	0	0	13	1	9	-	4	0	0	0	0	X	2
1-1	37	11	39	8	0	2	19	1	3	6	2	10	0	1	2	6	0	1	X

Four populations had seed sets of less than 1%, and in six populations seed set was less than 10%. There was no reason to suspect any lack of pollinator activity in these populations. These populations may lack the minimum number of S-alleles (four) to ensure compatible crosses in a sporophytic system. Two of these populations (Kletzsch Park and Roehl Co. Park I) have ramets in spatial distributions consistent with that of single large clones. When ramets were ranked in order of decreasing seed set, the distribution of seed set ranged from convex for those populations with high seed set, to concave in those with low mean seed set (Fig. 4). Our computer model generated seed set distributions ranging from concave to convex as the number of S-alleles in the population was varied from four to 20 (Fig. 5).

DISCUSSION

The rarity of *Aster furcatus* and the high probability of populations being founded by a small number of genets result in repeated genetic bottlenecks that are implicated in the origin of most extant populations (Les, Reinartz, and Esselman, 1991). There is direct evidence of the presence of a genetic incompatibility system in *Aster furcatus*. Sporophytic, multiallelic incompatibility systems are typical of the Asteraceae (Burt, 1977; Richards, 1986) and have been found in every species of *Aster* surveyed (Fryxell, 1957). The genotype of the pollen donor in our study has a dramatic effect on the seed set from crosses within single pollen recipients (Table 2). Several plants in our study were self-incompatible but had high rates of seed set when

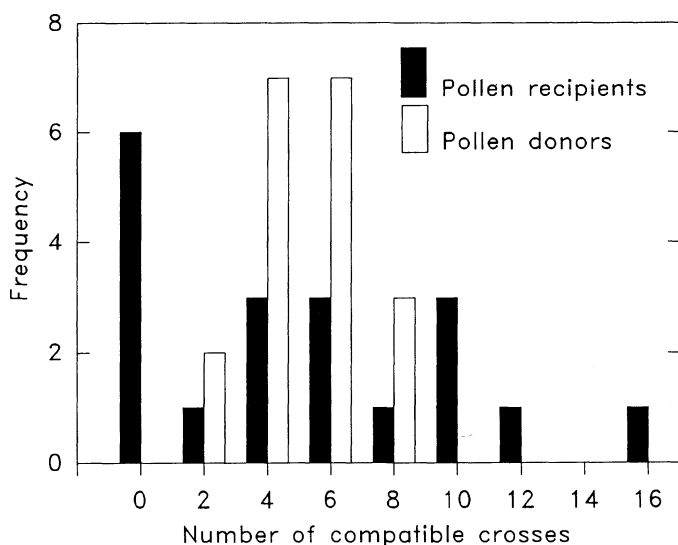


Fig. 3. Frequency distribution of the number of compatible crosses per plant among 19 individuals used either as pollen donors or recipients in one Wisconsin population of *Aster furcatus*.

crossed with specific compatible mates indicating a functional incompatibility system. Other plants were self-compatible. Imrie, Kirkman, and Ross (1972) modeled the loss of S-alleles from populations of a plant with the same breeding system as *A. furcatus* (sporophytic, multiallelic, monofactorial system with dominance series in the S-alleles). They found that genetic drift caused a rate of loss of S-alleles such that the maximum number that could be maintained in a population of 32 individuals was four. A population of 16 was unable to maintain the critical number of S-alleles. Our studies of the clonal structure and population biology of *A. furcatus* lead us to estimate that only six of the 14 known Wisconsin populations could have more than 30 genets, despite the fact that several populations number in the thousands of ramets (Reinartz and Les, unpublished data).

Loss of S-alleles through drift is probably slow in established populations of *A. furcatus* because most genets in populations are long-lived clones originally established during a disturbance event at the founding of the population. Loss of S-alleles is expected to be slow even in small, annual populations due to the tendency of rare alleles to increase toward equilibrium frequencies (Ewens, 1964; Ewens and Ewens, 1966). Perennation, however, reduces the equilibrium number of S-alleles and increases their rate of loss (Crosby, 1966). Sexual reproduction within populations is a rare event (Reinartz and Les, unpublished data). However, the loss of S-alleles resulting from severe bottlenecks during founding events substantially impairs seed set and thereby constitutes a strong selective force for self-compatibility.

