

# A Theoretical Investigation of the Evolution and Maintenance of Mirror-Image Flowers

Linley K. Jesson,<sup>1,\*</sup> Spencer C. H. Barrett,<sup>1</sup> and Troy Day<sup>2</sup>

1. Department of Botany, University of Toronto, 25 Willcocks Street, Toronto, Ontario M5S 3B2, Canada;

2. Department of Zoothelogy, University of Toronto, 25 Harbord Street, Toronto, Ontario M5S 3G5, Canada

Submitted January 17, 2002; Accepted November 14, 2002;  
Electronically published June 10, 2003

---

**ABSTRACT:** Enantiostyly is the deflection of the female sex organ either to the left or to the right side of the floral axis, resulting in mirror-image flowers. Two types of enantiostyly occur: in monomorphic enantiostyly, individuals exhibit both flower forms, whereas in dimorphic enantiostyly, the forms occur on separate plants. Monomorphic enantiostyly is known from at least 10 families, whereas dimorphic enantiostyly is reported in only three. Phylogenetic evidence suggests that monomorphic enantiostyly has evolved from a straight-styled ancestor and that dimorphic enantiostyly is derived from monomorphic enantiostyly. Here, we use theoretical models to investigate the role of pollen transfer in influencing these evolutionary transitions. We used numerical calculations to examine the evolution of monomorphic and dimorphic enantiostyly under different conditions of pollen transfer, inbreeding depression, and pollinator visitation. Our results demonstrate that in comparison to a putative straight-styled ancestor, both monomorphic and dimorphic enantiostyly function to reduce geitonogamous pollen transfer with a concomitant increase in pollen export. Our calculations suggest that the first stage in the evolution of monomorphic enantiostyly involves the deflection of the style only, followed by selection for reciprocity in anther position to promote more precise cross-pollination. Constraints associated with the developmental genetics of left-right asymmetries may account for the low number of evolutionary transitions from monomorphic to dimorphic enantiostyly, despite the evolutionary stability of this condition once it arises.

**Keywords:** enantiostyly, frequency-dependent selection, geitonogamy, plant sexual polymorphism, pollen transfer.

---

Many sexual polymorphisms in plants are maintained by negative frequency-dependent selection. Perhaps the best known example is heterostyly, where populations contain two (distyly) or three (tristyly) mating morphs differing in the reciprocal placement of stigmas and anthers (Darwin 1877; Barrett 1992). Sex-organ reciprocity is a mechanical device increasing the proficiency of cross-pollination as a result of segregated pollen deposition on different parts of the bodies of pollinators (Kohn and Barrett 1992; Lloyd and Webb 1992*a*, 1992*b*). Because of sex-organ reciprocity and the frequent occurrence of intramorph incompatibility, most mating in heterostylous species occurs between rather than within plants of different style morphs. This gives a mating advantage to the rare morph, increasing its frequency in populations.

Enantiostyly, the deflection of the female sex organ either to the left or the right side of a flower (Knuth 1906), is a plant sexual polymorphism for which the function is not well understood (Ornduff and Dulberger 1978; Graham and Barrett 1995; but see Jesson and Barrett 2002*c*). There are two distinct forms of mirror-image flowers in angiosperms (Barrett et al. 2000). In dimorphic enantiostyly, individuals are fixed for stylar direction and are either entirely left- or entirely right-styled and always have an anther deflected to the opposite side of the flower. In contrast, in monomorphic enantiostyly, individuals produce both left- and right-styled flowers, and both the style and an anther may be deflected to reciprocal positions (reciprocal monomorphic enantiostyly) or only the style may be deflected (nonreciprocal monomorphic enantiostyly). The main objective of this study is to use theoretical models to investigate the evolution and maintenance of these different forms of enantiostyly. Before we begin, however, we give a brief account of the principal features of the evolutionary biology of enantiostyly to provide the necessary conceptual background for model development.

Enantiostyly occurs in both the monocotyledons and dicotyledons, primarily in bee-pollinated species (Graham and Barrett 1995; Jesson 2002). Monomorphic enantiostyly is reported from at least 10 diverse families, including Pontederiaceae (*Heteranthera* and *Monochoria*; Graham

---

\* Corresponding author. Address for correspondence: School of Biological Sciences, Victoria University of Wellington, P.O. Box 600, Wellington, New Zealand; e-mail: linley.jesson@vuw.ac.nz.

and Barrett 1995), Haemodoraceae (*Dilatris* and *Schiekia*; Simpson 1990) and Tecophilaeaceae (*Cyanella*; Dulberger and Ornduff 1980) of the monocotyledons and Solanaceae (*Solanum*; Todd 1882; Bowers 1975), Caesalpiniaceae (*Cassia* and *Chamaecrista*; Todd 1882; Dulberger 1981), and Gesneriaceae (*Saintpaulia* and *Streptocarpus*; Harrison et al. 1999) of the dicotyledons. In contrast, dimorphic enantiostyly is exceedingly rare and is reported from only seven species in three monocotyledonous families: Haemodoraceae (*Wachendorfia* and *Barbaretta*; Ornduff 1974; Ornduff and Dulberger 1978), Pontederiaceae (*Heteranthera multiflora*; Jesson and Barrett 2002b); and Tecophilaeaceae (*Cyanella alba*; Dulberger and Ornduff 1980). This striking difference in the systematic distribution and frequency of occurrence between the two main forms of enantiostyly requires an evolutionary explanation.

Enantiostyly is commonly associated with a suite of floral traits including heteranthery, a division of labor into “feeding” anthers (from which pollinators collect pollen) and a reciprocally deflected, often large, cryptically colored “pollinating” anther, and nontubular, outwardly oriented, nectarless flowers (Dulberger 1981; Jesson 2002). The occurrence of these similar suites of traits in unrelated families suggests that enantiostyly has likely evolved as a result of convergent selective pressures associated with cross-pollination (Graham and Barrett 1995). Indeed, many researchers have suggested that enantiostyly promotes pollination between flowers of opposite type in a manner functionally analogous to heterostyly. Support for this hypothesis comes from two sources. First, observational and experimental evidence indicates that pollen removed from flowers of one style orientation is more likely to be deposited on stigmas of flowers with oppositely deflected styles (Bowers 1975; Wang et al. 1995; Jesson and Barrett 2002c). Second, surveys of morph ratios indicating equal ratios of left- and right-styled plants in natural populations of dimorphic enantiostylous species imply significant levels of intermorph mating (Ornduff 1974; Jesson and Barrett 2002a).

However, the cross-pollination hypothesis has been viewed as problematic for explaining the adaptive significance of monomorphic enantiostyly (Bowers 1975; Dulberger 1981; Fenster 1995; Graham and Barrett 1995). By having left- and right-styled flowers on the same plant, a pollinator visiting successive flowers could promote between-flower cross-pollination resulting in geitonogamous self-fertilization. Geitonogamy is generally viewed as a “nonadaptive” cost of large floral displays since it can result in inbreeding depression and losses in male fitness through pollen discounting (Lloyd 1992; Harder and Wilson 1998a). However, since geitonogamy and its associated costs are likely to be commonplace whenever species have large floral displays, adaptive explanations for the evolu-

tion of monomorphic enantiostyly require knowledge of the ancestral condition from which it evolved. Therefore, phylogenetic information on the sequences by which monomorphic and dimorphic enantiostyly may have developed are necessary for interpreting their evolutionary origins.

Phylogenetic evidence suggests that the most likely ancestral phenotype for monomorphic enantiostyly involves styles that are uniform in position and straight in direction with respect to the midplane of the flower. This morphology is probably the most common stylar condition in animal-pollinated species. For example, in *Solanum* a single clade (section *Androceras*) exhibits monomorphic enantiostyly, and all other sections, both more basal and derived, are either straight styled or have upwardly curved styles (Whalen 1984; Olmstead and Palmer 1997). Species with upwardly curved styles occur in two clades, both of which are more derived than section *Androceras*. Similarly, monomorphic enantiostyly in *Streptocarpus* and *Saintpaulia* (Gesneriaceae) appears to be derived from a straight-styled morphology (Harrison et al. 1999). Moreover, in this group reciprocal monomorphic enantiostyly is likely derived from the nonreciprocal condition in which the style is well deflected away from a central group of monomorphic stamens that exhibit no division of labor.

Dimorphic enantiostyly only occurs in families that also possess monomorphic enantiostyly. Phylogenetic reconstructions of the Haemodoraceae based on both morphological and molecular data (Simpson 1990; Hopper et al. 1999) indicate that dimorphic enantiostyly is most probably derived from monomorphic enantiostyly (and see Jesson 2002). Phylogenetic information is not available for the two other dimorphic enantiostylous species (Pontederiaceae—*H. multiflora*; Tecophilaeaceae—*C. alba*). However, their isolated occurrence in otherwise monomorphic enantiostylous clades, and the wide distribution of monomorphic enantiostyly both inside and outside of the families in which these two species occur, strongly suggests that dimorphic enantiostyly is derived from monomorphic enantiostyly in these two groups.

