

## CHAPTER 4

# Pollen and ovule fates and reproductive performance by flowering plants

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### Outline

Pollen and ovules experience diverse fates during pollination, pollen-tube growth, fertilization, and seed development that govern the male and female potential of flowering plants. This chapter identifies these fates and many of their interactions, and considers their theoretical implications for the evolution of pollen export and the production of selfed and outcrossed seeds. This analysis clarifies the importance of pollen quantity and quality for seed production. Our analysis emphasizes the asymmetry of pollen and ovule fates and considers its consequences for reproductive evolution. We also identify ovule limitation as a constraint on seed production, which has paradoxically not been recognized before, but is an implicit assumption of previous theoretical analysis of mating-system evolution. Ovule limitation increases the diversity of possible reproductive strategies. In addition to ovule limitation, we consider the implications of pollen and resource limitation for the evolution of self- and cross-fertilization. Resource limitation occurs only if plants produce more ovules than they can mature into seeds, which allows a mixture of selfing and outcrossing to be an optimal mating system in some circumstances. The chance of mixed mating being optimal is enhanced by trade-offs between self- and cross-pollination, and more diverse optimal combinations of selfing and outcrossing are possible should mixed mating be favoured. Our analysis illustrates the key role played by interactions between genetic and ecological influences on reproductive performance in the evolution of plant reproduction.

### 4.1 Introduction

Most plants produce millions of pollen grains and/or thousands of ovules during their lives, but only one of each will be represented in the next generation in a stable population, on average. Clearly, most pollen grains and ovules succumb to fates other than successful reproduction, which bear significant implications for the persistence and dynamics of plant populations (Morgan *et al.* 2005). These fates arise from diverse interactions with the abiotic environment, with other species, and with other pollen grains and seeds, as well as from a lack of such interactions. This dependence

on variable abiotic and biotic environments renders plant mating highly stochastic and context dependent (e.g., Herrera 2002; Herrera 2004; Johnson *et al.* 2005). Nevertheless the diversity of pollen and ovule fates provides many opportunities for reproductive adaptation (see Lloyd 1979, 1992; Harder and Wilson 1998; Harder 2000).

Pollen grains and ovules experience very different environments, resulting in dissimilar fates. The independence of pollen grains from their parental sporophytes during much of their functional lives exposes them to the vagaries of weather, predation by pollen-consuming animals,

misadventure during dispersal, competition with other pollen grains for access to ovules if they reach a stigma, and rejection by pistils. In contrast, angiosperm ovules are relatively protected within ovaries, so that their fates depend primarily on the quantity and quality of pollen entering the ovary and the availability of maternal resources for seed production, unless developing seeds are attacked by pre-dispersal seed predators. This asymmetry has many implications for the operation of sexual selection in plants (Skogsmyr and Lankinen 2002), the allocation of resources to the sex roles (Charnov 1982; Lloyd 1984; Chapter 3), and the evolution of floral and inflorescence characteristics (Lloyd 1984; Bell 1985; Bell and Cresswell 1998).

Despite the ecological and evolutionary importance of pollen and ovule fates, they have been subject to relatively little explicit integrated analysis. This neglect partly reflects the traditional separation of studies of the pollination, post-pollination, and seed-development phases of reproduction, so the dependence of later reproductive stages on earlier stages is often ignored. Lloyd (1979, 1992) initiated explicit studies of pollen and ovule fates after recognizing that when and how self-pollination occurs affects a plant's opportunities for cross-fertilization and pollen export, and the extent to which selfing provides reproductive assurance. This perspective has been incorporated increasingly in models of mating-system evolution (Schoen *et al.* 1996; Morgan *et al.* 1997; Harder and Wilson 1998; Chapter 2) and population dynamics (Morgan *et al.* 2005). Nevertheless, these studies have ignored ovule fates, other than whether they are fertilized or not. Lloyd's identification of alternative modes of selfing also motivated some empirical analysis of pollen fates (e.g., Eckert 2000; Harder 2000; Goodwillie *et al.* 2005; Johnson *et al.* 2005). However, these studies have been conducted largely independently of analyses of the relative incidence of pollen versus resource limitation of seed production (reviewed by Casper and Neisenbaum 1993; Ashman *et al.* 2004), or of studies of pollen-tube competition or seed development that were stimulated by interest in sexual selection during the 1980s and 1990s (reviewed by Korbecka *et al.* 2002; Skogsmyr and Lankinen 2002).

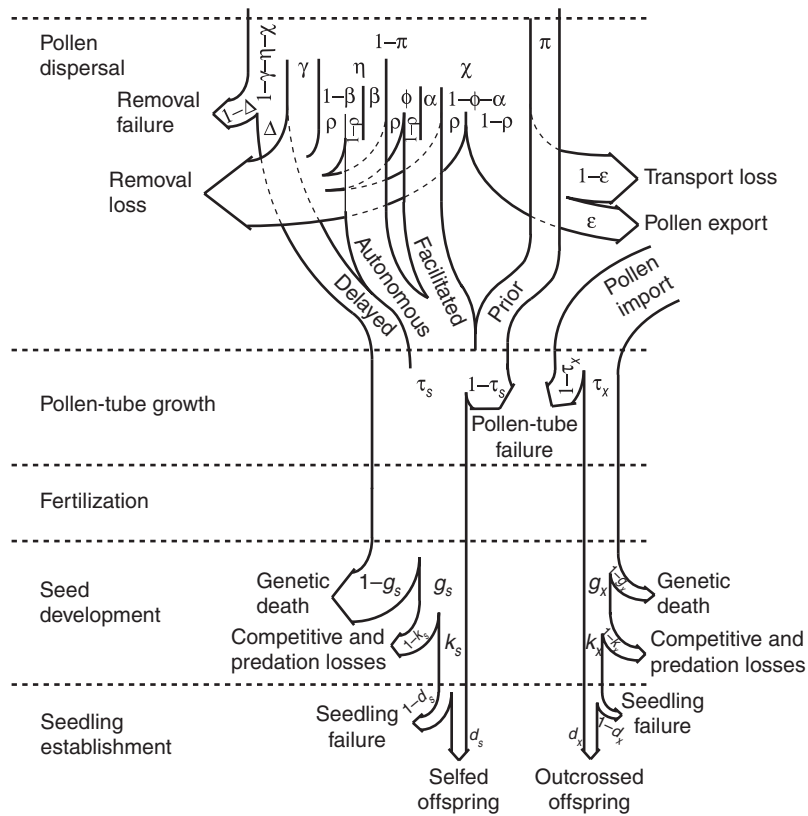
In this chapter, we characterize the diverse fates that await pollen and ovules and describe four theoretical examples of their implications for floral and mating-system evolution. We first identify pollen and ovule fates and many of their interactions and briefly review empirical estimates of the magnitudes of key fates. This overview clarifies the three limits on seed production: pollen receipt, ovule production, and resource availability. Given this foundation, we then consider the conditions under which a population could be invaded by a phenotype with novel floral traits that alter its pollen export or self-pollination. This analysis reveals that previous mating-system theory has largely ignored the consequences of resource limitation. Finally, we consider several implications of our results for current perspectives on angiosperm reproduction and its microevolution.

## 4.2 Pollen fates and ovule fates

The fates that await a plant's *P* pollen grains and *O* ovules are diverse and interact in complex ways (Fig. 4.1). These fates arise during five partially overlapping phases of the reproductive process: pollen dispersal, pollen-tube growth, ovule fertilization, seed development, and seedling establishment. Because of this sequential process, the opportunities for pollen and ovules to participate during each phase depend on the outcomes of all preceding phases. The conditional nature of pollen and ovule fates is particularly important because it determines the incidence and intensity of competition during two phases: competition among pollen tubes to fertilize ovules, and competition among developing seeds for maternal resources.

### 4.2.1 Pollen dispersal

Our characterization of pollen fates during dispersal (Table 4.1) integrates the perspectives of Lloyd (1979, 1992) and Harder (Harder and Wilson 1998; Harder 2000). Lloyd considered the pollen deposited on stigmas and emphasized the timing of self-pollination relative to cross-pollination and whether self-pollination occurred autonomously or with the aid of pollen vectors. In contrast, Harder considered all of the *P* pollen grains produced by a



**Figure 4.1** The general fates that await the *P* pollen grains and *O* ovules produced by a plant during pollen dispersal, pollen-tube growth, ovule fertilization, seed development, and seedling establishment. Each parameter indicates a proportion of the pollen grains, pollen tubes, zygotes, embryos, or seeds above it.

plant and explicitly divided self-pollination into components that reduce opportunities for pollen export (pollen discounting) or occur independently of export (non-discounting self-pollination). Similar to Lloyd, we recognize three phases of pollination (Table 4.1), although our terminology differs somewhat, as we explain below. Prior self-pollination occurs before flowers interact with pollen vectors and involves a fraction  $\pi$  of the *P* pollen grains produced by a plant. Processes that occur simultaneously with cross-pollination divide the remaining  $(1-\pi)P$  pollen grains into four fractions:  $\chi$ , pollen that could be exported to other plants;  $\eta$ , pollen that could be involved in self-pollination without affecting pollen export;  $\gamma$ , pollen that is displaced from flowers independently of self- or cross-pollination, such as by wind or rain; and any remainder,  $1-\chi-\eta-\gamma$ , which is involved

in processes that occur once opportunities for pollen export cease. We now characterize the distribution of pollen that arises after prior self-pollination.