We found a great deal of variability in the functioning of the sporophytic self-incompatibility system and in the level of self-compatibility among individuals in a population of *Aster furcatus*. Only a small proportion of genets in the studied population appeared to be fully self-compatible, but most individuals showed some level of partial self-compatibility. It is difficult to conduct detailed studies of the genetics of self-compatibility in sporophytic systems because mutations cannot be detected unless they are established in the entire plant, or at least, in an entire anther (de Nettancourt, 1977). In the only detailed genetic study of SI in the Asteraceae, Ronald and Ascher (1975) found that inheritance of self-compatibility in *Chrysanthemum morifolium* was associated with more than one major dominant gene unrelated to the S-locus, but involving a dependence on, or epistasis with, the S-genotype of the plant. Data from studies of kale have also shown that self-compatibility "factors" are inherited independently of the SI system (Thompson and Taylor, 1966).

Functional SI systems may involve several interactions among the S-alleles including independent action, partial or complete dominance, and mutual weakening (Sampson, 1960). Although sporophytic SI is usually a single locus system, it is frequently altered by modifiers resulting in incomplete dominance or mutual or independent weak-

TABLE 3. Seed production in natural populations of *Aster furcatus*.

Population	Flowers per head mean (SE) ^a	Seed set mean (SE) ^a	Seed weight (mg) mean (SE) ^a
Bachman Woods	32.0 (1.19)	0.056 (0.0130)	0.953 (0.0376)
Cambridge Avenue	36.5 (1.08)	0.324 (0.0373)	0.696 (0.0377)
Fond du Lac	37.6 (0.50)	0.008 (0.0010)	0.861 (0.0312)
Greendale Cemetery	36.5 (1.20)	0.428 (0.0519)	0.962 (0.0355)
Honey Creek	34.0 (1.54)	0.353 (0.0364)	0.955 (0.0365)
Jacobus Park	36.9 (0.92)	0.458 (0.0315)	0.788 (0.0292)
Jacobus II	33.3 (1.27)	0.526 (0.0373)	0.772 (0.0250)
Kletzsch Park	35.5 (0.85)	0.002 (0.0007)	1.317 (0.2483)
Lauderdale Lakes	32.9 (1.35)	0.009 (0.0033)	0.846 (0.0293)
Perkins Property	32.9 (1.63)	0.153 (0.0256)	1.130 (0.0434)
Riveredge Center	32.2 (0.91)	0.084 (0.0122)	0.807 (0.0331)
Roehl Co. Park I	35.6 (1.01)	0	—
Roehl Co. Park II ^b	25.4 (5.24)	0.209 (0.0718)	0.725 (0.0544)
Sheboygan Falls	38.0 (1.25)	0.457 (0.0266)	1.060 (0.0317)
<i>F</i> [12,234]	3.45	55.17	13.16
Significance	<i>P</i> < 0.001	<i>P</i> < 0.001	<i>P</i> < 0.001

^a *N* = 20 except Perkins Property = 15; Roehl Co. Park I = 12; Roehl Co. Park II = 4; Kletzsch Park = 6.

^b Some seed was lost from the Roehl Co. Park II heads before harvest. Since counts of florets per head were inaccurate, Roehl Co. Park II was excluded from ANOVA.