In light of this comparative evidence and as a means of motivating particular details of our models, we propose two main evolutionary scenarios for the origins of monomorphic and dimorphic enantiostyly. First, monomorphic enantiostyly has evolved from a straight-styled ancestor rather than one in which the style was already deflected. Reciprocal monomorphic enantiostyly could have evolved in a number of ways. A rare mutant could have arisen in a straight-styled population (possibly via an intermediate step involving a deflection of the style only), or there could have been gradual selection to increase stigma-anther separation (herkogamy), perhaps via selection on existing levels of asymmetry in the style and stamen (fluctuating

asymmetry). Second, we propose that dimorphic enantiostyly evolved from monomorphic enantiostyly. This could have occurred through two invasion events: the invasion of a mutant in which all styles are fixed for the same direction followed by the invasion of a mirror-image mutant or, alternatively, through disruptive selection on the proportion of left- and right-styled flowers on a plant in monomorphic enantiostyly populations. For reasons discussed more fully below, we assume in our models that monomorphic enantiostyly evolved through gradual selection on stigma-anther separation whereas the evolution of dimorphic enantiostyly occurred by the invasion of two mutants fixed for opposite directions of style deflection.

Our treatment of the evolution of enantiostyly uses numerical calculations to examine the two main evolutionary scenarios presented above: the invasion of a monomorphic enantiostyly plant into a straight-styled population and the subsequent invasion of dimorphic enantiostyly into a monomorphic enantiostyly population. To investigate the evolutionary stability of dimorphic enantiostyly, we also consider the reverse situation by examining the invasion of either a monomorphic enantiostyly mutant or a straight-styled mutant into a dimorphic enantiostyly population. These invasion scenarios are examined under different conditions, including changes in the precision of pollen transfer and different intensities of selfing and inbreeding depression. Following the presentation of results, we discuss their implications for the evolution and maintenance of enantiostyly, and we consider possible explanations as to why dimorphic enantiostyly is apparently so rare.

### The Selection of Enantiostyly

The appendix lists important parameters and their definitions for all models. Observations that bees visiting enantiostyly flowers pick up pollen on the sides of their bodies suggest that enantiostyly promotes pollen transfer between flowers of opposite style deflection (Bowers 1975; Jesson and Barrett 2002c). If we assume that in straight-styled plants pollen transfer can potentially occur between all flowers visited on the same inflorescence, while for enantiostyly plants pollen transfer occurs only between left- and right-styled flowers (and vice versa), then it is obvious that geitonogamy will always be lower for monomorphic enantiostyly, in comparison with the straight-styled condition, providing that pollinators visit equivalent numbers of flowers per inflorescence (and see Barrett et al. 2000, fig. 7). In fact, under the standard assumptions of previous pollen transfer models (de Jong et al. 1992; Iwasa et al. 1995; Harder and Wilson 1998b; see also “Gen-

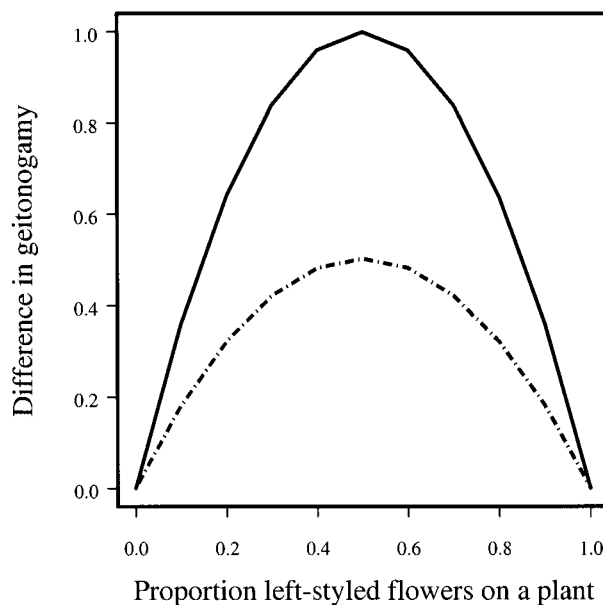
eral Features of the Numerical Calculations”), it can be shown that the amount of geitonogamy in a monomorphic enantiostyly plant relative to that of a straight-styled plant is

$$\frac{\bar{G}_T}{G_T^{NE}} = 2 \frac{v}{v-1} l(1-l), \quad (1)$$

where  $\bar{G}_T$  is the average total geitonogamy experienced by an enantiostyly plant,  $G_T^{NE}$  is the total geitonogamy experienced by a straight-styled plant,  $v$  is the number of flowers visited on an inflorescence, and  $l$  is the proportion of left-styled flowers on the plant (L. K. Jesson, S. C. H. Barrett, and T. Day, unpublished results). Thus, differences in geitonogamy are dependent on the number of flowers visited and the ratio of left- and right-styled flowers on an inflorescence of a monomorphic enantiostyly plant (fig. 1). If a plant is fixed for stylar direction (i.e., exhibits dimorphic enantiostyly) under these assumptions, the levels of geitonogamy should be close to zero.

### General Features of the Numerical Calculations

Although the intensity of geitonogamous pollen transfer is likely to vary among the three stylar conditions, it is



**Figure 1:** The relative amount of geitonogamy in a monomorphic enantiostyly plant compared with a straight-styled plant. The solid line indicates the difference in geitonogamy if two flowers are visited on a plant ( $v = 2$ ); the dashed line is  $v = 200$ . See appendix for definition of terms.

not intuitive how these differences translate in terms of fitness. We used numerical calculations involving pollen transfer within and between plants to examine contrasting scenarios for the evolution of enantiostyly. We assume in this model that all plants produce a similar mean number of pollen grains that are exported per flower ( $R$ ) as well as ovules per flower ( $o$ ). All ovules are fertilized, and self and outcross pollen compete equally for the same ovules. The fitness of a mutant individual ( $w^*$ ) is the sum of the number of ovules that are selfed ( $S^*$ , which contributes to both female and male fitness), the number of ovules that are outcrossed ( $O^*$ ), and the number of ovules of other plants that are fertilized by a given plant's pollen ( $M^*$ ). Selfed seeds incur some fixed cost ( $\delta$ ) because of inbreeding depression. The mean probability of survival of a zygote produced by self-fertilization is therefore  $(1 - \delta)$ . The fitness of the mutant individual ( $w^*$ ) is

$$w^* = O^* + 2S^*(1 - \delta) + M^*. \quad (2)$$

A pollinator visiting a mutant plant has  $A$  outcross pollen grains already on its body. When a pollinator visits a flower,  $R$  pollen is removed. A proportion of that pollen  $(1 - \pi)$  is lost as a result of the vagaries of the pollination process, and a proportion ( $\pi$ ) is deposited on the pollinator's body and is available for pollen transfer. At the same time, a proportion ( $\rho$ ) of the pollen that is on the pollinator's body is deposited on the stigma.

We calculated the number of ovules that are cross-fertilized on a plant ( $O^*$ ) and selfed ( $S^*$ ) as the proportion of outcross or self pollen grains deposited on the stigma of a flower times the total number of ovules in the flower. Selfing could occur through either geitonogamous or intrafloral self-pollination. We then summed the number of ovules cross- or self-fertilized over all flowers on a plant. We calculated the number of pollen grains deposited geitonogamously for each stylar condition differently (see below). Any pollen available for transfer on the pollinator's body that was not deposited on self stigmas was considered pollen exported from the plant.

To calculate male fitness, we assumed that the mutant plant invaded a population of size  $N$ . The proportion of all exported pollen coming from the mutant is given as

$$\frac{P^*}{(N - 1)P + P^*}, \quad (3)$$

where  $P^*$  is the number of pollen grains exported by the mutant plant and  $P$  is the number of pollen grains exported by a resident plant. We calculated the total number of ovules fertilized by pollen exported from the mutant plant ( $M$ ) as

$$\begin{aligned} M^* &= \frac{P^*}{P(N - 1) + P^*} \hat{o}(N - 1), \\ &\stackrel{N \rightarrow \infty}{=} \frac{P^*}{P} \hat{o}, \end{aligned} \quad (4)$$

where  $\hat{o}$  is the number of ovules available to be outcrossed (i.e., not selfed through geitonogamy or intrafloral self-pollination). The fitness of plants in the resident population was calculated in a similar manner as above.

To obtain fitnesses of monomorphic enantiostylous plants (either as mutant or resident plants; see below), we ran numerical calculations. An inflorescence was simulated by randomly determining  $v$  flowers as either left or right styled. Estimates of geitonogamy, pollen export, and fitness were then calculated for a single visitation to this inflorescence. This was repeated 100 times, and the results were averaged.