The  $(1-\pi)\chi P$  pollen grains that could be exported experience the most diverse fates. A fraction  $\alpha$  of exportable pollen may be involved in autonomous, intrafloral self-pollination. In addition, pollen vectors may deposit a fraction  $\phi$  of the exportable pollen on the plant's own stigmas, resulting in facilitated self-pollination either within or between flowers (note that this usage differs from Lloyd's facilitated self-pollination, which involved only the intrafloral component). The remaining fraction,  $1-\alpha-\phi$ , is poised to leave the plant, except that pollen vectors could displace a fraction,  $\rho$ , which is lost from dispersal, adding to the removal loss associated with  $\gamma$  ( $\rho$  also

**Table 4.1** Possible fates of the  $P$  pollen grains produced by a single plant and their effects on pollen export.

Timing relative to access to pollen vectors	Fate	Number of pollen grains involved	Effect on an individual's pollen export
Prior	Autonomous self-pollination	$P_p = \pi P$	Negative
Simultaneous <sup>a</sup>	Autonomous self-pollination	$P_a = (1 - \pi)(\alpha\chi + \beta\eta)P$	Negative if $\alpha > 0$ , strongly so if $\eta \cong 0$
	Facilitated self-pollination <sup>b</sup>	$P_f = (1 - \pi)(\phi\chi + [1 - \beta]\eta)(1 - \rho)P$	Negative if $\phi > 0$ , strongly so if $\eta \cong 0$
	Export	$P_e = (1 - \pi)(1 - \alpha - \phi)\chi(1 - \rho)\varepsilon P$	
	Removal loss	$P_l = (1 - \pi)\{[(1 - \alpha)\chi + (1 - \beta)\eta]\rho + \gamma\}P$	Negative, intensity increases with $\rho$
Delayed <sup>c</sup>	Transport loss	$P_t = (1 - \pi)(1 - \alpha - \phi)\chi(1 - \rho)(1 - \varepsilon)P$	Negative, intensity increases with $1 - \varepsilon$
	Autonomous self-pollination	$P_d = (1 - \pi)(1 - \chi - \eta - \gamma)\Delta P$	None
	Removal failure	$P_r = (1 - \pi)(1 - \chi - \eta - \gamma)(1 - \Delta)P$	None

Note that although pollen export cannot vary positively with simultaneous self-pollination, removal loss, or transport loss, for individual plants, such a relation is possible *among* plants if plants differ in their proportion of potentially exportable simultaneous pollen ( $\chi$ ). See Fig. 4.1 for parameter definitions.

<sup>a</sup>These fates require that  $\pi < 1$ .

<sup>b</sup>May include both intrafloral and interfloral (geitonogamous) components.

<sup>c</sup>These fates require that  $\pi < 1$  and  $\chi + \eta + \gamma < 1$ .

reduces facilitated self-pollination). Of the remaining  $(1 - \pi)\chi(1 - \alpha - \phi)(1 - \rho)P$  pollen grains that leave the plant, a fraction  $\varepsilon$  is exported to conspecific stigmas and the remainder is lost during transport (including deposition on hetero-specific stigmas).

The  $(1 - \pi)\eta P$  pollen grains that could be involved in self-pollination without affecting pollen export may participate in up to three fates. A fraction  $\beta$  of this pollen may be involved in autonomous, intrafloral self-pollination, and the remaining fraction  $(1 - \beta)$  could be displaced by pollinators and contribute to either facilitated self-pollination or removal loss with probability  $\rho$  (we assume the same  $\rho$  for discounting and non-discounting facilitated self-pollination for simplicity). Facilitated self-pollination could occur without pollen discounting if pollen that would otherwise have fallen from a flower during a pollinator visit instead lands on a stigma. Thus, facilitated self-pollination can include both discounting and non-discounting components, depending on the magnitudes of  $\phi$  and  $1 - \beta$ , respectively (Table 4.1).

Once cross-pollination ceases, a fraction  $\Delta$  of the remaining  $(1 - \pi)(1 - \chi - \eta - \gamma)P$  pollen grains could be involved in delayed self-pollination, with the remainder constituting pollen removal failure

(Table 4.1). Delayed self-pollination can occur through various mechanisms, including the anthers collapsing onto the stigma, the stigma growing to contact the anthers, or the anthers brushing the stigma as the corolla falls (Chapter 10).

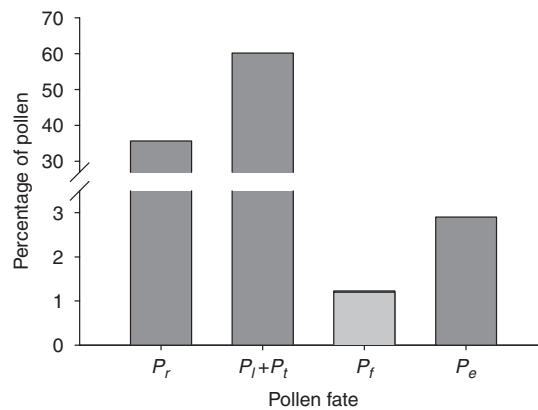
In summary, nine pollen fates arise during dispersal (Table 4.1). Self-pollination can occur autonomously before (*prior self-pollination*,  $P_p$ ), simultaneously with (*simultaneous, autonomous self-pollination*,  $P_a$ ), or after cross-pollination (*delayed self-pollination*,  $P_d$ ). Self-pollination can also involve the action of pollen vectors (*facilitated self-pollination*,  $P_f$ ) either within the flower that produced the pollen (*intrafloral, facilitated self-pollination*) or among flowers on the same plant (*geitonogamy*). In addition, *pollen export* ( $P_e$ ) results when pollen reaches stigmas on other conspecific plants. Finally, pollination can fail owing to three fates: *removal loss* ( $P_l$ ) while pollen vectors interact with the producing plant, *transport loss* as vectors move among plants ( $P_t$ ), and *removal failure* ( $P_r$ ). Pollen dispersal establishes the scope of potential reproductive output by determining the numbers of pollen grains involved in self-pollination ( $P_s = P_p + P_a + P_f + P_d$ ), pollen export ( $P_e$ ), and pollen import ( $P_i$ ). Pollen export equals import within a closed population; however, this equality need not hold for individual plants.

According to our characterization of pollen fates, all modes of self-pollination, except for delayed self-pollination, can reduce a plant's ability to export pollen (Table 4.1). As Lloyd (1979, 1992) noted, this pollen discounting is a universal consequence of prior self-pollination. The extent to which simultaneous self-pollination discounts pollen export depends on how it occurs. Simultaneous self-pollination does not affect export if  $\alpha = \phi = 0$ . In contrast, every self-deposited grain reduces export opportunities when  $\eta = 0$  and  $\alpha$  or  $\phi > 0$ . Given the diversity of ways in which a plant's pollen can be deposited on its own stigmas, reality may typically lie between these extremes. Nevertheless, a negative relation between discounting, simultaneous self-pollination, and pollen export within individual plants may be difficult to detect from comparisons among plants, because both outcomes vary positively with the proportion of pollen remaining after prior self-pollination that could be exported to other plants,  $\chi$  (see Table 4.1). For example, if all self-pollination is facilitated (i.e.,  $\pi = \alpha = \beta = \delta = 0$  and  $0 < \phi < 1$ ), then the number of pollen grains involved in self-pollination and export equals  $P_s = (\chi\phi + \eta)(1 - \rho)P$  and  $P_e = \chi(1 - \phi)(1 - \rho)\epsilon P$ , respectively. If plants differ in their proportions of exportable pollen  $\chi$ , self-pollination will vary positively and linearly with pollen export according to  $P_s = \eta(1 - \rho)P + (\phi/[1 - \phi]\epsilon)P_e$  (also see Harder 2000; Harder *et al.* 2000). A similar positive relation between pollen export and removal or transport loss may explain the paradox of species that use pollen-collecting bees to disperse their pollen (Harder and Wilson 1997).