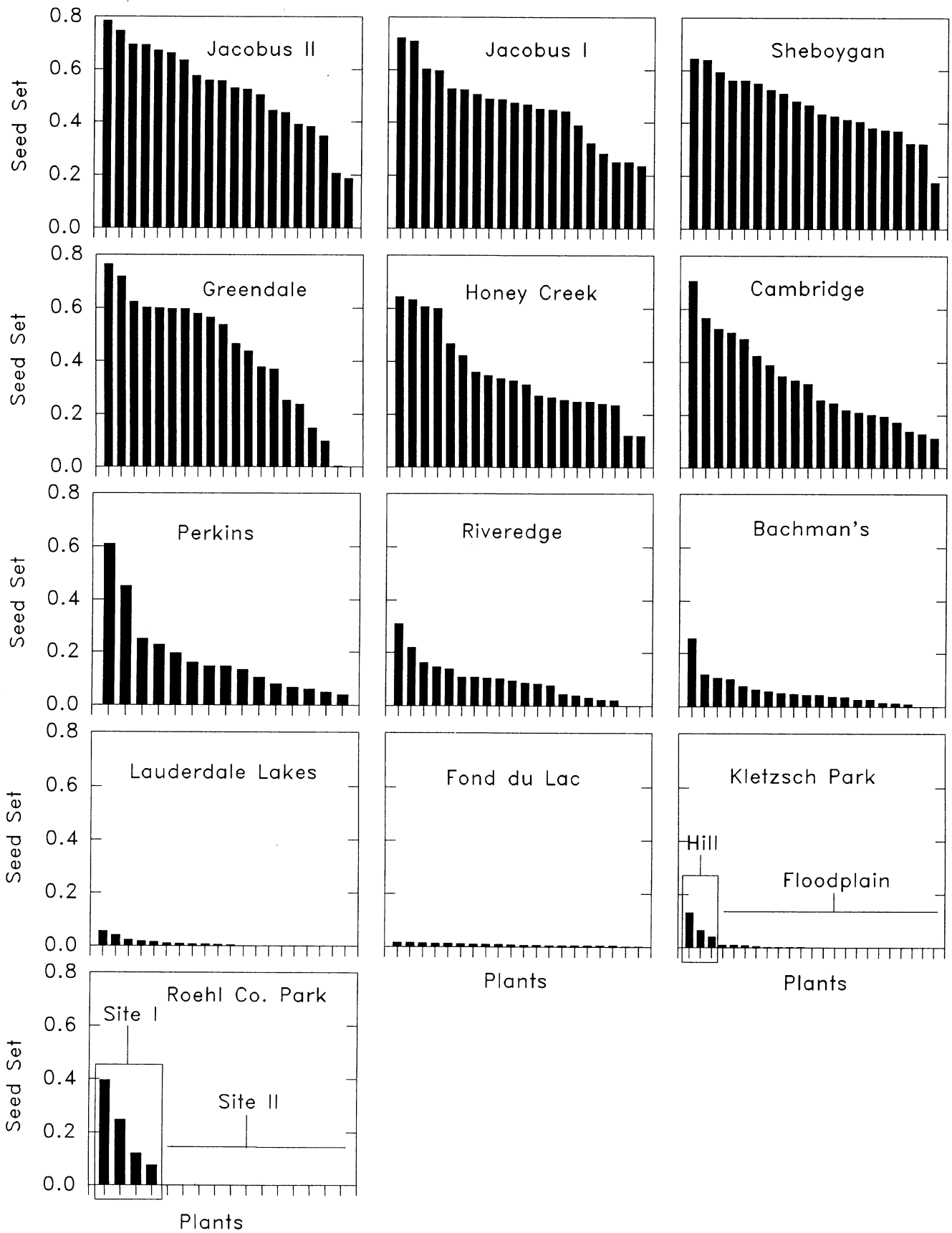


Fig. 4. Distribution of the mean proportion seed sets (developed achenes/floret) of 16 to 20 ramets from each of 13 natural populations of *Aster furcatus* in Wisconsin. The randomly sampled plants are ranked in order of decreasing seed set.

ening (Stevens and Kay, 1989). Selection for interaction of alleles at the S-locus is expected in small populations because of the large increase in the number of compatible matings conferred by these relationships when there is a decaying pool of S-alleles.

