EVOLUTIONARY INTERPRETATIONS OF DIFFERENCES IN POLLEN TUBE GROWTH RATES

N. E. WALSH AND D. CHARLESWORTH

Department of Ecology and Evolution, University of Chicago
Chicago, Illinois 60637 USA

ABSTRACT

The striking opportunities for competition in the pollination stage of the life cycle of flowering plants have been remarked on for many years, yet it is still not clearly known how much genetic variation for the rates of growth of pollen tubes exists in natural populations. Here we review the evidence for such variation, and discuss some of the possible mechanisms by which this type of variation could be maintained. If faster pollen tube growth rates tend to be correlated with higher fitness of the progeny sired, then most of the variation would be expected to be due to detrimental mutations, and therefore subject to purifying selection. If, however, variation is maintained in populations, it is likely that genetic factors for fast pollen tube growth would reduce fitness at other stages of the life cycle, resulting in negative genetic correlations between pollen and progeny quality. Future studies of pollen competition and its effects on progeny fitness components should be designed to avoid confounding effects of such factors as inbreeding depression and self-incompatibility types, enabling these possibilities to be distinguished.

INTRODUCTION

IN NATURE there may often be more pollen grains on the stigma than are necessary to fertilize all of a flower's ovules, and thus there is opportunity for competition to occur between pollen tubes. Buchholz (1922) first drew attention to the potential evolutionary significance of the haploid gametophytic stage in the life cycle of flowering plants, and Haldane (1932: 121) observed that "there is serious overcrowding at a stage in the life cycle where it can only be detected with the microscope, namely among the pollen grains."

Mulcahy (1979) has further suggested that intense selection differentials may exist among pollen grains because of the large numbers produced by plants, and that this could allow an increase in the speed of angiosperm evolution, compared with that in plants with less intense competition between male gametophytes. Haldane (1932), however, pointed out that when only a small proportion z of individuals survives some competitive process, the intensity of selection (i.e., the proportion of the more competitive type among the survivors after selection) does not increase proportionately with the number of competing individuals, but rather as the logarithm of z, a much slower increase (see Haldane, pp. 123 and 176–179). Thus, even though recessive alleles are exposed to selection at the pollination stage, and there may be competition between pollen genotypes, this stage will not necessarily be the most important selective stage in the life cycle of plants, especially if there is a proportion of loci that are not expressed during this stage.

The view that selection among pollen is important in plant evolution also assumes that

The Quarterly Review of Biology, March 1992, Vol. 67, No. 1

0033-5770/92/6701-0002$01.00

pollen genotypes with fast pollen tube growth rates tend to produce progeny with high fitness (Mulcahy and Mulcahy, 1975, 1987; Mulcahy, 1979). If pollen grains that grow more rapidly than others have an advantage in access to unfertilized ovules, genes for accelerated growth should spread in a population, even if these effects were opposed by selection in the opposite direction at some other life stage. Conversely, mutations causing slower growth should be eliminated. Consequently, selection in the pollination stage of the life cycle should lead to fixation of alleles giving maximal pollen tube growth rates, as well as being a form of purifying selection against mutations that cause slower growth. In either case, there should be little extant genetic variation in growth rates.

Attention has been focused recently on the possibility that variation is nevertheless present, and that genetically determined interactions between pollen and the style may thus play an important role in angiosperm evolution. These discussions have often been in terms of sexual selection, with female choice between, and competition among, pollen grains (Stephenson and Bertin, 1983; Willson and Burley, 1983; Bookman; 1984; Bertin, 1986; Marshall and Ellstrand, 1986; Mulcahy and Mulcahy, 1987; Queller, 1987). These ideas have motivated several empirical studies of pollen tube competition. Here we review the current state of knowledge about differences in pollen tube growth.

POSSIBILITIES FOR THE MAINTENANCE OF GENETIC VARIATION AFFECTING POLLEN CHARACTERISTICS

For pollen competition to have evolutionary significance, genetic variation in pollen tube growth rates must exist. As mentioned above, however, selection in the pollination stage of the life cycle would seem most likely to lead to fixation of alleles for the best-competing phenotype, and thus to loss of any genetic variation that might be present. It is known from theoretical studies that a population at equilibrium under a constant selection regime with no sex differences in fitness should exhibit no additive genetic variance in fitness components even with nonrandom mating, frequency-dependent selection, epistasis, linkage disequilibrium, and strong selection (Fisher, 1930; Kimura, 1958; Maynard Smith, 1978; B. Charlesworth, 1987). The mechanisms that could maintain additive genetic variance in pollen tube growth rate are therefore limited. Two of the possibilities, migration and environmental heterogeneity, are limited to specific environmental conditions. These may be quite frequent in natural populations, but it is not clear that they will operate generally for a high proportion of loci (Lande, 1988). Although selection in the haploid stage has not received much attention, the theoretical conclusion that variation is unlikely to be maintained by such selection remains valid (Haldane and Jayakar, 1963; Gliddon and Strobeck, 1975).

Mutation, however, "is a ubiquitous factor acting to maintain genetic variance in virtually every character in all populations" (Lande, 1976, 1988: 83), and could potentially maintain additive genetic variance in pollen tube growth rates. The magnitude of the variance maintained by mutation depends on the per genome mutation rate for loci affecting pollen tube growth, as well as on the magnitude of the effects of the mutations (B. Charlesworth, 1987). Mutational variation might be expected to lead to better offspring fitness of progeny sired by the fastest pollen tubes, because the offspring would also benefit from the lower genetic load of the faster pollen genotypes (Charlesworth and Charlesworth, in press). In other words, according to this hypothesis, fast pollen tube growth will be an indication of overall high fitness in all life stages, as assumed by Mulcahy (1979), to the extent that loci expressed in pollen are expressed at other life stages. The evolutionary importance of this possibility means that we should examine critically the large body of evidence (see references in Snow, 1990a) that situations involving pollen competition lead to superior progeny, to see whether these effects are genetic.