The incidences of alternative pollen fates have not been measured completely for any plant species, but it is clear that most pollen does not reach stigmas (e.g., Fig. 4.2). Harder (2000) surveyed studies of monocots and found a strong dichotomy between species with granular pollen and orchids with pollen aggregated into pollinia. Species with granular pollen experienced relatively low removal failure (median = 7%), but only 1% of pollen removed from anthers reached conspecific stigmas (self- and cross-pollination), because of removal and transport losses (13 species). In contrast, orchids have much higher removal failure (median = 49%), but a median of 17% of the pollen

removed from anthers reaches stigmas (11 species). Therefore, the evolution of pollinia seems to reduce transport loss considerably. Of the relatively small amount of pollen deposited on stigmas, self-pollination can contribute a variable fraction, as is illustrated by the extensive variation among species in the proportion of seeds that are self-fertilized (reviewed by Goodwillie *et al.* 2005).

Pollen fates also vary extensively within species. In the most complete study of pollen fates to date, Johnson *et al.* (2005) found that *Disa cooperi* (Orchidaceae) plants experienced large differences in pollination between two consecutive years, including 76 and 37% declines in self-pollination and pollen export, respectively. During both years, the percentage of pollen on stigmas attributed to geitonogamous self-pollination ranged from 0 to 100% among plants, with most plants exporting no pollen during two days of observation, but one plant exporting pollen to eight recipients. Similarly, several studies have shown that the incidence of geitonogamy varies positively among plants with the number of flowers that they display simultaneously (reviewed by Harder *et al.* 2004). These results illustrate that pollen fates during dispersal depend strongly on the characteristics of individual plants



**Figure 4.2** Fates of *Disa cooperi* (Orchidaceae) pollen during two nights' exposure to hawk-moth pollination, including pollen removal failure ( $P_r$ ); pollen removal loss ( $P_l$ ); pollen transport loss ( $P_t$ ); facilitated self-pollination ( $P_s$ ), including both geitonogamous (light grey bar) and intrafloral deposition (dark grey bar); and pollen export ( $P_e$ ). This species cannot self-pollinate autonomously. Based on data from Johnson *et al.* 2005. Note the different scaling on either side of the break along the ordinate.

and their pollination environment. However, some plants can adjust their characteristics to alter pollen fates adaptively in response to their recent pollination history. Specifically, if pollination shortens floral longevity, frequent pollinator visits reduce floral display size, limiting geitonogamy and its associated pollen discounting (Harder and Johnson 2005).

#### 4.2.2 Pollen-tube growth

Once pollen reaches a stigma it must germinate and grow a pollen tube into the ovary. This process involves complex interactions between the pollen tube and pistil (Wheeler *et al.* 2001; Skogsmyr and Lankinen 2002; Stephenson *et al.* 2003), and competition among pollen tubes from the same or different pollen donors (Skogsmyr and Lankinen 2002; Armbruster and Rogers 2004; Bernasconi 2004). Pollen from some donors may fail completely in a particular pistil because of incompatibility reactions based on the genotype of the haploid pollen grain, or its diploid parent (homomorphic incompatibility), or, in heterostylous species, based on the anther level that produced a pollen grain (heteromorphic incompatibility: de Nettancourt 2001). However, self-incompatibility can weaken as flowers age (Good-Avila and Stephenson 2002; Goodwillie *et al.* 2004; Travers *et al.* 2004), allowing reproductive assurance if cross-fertilization is incomplete and adaptive implementation of mixed mating (Vallejo-Marín and Uyenoyama 2004). Nevertheless, the number of viable male gametophytes undergoes considerable attrition between pollination and fertilization. Indeed, the amount of pollen needed to maximize ovule fertilization typically exceeds the number of ovules in an ovary 5- to 10-fold (e.g., Mitchell 1997 and papers cited therein). Furthermore, relatively less self-pollen than cross-pollen survives during this post-pollination phase, even for species with weak self-incompatibility mechanisms (Bernasconi 2004). We represent the probability that a self- or cross-pollen grain on a stigma produces a pollen tube that reaches the ovary by  $\tau_s$  and  $\tau_x$ , respectively, where  $\tau_s=0$  for a self-incompatible species.  $\tau_s$  and  $\tau_x$  do not incorporate competition among pollen tubes, which we incorporate during the next phase of reproduction.

#### 4.2.3 Ovule fertilization

The composition of a plant's zygotes depends on the numbers of prior, simultaneous, and delayed self-pollen tubes and cross-pollen tubes entering the ovary and the number of ovules that remain unfertilized (Table 4.2). We assume that all pollen grains germinate and grow pollen tubes at the same rate, so that fertilization occurs in the same order that pollen grains arrive on stigmas. Consequently, prior self-pollen (if present) fertilizes ovules first, followed by simultaneous self- and imported pollen, and then delayed self-pollen. As Table 4.2 summarizes, two or three quantitative outcomes are possible for each fertilization fate, depending on whether ovules remain unfertilized when pollen tubes enter the ovary and, if so, whether fewer or more pollen tubes enter the ovary than are needed to fertilize the remaining ovules.

Table 4.2 reveals several features of ovule fates. First, fertilization outcomes depend on pollen fates during pollen dispersal and pollen-tube growth. Second, the distinction between different modes of simultaneous self-pollination becomes irrelevant during fertilization, as the resulting self-pollen tubes are functionally identical. Third, the number of fertilized ovules can be limited by either pollen receipt, if  $\tau_s(P_p + P_a + P_f + P_d) + \tau_x P_i < O$ , or ovule production, if  $\tau_s(P_p + P_a + P_f + P_d) + \tau_x P_i \geq O$ . Note that pollen tubes compete only when fertilization is ovule-limited and that the likelihood of competition increases from prior to simultaneous through delayed phases. We assume that the probability that a pollen tube fertilizes an ovule equals 1 when fertilization is pollen-limited but equals the inverse of the number of competing pollen tubes when fertilization is ovule-limited (i.e.,  $1/[P_s + P_i]$ ; Table 4.2).

#### 4.2.4 Seed development

After fertilization, zygotes become embryos and, together with associated tissues, consume maternal resources and develop into seeds. This process may fail for three reasons: death of zygotes or young embryos from the expression of lethal alleles (genetic death: Charlesworth and Charlesworth

**Table 4.2** Possible fertilization fates of the *O* ovules produced by a single plant.

Timing relative to cross-fertilization	Fate	Number of ovules involved	Condition(s)
Prior	Self-fertilized	$F_p = \tau_s P_p$ , or $F_p = O$	(1) $\tau_s P_p < O$ (2) $\tau_s P_p \geq O$
Simultaneous	Self-fertilized	$F_{af} = 0$ $F_{af} = \tau_s(P_a + P_f)$ , or $F_{af} = \frac{\tau_s(P_a + P_f)(O - \tau_s P_p)}{\tau_s(P_a + P_f) + \tau_x P_i}$	(2) (3) $\tau_s(P_p + P_a + P_f) + \tau_x P_i < O$ (4) $\tau_s(P_p + P_a + P_f) + \tau_x P_i \geq O$
	Cross-fertilized	$F_x = 0$ $F_x = \tau_x P_i$ , or $F_x = \frac{\tau_x P_i(O - \tau_s P_p)}{\tau_s(P_a + P_f) + \tau_x P_i}$	(2) (3) (4)
Delayed	Self-fertilized	$F_d = 0$ $F_d = \tau_s P_d$ , or $F_d = O - \tau_s(P_p + P_a + P_f) - \tau_x P_i$	(4) (5) $\tau_s(P_p + P_a + P_f + P_d) + \tau_x P_i < O$ (6) $\tau_s(P_p + P_a + P_f + P_d) + \tau_x P_i \geq O$
	Unfertilized	$F_u = 0$ $F_u = O - \tau_s(P_p + P_a + P_f + P_d) - \tau_x P_i$	(6) (5)

All fates, except for prior self-fertilization, depend on ovules remaining unfertilized after all preceding fates are fulfilled. The second condition for a fate (first for prior) applies when fewer pollen tubes enter the ovary than are needed to fertilize the remaining ovules, and the third condition (second for prior) applies when pollen tubes compete for fertilizations.  $P_p$ ,  $P_a$ ,  $P_f$ ,  $P_d$ , and  $P_i$  are defined in Table 4.1 (except  $P_i$ ) and represent the numbers of a plant's *P* pollen grains that are involved in: prior self-pollination; autonomous, simultaneous self-pollination; facilitated self-pollination; delayed self-pollination; and pollen import, respectively.  $\tau_s$  and  $\tau_x$  are the proportions of self-pollen and outcrossed pollen grains on a stigma whose pollen tubes enter the ovary, respectively.