#### *Evolution of Nonreciprocal and Reciprocal Monomorphic Enantiostyly*

Here, we consider the fate of a rare mutant with a slight deflection in sexual organs that arises in a resident population. Changes in the deflection of the sexual organs of a mutant will influence pollen transfer through both the receipt of pollen by the mutant plant and the donation of pollen to resident plants. We defined three parameters ( $\alpha$ ,  $\beta$ , and  $\gamma$ ) to reflect these changes. The parameter  $\alpha$  is the deflection of a plant's stigma position from the position of the resident plant's anther as measured by the proportional reduction in pollen transfer between the two. If  $\alpha = 0$ , it means that the stigma is in the same position as a resident plant's anther, and there is no reduction in pollen transfer. If  $\alpha = 1$ , there is a deflection of the style such that there is no pollen flow from the resident population to the mutant. In all situations, the resident population will have  $\alpha = 0$ . The parameter  $\beta$  is defined as the deflection of a plant's stigma position from its own anther position (again measured as the proportional reduction in pollen transfer). Thus, for a straight-styled plant,  $\beta = 0$ , and for a plant with a deflected style,  $\beta > 0$ . The third parameter,  $\gamma$ , is the deflection of the plant's anther position from the stylar position of a resident. We specified  $\alpha$ ,  $\beta$ , and  $\gamma$  for both the mutant and the resident plants.

We examined the fitness consequences of changes in deflection in three ways (table 1). In the first situation, a mutant had both the style and stamen reciprocally deflected either to the left or the right side (influencing  $\alpha$ ,  $\beta$ , and  $\gamma$ ). We next examined a situation where the rare

**Table 1:** Combinations of parameters used in numerical calculations for the evolution of monomorphic enantiostyly

Sex organ arrangement		$\alpha$	$\beta$	$\gamma$
Situation 1:				
Resident	Straight styled	0	0	0
Mutant	Both style and anther deflected	>0	$0 < \beta \leq \alpha$	>0
Situation 2:				
Resident	Straight styled	0	0	0
Mutant	Style deflected only	>0	$0 < \beta \leq \alpha$	0
Situation 3:				
Resident	Style deflected only	>0	>0	>0
Mutant	Both style and anther deflected	>0 or $\alpha = 0^a$	>0 or $\beta = 0^a$	>0 or $\gamma = 0^a$

Note:  $\alpha$  is the deflection of a plant's stigma position from the position of the resident plant's anther,  $\beta$  is the deflection of a plant's stigma position from its own anther position, and  $\gamma$  is the deflection of the plant's anther position from the stylar position of a resident as measured by the proportional reduction in pollen flow between the two.

<sup>a</sup> Depending on combination of floral forms.

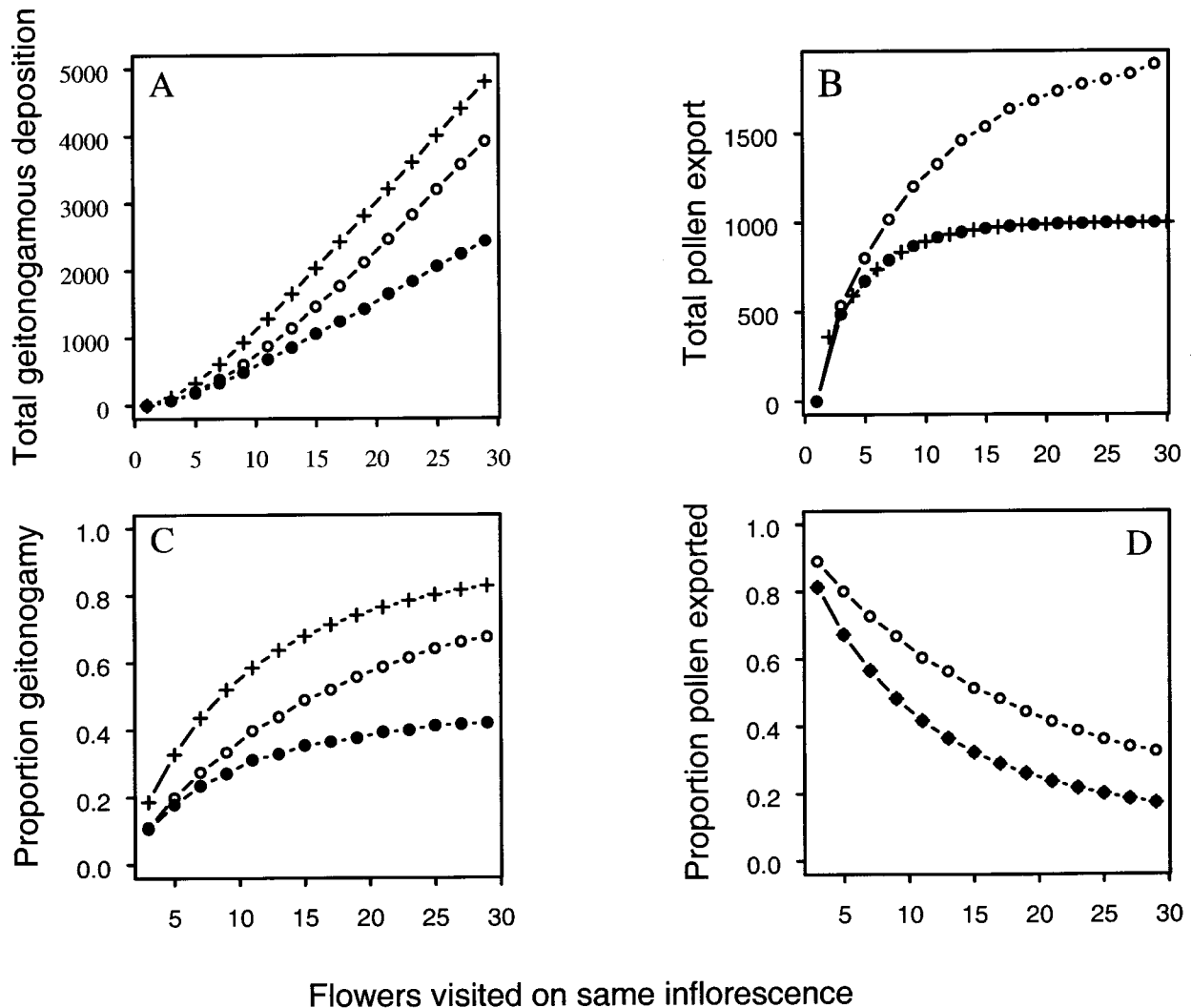
mutant had only the style deflected. In this situation, changes in stigma position influenced the amount of outcross and self pollen deposited on the stigma of the mutant ( $\alpha$  and  $\beta$ ), but there was no effect on the amount of pollen exported from the mutant that was deposited on outcross stigmas ( $\gamma$ ). In the third situation, we examined the fitness of a rare mutant with reciprocal monomorphic enantiostyly invading a population that had stylar deflection only. Plants in the resident population had lower pollen transfer ( $\alpha$  and  $\beta$ ) compared with the mutant plant because of the centrally placed anthers.

In calculating geitonogamy for a mutant with reciprocal monomorphic enantiostyly, we examined two scenarios of pollen deposition. In the first, every time a pollinator visited a flower, pollen was deposited on one side of the pollinator, and if pollen was present on the opposite side, a proportion was deposited from the pollinator to the stigma of the flower. This scenario assumes no loss of pollen from a pollinator's body except when that pollen comes in contact with a stigma. For example, if a pollinator visited five left-styled flowers in a row,  $\pi\rho R$  pollen grains would accumulate on the pollinator's body each time. This can be considered a high pollen export scenario. In the second scenario, pollen was assumed to be removed from a pollinator on every flower visited, either deposited on the stigma of the flower if it was the appropriate stylar orientation or lost from the flower if it was the inappropriate orientation. Thus, if a pollinator visited five left-styled flowers consecutively, the pollinator would lose  $\pi\rho$  pollen grains from the pool on the pollinator's body as well as accumulating  $\pi\rho R$  pollen grains. In this scenario, the pollen exported from the plant is identical to that of a straight-styled plant. This means that a pollinator visiting a sequence of all left-styled flowers would not accumulate pollen on its body, and therefore, this situation can be

considered a low pollen export scenario. The low pollen export scenario was not examined for nonreciprocal monomorphic enantiostyly because anther position was the same as for a straight-styled plant.

Figure 2 compares predicted levels of geitonogamy and pollen export on a straight-styled plant with these quantities for a reciprocal monomorphic enantiostylous plant, averaged over all possible combinations of left- and right-styled flowers visited. For a monomorphic enantiostylous plant, in the high pollen export scenario, pollen is only removed from a pollinator by a stigma of the appropriate orientation, leading to more pollen exported to other plants. In the low pollen export scenario, pollen is removed from the pollinator on every flower visited. Geitonogamous pollen deposition was highest for a straight-styled plant and lowest for a monomorphic enantiostylous plant where pollen export is low.