The large number of nonreciprocal compatibilities in our crossing experiment must be the result of dominance, interaction, or weakening relationships among the S-alleles in this population. Dominance factors at the SI locus have been reported in other Asteraceae including the genera *Carthamus*, *Chrysanthemum*, *Cosmos*, *Crepis*, and *Parthenium* (Bateman, 1954; Crowe, 1954; Imrie, Kirkman, and Ross, 1972; Ronald and Ascher, 1975). A high frequency of recessive S-alleles in populations has been found to be associated with a higher proportion of partly to fully self-compatible plants (Thompson and Taylor, 1966). Dominance increases the proportion of compatible crosses in sporophytic incompatible populations with a small number of S-alleles by raising levels of sib-compatibility (Knight and Rogers, 1955; Brewbaker, 1957). With four equally frequent S-alleles in a population and no recessive alleles, only 20% of crosses will be compatible (Ockendon, 1974). In this same example, cross-compatibility increases to 40% if one allele is recessive and to 60% if two alleles are recessive (Ockendon, 1974). Of 342 crosses with 19 self-incompatible *Aster furcatus* from the Sheboygan Falls population, 93 (27%) were compatible, but only 13 of 171 pairings (8%) were reciprocally compatible.

Our application of the Bateman method for estimating the number of S-alleles assumes that those crosses that were compatible, but not reciprocally compatible, produced seed because of dominance relationships despite sharing an S-allele. Bateman's method for estimating the total number of S-alleles in a sample from crossing data is a rough approximation and is, in a strict sense, inappropriate for sporophytic systems because they can violate two assumptions: 1) that there are no homozygotes for S-alleles present; and 2) that the S-allele frequencies are equal (O'Donnell and Lawrence, 1984; Stevens and Kay, 1989). However, violation of the assumption of no homozygotes can result only in an overestimate of the number of alleles; and unequal S-allele frequencies has only a small effect on the estimate when the number of alleles is low (O'Donnell and Lawrence, 1984). Assuming that only pairs of plants that are reciprocally compatible share no S-allele, we could use Bateman's method to obtain an estimate of the number of S-alleles in our sample of 19 plants. A rough calculation of the number of S-alleles in this sample of 19 plants (Bateman, 1947) yields an estimate of only four or five alleles. However, compatibility is increased by the presence of recessive alleles. Most of the S-allele recessiveness is probably expressed at the stigma as opposed to the pollen grain because plants exhibit much greater variance in compatibility as pollen recipients than they do as pollen donors.

Our previous estimate of self-compatibility in the Sheboygan Falls population of *Aster furcatus* based on a much smaller number of plants indicated less than 1% selfed seed set (Les, Reinartz, and Esselman, 1991). This, more detailed, study of the same population has revealed that there is extensive variation in self-compatibility with some individuals surpassing 29% selfed seed set (Table 1).

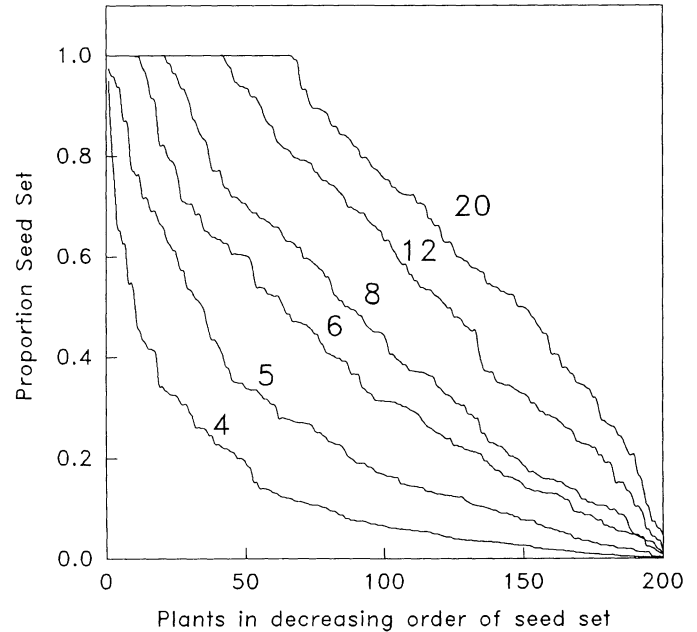


Fig. 5. Seed set distributions modeled for four to 20 S-alleles in a population of 200 sporophytic, self-incompatible plants. See text for a description of the model.

This study also revealed a strong nongenetic influence on self-compatibility in *Aster furcatus*. The proportion of florets developing into achenes after self pollinations was highly correlated with the number of florets per head. Differential allocation of resources within plants probably affects the relative amount of resources available to individual heads. In this way, resource availability could be positively correlated with both number of florets per head and also the degree of self-compatibility. The fact that number of florets per head and self-compatibility level changed in a correlated fashion over the 2 years of study (Fig. 2) is strong evidence for nongenetic influences on self-compatibility level.