1987; Husband and Schemske 1996); competition among embryos for maternal resources (including preferential maternal allocation: Casper and Nienbaum 1993; Korbecka *et al.* 2002); and consumption by pre-dispersal predators (Fenner and Thompson 2005). We propose that genetic death occurs before embryos consume appreciable maternal resources and that proportions  $g_s$  and  $g_x$  of the  $F_s = F_p + F_{af} + F_d$  selfed and  $F_x$  outcrossed embryos, respectively, survive this phase. Because selfed offspring are homozygous at more loci than outcrossed offspring, they are more likely to express recessive lethal traits, so that  $g_s < g_x$  unless the population bears negligible genetic load (Charlesworth and Charlesworth 1987; Husband and Schemske 1996). This characterization of early embryo mortality includes late-acting self-incompatibility, whereby interaction with a self-pollen tube disables an ovule, even if fertilization does not occur (de Nettancourt 2001). Embryos that survive genetic death then consume maternal resources. We assume that a maternal plant has sufficient resources to mature a fraction,  $m$ , of its ovules into seeds, so embryos

compete for resources if the number of embryos surviving genetic death exceeds  $mO$  (i.e.,  $F_s g_s + F_x g_x > mO$ ). In general, the probabilities of selfed and outcrossed embryos becoming seeds after they survive genetic death are  $k_s$  and  $k_x$ , respectively, with  $k_s = k_x = 1$  in the absence of resource competition.  $k_s$  and  $k_x$  can incorporate survival of pre-dispersal seed predation, but we ignore this process. Therefore, pre-dispersal, or early-acting, inbreeding depression equals  $1 - (g_s k_s / g_x k_x)$ .

For simplicity, we make two assumptions concerning resource competition. First, we assume that the timing of fertilization does not affect competitive outcomes. In contrast, embryos fertilized early compete more successfully than later embryos if competitive ability depends on the size of a developing seed (Ganeshiah and Uma Shaanker 1994; Uma Shaanker *et al.* 1995). This priority necessarily alters mating outcomes, although whether it affects the optimal mating patterns is uncertain. Second, we assume that selfed and outcrossed embryos compete equally, surviving resource competition with a probability

equal to the inverse of the total number of embryos that survive genetic death (i.e.,  $k_s = k_x = 1/[F_s g_s + F_x g_x]$ ). Although selfed embryos may compete less successfully than outcrossed embryos owing to greater expression of non-lethal deleterious alleles (Korbecka *et al.* 2002), this simplification has little effect on the optimal mating system (L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript).

The concepts of seed and ovule discounting explicitly recognize the ability of selfing to reduce seed production. Lloyd (1992) defined seed discounting as reduced production of outcrossed seeds caused by self-fertilization. Barrett *et al.* (1996) distinguished this post-zygotic process from ovule discounting, a reduction in seed production caused when self-pollen tubes disable some ovules (see Chapter 13). However, neither ovule nor seed discounting is an inevitable outcome of late-acting self-incompatibility or self-fertilization in plants that produce more ovules than they can mature into seeds. Indeed, production of “excess” ovules may serve specifically to compensate for such losses with limited impact on female fertility (Porcher and Lande 2005; L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript).

#### 4.2.5 Seedling establishment and parental fitness

Once their development is complete, seeds disperse and some germinate and establish reproductive offspring. As with pre-dispersal survival, the probability of post-dispersal survival of outcrossed seeds ( $d_x$ ) generally exceeds that of selfed seeds ( $d_s$ ), which are more likely to express deleterious traits due to their higher homozygosity (Husband and Schemske 1996). Therefore, post-dispersal or late-acting inbreeding depression equals  $1 - d_s/d_x$ . In general,  $d_s$  and  $d_x$  are on the order of the inverse of a plant's lifetime seed production, because one successful seed is sufficient to replace a parental plant.

A plant's fitness ( $w$ ) depends on its genetic contributions to the next generation through selfed seeds ( $S_s$ —two contributions per seed), its own outcrossed seeds ( $S_\varphi$ —one contribution per seed),

and seeds sired on other plants ( $S_\delta$ —one contribution per seed),

$$w = 2S_s d_s + S_\varphi d_x + S_\delta d_x.$$

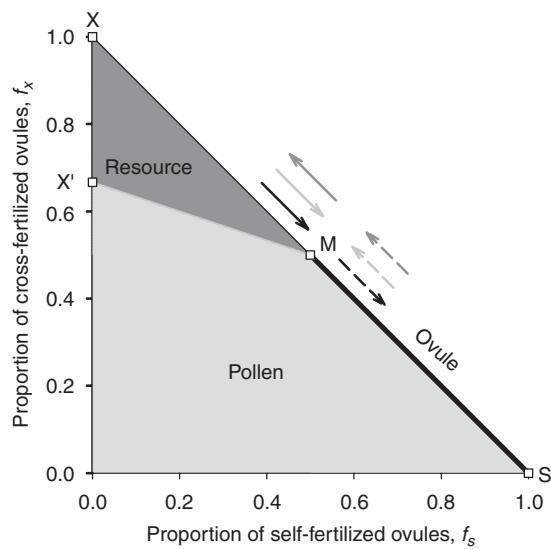
The ovule and pollen fates outlined above govern the details of these three fitness components.

### 4.3 Limits on seed production

Pollen and ovules contribute to the next generation only if they are represented in seeds from the producing plant and plants that import its pollen. Consequently, limits on seed production fundamentally govern population dynamics and reproductive evolution. To date, studies of seed-production constraints have considered two factors: pollen receipt, which affects fertilization success, and the availability of maternal resources during seed development, which determines maximum fecundity (reviewed by Casper and Niesenbaum 1993; Ashman *et al.* 2004). However, analysis of ovule fates exposes a third constraint, ovule limitation (L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript), which bears important implications for the evolution of ovule production and mating-system evolution. Before considering these implications, we clarify the conditions that result in pollen, ovule, and resource limitation.

#### 4.3.1 Pollen limitation

Pollen limitation occurs when (1) some of an individual's ovules remain unfertilized ( $F_s + F_x < O$ ) and (2) too few embryos avoid genetic death and predation to compete for maternal resources ( $F_s g_s + F_x g_x < mO$ ; Fig. 4.3). The first condition involves pollination quantity and indicates that plants do not compete for ovule fertilization, so an individual's fertilization success as a maternal and paternal parent depends only on the absolute numbers of pollen tubes entering its ovaries and those on other plants, respectively. This lack of competition is one reason why we refer to “simultaneous, autonomous self-pollination,” rather than Lloyd's (1992) “competing self-pollination.” This pollen-quantity aspect also



**Figure 4.3** Relations of pollen limitation (light grey area), ovule limitation (heavy line), and resource limitation (dark grey area) of seed production to the proportions of self- and cross-fertilized ovules. In this example, the probability of a cross-fertilized zygote surviving genetic death exceeds the proportion of ovules that the plant can mature into seeds, given available resources (i.e.,  $m < g_x < 1$ ). The diagonal line (X – M – S) depicts combinations of self- and cross-fertilizations that result in fertilization of all ovules ( $f_s + f_x = 1$ ). Along the transition from pollen to resource limitation (line X' – M) the proportion of zygotes surviving genetic death equals the proportion of ovules that can mature into seeds ( $f_s g_s + f_x g_x = m$ ). The arrows indicate the direction of mating-system evolution given either resource limitation (solid arrows), or ovule limitation (dashed arrows). The directions of the arrows depend on the conditions that allow increased self-pollination, given these limits on seed production, as listed in Table 4.3 (black arrows, both conditions satisfied; dark grey arrows, neither condition satisfied; light grey arrow, resource-limited condition satisfied, but ovule-limited condition not satisfied).

clarifies that pollen limitation results from poor pollen dispersal (any of small  $\chi$ , large  $\rho$ , or small  $\epsilon$ ) and/or limited self-pollination (any of small  $\pi$ ,  $\eta$ ,  $\phi$ , or  $\Delta$ ) and/or poor pollen-tube performance (small  $\tau_s$  and/or  $\tau_x$ ), and so is initiated by partial failure of male function, which precipitates subsequent partial failure of female function. Increased autonomous self-pollination can alleviate this aspect of pollen limitation from the female perspective (Chapter 10); however, it does little to relax the limitation of male performance. Therefore, reproductive assurance does not solve pollen limitation completely.

The second condition for pollen limitation involves aspects of pollination quality that determine the survival of young embryos ( $g_s$  and  $g_x$ ), which has generally been overlooked (although see Ramsey 1995; Ramsey and Vaughton 2000; Ashman *et al.* 2004; Chapter 9). This aspect of pollen limitation has important consequences for the detection of pollen limitation, which is typically assessed by the addition of outcross pollen to flowers that are otherwise subject to natural pollination conditions. Consider a self-compatible plant with  $F_x$  cross-fertilized ovules and the remainder,  $O - F_x$ , self-fertilized, such that fertilization is not pollen-limited. In the absence of resource limitation, this plant would produce a total of  $F_x g_x + (O - F_x) g_s = F_x(g_x - g_s) + O g_s$  seeds. Suppose that supplemental cross-pollen is applied to stigmas while flowers are young so that the number of cross-fertilized zygotes increases to  $F_x + \Sigma$ , decreasing the number of self-fertilized zygotes to  $O - F_x - \Sigma$ . Because outcrossed zygotes survive better than selfed zygotes ( $g_x > g_s$ ), the replacement of self-fertilizations by cross-fertilizations caused by supplemental pollination increases seed production by  $\Sigma(g_x - g_s)$  seeds. Such an elevation of seed production in response to supplemental cross-pollination would typically be interpreted as an indication of pollen limitation under natural conditions, even though fertilization was complete in both cases. This effect of pollen quality raises questions about whether pollen limitation is as common as supplementation experiments suggest (reviewed by Burd 1994; Ashman *et al.* 2004).