Another possibility is that alleles for faster pollen tube growth may tend to have unfavorable effects at other stages in the life cycle (Falconer, 1981). Antagonistic pleiotropic effects of this kind can maintain additive genetic variance in components of fitness, even when fitness itself has no additive genetic variance

(Lande, 1982; Rose, 1982; B. Charlesworth, 1987). In this case, the expectation would be that genotypes with fast pollen tube growth would have lower fitness at some other life stage, the opposite of Mulcahy's (1979) view. At present, we have no evidence for any such effects, though a mutation in maize has been found that not only causes increased pollination ability of the pollen, but also very low survival of the female gametophyte (Singleton and Mangelsdorf, 1940).

For competition among pollen tubes to have evolutionary importance, several conditions must be met. We will review empirical evidence pertaining to each of the following questions:

- Is there more pollen present on stigmas than is necessary to fertilize all of a flower's ovules?
- Does pollen express its own or its paternal parent's genotype during pollen tube growth?
- Is there genetic variation in pollen tube growth rates, and do faster-growing pollen tubes fertilize more ovules?
- Are the offspring of faster-growing pollen tubes more fit than those of slower-growing pollen tubes?

EVIDENCE FOR EXCESS POLLEN ON STIGMAS

For pollen tubes to experience a potentially competitive environment in the style, some pollen tubes must be excluded from access to unfertilized ovules, and only the fastest growing tubes should be successful in fertilizing ovules. Clearly, if seed set is limited by the amount of pollen that a given flower receives, neither pollen tube competition nor sexual selection are likely to occur (Snow, 1986). Typically, flowers produce large numbers of pollen grains. The common assumption that most flowers receive more than enough pollen to produce a full complement of seeds is supported by evidence that in some species pollen is in excess of the amount needed for fertilization of the ovules, at least in some seasons (Waser, 1978; Willson et al., 1979; Hoekstra, 1983; Mulcahy et al., 1983; Stephenson and Bertin, 1983; Namai and Ohsawa, 1986; Levin, 1990). In other species, however, seed set is pollen limited, such as *Arisaema triphyllum* (Bierzychudek, 1981), *Erythronium albidum* (Schemske et al., 1978), *Phlox divaricata* (Willson et al., 1979), and *Campsis radicans* (Bertin, 1982). In a summary of this literature, Snow (1986) found that the seed set of only 6 of 25 species investigated was limited by adequate pollen deposition.

Snow (1986), however, has pointed out that we must view these data with caution for a number of reasons. First, even in species that are pollen limited, some individual flowers may receive excess pollen, and thus there may be an opportunity for pollen competition to occur. Second, there may be competition even when pollen receipt is less than the level that ensures full seed set, because of differences in the probability of seed maturation of different ovules (Nakamura, 1988). If the fastest pollen tubes can preferentially fertilize the ovules that are most likely to be matured, they would enjoy an advantage (reviewed by Rocha and Stephenson, 1990).

Finally, the opportunity for pollen competition is affected not only by the absolute number of pollen grains that reach the stigma, but also by the rate at which they arrive. If pollen arrives gradually over time, the first cohort to arrive on the stigma will grow in a noncompetitive situation if it contains fewer pollen grains than available ovules. In this case, all pollen grains, regardless of the speed of their growth, will fertilize ovules. The timing of pollen arrival, as opposed to the growth rate of pollen tubes, may thus determine which pollen tubes are successful (Mulcahy et al., 1983; Snow, 1986). In *Epilobium canum*, 50 to 70 percent of the flowers in a natural population were not pollen limited in their seed set but, because of differences in pollen arrival, Snow (1986) calculated that only 17 to 20 percent of the fruits were produced from crowded and potentially competitive styles. For *Phlox drummondii*, an outcrossing species, Levin (1990) found in a study of 19 natural populations that 69 percent of flowers had more pollen grains in their stigmatic lobes than the number of ovules in the ovary. Mulcahy et al. (1983), however, found that although pollen arrives discontinuously in *Geranium maculatum*, phenotypic variance in pollen tube growth rates was sufficiently large to allow pollen from later pollinations to outgrow some pollen from earlier pollinations. Even

when pollen arrives simultaneously, it may not all germinate and begin to grow down the style at the same time (Thomson, 1989).

THE ROLES OF POLLEN AND MATERNAL PLANT GENE EXPRESSION IN THE POLLINATION PROCESS

Gene expression in pollen may be either gametophytic (expressing the genes carried by the individual pollen grains) or sporophytic (expressing the parent's genes), or both. Competition among gametophytes depends on gametophytic gene expression in the pollen, or on sporophytic expression of characters affecting pollen function, whereas sexual selection, defined in terms of selection based on differences in the phenotypes of pollen donors (Queller, 1987), would depend upon sporophytic expression of the pollen donors' genotypes.

Before reviewing evidence for genetic differences in pollen performance, we will briefly discuss the importance of the maternal sporophyte. Among other factors, including the paternal sporophyte, the pollen genotype, and environmental variables, the maternal sporophyte may exert an influence on pollen tube growth in angiosperms. After germination and the early stages of pollen tube growth, further growth depends on both the pollen and the stylar tissue (Ottaviano et al., 1980, 1988), so the male gametophyte does not function in an entirely passive environment.

Effects of the Stylar Environment

Pollen that has been subjected to several types of damage may still perform many of the pollination functions, perhaps by expression of preformed pollen mRNAs. Studies of pollen exposed to X rays and gamma radiation have found little or no effect on pollen tube growth rates (Brown and Cave, 1954; Pfahler et al., 1981). As Searcy and Mulcahy (1985) point out, ovules can sometimes be fertilized by such pollen even when the seeds formed are aborted (den Nijs and van den Boom, 1983). Even inhibition of all RNA synthesis does not appear to prevent germination or early pollen tube growth of *Tradescantia* pollen (Mascarenhas, 1966), though some mRNAs are normally synthesized during this stage and, as we discuss below, some X-ray-induced mutations can affect transmission by the male gametophyte.

Results with pollen substitutes confirm a strong role of the stylar environment. In three species representing open, intermediate and closed stylar types, Sanders and Lord (1989) demonstrated that latex beads moved along the same secretory matrix pathways and at the same rate as normal pollen tubes in mature styles. In two of the three species they studied, however, cutting off the styles and part of the stigma was necessary for the beads to progress down the style, indicating that pollen plays a role in the germination process in these two species.