The relative amounts of geitonogamy and pollen export influenced the relative fitness of a rare, invading mutant (fig. 3). When a mutant with reciprocal deflection of both style and stamen arose in a straight-styled population, the mutant could not invade (fig. 3A, 3B). However, if only the style of the mutant was deflected, the mutant could invade, especially when large numbers of flowers were visited on a plant (fig. 3C, 3D). Although high levels of inbreeding depression influenced whether the mutant could invade, it did not influence the qualitative results (results not shown). A mutant that had a reciprocal deflection of style and stamen could invade a population in which the style was deflected, although the strength of selection decreased as more flowers were visited or if less pollen was exported (fig. 3E, 3F). The difficulty that a mutant with reciprocal monomorphic enantiostyly has in invading a straight-styled population suggests that reciprocal enan-



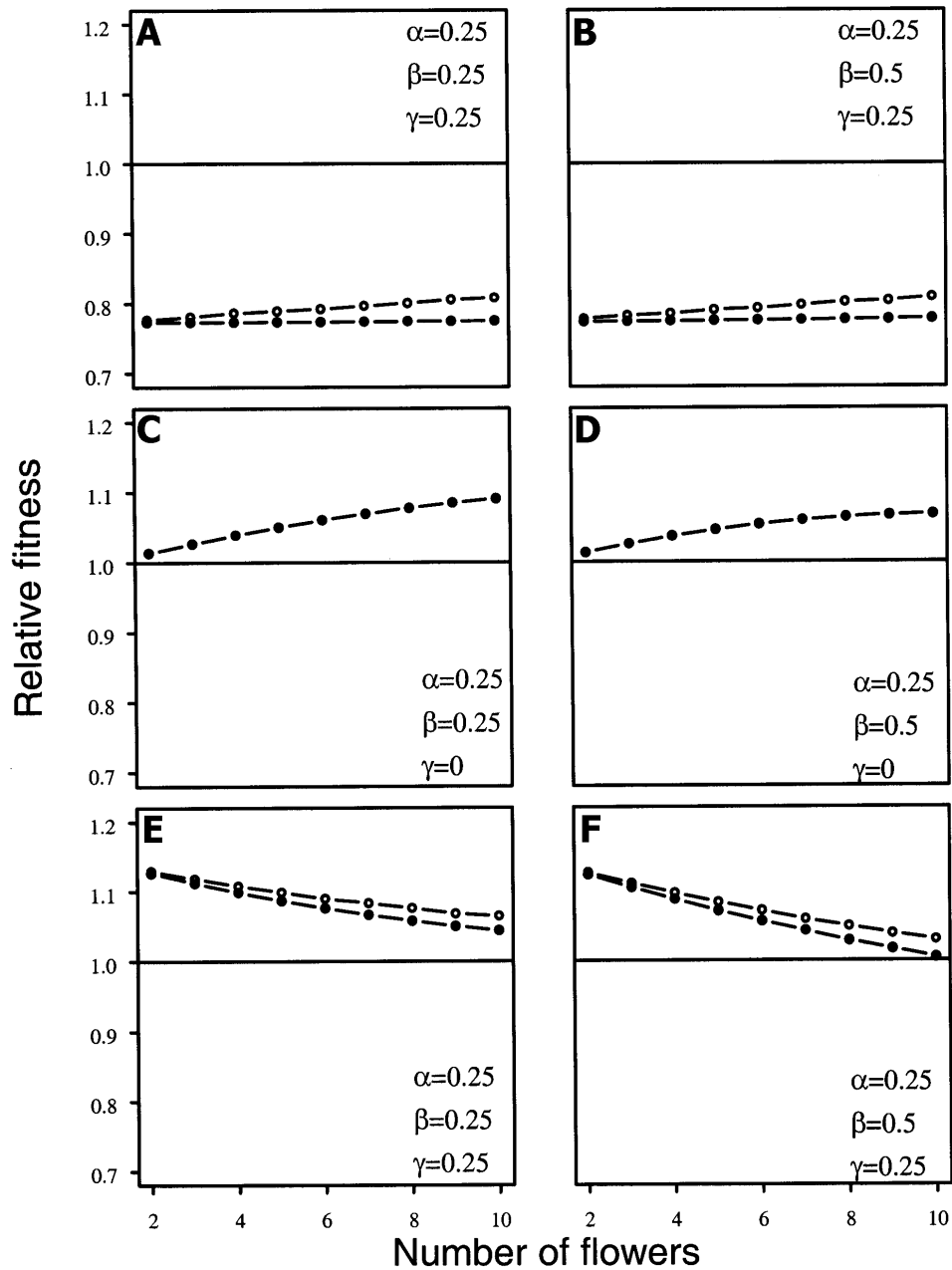
**Figure 2:** A, Average number of pollen grains received by a stigma that are deposited by geitonogamous pollination. B, Average number of pollen grains produced by a plant that are exported. C, Proportion of all pollen grains that are deposited by geitonogamous pollination. D, Proportion of all pollen grains produced by a plant that are exported. Three scenarios were modeled: a straight-styled plant (*crosses*), monomorphic enantiostyly with high pollen export (*open circles*), and monomorphic enantiostyly with low pollen export (*filled circles*). Values are means based on 100 simulations. Parameters used were  $\pi R = 100$  and  $\rho = 0.1$ . Standard errors (not shown) were  $<1\%$  of values for all calculations.

tiostyly is more likely to have arisen via nonreciprocal enantiostyly.

#### *Invasion of Plants Fixed for Styler Deflection*

We conducted numerical calculations of the evolution of dimorphic enantiostyly by modeling the case of a rare, left-styled mutant (which had an anther deflected to the opposite side of the flower) invading a population of

monomorphic enantiostylous plants. The same results apply if the mutant was right styled. A pollinator visiting a left-styled mutant in a monomorphic enantiostylous population would have two pools of outcross pollen on its body ( $A_1$  and  $A_r$ ). Only one of these pollen pools would be available to fertilize ovules of the mutant. Geitonogamous selfing of the mutant was considered to be zero, so ovules were fertilized either as a result of autonomous selfing or from outcrossing. As for the situation with