### 4.3.2 Ovule limitation

Ovule limitation occurs when all of a plant's ovules are fertilized ( $F_s + F_x = O$ ) but too few zygotes avoid genetic death and predation to compete for maternal resources ( $F_s g_s + F_x g_x \leq mO$ ; Fig. 4.3). This limit results when plants invest too few resources in ovule production during flower production, perhaps because resource availability improves between flower initiation and seed production. In this case, pollen tubes compete for fertilizations. In our model, pollen from different plants fertilizes ovules in proportion to its contribution to the pollen tubes that enter an ovary

simultaneously (as in Holsinger's [1991] "mass-action" models: also see Lloyd 1992; Chapter 2), although biased competition is also possible. In addition, self- and cross-pollen tubes compete for access to ovules in a manner that depends on their relative timing, as outlined in Table 4.2. However, because of poor zygote survival, embryos develop without competition for maternal resources when ovule availability limits seed production, so  $k_s = k_x = 1$ . By not incorporating the possibility of differential survival of selfed and outcrossed embryos, all previous models of the consequences of different modes of self-pollination (e.g., Lloyd 1979, 1992; Schoen *et al.* 1996; Morgan and Wilson 2005) have implicitly considered this situation (see Section 4.4.2).

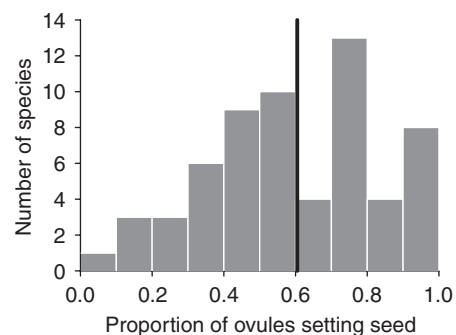
### 4.3.3 Resource limitation

Resource limitation occurs when more zygotes avoid genetic death than can mature into seeds, given the available maternal resources ( $F_{sgs} + F_{xgx} > mO$ ; Fig. 4.3), whether or not fertilization is complete. Resource limitation imposes competition among developing embryos, so the probabilities that selfed and outcrossed embryos develop into seeds,  $k_s$  and  $k_x$  respectively, are  $< 1$ . In general, these probabilities depend on the number of competing embryos ( $F_{sgs} + F_{xgx}$ ), the maximum number of seeds that can be produced ( $mO$ ), and the relative competitive ability of selfed versus outcrossed embryos. Because resource limitation can occur without complete fertilization (Fig. 4.3), pollen tubes may fertilize ovules independently as described for pollen limitation, or they may compete for fertilizations as described for ovule limitation. Curiously, although resource limitation is a widely recognized constraint on seed production (e.g., Haig and Westoby 1988), its consequences for mating-system evolution have been considered only recently (Sakai and Ishii 1999; Porcher and Lande 2005; L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript).

Resource competition occurs *only* if plants produce more ovules than they can mature into seeds (i.e.,  $m < 1$ ). Specifically, resource competition requires that the proportion of outcrossed zygotes surviving genetic death exceeds the maximum

proportion of ovules that can mature into seeds ( $g_x > m$ ). Such overproduction of ovules can occur for two reasons. The first cause is ecological and results from a mistake in resource allocation to ovule production versus seed development, which could occur if resource conditions decline between flower initiation and seed production. The second cause is adaptive and occurs, for example, if plants produce "extra" ovules to take advantage of unpredictably good pollination (Burd 1995), or resource availability, or to compensate for embryo losses during development to genetic death and/or predation (Porcher and Lande 2005; L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript). The latter, evolutionary explanation may apply commonly, as a literature survey of 65 species found an average seed:ovule ratio of 0.6 for plants subject to excess hand cross-pollination, indicating that plants typically produce many more ovules than they mature into seeds (Fig. 4.4).

Reproductive compensation has important consequences for mating-system evolution (see Section 4.4) and the genetic load within populations (Porcher and Lande 2005; L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript). Production of more ovules than can mature into seeds allows the genetic death of embryos soon after fertilization to have a limited impact on seed production by the maternal plant. By compensating for genetic deaths, extra ovules allow maternal plants to screen embryos *passively* for viable offspring, at the cost of producing the failed



**Figure 4.4** The proportion of ovules that develop into seeds for 65 species that were subjected to supplemental cross-pollination. The vertical line indicates the mean seed:ovule ratio.

ovules. This cost is probably relatively small, given that an ovule costs approximately 1% as much as a seed (C. A. Greenway and L. D. Harder unpublished data). Compensatory ovules would also facilitate *active* maternal choice among developing embryos with little cost (see Korbecka *et al.* 2002). Of particular relevance to mating-system evolution is the opportunity that compensatory ovules create for producing viable selfed offspring, which carry two haploid maternal genomes rather than one for outcrossed offspring, despite considerable genetic load in a population. Interestingly, this success of selfed offspring helps maintain genetic load, because parents produce a higher frequency of offspring that are heterozygous carriers for recessive lethal alleles than they would in the absence of reproductive compensation (Porcher and Lande 2005).

#### 4.4 Examples of the roles of pollen and ovule fates in floral and mating-system evolution

To illustrate the evolutionary consequences of pollen and ovule fates, we consider the fitness differential between two phenotypes with different pollination and fertilization patterns,

$$w_2 - w_1 = 2(S_{s2} - S_{s1})d_s + (S_{\varrho 2} - S_{\varrho 1})d_x + (S_{j2} - S_{j1})d_x,$$

and identify circumstances in which phenotype 2 has an advantage (i.e.,  $w_2 - w_1 > 0$ ). The characteristics of this fitness differential depend on the details of pollination, whether pollen tubes compete for fertilization, whether embryos compete for maternal resources, and the proportion of the population comprised of phenotype 1 ( $z$ : phenotype 2 represents proportion  $1 - z$ ). We now consider four of many possible evolutionary scenarios, showing the detailed derivation in the first case and simply summarizing the results for subsequent examples. Because the outcomes in several cases are frequency dependent, we focus on conditions that allow phenotype 2 to invade a population of phenotype 1 (i.e.,  $z \cong 1$ ) and consider equilibrium outcomes only when they are not frequency dependent. In all cases of

resource competition, we assume that selfed and outcrossed embryos compete equally for maternal resources.

##### 4.4.1 Improvements in pollen export

We illustrate the evolution of improved pollen export for plants that self-pollinate without affecting export, although the general conclusions also apply when self-pollination causes pollen discounting (results not shown). In the absence of pollen discounting, self-pollination equals  $P_s = \eta P$ , pollen export for the two phenotypes equals  $P_{e1} = \chi_1(1 - \rho_1)\varepsilon_1 P$  and  $P_{e2} = \chi_2(1 - \rho_2)\varepsilon_2 P$ , and each plant of either phenotype imports  $P_i = zP_{e1} + (1 - z)P_{e2}$  pollen grains. Thus, phenotype 2 might export pollen more successfully because its flowers attract more pollinators (increased  $\chi$ ) or they place pollen on pollinators' bodies where it is less susceptible to removal or transport losses (reduced  $\rho$ , increased  $\varepsilon$ ).

In general, fitness depends on whether seed production is pollen-, ovule-, or resource-limited. With pollen limitation, the fitnesses of the two genotypes are

$$w_1 = 2\tau_s P_s g_s d_s + (P_i + P_{e1})\tau_x g_x d_x \text{ and} \\ w_2 = 2\tau_s P_s g_s d_s + (P_i + P_{e2})\tau_x g_x d_x,$$

resulting in a fitness differential of  $w_2 - w_1 = (P_{e2} - P_{e1})\tau_x g_x d_x$ . Consequently, phenotype 2 can invade a pollen-limited population of phenotype 1 (i.e.,  $w_2 > w_1$ ) as long as it exports more pollen (i.e.,  $P_{e2} > P_{e1}$ ; Table 4.3). In contrast, in an ovule-limited population, plants import enough pollen to fertilize all their  $O$  ovules, so the  $\tau_s P_s + \tau_x P_i$  pollen tubes compete for fertilizations. If too few embryos survive genetic death to cause competition of maternal resources, the fitnesses of the two genotypes are

$$w_1 = \frac{2\tau_s P_s g_s d_s + \tau_x (P_i + P_{e1}) g_x d_x}{\tau_s P_s + \tau_x P_i} O \text{ and} \\ w_2 = \frac{2\tau_s P_s g_s d_s + \tau_x (P_i + P_{e2}) g_x d_x}{\tau_s P_s + \tau_x P_i} O,$$