Pollen tube growth often depends on style maturity. In the peach (*Prunus persica*), pollen tubes depend on stylar nutrients and their growth is controlled by, and correlated with, maturation of the pistil (Herrero and Arboloa, 1989). Once pollen tubes reach the base of the style, their growth is halted for several days until the pistil has matured and the ovules are receptive. A similar situation is found in almonds (*Prunus dulcis*) and pears (*Pyrus communis*). Although pollen rapidly reaches the base of the style in these species, fertilization may not occur for up to four days after all the pollen is present at the base of the style (Polito and Pimienta, 1982). Murdy and Carter (1987) found interpopulational variation for a stigma trait that delays pollen germination for two hours in *Talinum mengesii*, showing that the pollen is under the control of the style in this case.

Another influence on both germination and pollen tube growth rates is the density of pollen placed on the stigma (Jennings and Topham, 1971; Ter-Avanesian, 1978a,b; Schemske and Fenster, 1983; Cruzan, 1986; Ganeshaiah et al., 1986), though such effects are not always found (Snow, 1986). In other cases, the relationship between pollen grain or tube number and seed number may show diminishing returns (Modlibowska, 1942; Cruzan, 1989), and in walnuts high pollen loads appear to cause failure of fruit set (Kavetskaya and Tokar, 1963). Some studies indicate that secretions of both the pollen and the stigma cause this pollen population effect (Brewbaker and Majumder, 1961; Jennings and Topham, 1971), but sometimes the maternal sporophyte alone is involved (Ter-Avanesian,

1978a,b; Ganeshaiah et al., 1986). Ganeshaiah et al. (1986) found that the percent germination in vitro of *Leucaena leucocephala* pollen was relatively constant in water, despite wide variation in pollen grain number, but germination varied greatly in stigmatic fluid. Thus the stigmatic secretions affected the relationship between number of pollen grains per stigma and percent germination. Malti and Shivanna (1985) also found an increase in the mean pollen tube length of *Crotalaria retusa*, and greater variance in pollen tube growth rates in media containing pistil leachates.

Perhaps the strongest evidence implicating the pistil as an important factor in the growth of pollen tubes comes from studies that indicate that the outcomes of pollen tube growth competition differ among maternal sporophytes (reviewed by Stephenson and Bertin, 1983). The role of the maternal sporophyte in preventing the growth of incompatible pollen in self-incompatible species is well known (de Nettancourt, 1977). Studies in self-compatible species have also found differences between the success of two pollen types in competitive situations, depending on the maternal plants used. For example, self pollen has been shown to be competitively inferior to pollen from unrelated donor plants, even in self-compatible species (Darwin, 1876: 394, 398–399; Bateman, 1956; Pfahler, 1965), and in *Dianthus chinensis* this has been shown to be due to slower growth of self pollen (Aizen et al., 1990). Jones (1928) showed different fertilization ability of pollen from different maize strains, using standard recipients, and Pfahler (1967) found that differences between different nonself donors depend on the recipient maternal genotypes.

These studies show that pollen tube growth is not merely a reflection of pollen genotype. Certainly any study that attempts to document differences in pollen tube growth rates must use a sufficiently large number of maternal plants to be able to distinguish between maternal and pollen effects.

Expression of Genetic Functions by Pollen

The studies of pollen competition just mentioned also show that pollen functions can be important in the pollination process (see review by Heslop-Harrison and Heslop-Harrison, 1989). The classical genetic work on mutant alleles with gametophytic effects on pollen function provides further evidence (reviewed by Haldane, 1932 and Tanksley et al., 1981). Brink and MacGillivray (1924) and Demerec (1924) found that maize plants heterozygous for the "waxy" endosperm character produced waxy and starchy pollen grains in approximately equal numbers, indicating gametophytic expression of these genes. Similar results were found for the mutant "Tricarpel" in *Datura* (Buchholz and Blakeslee, 1927), for small pollen size in maize (Singleton and Mangelsdorf, 1940), and for other pollen lethals in several species (e.g., Williams and Rouse, 1990, who showed 1:1 segregation in tetrads in a *Rhododendron* species). Evidence for sporophytic effects on pollen function comes from findings of improved fertilization ability of pollen of F1 hybrids between maize strains (Murakami and Yamada, 1972), and of lower pollen function when the donor plants are inbred (Johnson and Mulcahy, 1978; Willis, pers. commun.). It is not known in these cases, however, whether the effects are due to differences in pollen viability, germination ability, or growth rates, or even whether the effects are due to sporophyte influences on the pollen produced (Johnson and Mulcahy, 1978). Gametophytic and sporophytic self-incompatibility systems also show that both these forms of gene expression can occur (see De Nettancourt, 1977; D. Charlesworth, 1987; and Bernatsky et al., 1988 for reviews).

Here, however, we are interested in whether many loci are expressed in pollen and are important in pollen function, and also whether the same loci are expressed in the sporophyte stage. Studies of gene expression in pollen give us the clearest evidence for gametophytic gene expression, and can provide quantitative estimates of the numbers of loci that are expressed in pollen and the proportion that are also expressed in the sporophyte tissues.