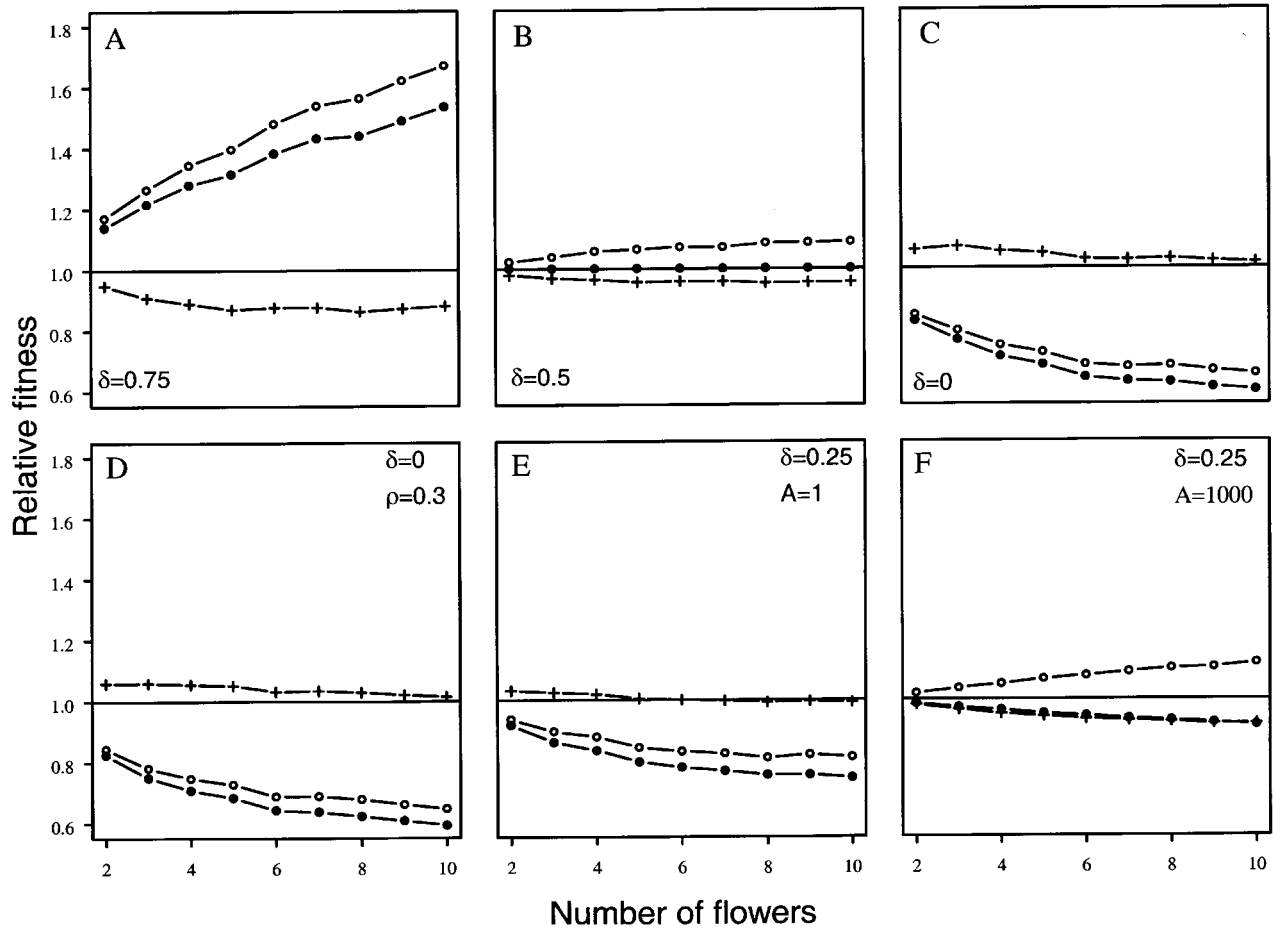


**Figure 3:** Fitness of a mutant enantiostylous individual relative to the fitness of an individual in the resident straight-styled population. *A* and *B* depict a situation where the mutant plant has a deflection of both style and anther. Changes in stigma-anther separation of the mutant ( $\alpha$ ,  $\beta$ , and  $\gamma$ ) will influence the amount of pollen deposited on a stigma and the amount of pollen deposited on outcrossed stigmas. *C* and *D* depict a situation where the mutant plant has deflection of the style only and will not influence subsequent pollen deposition on outcross stigmas ( $\gamma = 0$ ). *E* and *F* depict the invasion of a mutant with reciprocal monomorphic enantiostyly into a population of nonreciprocal enantiostylous plants. For *A–D*, values of  $\alpha$ ,  $\beta$ , and  $\gamma$  are those of the mutant plant; for *E* and *F*, values of  $\alpha$ ,  $\beta$ , and  $\gamma$  are those for the resident plants. All other plants have  $\alpha$ ,  $\beta$ , and  $\gamma = 0$ . Open circles are the fitness of a mutant with high pollen export; closed circles are the fitness of a mutant with low pollen export (not calculated for *C* and *D* because anther position of the mutant is the same as for straight-styled plants). If the relative fitness of the mutant is  $>1$ , then the mutant can invade. Unless otherwise stated, parameters are  $A = 10,000$ ,  $\pi R = 200$ ,  $\sigma = 30$ ,  $\delta = 0.5$ ,  $I = 0$ , and  $\rho = 0.2$ . Standard errors (not shown) were  $<1\%$  of values for all calculations.

monomorphic enantiostyly, we calculated pollen export for both dimorphic and monomorphic enantiostylous plants using both high and low export scenarios (see above). To examine conditions where monomorphic enantiostyly would be resistant to invasion, we also examined the invasion of a rare, straight-styled mutant into the resident population. Here, we considered both high and low pollen export scenarios and varied the parameters  $\alpha$  and  $\gamma$  to examine the effect of changes in pollen transfer between individuals in the population.

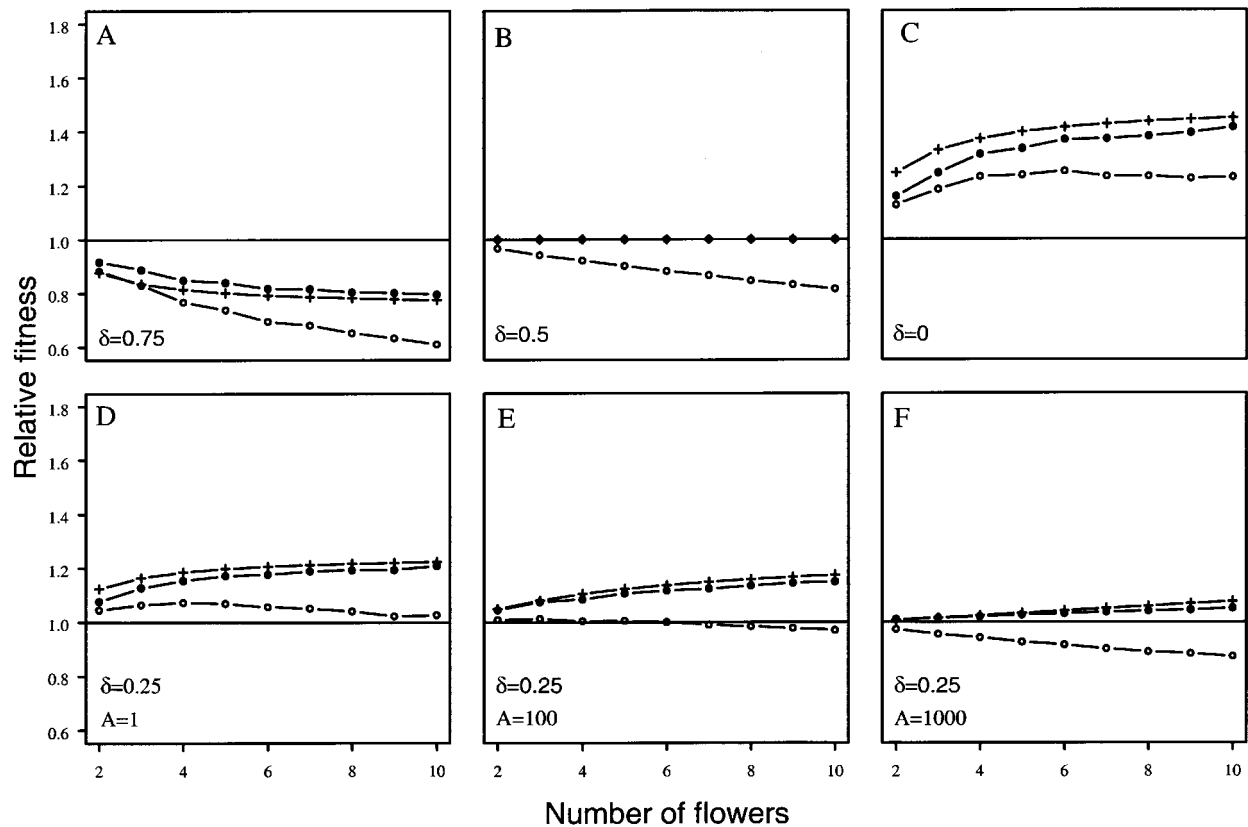
The invasion of a rare, left-styled mutant into a monomorphic enantiostylous population occurred under all pa-

rameter combinations examined when inbreeding depression was greater than 0.5 (e.g., fig. 4A, 4B). If inbreeding depression was less than 0.5, monomorphic enantiostyly was resistant to invasion by a left-styled mutant under most conditions, including variation in the proportion of pollen grains that are deposited onto a stigma (fig. 4C, 4D). If the number of outcross pollen grains on the pollinator's body was high, the left-styled mutant was able to invade if pollen export was high (fig. 4E, 4F). It was more difficult for a left-styled mutant to invade if pollen export was low than if it was high (fig. 4, closed circles vs. open circles). This indicates that most of the advantages to di-



**Figure 4:** Fitness of a rare, invading left-styled mutant relative to the fitness of an individual in the resident reciprocal monomorphic enantiostylous population. Open circles are the fitness of a mutant with high pollen export; closed circles are the fitness of a mutant and resident population with low pollen export. Crosses depict a straight-styled mutant invading a monomorphic enantiostylous population with low pollen export. A–C show effects of varying inbreeding depression ( $\delta = 0.75, 0.5,$  and  $0,$  respectively), D shows effects of varying the proportion of pollen grains deposited on a stigma ( $\rho$ ), and E and F show the influence of the number of outcross pollen grains on a pollinator's body ( $A$ ). Unless otherwise stated,  $A = 1, \pi R = 200, o = 30, I = 0,$  and  $\rho = 0.2$ . Standard errors (not shown) were  $<1\%$  of values for all calculations. For the straight-styled mutant, the parameters  $\alpha$  and  $\gamma$  were  $0.25$ .





**Figure 5:** Fitness of a rare, reciprocal monomorphic enantiostylous mutant relative to the fitness of an individual in the resident dimorphic enantiostylous population. Open circles are the fitness of a mutant with high pollen export; closed circles are the fitness of a mutant and resident population with low pollen export. Crosses depict a straight-styled mutant invading a dimorphic enantiostylous population with low pollen export. A–C show effects of varying inbreeding depression ( $\delta = 0.75, 0.5,$  and  $0$ , respectively). D–F show the influence of the number of outcross pollen grains on a pollinator's body ( $A = 1, 100,$  and  $1,000$  respectively). Unless stated,  $A = 100, \pi R = 200, o = 30, I = 0,$  and  $\rho = 0.2$ . For the straight-styled mutant, the parameters  $\alpha$  and  $\gamma$  were  $0.25$ . Standard errors (not shown) were  $<1\%$  of the value for all calculations.

morphic enantiostyly occur through a reduction in the cost of inbreeding depression and increased pollen export. Once a left-styled mutant has invaded, these advantages indicate that a mutant of opposite stylar deflection is also able to invade.

Monomorphic enantiostyly was stable against the invasion of either a left-styled or a straight-styled mutant under restricted conditions. Monomorphic enantiostyly could not be invaded by either mutant when there were high levels of pollen export from a plant, low inbreeding depression, and low numbers of outcrossed pollen grains on a pollinator. The strength of selection increased as the number of flowers visited also increased (fig. 4E). If pollen export from a monomorphic enantiostylous plant was low, a monomorphic enantiostylous population could always be invaded (results not shown).