resulting in a fitness differential of  $w_2 - w_1 = \tau_x (P_{e2} - P_{e1}) g_x d_x O / (\tau_s P_s + \tau_x P_i)$ . Increased pollen export is again favoured (Table 4.3);

in this case because cross-fertilizations by phenotype 2 displace self- and cross-fertilizations by phenotype 1. Finally, in a resource-limited population the  $\tau_s P_s g_s + \tau_x P_i g_x$  embryos that survive genetic death exceed the number that can mature into seeds ( $mO$ ), causing competition for maternal resources (i.e.,  $k_s = k_x = 1/[\tau_s P_s g_s + \tau_x P_i g_x]$ ). Now the fitnesses of the two genotypes are

$$w_1 = \frac{2\tau_s P_s g_s d_s + \tau_x (P_i + P_{e1}) g_x d_x}{\tau_s P_s g_s + \tau_x P_i g_x} mO \quad \text{and}$$

$$w_2 = \frac{2\tau_s P_s g_s d_s + \tau_x (P_i + P_{e2}) g_x d_x}{\tau_s P_s g_s + \tau_x P_i g_x} mO,$$

whether or not pollen tubes compete for fertilizations, and the fitness differential is  $w_2 - w_1 = \tau_x (P_{e2} - P_{e1}) g_x d_x mO / (\tau_s P_s g_s + \tau_x P_i g_x)$ . Increased pollen export is again favoured (Table 4.3), in this case because an increase in the number of embryos sired by phenotype 2 displaces embryos sired by phenotype 1 during resource competition.

The preceding results demonstrate a general advantage to increased pollen export resulting from altered floral mechanisms or pollinator shifts that increase the proportion of exportable pollen ( $\chi$ ) or reduce removal or transport losses (decreased  $\rho$  or increased  $\varepsilon$ , respectively). Furthermore, because the fitness differential does not depend on the relative frequencies of the two phenotypes ( $z$ ), a phenotype

with higher pollen export should replace one with lesser export. This universal advantage arises in our model because increased pollen export alters the number and/or mixture of seeds produced on other plants, without affecting a plant's own seed production. In contrast, a change in a floral trait, such as increased dichogamy, that improves pollen export but reduces non-discounting self-pollination can be detrimental (results not shown). Nevertheless, the contrast between the expected general benefit of enhanced pollen export and the observation that plants with granular pollen export only 1% of their pollen, on average (see Section 4.2.1), suggests that the evolution of cross-pollination is subject to strong functional constraints.

#### 4.4.2 Simultaneous, autonomous self-pollination without pollen discounting

Now consider a situation in which the two phenotypes differ in their ability to self-pollinate autonomously without pollen discounting, so  $P_{s1} = \eta_1 P$  and  $P_{s2} = \eta_2 P$ , but they export equivalent amounts of pollen,  $P_e = \chi(1 - \rho)\varepsilon P$ . For example, suppose the petals of phenotype 2 retain more pollen that has fallen from anthers and would otherwise be lost, which is then transferred onto stigmas when flowers close at night. If seed

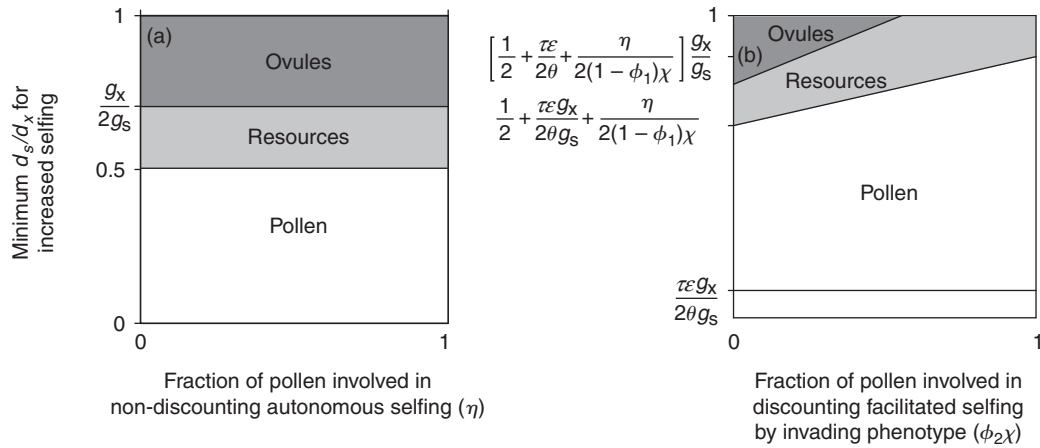
**Table 4.3** Conditions for the invasion of a population of self-compatible plants (phenotype 1) by a phenotype (phenotype 2) that either exports more pollen, or self-pollinates more than the resident phenotype.

Limit on seed production	Pollination contrast			
	Pollen export <sup>a</sup>	Simultaneous, non-discounting self-pollination	Simultaneous, facilitated self-pollination	Delayed, autonomous self-pollination <sup>b</sup>
Pollen	$\frac{d_s}{d_x} > 0$	$\frac{d_s}{d_x} > 0$	$\frac{d_s}{d_x} > \frac{\tau_x \varepsilon}{2\tau_s} \cdot \frac{g_x}{g_s}$	$\frac{d_s}{d_x} > 0$
Resources	$\frac{d_s}{d_x} > 0$	$\frac{d_s}{d_x} > \frac{1}{2}$	$\frac{d_s}{d_x} > \frac{1}{2} + \frac{\tau_x \varepsilon}{2\tau_s} \cdot \frac{g_x}{g_s} + \frac{\eta + \phi_2 \chi}{2(1 - \phi_1) \chi}$	$\frac{d_s}{d_x} > \frac{1}{2}$
Ovules	$\frac{d_s}{d_x} > 0$	$\frac{d_s}{d_x} > \frac{1}{2} \cdot \frac{g_x}{g_s}$	$\frac{d_s}{d_x} > \left[ \frac{1}{2} + \frac{\tau_x \varepsilon}{2\tau_s} + \frac{\eta + \phi_2 \chi}{2(1 - \phi_1) \chi} \right] \frac{g_x}{g_s}$	

See Fig. 4.3 for the implications of the conditions for resource and ovule limitation on mating-system evolution.

<sup>a</sup> Increased pollen export is favoured universally, because  $d_x$  always exceeds  $d_s$ .

<sup>b</sup> Increased delayed selfing cannot evolve if plants already experience enough self- and cross-pollination to fertilize all ovules.



**Figure 4.5** Relations of the minimum ratio of post-dispersal survival of selfed ( $d_s$ ) and outcrossed seeds ( $d_x$ ) that allows the invasion of a phenotype with increased self-pollination to aspects of self-pollination and the incidence of pollen limitation (white area and above), ovule limitation (light grey area and above), and resource limitation (dark grey area). Two contrasting situations are illustrated: (a) the novel phenotype has elevated autonomous, non-discounting self-pollination ( $\eta$ ); (b) elevated facilitated self-pollination (both non-discounting and discounting;  $\phi$ ).

production is pollen-limited (i.e., no pollen-tube or embryo competition), phenotype 2 increases in frequency ( $w_2 - w_1 > 0$ ) if it self-pollinates more than phenotype 1 ( $\eta_2 > \eta_1$ ; Table 4.3, Fig. 4.5a). As demonstrated in Section 4.4.1, this situation also promotes increased pollen export. Therefore, any floral trait that alleviates pollen limitation is favoured, whether or not it increases self-pollination or pollen export, as long as the two pollination modes do not interact negatively.

Selection is more complex when pollen tubes compete for fertilizations and/or embryos compete for maternal resources. With competition for fertilization, but not for maternal resources (i.e., ovule limitation), increased non-discounting self-pollination is favoured if the survival of selfed *zygotes* relative to outcrossed *zygotes* exceeds 0.5 ( $g_s d_s / g_x d_x > 0.5$ ; Table 4.3, Fig. 4.5a). In contrast, when resource availability limits seed production, phenotype 2 is favoured if the survival of selfed *seeds* relative to outcrossed *seeds* exceeds 0.5 ( $d_s / d_x > 0.5$ ; Table 4.3, Fig. 4.5a).

Comparison of results for resource- and ovule-limited cases reveals a feature of classic mating-system theory that has not been recognized previously. Standard mating-system models (e.g., Lande and Schemske 1985) and Lloyd's (1979) model of competing selfing assumed that self-

pollination did not affect pollen export and so are equivalent to the case modelled here. These models suggested that self-pollination is favoured if inbreeding depression ( $\delta$ ) after self-fertilization is  $< 0.5$ . In terms of our notation,  $\delta = 1 - (g_s d_s / g_x d_x)$ . Therefore, the classic result of mating-system theory is identical to our result for ovule limitation. In contrast, when seed production is resource-limited, we find a less stringent condition for increased selfing, which involves inbreeding depression only after seed production (post-dispersal inbreeding depression). Therefore, both classical mating-system models and Lloyd's more mechanistic model implicitly assume no resource limitation.