Gel electrophoresis of pollen enzymes from heterozygous parents is one such source of evidence. Dimeric enzymes contain two subunits. If the enzymes found in pollen are products of the diploid sporophytic tissues, one would expect to see both the homodimeric and the heterodimeric forms of such enzymes. If, however, the enzymes are synthesized by the pollen grains expressing their own ge-

TABLE 1
Results of tests for gametophytic expression of genes in pollen

Species	Total no. of enzymes	No. of enzymes gametophytic	References
ANGIOSPERMS			
Zea mays	1	1	Schwartz, 1971
	29	12	Sari Gorla et al., 1986
Lycopersicon esculentum	7	7	Tanksley et al., 1981
Clarkia spp.	2	2	Weeden and Gottlieb, 1979
Malus domesticus	5	5	Weeden, 1983
Brassica campestris	1	1	Singh and Knox, 1986
Populus spp.	6	6	Rajora and Zsuffa, 1986
Hordeum spp.	2	2	Pederson et al., 1988
FERNS			
Pellaea andromedaefolia	3	3	Gastony and Gottlieb, 1982

nomes, only homodimers are expected for these loci. Results from studies using dimeric enzymes are summarized in Table 1, showing clearly that pollen expresses some of the same genes as are expressed in the sporophyte stage. Although these studies give the most conclusive evidence to date for considerable overlap in the sets of loci expressed in the two stages, isozymic genes may not be representative of all genes in pollen, especially as many of these are probably "housekeeping genes" that may be particularly likely to be expressed in all life stages (Tanksley et al., 1981). Certainly, it is not possible to generalize these results from a few specific genes to the entire gametophytic genome.

The results from isozymes are supported by studies of mRNA. At anthesis, a mature pollen grain contains stored mRNAs that were synthesized during its development (Mascarenhas, 1966, 1975, 1989; Tupy, 1982; Mascarenhas et al., 1984; Stinson et al., 1987). During early pollen tube growth in tomato, pollen-specific mRNAs not present in sporophytic tissues have been found (Mascarenhas, 1966), and some pollen mRNA species are known to be synthesized after meiosis (Mascarenhas, 1966; Stinson et al., 1987; Schrauwen et al., 1988). In many taxa, after microspore meiosis, a thick callose wall surrounds the pollen grains, and major physiological contact between tapetal (sporophytic) tissues and the pollen cells has ceased (Mascarenhas, 1975; Shivanna and Johri, 1985). This has been suggested to be a mechanism for keeping haploid development independent of sporophytic influence (see Heslop-Harrison and Heslop-Harrison, 1989; Ottaviano and Mulcahy, 1989).

The numbers of genes expressed in the microgametophyte have been estimated from rates of hybridization between cDNA prepared from mature pollen RNA. Willing and Mascarenhas (1984) found that the total complexity of poly(A)RNA in *Tradescantia paludosa* pollen was 2.3×10^7 nucleotides, corresponding to approximately 20,000 transcripts, mostly of low abundance. *Zea mays* pollen has a total complexity of 2.4×10^7 nucleotides (Willing et al., 1988).

Clearly, therefore, pollen is capable of producing mRNA. Although the production of some mRNAs by pollen does not prove that all mRNAs found in pollen are produced gametophytically, this evidence, like the isoenzyme evidence, supports the view that some loci have gametophytic expression. The proportion of mRNA species that correspond to loci expressed only in pollen, versus loci expressed in both pollen and sporophytic tissues, can be estimated by hybridization studies using randomly chosen cDNA clones. Stinson et al. (1987) found by performing colony hybridization tests that about 20 percent of the clones derived from *Tradescantia paludosa* pollen and 10 percent of maize clones are pollen specific, though the the total numbers of clones tested are not stated. It would also be interesting to study mRNAs of different abundance categories, because low-abundance types may be the ones most likely to show tissue- and stage-specific expression. They also form the majority of loci.

Overall, these studies provide good evi-

dence for gametophytic expression of many genes. Studies of transmission of genetic markers through the male gametophyte can provide evidence about the frequency with which genomic regions contain factors with effects on pollen function. It has long been known that heterozygotes for chromosomes carrying deletions (unless these remove only heterochromatic regions) frequently produce 50 percent abnormal pollen, which is usually nonfunctional, but sometimes viable and associated with lower than normal transmission via pollen (Stadler and Roman, 1948; Khush and Rick, 1967). Even when homozygotes for the deletions are lethal in the sporophyte stage, deletion chromosomes can sometimes be transmitted via pollen, at least in *Arabidopsis*, which has short styles (Meinke, 1982; Meinke et al., 1985). This shows that fewer loci are needed for the male gametophyte stage than for the sporophyte, but the results nevertheless suggest that many regions of the genome carry loci that are important for pollen function. Smith (1963) found a case of a deletion of part of the X chromosome of the dioecious plant *Silene alba* that caused failure of pollen bearing this chromosome to function, so that pollination by a male plant with this chromosome produced all male progeny. As Haldane (1933) first pointed out, recessive mutations at loci that are expressed during the haploid stage of plant growth will be exposed to selection. This may account for the rarity of degenerated sex chromosomes in dioecious species of angiosperms.

Further support comes from the finding that a high proportion of induced mutations affect transmission by the male gametophyte in *Arabadopsis thaliana* (Meinke, 1982; Dellaert, 1980) and *Hordeum vulgare* (Moh and Nilan, 1956). Indeed, Stadler and Roman (1948: 278) state that "trials of male transmission frequency are complicated by the fact that there are many unidentified pollen tube growth factors in maize. . . ." Some cases have been found of apparent unequal segregation of alleles in heterozygotes and, using electrophoretic markers, the possibility of postzygotic survival differences has sometimes been ruled out (Wendel et al., 1987). These interesting effects, however, were detected only in the F2, but not in backcrosses, perhaps because of the much smaller numbers of backcross progeny studied. These data also suggest that many genomic regions may carry genes with effects on fertilization ability of pollen. Further studies of this kind would be valuable.

Good evidence for the expression of different sets of loci in the sporophyte and gametophyte stages has been obtained in ferns, in which it has been shown that some gametophytes carry loci that are lethal to sporophytes when homozygous, even though the haploid carrier gametophytes are both viable and fertile (reviewed by Klekowski, 1988). More studies of ferns, and perhaps mosses, would be very valuable and could give evidence about the numbers of loci expressed in these two stages. As far as we are aware, there are at present very few studies of the expression of isozyme loci in these two stages for comparison with those cited above for pollen and sporophytes of angiosperms, other than the work of Gastony and Gottlieb (1982), even though isozyme studies of ferns and mosses have shown that they often have considerable genetic variation (e.g., Soltis and Soltis, 1986; Wyatt et al., 1989).