#### *Evolutionary Stability of Dimorphic Enantiostyly*

To establish conditions for the evolutionary stability of dimorphic enantiostyly, we simulated the invasion of either a rare monomorphic enantiostylous mutant or a straight-styled mutant into a dimorphic population in which the ratio of left- and right-styled plants was  $1 : 1$ . In this situation, the monomorphic enantiostylous plant was potentially able to mate with all individuals in the population, whereas resident plants could only mate with half of the individuals. For the straight-styled mutant, we varied the parameters  $\alpha$  and  $\gamma$  to examine changes in pollen transfer between the mutant and other individuals in the population.

The results indicate that both a monomorphic enantiostylous and a straight-styled mutant can invade a di-

morphic enantiostylous population under certain conditions (fig. 5). For example, it was easier for either mutant to invade a dimorphic enantiostylous population when pollen export from the mutant was low (fig. 5, closed circles). In this situation, a mutant could not invade if inbreeding depression was greater than or equal to 0.5 (fig. 5A, 5B) but could invade as long as inbreeding depression was low (fig. 5C). In the high pollen export scenario, a mutant was able to invade only when inbreeding depression was low (fig. 5C) or if a small number of outcrossed pollen grains were present on a pollinator's body before visiting the mutant plant (fig. 5D, 5E). Invasion of the mutant was much more difficult if there were large numbers of outcrossed pollen grains on the pollinator's body (fig. 5F). There was no effect of changes in the parameters  $\alpha$  and  $\gamma$  on invasion of a straight-styled mutant (results not shown). It was much more difficult for a nonenantiostylous mutant to invade a dimorphic population if enantiostyly resulted in high pollen export (results not shown). These results indicate that in situations with high pollen export and high pollinator visitation, dimorphic enantiostyly is likely to be quite resistant to invasion.

### Discussion

Hermaphroditic flowering plants with large floral displays suffer from a fundamental conflict. Male and female sexual organs located in similar positions within a flower increase the precision of cross-pollen transfer, but this arrangement increases the likelihood of intrafloral and geitonogamous self-pollination (Lloyd and Webb 1986; Webb and Lloyd 1986; Harder and Wilson 1998a; Barrett 2002). Self-pollination can represent a major obstacle to plant fitness since it can lead to inbreeding depression (Charlesworth and Charlesworth 1987) and reduced siring success through male function (Harder and Barrett 1996). Thus, aspects of both the design of individual flowers and the manner in which they are displayed within inflorescences are likely to have been selected to minimize constraints associated with sexual interference between female and male function. The reciprocal placement of reproductive organs found in the plant sexual polymorphisms heterostyly and enantiostyly can be considered mechanisms that simultaneously maintain precise pollen transfer while reducing mating costs associated with self-pollination.

Like heterostyly, enantiostyly has long been considered a floral device promoting effective transfer of pollen between flowers of different style forms (Todd 1882; Webb and Lloyd 1986; Barrett et al. 2000). Models of the selection of heterostyly indicate that this polymorphism is likely to be evolutionarily stable as long as pollen transfer between

morphs is greater than pollen transfer within morphs (Charlesworth and Charlesworth 1979; Lloyd and Webb 1992b). Empirical evidence from natural populations indicates that heterostyly does indeed promote intermorph pollen transfer (reviewed in Lloyd and Webb 1992b). However, a fundamental distinction between heterostyly and enantiostyly is that in monomorphic enantiostyly, both forms of flowers occur on the same plant, a situation that never occurs in heterostylous species. This has led to suggestions that monomorphic enantiostyly might increase geitonogamy, thus potentially reducing cross-pollination (Bowers 1975; Dulberger 1981; Fenster 1995; Graham and Barrett 1995). Thus, any theoretical examination of the evolution of enantiostyly needs to consider this unique feature of the polymorphism and how "mixed versus fixed" flower forms on a plant influences pollination and mating.

Unlike earlier theoretical investigations of the selection of heterostyly (Charlesworth and Charlesworth 1979; Lloyd and Webb 1992b), we chose to specifically model both geitonogamous pollen transfer and pollen transfer to other plants in the population. This was necessary so we could explore the influence of monomorphic enantiostyly on both geitonogamy and pollen export. Our results demonstrate that in comparison to a putative straight-styled ancestor, both monomorphic and dimorphic enantiostyly function to both reduce geitonogamous pollen transfer and increase pollen export.

### *Male and Female Fitness in the Selection of Monomorphic and Dimorphic Enantiostyly*

Our models indicate that fitness differences between straight-styled monomorphic enantiostyly and dimorphic enantiostyly result from changes in both the relative levels of geitonogamy and pollen export. In numerical calculations of the evolution of monomorphic enantiostyly, a mutant was able to invade a straight-styled population when only the style was deflected, not both the anther and the style. In this situation, geitonogamy was lower than the straight-styled ancestor, and there was no loss of pollen transfer to other plants in the population. When changes in the deflection of an anther resulted in less pollen deposition on outcrossed stigmas, a monomorphic enantiostylous plant could not invade. This suggests that costs associated with a loss in male fitness represent a serious obstacle for the spread of monomorphic enantiostyly. In situations where stylar deflection led to an increase in pollen export, there was a fitness advantage at all levels of inbreeding depression (results not shown). The stylar deflection found in nonreciprocal monomorphic enantiostyly may have initially evolved as a mechanism to reduce

the intensity of self-pollination. Subsequent selection for precise pollen transfer may then have enabled the invasion of a mutant with a reciprocally deflected anther. Barrett et al. (2000) proposed that the function of all stylar polymorphisms in plants is to increase male fitness through more proficient pollen dispersal. The results of these numerical calculations support the hypothesis of an advantage through male function in the selection and maintenance of the forms of enantiostyly with sex-organ reciprocity.

The differences in geitonogamy and pollen export between straight-styled plants and the various enantiostylous mutants examined here are likely to be particularly important in species with multiflowered inflorescences. While occasional enantiostylous species in which one or a few flowers are produced each day do occur (e.g., *Cyanella alba*; Dulberger and Ornduff 1980), most enantiostylous species exhibit large floral displays (Jesson and Barrett 2002a; L. K. Jesson, unpublished data). Thus, by reducing levels of geitonogamous pollination, enantiostyly may allow plants to display more flowers simultaneously than could be achieved in straight-styled plants. Similar arguments have also been made in explaining the function of other floral strategies like dichogamy and heterostyly that decouple the benefit of large floral display from the mating costs associated with geitonogamy (Harder and Barrett 1996; Harder et al. 2000).

Pollen discounting is likely to have an important role in selection associated with the occurrence of geitonogamy. In our models, we examined two contrasting scenarios: high pollen export and low pollen export. Recall that in the first, pollen is removed from a pollinator only when it visits a flower of appropriate stylar direction, whereas in the second, pollen is removed at every flower, regardless of its stylar direction. In the high pollen export situation, enantiostyly influenced both female and male fitness, whereas in the low pollen export scenario, enantiostyly only benefits female fitness. Although the female fitness of the two scenarios differed to some degree, by far the greatest difference between these two situations resulted from the benefits that enantiostyly provided through male fitness.

The results from these numerical calculations also indicated that monomorphic enantiostylous populations suffer from a cost of geitonogamy and can be invaded by a mutant fixed for style deflection. This invasion is much more likely to occur if geitonogamy also leads to a reduction in pollen export (fig. 4, open circles) rather than through advantages to female fitness alone. If the levels of pollen exported by monomorphic versus dimorphic enantiostylous plants are the same (fig. 4, closed circles), dimorphic enantiostyly experiences higher fitness if inbreeding depression is greater than 0.5. This is because

selfed ovules contribute to both female and male fitness, and plants that experience selfing have a twofold fitness advantage in the absence of inbreeding depression (Fisher 1941). The advantages for a mutant with styles fixed in one direction are much more pronounced if there are also a large number of outcross pollen grains on a pollinator's body (fig. 4E, 4F). Thus, in these circumstances, the benefits for a mutant that is fixed for stylar deflection can come from both the female and/or male components of fitness.

On the basis of phylogenetic evidence, we assumed in our models that the ancestral condition for monomorphic enantiostyly is straight-styled and that dimorphic enantiostyly is derived from monomorphic enantiostyly. In common with earlier models of the evolution of heterostyly (Charlesworth and Charlesworth 1979; Lloyd and Webb 1992b), we also assumed that differences in the relative arrangements of sex organs of the phenotypes directly influenced pollen transfer between plants, an assumption that seems reasonable given the empirical evidence (Lloyd and Webb 1992b; and see below). It is possible that the ancestral condition was not completely straight-styled and that flowers also exhibited some stigma-anther separation. This would lower the values involving the parameters  $\alpha$ ,  $\beta$ , and  $\gamma$ , as well as the amount of self-pollination ( $I$ ), likely relaxing selection against geitonogamous pollination and self-fertilization but simultaneously reducing the amount of pollen exported to other stigmas. Selection for monomorphic enantiostyly will be strongest when pollen export is high (fig. 3, open circles), and hence it is likely that even in situations in which straight-styled plants are moderately herkogamous, monomorphic enantiostyly would still be able to invade.