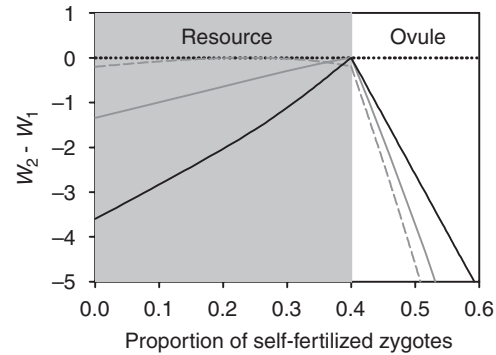
To appreciate the consequences of resource limitation for mating-system evolution, consider Fig. 4.3, which illustrates all possible combinations of self- and cross-fertilization. As we have demonstrated, any independent increase in self- or cross-pollination that lessens pollen limitation is always favoured, so that the optimal mating system will not be pollen-limited. Outcomes are more complex when enough ovules are fertilized to cause either ovule limitation (between points S and M in Fig. 4.3) or resource limitation (within triangle X-M-X' in Fig. 4.3). In these cases, the ultimate outcome of mating-system evolution depends on

the survival of selfed zygotes relative to outcrossed zygotes ( $g_s d_s / g_x d_x$ ) and of selfed seeds relative to outcrossed seeds ( $d_s / d_x$ ). If both aspects of relative survival exceed 0.5, then increased selfing is favoured, whether or not seed production is ovule- or resource-limited (dashed and solid black arrows in Fig. 4.3), leading eventually to exclusive selfing (point S in Fig. 4.3). If neither aspect of relative survival exceeds 0.5, then increased outcrossing is always favoured (dark grey arrows in Fig. 4.3), resulting in exclusive outcrossing (any point between X and X' in Fig. 4.3). Finally, if genetic death causes low survival of selfed zygotes compared with outcrossed zygotes ( $g_s d_s / g_x d_x < 0.5$ ), but selfed seeds have high relative survival ( $d_s / d_x > 0.5$ ), then mixed mating is favoured (light grey arrows in Fig. 4.3). In this case, the optimal mixture of self- and cross-fertilization lies at the transition from ovule to resource limitation (point M in Fig. 4.3), or

$$f_s^* = \frac{m - g_s}{g_x - g_s} \quad \text{and} \quad f_x^* = \frac{g_x - m}{g_x - g_s},$$

and so depends only on the probabilities that selfed and outcrossed zygotes survive genetic death ( $g_s$  and  $g_x$ , respectively) and the proportion of ovules that can mature into seeds given the available maternal resources ( $m$ ). Thus mixed mating can be an optimal mating system in the absence of pollen discounting if selfed zygotes are more susceptible to genetic death than outcrossed zygotes and seed production can be resource-limited. (The fourth combination of high relative survival of selfed zygotes and low relative survival of selfed seeds is mathematically impossible.) Note that none of the preceding results depends on the relative frequencies of competing phenotypes and so they apply to both invasion and equilibrium situations.

The solid black curve in Fig. 4.6 illustrates a specific case in which mixed mating is an evolutionary stable strategy (ESS), whereby no alternative mating pattern results in higher fitness. In addition to illustrating that the optimal mating system occurs at the transition between resource and ovule limitation, this figure reveals that the fitness differential between the resident and invading phenotypes declines more steeply away



**Figure 4.6** Examples of evolutionarily stable mixed mating when all ovules are fertilized and self-pollination either does not affect pollen export (black solid line) or causes pollen discounting (grey curves). The ESS occurs at the proportion of self-fertilization that equalizes the fitnesses of the resident and invading phenotypes ( $w_1$  and  $w_2$ , respectively), so  $w_2 - w_1 = 0$ . For non-discounting self-pollination (black solid line) and some cases of discounting self-pollination (e.g., grey solid line), the ESS lies at the transition between resource limitation (grey area) and ovule limitation (white area). Other cases of discounting self-pollination result in other optimal mating systems (e.g., grey dashed line). For all examples,  $P = 50\,000$  pollen grains,  $O = 100$  ovules,  $\chi(1 - \rho)\epsilon\tau_x = 0.01$ ,  $g_s = 0.3$ ,  $g_x = 0.8$ ,  $d_s/d_x = 0.71$ , and  $m = 0.6$ . For the solid lines  $\tau_x = 0.1$ , whereas  $\tau_x = 0.07$  for the dashed line.

from the ESS in the direction of ovule limitation, rather than resource limitation. Thus, ovule limitation imposes stronger selection toward the ESS than does resource limitation. Nevertheless the steep declines in fitness on either side of the ESS should result in strong stabilizing selection on the mating system.

#### 4.4.3 Facilitated self-pollination

We now consider two phenotypes that rely on pollen vectors to facilitate both self-pollination ( $P_s = [\phi\chi + \eta][1 - \rho]P$ ) and pollen export ( $P_e = [1 - \phi]\chi[1 - \rho]\epsilon P$ ). Specifically, we suppose that the phenotypes differ in the discounting component of self-pollination ( $\phi_1$  versus  $\phi_2$ ), which then alters pollen export. This situation could arise if phenotype 2 displays more flowers simultaneously than phenotype 1, so each pollinator visits more flowers, resulting in greater geitonogamy. Note that the  $\phi\chi P$  pollen grains involved in pollen discounting cause a one-to-one

reduction in pollen-export *opportunities*, as determined by the amount of pollen carried away from a plant ( $[1 - \phi]\chi[1 - \rho]P$ ), but have a weaker effect on *realized* export, because of the lower probability of reaching a stigma owing to transport losses.

Not surprisingly, pollen discounting affects the outcomes of mating-system evolution. Two general outcomes are possible, compared with those observed when simultaneous self-pollination does not affect pollen export. One possibility involves the same ESS outcomes as observed for non-discounting self-pollination, namely, exclusive outcrossing, exclusive selfing, or mixed mating at the transition between resource and ovule limitation (Fig. 4.6, solid grey curve), although pollen discounting restricts the range of conditions that favour increased self-pollination, whether or not seed production is pollen-, ovule-, or resource-limited (Table 4.3, Fig. 4.5b). This situation leads to relatively straightforward explicit solutions, which enable comparison with the non-discounting case. We describe these solutions below. In contrast, the second possibility is more complex and we leave detailed analysis of it to a subsequent paper (L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript). This case allows for a much greater range of mixed-mating systems, which are not constrained to lie at the transition between resource- and ovule-limitation (e.g., Fig. 4.6, dashed grey curve) and can even occur when resource limitation is not possible (i.e.,  $m = 1$ ).

In cases of discounting self-pollination that allow explicit solutions, the criterion for increased selfing observed in the absence of pollen discounting is incremented by at least  $\tau_x \varepsilon g_x / 2\tau_s g_s$ . This ratio includes all aspects of mating that are unique to outcrossing in the numerator and unique to selfing in the denominator, including the proportion of pollen grains that survive transport loss ( $\varepsilon$ ), the proportions of self- and cross-pollen grains on stigmas that are represented by pollen tubes in the ovary ( $\tau_s$  and  $\tau_x$ , respectively), the proportions of selfed and outcrossed zygotes that survive genetic death ( $g_s$  and  $g_x$ , respectively), and the two-fold transmission advantage of selfed offspring. In general, we expect  $\tau_x / \tau_s$  and  $g_x / g_s$  to exceed 1, whereas  $\varepsilon$  is approximately 0.01 for species with granular pollen and 0.1 for species with pollinia (see Section

4.2.1). Therefore, for species with granular pollen  $\tau_x \varepsilon g_x / 2\tau_s g_s$  probably lies between approximately 0.02 and 0.05, and so has a small effect on the threshold for increased self-fertilization. In contrast, for species with pollinia,  $\tau_x \varepsilon g_x / 2\tau_s g_s$  probably lies between approximately 0.2 and 0.5, which greatly elevates the selfing threshold and could even exclude any possibility of increased discounting self-pollination. The common occurrence among orchids of features of the pollinarium that preclude self-pollen deposition until after a pollinator has left a plant (Darwin 1862; Peter and Johnson 2006) is consistent with this expectation.

When pollen tubes compete for fertilization and/or embryos compete for maternal resources, the threshold for increased selfing is further increased by  $(\eta + \phi_2 \chi) / (2[1 - \phi_1 \chi])$ , which includes details of self-pollination (Fig. 4.5b). In general, this term renders increased selfing less likely to invade if the resident already self-pollinates extensively whether or not it involves pollen discounting (large  $\eta$  or  $\phi_1$ ), or if the floral or inflorescence traits of the variant greatly increase discounting above the prevailing level (large  $\phi_2$ ; Fig. 4.5b). Note that with pollen-tube competition this term is also multiplied by  $g_x / g_s$ , which is greater than 1, so that increased selfing is more likely to be favoured with resource limitation than with ovule limitation (Fig. 4.5b).