GENETIC VARIATION IN POLLEN TUBE GROWTH RATES AND FERTILIZATION ABILITIES

Pollen tubes that grow faster than others should reach the ovary in less time and should, therefore, enjoy increased access to unfertilized ovules causing nonrandom fertilization based upon differences in pollen genotypes expressed during pollen tube growth. Evidence for differences in pollen tube growth rates and siring ability comes from several different kinds of studies. It is important to distinguish between those that can rigorously show differences in the growth rates themselves, estimated by one of several techniques, and those that infer growth rate differences from differing representation in the seeds or seedlings of progeny of the different pollen donors.

Results from the second type of study, using genetic markers, must be evaluated critically to be sure that the differences are not due to differences in pollen or zygote survival. If it is shown that all, or nearly all, ovules mature into seed, which is fully germinable and viable up to the stage when the identities

of the sires are scored, this type of data can satisfactorily demonstrate differences in the pollination stage. If, however, there is the possibility of differential survival or germination of seeds of the different male parents, the data are inadmissible as evidence of differences in pollen tube growth rates. In the rest of this section, we shall use the phrase "differences in pollen tube growth rates" only when the experiments actually involved measures of these rates.

It is also important to realize that differences in pollen success and competitive ability may not necessarily be genetic. When the pollen of individual males is tested, it may differ due to such differences as time of shedding (Pfahler, 1967; Pfahler et al., 1986) or environmental differences in the growth conditions of the donor plants (Young and Stanton, 1990). Studies involving comparisons between self and outcross pollen sources, or between cultivars or pollen with known genotypic differences (see below), however, do suggest that genetic differences in pollen quality must exist. After discussing some of the evidence based on individual male comparisons, we will review the data from pollen with known genetic differences, including the pertinent evidence from certation studies (see below), and from mutagenized plants. These results support the conclusion from the molecular studies discussed above, that pollen gene expression is essential for pollen function, and that genetic damage to pollen can impair its competitive ability.

Differential Success of Self and Outcross Pollen

One of the situations that would seem the most promising for detection of different pollen tube growth rates is in comparisons of the success of self pollen versus that of pollen from unrelated donors. There is abundant evidence that seed set is often lower on selfing than after cross-pollination, even in species that are not strictly self-incompatible (e.g., Ramirez and Brito, 1990), but studies of seed set, without evidence on pollen tube growth rates, cannot rule out the possibility that differential abortion of self versus outcrossed seeds causes the differences observed. We have already mentioned differences between self and nonself pollen in the self-compatible species *Dianthus chinensis* (Aizen et al., 1990). Other studies of self-compatible plants have found no evidence for differences, based on transmission of genetic markers to progeny seeds, using pollen mixtures. This has been shown in *Geranium caespitosum* (Hessing, 1986) and *Chamaecrista fasciculata* (Fenster and Sork, 1988). Differences in rates were found by direct measurement of pollen tube growth in *Amsinckia grandiflora* (Weller and Ornduff, 1989), *Erythronium grandiflorum* (Cruzan, 1989), and *Delphinium nelsonii* (Waser et al., 1987), and also by studying the transmission of a genetic marker to progeny in *Clarkia unguiculata*, a species with no seed abortion, so that differential abortion of self versus outcrossed seeds could not have caused the observed differences in paternal success in seed production (Bowman, 1987). In these cases, however, the differences may be due to weak self-incompatibility systems, as these four species are known or thought to be self-incompatible in at least some populations, or to be closely related to self-incompatible species. Alternatively, there may be cryptic self-incompatibility, i.e., expression of an incompatibility reaction that is detectable only when compatible pollen is also present. This probably happens because growth of incompatible pollen tubes is slower than that of compatible ones, rather than being completely inhibited. It may thus be a phenomenon similar to weak incompatibility.

It is important to realize that differential success of pollen genotypes can also arise during the stage of pollen germination, before pollen tube growth begins (Sprague, 1933). Preferential cross-fertilization, when self and foreign pollen are applied to stigmas, has also been found in *Phlox*, but the differences in self versus outcross pollen fertilization ability were due to differences in germination, not in pollen tube growth rates (Levin, 1975).

Johnson and Mulcahy (1978) found that in maize the competitive ability of self pollen increased as the number of generations of selfing increased. They further determined that differences in pollen tube growth rates, as opposed to germination, were the cause of the increased fertilization ability of the self pollen, but the cause of this change is not known. Pfahler (1967) and Mulcahy (1974) suggested that this ability of self pollen to outcompete nonself pollen stems from the evolution by

pollen of specialization to the stylar environment of particular inbred strains. Alternatively, it could be due to elimination of deleterious alleles in the selfed populations.

Differential Success of Pollen from Different Donors

Differences in pollen success have been found between different compatible donors in several species with self-incompatibility systems, using genetic markers to estimate the ability of different plants to sire seeds (Bertin, 1982, 1985; Marshall and Ellstrand, 1986). In *Rhaphanus raphanistrum*, differential abortion was ruled out, but not the possibility that differences, which were sometimes absolute, were due to cryptically acting self-incompatibility alleles (Snow and Mazer, 1988). Furthermore, the variation observed was not heritable. Recently, Snow and Spira (1991) have combined the methods of pollen tube growth rate estimation and marker transmission to the seeds in *Hibiscus moscheutos* plants from a natural population, to show that plants whose pollen grows fastest also sire higher proportions of the progeny. Interestingly, they found that rankings of pollen tube growth rates were consistent in the styles of different recipient plants, unlike the findings in some other species (Bertin, 1982). Numerous in vitro studies of pollen have also found differences among plants in pollen tube growth rates (Schemske and Fenster, 1983; Stephenson and Bertin, 1983). In vivo verification of in vitro results, however, tends to be difficult (Sari Gorla et al., 1975; Stephenson and Bertin, 1983; Mazer, 1987).