One of the main assumptions of these models is that there is no pollen transfer between flowers of the same stylar orientation because of the segregated pollen pool on pollinator's bodies. While this is an ideal situation and therefore is unlikely to be found under natural conditions, some empirical evidence from enantiostylous species supports the assumption that pollen transfer between floral forms is greater than pollen transfer within forms (Bowers 1975; Wang et al. 1995). Moreover, a recent marker gene study of *Solanum rostratum* plants manipulated to be dimorphic enantiostylous revealed that ~75% of all outcrossed matings occurred between rather than within plants of opposite style orientation (Jesson and Barrett 2002c). We examined the importance of the segregated pollen pool assumption for geitonogamy in monomorphic enantiostyly and straight-styled plants (fig. 3; changes in  $\beta$ ). We found that as long as mating between flower forms is greater than mating within flower forms, geitonogamy will be lower in monomorphic enantiostylous plants (results not shown). Hence, we believe this simplifying as-

sumption does not change the qualitative conclusions of our models.

#### *Evolution of Dimorphic Enantiostyly*

We have demonstrated that a plant fixed for stylar deflection is able to invade a monomorphic enantiostylous population under a wide range of conditions; however, dimorphic enantiostyly is not always evolutionarily stable. For example, if pollen export is low and there is either weak inbreeding depression or few outcross pollen grains on a pollinator's body, then a monomorphic enantiostylous plant can invade a dimorphic population. In situations of low pollinator visitation and few outcross pollen grains on a pollinator's body, much of the female fitness of monomorphic enantiostylous plants comes from geitonogamous selfing. Moreover, when inbreeding depression is weak, the increased level of selfing that occurs in such plants further enhances their reproductive success because selfing is then a more profitable route for reproducing than is outcrossing (Fisher 1941).

These results illustrate that certain selective regimes preclude the evolutionary stability of dimorphic enantiostyly, and therefore this polymorphism need not be the expected long-term evolutionary outcome under all conditions. In such cases, our results reveal that instead, one of two possibilities can occur: either monomorphic enantiostyly or a straight-styled condition is evolutionarily stable. Although circumstances can be found for the occurrence of either of these outcomes, our results demonstrate that the latter situation is much more likely than the former. For most parameter values examined, if dimorphic enantiostyly is vulnerable to invasion by monomorphic enantiostyly, then it is also vulnerable to invasion by straight-styled plants (fig. 5). Additionally, these conditions tend to result in monomorphic enantiostylous populations being vulnerable to invasion by straight-styled plants (fig. 4). Thus, it appears that, under most conditions, we expect either the evolution of dimorphic enantiostyly or the evolution of plants with straight styles; it is rare for the intermediate form of monomorphic enantiostyly to be evolutionarily stable.

It is also worth noting that the conditions identified here as favoring plants with straight styles through the benefits of geitonogamous pollination would also likely be conducive to the evolution of other floral strategies not considered by us. As Lloyd (1992) has shown, geitonogamy can never be selected, and thus in such situations it is more likely that selection for autonomous selfing would occur. A plant with autonomous selfing does not require the services of a pollinator and thus benefits from repro-

ductive assurance (Lloyd 1992). In situations with low pollinator visitation and weak inbreeding depression, we found a monomorphic enantiostylous plant with high levels of intrafloral selfing always had higher fitness than a monomorphic enantiostylous plant with no selfing (results not shown). This suggests that when pollinator service is infrequent, a selfing variant would invade.

The above considerations suggest that populations currently displaying monomorphic enantiostyly remain vulnerable to invasion by a plant fixed for stylar deflection. Thus, the critical difficulty in establishing dimorphic enantiostyly might be related to developmental constraints. Major structural reorganization of inflorescence architecture would likely be required to produce a mutant with fixed stylar deflection from plants with monomorphic enantiostyly. The origin of one of these mutations, let alone two involving plants with opposite stylar deflection, may be exceedingly rare. Conversely, the origin of a monomorphic enantiostylous mutant in a dimorphic enantiostylous population may also be limited by similar constraints. Indeed, a survey of 54 populations of four species of *Wachendorfia* with dimorphic enantiostyly by Jesson and Barrett (2002a) failed to detect a single monomorphic enantiostylous individual, suggesting that, at least in this group, dimorphic enantiostyly is quite resistant to invasion by plants with both flower types. These results suggest that most populations currently displaying dimorphic enantiostyly are evolutionarily stable.

#### *Pathways for the Evolution of Floral Asymmetries*

Our results indicate that if a mutant with a deflection of both style and anther arises in a straight-styled population, it is not likely to invade because of losses in male fitness. However, if the mutant has a small deflection of the style only, it is more likely to invade. This suggests that the first stage in the evolution of enantiostyly is the deflection of a style alone, and only then does the reciprocal placement of an anther evolve, increasing the precision of cross-pollination. Phylogenetic evidence from Gesneriaceae is consistent with this pathway (Harrison et al. 1999). This situation resembles Lloyd and Webb's (1992b) model of the selection of heterostyly in which the first stage in the evolution of heterostyly involves the invasion of a mutant with altered stylar position into a resident population with uniform style length. Only later do subsequent changes to anther position occur through selection to increase the proficiency of cross-pollination.

Unlike this scenario for the evolution of monomorphic enantiostyly, our models of the invasion of dimorphic enantiostyly assume the spread of mutants in which one or

a few genes of large effect control the direction of deflection. It is also conceivable that dimorphic enantiostyly could evolve through selection on the proportion of left- or right-styled flowers in populations with monomorphic enantiostyly. As shown in figure 2, geitonogamy in a monomorphic enantiostylous plant is reduced as the proportion of left-styled flowers on a plant deviates from 0.5 and is at a minimum in a plant fixed for stylar deflection. Models examining selection on the proportion of left-styled (or right-styled) flowers on an inflorescence would likely yield similar qualitative results and indicate that when pollinator service is high, dimorphic enantiostyly has higher fitness than the monomorphic condition.

In most monomorphic enantiostylous species, the ratios of left- and right-styled flowers on an individual are consistently 1 : 1, implying that there may be little heritable variation in stylar direction (Barrett et al. 2000; Jesson et al. 2003). In dimorphic enantiostylous *Heteranthera multiflora*, left- and right-deflected plants are governed by a single diallelic Mendelian locus (Jesson and Barrett 2002b), supporting the scenario in which major gene mutants fixed for alternate stylar orientation invade monomorphic enantiostylous populations. Other evidence indicates that there may occasionally be selection on the ratio of left- and right-styled flowers in monomorphic enantiostylous species. For example, in *Heteranthera reniformis*, the ratios of left- and right-styled flowers on an inflorescence are highly skewed, with more left-styled flowers than right-styled flowers (S. C. H. Barrett, unpublished data). Also in *C. alba*, an apparently dimorphic enantiostylous species, Dulberger and Ornduff (1980) observed that approximately 10% of plants grown in glasshouse populations produced mixed inflorescences rather than producing flowers of uniform stylar orientation. It is not clear whether this variation is associated with the buildup or breakdown of dimorphic enantiostyly, but further study of this species may provide clues regarding the evolutionary pathways involved in the origin of dimorphic enantiostyly.

The evolution of dimorphic enantiostyly through two separate origins of plants fixed for styles in opposite directions seems likely to be rather infrequent, and this may

explain, in part, the extreme rarity of dimorphic enantiostyly in the angiosperms. The origin of dimorphic enantiostyly may also be further constrained by a lack of positional information determining left from right in the developing bud. Dimorphic enantiostyly may be more likely to originate in plant families with existing positional gradients related to asymmetry such as zygomorphy (Luo et al. 1999), or an established left-right gradient, rather than in families with radial symmetry. It is perhaps no coincidence that the three monocotyledonous groups in which dimorphic enantiostyly occurs are moderately zygomorphic, whereas monomorphic enantiostyly can occur in species with radially symmetric flowers.

Further work is needed to determine both the evolutionary and developmental mechanisms responsible for the origin of mirror-image flowers. Until then, our work on these curious floral polymorphisms represents the first attempt to use theoretical approaches to understand the evolution of enantiostyly. A final line of enquiry that could be profitable concerns possible analogies between monomorphic and dimorphic enantiostyly and the "mixed" and "pure strategies" of game theory. Although these strategies are often treated as equivalent ways of achieving the same evolutionary outcome, recent attempts have been made to identify general factors that might favor one condition over another (see Bergstrom and Godfrey-Smith 1998). It may be useful to explore similar ideas in the context of models for the evolution of monomorphic and dimorphic enantiostyly.

#### Acknowledgments

We thank L. Harder, D. Houle, R. Palmer, D. Schoen, and J. Thomson for helpful advice; two anonymous reviewers for valuable comments; and D. Lloyd for early encouragement. This research was funded by a Natural Sciences and Engineering Research Council research grant to S.C.H.B. and by student scholarships to L.K.J. from the Connaught Foundation of the University of Toronto and the Ontario government.