Our analysis of all outcomes of discounting, simultaneous self-pollination leads to a general conclusion about its effect on the evolution of mixed mating. This mating system is possible only with relatively strong inbreeding depression before seed dispersal ( $2g_s d_s > g_x d_x$ ), but not later ( $2d_s > d_x$ ). With pollen discounting, each pollen grain can be used in either self-pollination or potential export. If it is used in selfing, it has a high chance of reaching a stigma, but a low chance of becoming a viable offspring, because of pre-dispersal inbreeding depression. If it is used in potential export, the pollen grain has a low chance of reaching a stigma, because of losses during transport, but a high(er) chance of becoming a viable offspring if it does reach a stigma. In this case, a plant could sire more offspring through partial selfing if the chance of export is lower than the losses due to inbreeding depression.

In contrast, pure outcrossing should be favoured if pollen transport losses are relatively low, whereas some selfing is favoured when transport losses are relatively high. This mechanism can occur regardless of the proportion of ovules that can mature into seeds ( $m$ ).

#### 4.4.4 Delayed self-pollination

That delayed self-pollination occurs after cross-pollination, such as when the stamens of wilting flowers collapse on stigmas, bears two immediate consequences for mating-system evolution. First, delayed self-pollination cannot affect a plant's outcross siring success, so the fitness differential between competing phenotypes depends only on their relative maternal contributions (also see Lloyd 1979). Second, delayed self-fertilization cannot alter the mixture of selfed and outcrossed zygotes if all ovules are fertilized, so it cannot mitigate the effects of poor pollen quality on seed production for ovule-limited plants. We now consider additional consequences of delayed self-pollination for pollen- and resource-limited plants.

Our analysis partially contradicts Lloyd's (1979) assertion that "delayed self-fertilization... is always advantageous whenever it is possible" (p. 71). Our results identify that delayed selfing is advantageous either (1) if it provides reproductive assurance when seed production is pollen-limited or (2) if it increases the number of selfed embryos competing for maternal resources when selfed seeds survive at least half as well as outcrossed seeds (i.e.,  $2d_s > d_x$ ; Table 4.3), or equivalently *post-dispersal* inbreeding depression is  $< 0.5$ . When selfed seeds survive poorly, delayed selfing is disadvantageous if it aggravates resource competition among developing seeds. Because predominantly outcrossing species suffer less *post-dispersal* inbreeding depression than selfing species (Husband and Schemske 1996), delayed selfing should be a more common mode of selfing for outcrossing species. The incompleteness of Lloyd's conclusions resulted because, as with most other analyses of mating-system evolution, he did not consider the direct consequences of resource competition, and so implicitly assumed that this mode of self-fertilization bears no resource

consequences. Interestingly, Lloyd (1992) raised the possibility that resource consumption by seeds produced by delayed selfing could reduce a perennial parent's fitness by reducing its future survival and/or productivity during subsequent breeding seasons (also see Morgan *et al.* 1997).

## 4.5 Concluding discussion

### 4.5.1 The asymmetry of pollen and ovule fates

This review confirms the asymmetry of pollen and ovule fates that is obvious from the imbalance in pollen and ovule production, even though every sexually produced seed involves one pollen grain and one ovule. This asymmetry occurs for two reasons. Most important, outcrossing exposes pollen grains to many risks that are not experienced by ovules, particularly removal and transport losses. In addition, pollen fates depend on both the timing of alternative pollination modes and their dependence on pollen vectors, whereas ovule fates depend largely on the timing of fertilization. As a result, pollen is subject to a greater variety of fates, many of which do not result in direct genetic contributions to the next generation. We now briefly consider two implications of this asymmetry for the evolution of reproductive traits and its analysis.

The greater diversity and severity of risks confronted by pollen grains than by ovules requires plants to produce many more pollen grains than ovules. Following Lloyd (1965), Cruden (1977) used similar reasoning to explain the higher pollen:ovule ratios of outcrossing species than selfing species, although he focused on the delivery of pollen to fertilize seeds. Charnov (1982) criticized this argument, claiming that Cruden's explanation largely ignored paternal contributions to the fitness of hermaphrodites. Instead, Charnov proposed that elevated pollen production benefits outcrossing species by increasing competitive opportunities in pistils, whereas a similar increase in pollen production for a selfing species would simply aggravate competition among sibling grains (local mate competition). Although true, this argument ignores the contrasting risks involved in self-pollination versus pollen export. As a result, the relatively high

pollen production of outcrossing species is undoubtedly necessary to compensate for pollen losses during transport. For example, Kjellberg *et al.* (2001) found that fig species pollinated by wasp species that collect pollen actively and carry it to receptive trees in pollen pockets on their thoraces, presumably limiting transport losses, produce significantly fewer anthers per ovule (and presumably a lower pollen:ovule ratio) than species pollinated by wasps that carry pollen passively and often groom it from their bodies.

Bateman's (1948) principle also considers the consequences of mating asymmetries between the sexes, but its applicability to the evolution of plant reproduction has been questioned (Burd 1994; Wilson *et al.* 1994; Ashman and Morgan 2004). Based on insightful experiments with *Drosophila*, Bateman proposed that resource availability typically limits female fertility, whereas mating opportunities limit male fertility, so sexual selection should favour female traits that promote mate quality and male traits that enhance mating frequency. In contrast to Bateman's expectation that female fertility is resource-limited, seed production often seems to be limited by pollen receipt (Knight *et al.* 2006; although see Section 4.3.1), stimulating the claim that Bateman's principle often may not apply to plants. Although this claim is strictly correct, pollen limitation need not imply that female and male traits therefore experience equal selection for increased mating opportunities. In particular, pollen limitation changes the nature, but not the existence, of asymmetries in the mating prospects of individual ovules and pollen grains. In particular, the greater diversity of mechanisms that can lead to pollen failure allow for greater variation in mating success through male function than through female function. Such variance differences underlie the role of sexual selection in the evolution of mating traits (Shuster and Wade 2003), including floral characteristics.

#### 4.5.2 Limits on seed production and reproductive evolution

In addition to the widely recognized roles of pollen receipt and resource availability in limiting seed production, our model exposes the possibility

of ovule limitation, which has several implications for reproductive evolution. Recognition of ovule limitation as an error in the allocation of reproductive resources to ovules versus seed production raises intriguing questions about the selection of ovule number that are beyond the scope of this chapter. In addition, identification of the possibility of ovule limitation broadens the variety of expectations for evolution of plant reproduction. The adaptive balance between pollen and resource limitation proposed by Haig and Westoby (1988; also see Ashman *et al.* 2004) is expected when selfed zygotes and seeds both have poor survival prospects compared with outcrossed zygotes and seeds (Fig. 4.3, point X'). In contrast, when selfed zygotes and seeds both survive relatively well, plants should be completely ovule-limited (Fig. 4.3, point S). Finally, if selfed zygotes survive relatively poorly, but selfed seeds are relatively successful, compared with outcrossed seeds, then the optimal reproductive policy can balance pollen, ovule, and resource limitation (Fig. 4.3, point M). In the latter case, plants benefit from producing extra ovules that allow them to identify viable selfed offspring that have survived genetic death (Porcher and Lande 2005; L. D. Harder, M. B. Routley, and S. A. Richards unpublished manuscript).

Our models of mating-system evolution differ from preceding analyses because they explicitly consider the consequences of resource limitation of seed production. Intriguingly, resource limitation imposes less stringent conditions on the evolution of self-fertilization than does the ovule limitation which is implicit, if unrecognized, in most mating-system models (Table 4.3). This contrast creates the opportunity for mixed mating to maximize parental fitness, especially if pollen discounting is limited. Given that most plants produce "extra" ovules (i.e.,  $m < 1$ ), which is a necessary condition for resource limitation, this mechanism may provide one of the few general, adaptive explanations for the common occurrence of mixed mating (see Goodwillie *et al.* 2005).

The theory presented in this chapter follows Lloyd's (1979, 1992) lead of expanding the analysis of mating-system evolution beyond the traditional genetic approach, which emphasized inbreeding

depression (e.g., Lande and Schemske 1985; Charlesworth and Charlesworth 1987), to consider the influences of ecological factors (also see Uyenoyama *et al.* 1993; Goodwillie *et al.* 2005; Chapters 2, 6, 8, 10 and 12). A specific contribution of our models is the recognition that the post-dispersal performance of selfed seeds relative to outcrossed seeds ( $d_s/d_x$ ) provides a more general criterion for mating system evolution than does lifetime inbreeding depression (see Table 4.3). In addition, our models link mating-system evolution directly to the fates of pollen and ovules and the alternative limits on seed production. This theory, and that of others, integrates the evolution of floral, fruit, and seed characteristics with that of the mating system, illustrating the interplay between reproductive ecology and evolution.

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