Different Success of Pollen from Different Cultivars or Species

Differences in pollen tube growth rates have been found among various strains of inbred cultivated plants, or produced by selecting for fast growing pollen tubes (Pfahler, 1967; Mulcahy, 1971; Sari Gorla et al., 1975; Johnson and Mulcahy, 1978; Ottaviano et al., 1980; Gawel and Robacker, 1986). Similar differences in pollen tube growth rates between plants from different populations could be due to genetic variation that exists between populations or to partial reproductive isolation between populations (Stebbins, 1950; D. Charlesworth et al., 1987). Studies such as Bookman's (1984), which compared pollinations between plants of different populations, may involve such wide crossing effects. Both these types of differences are likely to be due to differences at many loci. Despite the fact that we therefore cannot expect these results to represent accurately the differences in pollen tube growth rates within natural plant populations, they nonetheless establish that genetic differences in pollen tube growth rates are possible.

In studies of *Zea mays* (Jones, 1928) and *Lycopersicon esculentum* (Hornby and Li, 1975), within-strain pollen outcompetes pollen from other strains with respect to fertilization of ovules. Jones (1928) showed that the differences in fertilization ability were due to differences in pollen tube growth rates. Ottaviano et al. (1975) also found differing fertilization abilities attributable to variation in pollen tube growth rates among strains of *Zea mays*. Differences in siring ability associated with genetic markers have also been found, using pollen of hybrids between two tomato species, thus showing that the pollen behavior was determined gametophytically, rather than by the donor genotypes (Zamir et al., 1982).

Certation

Certation is frequently defined as a shifting of the sex ratio due to differences in fertilization success through competition in pollen tube growth rates between pollen tubes with different mechanisms of sex determination (Rieger and Michaelis, 1958), or more generally as pollen tube competition between different pollen genotypes resulting in unequal chances of fertilization success (Van Breukelen, 1982). It is thus simply another example of the type of phenomenon we are discussing here, and is similar to differences between cultivars or races, in that multiple genetic differences must exist between the competing genotypes. A number of studies have demonstrated certation in several different types of situation.

The first study of this phenomenon involved pollen carrying different translocation complexes in *Oenothera*. Pollen with the *rubrinervis* translocation complex fertilizes more ovules than pollen with a different complex. Although different pollen tube growth rates had earlier been suspected to be the cause, the

first convincing proof of this was by Heribert-Nielsson (1923), who showed by experiments involving cutting off styles at different times after pollination, that pollen with the *rubrinervis* translocation complex grows faster than pollen with the competing complex. The difference in segregation ratios of the complexes does not occur when the heterozygous parent is the female parent, further demonstrating that the effect is not caused by any difference in zygote survival. This type of effect has been found with other translocation complexes in *Oenothera*, and it has been shown that the degree to which the progeny were biased depends on the length of the style in which they grew and, as in the dioecious species discussed below, heavy loads of pollen resulted in the most skewed ratios (reviewed by Cleland, 1972).

Correns (1928) found that changing the amount of pollen that maternal plants received changed the sex ratio in the offspring of two dioecious species (*Silene alba* and *Rumex acetosa*). When pollen loads were low, the sex ratios were close to 1:1, but with high loads more than 50 percent of the offspring produced were female (Correns, 1928). Other cases are reviewed by Lloyd (1974). Increased pollen loads in *Rumex* (Smith, 1963; Rychlewski and Zarzycki, 1975; Conn and Blum, 1981) also led to female-biased sex ratios. Since the biased sex ratios were present in seedlings with close to 100 percent germination, differential pollen tube growth rates are the most likely explanation for the female-biased ratios.

Correns attributed this skewing of the sex ratio toward females to slower growth of pollen tubes of male- rather than of female-determining pollen grains, i.e., to different growth rates of pollen tubes bearing Y and X chromosomes, respectively. Since the Y chromosomes of many organisms are genetically degenerated, this may lead to their having lower competitive ability (Smith, 1963; Charlesworth, 1978). Evidence for loss of genetic functions of the Y chromosomes of plants comes from a few species in which the YY genotype is known to be inviable, but there is no direct evidence that this loss of functions affects the pollen. Heterochromatinization of the Y chromosomes of plants has hardly been studied (Zuk, 1969). It would be very worthwhile to have more information on plant Y chromosomes. Further studies of the species investigated by Correns have sometimes failed to replicate his results (Mulcahy, 1967), and a direct study of pollen tube growth found no differences in pollen tube growth rates between male- and female-determining pollen grains (Carroll and Mulcahy, 1990). These discordant results may be due to different degrees of degeneration of different Y chromosomes, and there is some evidence for effects of different male plants in the sex ratios produced (Correns, 1928: 56–57; Lawrence ,1963), though it is not known whether the variation is genetic or environmental. There is also karyotypic variation in Y chromosomes in *Rumex* (Zuk, 1969).

Certation is also known in some other situations. Some studies have found differences in pollen tube growth rates between pollen of different ploidy levels. In *Solanum tuberosum* and *Solanum phureja*, Van Breukelen (1982) found that 2x pollen grew faster than normal haploid pollen in both 4x and 2x styles. These results, however, do not indicate a general trend across species with polyploidy. Van Bruekelen (1982) reviewed evidence from numerous studies that found that haploid pollen grows faster, and others that found no differences between pollen tube growth rates of pollen with different ploidy.

ARE OFFSPRING OF FASTER-GROWING POLLEN TUBES MORE VIGOROUS?

In this section, we review both evidence that faster-growing pollen tubes produce more vigorous offspring and studies that have examined the heritability of increased vigor in the offspring.

Many studies have found a positive relationship between pollination intensity and progeny fitness (Mulcahy, 1974; Mulcahy and Mulcahy, 1975; Ottaviano et al., 1980; McKenna and Mulcahy, 1983; Ottaviano et al., 1983; McKenna, 1986; Stephenson et al., 1986; Davis et al., 1987; Winsor et al., 1987; Marshall and Whittaker, 1989; Bertin, 1990). Other studies have not found this positive correlation (Ter-Avenesian, 1978a; Snow, 1990a,b). Finally, two studies found inconsistent effects (Lee and Hartgerink, 1986; McKenna, 1986).

Studies of this kind have been criticized for a number of reasons. Charlesworth et al.