## APPENDIX

Table A1: Parameters used in models of the evolution of enantiostyly

	Definition
$\alpha$	The deflection of a plant's stigma position from the position of the resident plant's anther as measured by the proportional reduction in pollen transfer between the two
$\beta$	The deflection of a plant's stigma position from its own anther position measured as the proportional reduction in pollen transfer
$\delta$	The proportional reduction in survival of a zygote produced by self-fertilization
$\pi$	Proportion of pollen removed from a flower that is available for transfer
$\rho$	Proportion of pollen from the pollinator's body that is deposited on the stigma
$v$	Total number of flowers visited on a plant
$\gamma$	The deflection of the plant's anther position from the stigma position of a resident measured as the proportional reduction in pollen transfer
$A$	The number of outcross pollen grains on the pollinator's body before visiting the mutant plant. Subscripts l and r denote outcross pollen grains on the left and right sides of a pollinator's body, respectively
$I$	The number of self pollen grains deposited on a stigma from a flower's own anthers
$\bar{G}_T$	Average total geitonogamy experienced by an enantiostylous plant
$G_T^{NE}$	Total geitonogamous pollen deposited on all flowers on a straight-styled plant
$l$	Proportion of left-styled flowers on a plant
$M^*$	The number of ovules of other plants that are fertilized by a given plant's pollen
$O$	Mean number of ovules produced by a flower
$\hat{o}$	The number of ovules available to be outcrossed
$O^*$	The number of ovules produced by a mutant plant that are fertilized by outcross pollen
$N$	Size of resident population invaded by mutant plant
$P$	The number of pollen grains exported from a resident plant
$P^*$	The number of pollen grains exported from a mutant flower
$R$	The number of pollen grains that are removed from each flower
$S^*$	The number of ovules that are selfed (which contributes to both male and female fitness)
$w^*$	Fitness of a mutant individual

## Literature Cited

- Barrett, S. C. H., ed. 1992. Evolution and function of heterostyly. Springer, Berlin.
- . 2002. Sexual interference of the floral kind. *Heredity* 88:154–159.
- Barrett, S. C. H., L. K. Jesson, and A. M. Baker. 2000. The evolution and functional significance of stylar polymorphism in flowering plants. *Annals of Botany* 85(suppl. A):253–265.
- Bergstrom, C. T., and P. Godfrey-Smith. 1998. On the evolution of behavioural heterogeneity in individuals and populations. *Biology and Philosophy* 13:205–231.
- Bowers, K. 1975. The pollination ecology of *Solanum rostratum* (Solanaceae). *American Journal of Botany* 62: 633–638.
- Charlesworth, D., and B. Charlesworth. 1979. A model for evolution of distyly. *American Naturalist* 114:467–498.
- . 1987. Inbreeding depression and its evolutionary consequences. *Annual Review of Ecology and Systematics* 18:237–268.
- Darwin, C. 1877. The different forms of flowers on plants of the same species. J. Murray, London.
- de Jong, T., P. Kinkhamer, and M. Van Staaldunin. 1992. The consequences of pollination biology for selection of mass or extended blooming. *Functional Ecology* 6: 606–615.
- Dulberger, R. 1981. The floral biology of *Cassia didymobotrya* and *C. auriculata* (Caesalpinaceae). *American Journal of Botany* 68:1350–1360.
- Dulberger, R., and R. Ornduff. 1980. Floral morphology and reproductive biology of four species of *Cyanella* (Tecophilaeaceae). *New Phytologist* 86:45–56.
- Fenster, C. B. 1995. Mirror-image flowers and their effect on outcrossing rate in *Chamaecrista fasciculata* (Leguminosae). *American Journal of Botany* 82:46–50.
- Fisher, R. A. 1941. Average excess and average effect of a gene substitution. *Annals of Eugenics* 11:53–63.
- Graham, S. W., and S. C. H. Barrett. 1995. Phylogenetic systematics of the Pontederiales: implications for breeding-system evolution. Pages 415–441 in P. J. Rudall, P. J. Cribb, D. F. Cutler, and C. J. Humphries, eds. *Monocotyledons: systematics and evolution*. Royal Botanic Gardens, Kew.
- Harder, L. D., and S. C. H. Barrett. 1996. Pollen dispersal

- and mating patterns in animal pollinated plants. Pages 140–190 in D. G. Lloyd and S. C. H. Barrett, eds. *Floral biology: studies on floral evolution in animal-pollinated plants*. Chapman & Hall, New York.
- Harder, L. D., and W. G. Wilson. 1998a. A clarification of pollen discounting and its joint effects with inbreeding depression on mating system evolution. *American Naturalist* 152:684–695.
- . 1998b. Theoretical consequences of heterogeneous transport conditions for pollen dispersal by animals. *Ecology* 79:2789–2807.
- Harder, L. D., S. C. H. Barrett, and W. W. Cole. 2000. The mating consequences of sexual segregation within inflorescences of flowering plants. *Proceedings of the Royal Society of London Series B, Biological Sciences* 267:315–320.
- Harrison, C., M. Möller, and Q. C. B. Cronk. 1999. Evolution and development of floral diversity in *Streptocarpus* and *Saintpaulia*. *Annals of Botany* 84:49–60.
- Hopper, S. D., M. F. Fay, M. Rossetto, and M. W. Chase. 1999. A molecular phylogenetic analysis of the bloodroot and kangaroo paw family, Haemodoraceae: taxonomic, biogeographic and conservation implications. *Botanical Journal of the Linnean Society* 131:285–299.
- Iwasa, Y., T. J. de Jong, and P. G. L. Klinkhamer. 1995. Why pollinators visit only a fraction of the open flowers on a plant: the plant's point of view. *Journal of Evolutionary Biology* 8:439–453.
- Jesson, L. K. 2002. The evolution and functional significance of enantiostyly in flowering plants. Ph.D. thesis. University of Toronto.
- Jesson, L. K., and S. C. H. Barrett. 2002a. Enantiostyly in *Wachendorfia* (Haemodoraceae): the influence of reproductive systems on the maintenance of the polymorphism. *American Journal of Botany* 89:253–263.
- . 2002b. The genetics of mirror-image flowers. *Proceedings of the Royal Society of London B, Biological Sciences* 269:1835–1839.
- . 2002c. Solving the puzzle of mirror-image flowers. *Nature* 417:707.
- Jesson, L. K., J. Kang, S. L. Wagner, S. C. H. Barrett, and N. G. Dengler. 2003. The development of enantiostyly. *American Journal of Botany* 90:183–195.
- Knuth, P. 1906. *Handbook of flower pollination*. Clarendon, Oxford.
- Kohn, J. R., and S. C. H. Barrett. 1992. Experimental studies on the functional significance of heterostyly. *Evolution* 46:43–55.
- Lloyd, D. G. 1992. Self- and cross-fertilization in plants. II. The selection of self-fertilization. *International Journal of Plant Sciences* 153:370–380.
- Lloyd, D. G., and C. J. Webb. 1986. The avoidance of interference between the presentation of pollen and stigmas in angiosperms. I. Dichogamy. *New Zealand Journal of Botany* 24:135–162.
- . 1992a. The evolution of heterostyly. Pages 151–178 in S. C. H. Barrett, ed. *Evolution and function of heterostyly*. Springer, Berlin.
- . 1992b. The selection of heterostyly. Pages 179–208 in S. C. H. Barrett, ed. *Evolution and function of heterostyly*. Springer, Berlin.
- Luo, D., R. Carpenter, L. Copsey, C. Vincent, J. Clark, and E. Coen. 1999. Control of organ asymmetry in flowers of *Antirrhinum*. *Cell* 99:367–376.
- Olmstead, R. G., and J. D. Palmer. 1997. Implications for the phylogeny, classification, and biogeography of *Solanum* from cpDNA restriction site variation. *Systematic Botany* 22:19–29.
- Ornduff, R. 1974. Heterostyly in South African flowering plants: a conspectus. *Journal of South African Botany* 40:169–187.
- Ornduff, R., and R. Dulberger. 1978. Floral enantiomorphy and the reproductive system of *Wachendorfia paniculata* (Haemodoraceae). *New Phytologist* 80:427–434.
- Simpson, M. 1990. Phylogeny and classification of the Haemodoraceae. *Annals of the Missouri Botanical Garden* 77:722–784.
- Todd, J. E. 1882. On the flowers of *Solanum rostratum* and *Cassia chamaecrista*. *American Naturalist* 16:281–287.
- Wang, G., R. Muira, and T. Kusangai. 1995. The enantiostyly and pollination biology of *Monochoria korsakowii* (Pontederiaceae). *Acta Phytotaxonomica et Geobotanica* 46:55–65.
- Webb, C. J., and D. G. Lloyd. 1986. The avoidance of interference between the presentation of pollen and stigmas in angiosperms. II. Herkogamy. *New Zealand Journal of Botany* 34:164–178.
- Whalen, M. D. 1984. Conspectus of species groups in *Solanum* subgenus *Leptostemonum*. *Gentes Herbarum* 12: 179–282.