We cannot entirely dismiss inbreeding depression as a partial explanation for the variation in self-compatibility among individuals of *Aster furcatus*. Krebs and Hancock (1991) have shown that the more inbred an individual of *Vaccinium corymbosum* the lower its fertility; and the more related two mates the lower the fertility of their cross. We have observed an apparent correlation between plant vigor (head size) and self-compatibility. However, the correlated changes in head size and self-compatibility observed over the 2 years of this study would not be expected if vigor and fertility were both related to inbreeding coefficient. If inbreeding depression was the primary cause of variation in self-fertility, we would not expect variation from year to year in a plant's selfing rate relative to other individuals in the populations; we would expect strong correlations between self- and outcross fertility (Krebs and Hancock, 1991). *Aster furcatus*, in our experiments, showed no significant correlation between selfed seed set and outcrossed seed set ($r^2 = 0.161$; $P > 0.05$).

Many plants have lower fecundity after self-pollination than after outcrossing, and these systems have been re-

ferred to as "leaky" self-incompatibility (Lovett-Doust and Lovett-Doust, 1988). In these systems, however, there is usually no evidence of a narrowly defined genetic incompatibility system. Genetic incompatibility systems are known to be "leaky" with respect to flower age and environmental factors such as humidity and temperature (Vaquero et al., 1989; Hinata and Nishio, 1980). These previous reports of environmentally induced breakdown of genetic SI systems involve relatively more proximate causes in the sense that self-compatibility is brought on by environmental factors directly affecting (often stressing) the stigmatic surface (de Nettancourt, 1977). Our observation of nongenetic lability of self-incompatibility in *Aster furcatus* is novel in that the "leakiness" of the SI system is correlated with a resource-associated, growth characteristic of the plant. In this system, there exists the possibility of an adaptive association between resource availability and selfing rate.

Natural Wisconsin populations of *Aster furcatus* vary greatly in their mean seed set (0 to over 45%). Of the 14 populations we examined, 30% to 40% had seed sets that suggested a lack of the minimum pool of S-alleles required to ensure successful compatible crosses. We expect selection for self-compatibility in these populations to be intense. Most of the sampled populations may have a very small number of S-alleles, thus providing strong selection for dominance relationships that would dramatically increase cross-compatibility. In two populations with less than 1% seed set (Kletzsch Park, Roehl Co. Park), small isolated patches consisting of only a few ramets showed fairly high seed set (Fig. 4). These small patches probably represent largely self-compatible plants.

Overall, populations with low seed set tended to have concave seed set distributions, whereas populations having high seed set had convex distributions. Our computer simulation verified that a difference in the number of S-alleles among populations is sufficient to explain the changes in the overall shape of the seed set distribution curve. Obviously, the effects of dominance, frequency of self-compatible individuals, occurrence of SI-locus homozygotes, and other factors in the population interact to ultimately determine the exact seed set distribution. The Sheboygan Falls population that we studied in some detail probably contains only four to five S-alleles, yet generates a convex seed set distribution with a relatively high seed set of 46%. Although our selection of genets for crossing studies may have led to an undersampling of S-alleles in the population, much of the seed set in the Sheboygan Falls population must be due to the self-compatibility and dominance factors that we observed therein.

Schemske and Lande (1985) observed that the distribution of outcrossing rates among natural populations of plant species is strongly bimodal, with few species possessing intermediate rates. *Aster furcatus* appears to be actively evolving self-compatibility because bottlenecks during population founding events cause most populations to have a very small number of S-alleles. Self-compatibility and recessive S-alleles are required in many populations to ensure even a modest seed set. The selective forces acting on *A. furcatus* may be common to most rare, self-incompatible plants (especially the Asteraceae), and we predict a high frequency of poorly functioning or nonfunctional incompatibility systems in this group of

plants. The evolution of self-compatibility will lead to increased levels of selfing and inbreeding. Species capable of surviving the shift toward inbreeding may represent a substantial portion of derived self-compatible plants in the angiosperms.

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