(1987) pointed out that differences in progeny variance (e.g., Ter-Avenesian, 1978a) or quality have been found even in inbred strains and cultivars, where one would expect genetic differences to be minimal (Mulcahy, 1971; Mulcahy and Mulcahy, 1975; Ottaviano et al., 1980; Stephenson et al., 1986; Davis et al; 1987). Schlichting et al. (1990) found that the effects of pollen loads on progeny vigor found by Stephenson et al. (1986) in zucchini were not strongly heritable, and similar conclusions were reached for *Raphanus* (Snow and Mazer, 1988; Snow, 1990a). This suggests that nongenetic causes may affect these progeny measures. Furthermore, Mazer (1987), using single pollen donors rather than mixed pollinations, found no significant additive genetic variance in male performance for ovule fertilization, ovule growth, number of seeds per fruit, or seed weight per fruit in wild radish. Her data from this naturally occurring plant seem to indicate that nongenetic maternal and other environmental effects may be a major cause of such differences.

If possible, studies of the effect on progeny of pollen competition should endeavor to create situations with different degrees of competition, but no other differences. Clearly, this is not easy to achieve. Many studies failed to consider that high pollen loads alone may stimulate the maternal parent to allocate more resources to fruits (see Charlesworth, 1988 for discussion). Thus better quality offspring may be a result of the pollen load itself, rather than competition among pollen tubes. It is possible that flowers can respond to high pollen tube growth rates, and that similar effects on progeny quality may occur from this cause (see Becerra and Lloyd, unpub.).

A further problem arises from differences in seed sizes and numbers between the two treatments (e.g., Mulcahy and Mulcahy, 1975). In many plants, average seed weight decreases as the number of seeds within a fruit increases (see Stanton, 1984a for further references). Thus seeds from many-seeded fruits may be smaller on average than seeds from few-seeded fruits. If heavier seeds (low pollination treatments) produce the most vigorous offspring, this could merely be an effect of seed size on offspring fitness. It has been well documented that differences in seed size are known to affect seedling fitness characters such as germination, seedling size and survivorship (Schaal, 1980; Weis, 1982; Zimmerman and Weis, 1983; Winn, 1985; Gross and Kromer, 1986). Some studies have gone on to document seed-size effects on adult life stages (Mulcahy, 1979; Hutchinson, 1984; Stanton, 1984b). In all these instances, seed size was positively correlated with the fitness measures.

In order to avoid possible differences due to seed size, seed size is sometimes matched between the two pollination treatments (Winsor et al., 1987). This, however, will not always solve the difficulty (Charlesworth et al., 1987; Charlesworth, 1988). By matching seed size, the largest seeds from the many-seeded fruits (high pollination treatment) will usually be compared to the smallest seeds from the the few-seeded fruit (low pollination treatment). Thus a comparison intended to illuminate the effects of gametophytic competition may in fact be documenting the effects of healthy versus less healthy zygotes (Charlesworth, 1988). When average-weight seeds for both treatments in the experiment of Winsor et al. (1987) on zucchini were compared, however, the average-weight seed from the high pollen load treatment had equal or superior measures of vigor than average-weight seeds from the low pollen load treatment (Stephenson et al., 1988).

The best type of comparison will therefore be between fruits containing the same numbers of seeds. This was done in a study of the effects of pollen competition in *Dianthus chinensis* cultivars by Mulcahy and Mulcahy (1975). The extent of pollen competition was manipulated by placing pollen at different positions on the long stigmatic surface. Pollination at the tip of the style should be the more competitive situation, and indeed it yielded superior progeny. In this experiment, the base-pollinated pistils yielded low average seed numbers, so the seeds from the fruits used in the progeny trials should have come from the best such fruits. Alternatively, one could work with species in which seed size does not vary with seed number. McKenna (1986) did this in her work on the effect of long versus short style length in the heterostylous species *Turnera ulmifolia* and *Anchusa officinalis*.

Furthermore, this type of study must take into account the possibility of differential

abortion before seeds fully develop. If this occurs, differences in sporophyte quality could be due to undetected differential abortion, which might not be due to individual seed quality (Charlesworth, 1988).

Bertin's (1990) recent study of the effect of pollen load on offspring quality in *Campsis radicans* avoided most of these problems. In this species seed germination rates were higher when seeds were from high, rather than low, pollen loads. Because pollen load did not affect the weight of individual seeds, this study avoided problems due to differences in seed weight. Thus the range of explanations for these results narrows to two. One possibility is that differential abortion may be occurring in the high pollen load treatments, eliminating less vigorous zygotes. Alternatively, Bertin's results may support the hypothesis that gametophytic competition among pollen tubes produces more vigorous offspring. Bertin (1990), however, does not demonstrate the genetic transmission of the improved quality of the offspring to further generations.

The vast majority of studies of offspring quality still suffer from one or more of the problems outlined above (Snow, 1990a). Also, very few attempts have been made to document the heritability of these differences (Charlesworth, 1988; Snow, 1990a). Studies that have attempted to do so have thus far not found significant heritabilities of these traits, indicating that nongenetic causes, in addition to the problems discussed above, may be the cause of the observed differences (Mazer, 1987; Snow and Mazer, 1988; Snow, 1990a,b). If the increased vigor of offspring is heritable, it will be important to find out whether the additive genetic correlation between pollen tube growth rate and offspring vigor is positive.

CONCLUDING REMARKS

The evidence reviewed here makes it clear that gene expression is important in pollen and also that, given the frequent presence of excess pollen on stigmas in nature, there may be competition between different pollen genotypes. The chief questions concern the magnitude of the genetic variation in pollen. Is this variation mainly mutational in origin, i.e., is it an expression of genetic load, so that pollen selection acts as a purifying selection process to reduce the level of variation? In this case, the fastest growing pollen may sire the best quality offspring. Or is genetic variation maintained in pollen by some balancing form of selection, most likely by negative pleiotropic effects at other stages of the life cycle? Of course, both types of genetic variation may be present. If much genetic variation is of the second type, the fastest-growing pollen may not produce progeny with the highest fitness.

Experimental tests to distinguish between these possibilities are not easy to devise. In the first place, direct evidence for the existence of genetic variation in pollen growth rates is badly needed, preferably from self-compatible species where there is no possibility that effects could be due to cryptic self-incompatibility. Evidence already exists for considerable variability in these rates (Cruzan, 1989). Genetic variation in pollen tube growth rates can be investigated using standard breeding designs, like those used in other quantitative genetic studies (Kempthorne, 1957; Falconer, 1981), or by artificial selection for faster tube growth over one or more generations (Snow and Mazer, 1988). Only when we have this information will we be in a position to proceed to study the possible interactions with different maternal genotypes and the effects on the progeny generation. If no genetic variation can be detected, there will, of course, be no need to study the causes of maintenance of the variation, though the physiological causes of the differences, and the effects on progeny quality will still be interesting questions.

To determine if faster growing pollen tubes have more or less vigorous offspring—in other words, to distinguish between the two hypotheses discussed above for the maintenance of genetic variation in pollen tube growth rates—we must be able to distinguish the effects on progeny vigor due to fast versus slow pollen tube growth rates from those due to differences in pollen loads. Although some experiments involve differences in the length of the style to be traversed, rather than differences in the amounts of pollen (Mulcahy and Mulcahy, 1975; McKenna and Mulcahy, 1983), most studies inevitably involve differences in the environment of the pollen tubes as well as possible genotypic differences. Furthermore, differences in pollen tube growth rates could cause two different types of effects

on the progeny seeds and plants. The main focus of interest here is the possibility of genetic differences. We must also bear in mind, however, the possibility that the mere presence in styles of rapidly growing pollen tubes in some, but not all flowers may stimulate the maternal plant to preferentially provision those flowers and the fruits that develop from them (Becerra and Lloyd, unpub.). Thus if we find that better quality offspring are produced by pollen with fast tube growth, compared with pollen that generates slower tube growth rates, we cannot be sure that the effect is due to genetic differences in the pollen, even if these have been shown to exist. It will evidently be difficult to detect differences due to genetic effects if there are also differences due to maternal plant responses to tube growth rates per se.

These considerations suggest that we need to study both the effects of pollen tube growth rate differences, using maternal genotypes in whose styles pollen of a single donor shows a marked difference in growth rates, and also the effects on progeny of genetic differences in pollen, using single maternal plant genotypes in which genetic differences in pollen tube growth rates are expressed. The first type of experiment could enable us to detect differences in progeny quality due to the effects of growth rate differences, in the absence of paternal genetic differences, at least if we could achieve sufficient replication over a variety of maternal plant genotypes to be able to rule out systematic differences in progeny quality due to those genotypes. The second type of experiment would estimate the sum of the effects of both genetic and nongenetic factors. Clearly, interesting results could be obtained with just the second type of experiment, but the interpretation of the results might remain uncertain.

Evidently the experiments necessary to answer the questions posed here are likely to be extremely laborious, as pollen tube growth rate is a difficult character to measure. Although the use of genetic markers to study siring ability appears to offer a means to do the necessary types of experiments with less labor than microscopic counting of pollen tubes, there may be problems with this approach. In particular, as discussed above, even if the marker alleles are neutral, they may be linked to pollen tube growth factors that may have detectable effects. This is unlikely to be a problem in studies of differences between plants in natural populations, as there should not be strong linkage disequilibrium between the two types of loci (at least based on our information from outcrossing species, see Lewontin, 1964), but it could be very important when derived strains, or different populations, are crossed. Furthermore, it is also important to ensure that there are no differences due to post-zygotic differences in progeny survival that could produce the appearance of differential transmission of markers. It should usually be possible to check for this possibility by showing that the effects are found only when the allele with low apparent transmission to the progeny comes from the male parent. Nevertheless, it is clear that marker-based methods present some difficulties and that direct measures of pollen tube growth rates would be preferable. It will therefore be important to develop new methods for such measurements, so that they can be done reliably on numerous samples (e.g., Harris et al., 1990). One possibility is the method of Snow and Spira (1991), which allows comparisons of growth rates by counting the numbers of callose plugs in style sections.

Finally, selection in the haploid stages of the life cycle is not confined to the male reproductive functions, but there may also be differential survival of the female gametophytes of angiosperms and other plants. This is rarely studied in angiosperms (see Singleton and Mangelsdorf, 1940), but inviable gametophytes are found at detectable frequencies among germinating spores in some species of ferns (Klekowski, 1984). This must evidently be due to mutations arising after the gametophyte stage of the previous generation, since both gametophyte parents must have been viable in the preceding generation.

ACKNOWLEDGMENTS

We thank A. A. Snow, T. Spira, J. Willis, J. Becerra, and D. G. Lloyd for permission to cite unpublished work, and B. Charlesworth for comments on this manuscript. This work was supported by NSF grant BSR 8817976.

REFERENCES

Aizen, M. A., K. B. Searcy, and D. L. Mulcahy. 1990. Among- and within-flower comparisons of pollen tube growth following self- and cross-pollinations in *Dianthus chinensis* (Caryophyllaceae). *Am. J. Bot.*, 77: 671-676.

Bateman, A. J. 1956. Cryptic self-incompatibility in the wall flower: *Cheiranthus cheiri* L. *Heredity*, 10: 257-261.

Bernatzky, R., M. A. Anderson, and A. E. Clarke. 1988. Molecular genetics of self-incompatibility in flowering plants. *Dev. Genet.*, 9: 1-12.

Bertin, R. I. 1982. Paternity and fruit production in trumpet creeper (*Campsis radicans*). *Am. Nat.*, 119: 694-709.

———. 1985. Nonrandom fruit production in *Campsis radicans*: between year consistency and effects of prior pollination. *Am. Nat.*, 126: 750-759.

———. 1986. Consequences of mixed pollinations in *Campsis radicans*. *Oecologia*, 70: 1-5.

———. 1990. Effects of pollination intensity in *Campsis radicans*. *Am. J. Bot.*, 77: 178-187.

Bierzychudek, P. 1981. Pollinator limitation of plant reproductive effort. *Am. Nat.*, 117: 838-840.

Bookman, S. S. 1984. Evidence for selective fruit production in *Asclepias*. *Evolution*, 38: 72-86.

Bowman, R. N. 1987. Cryptic self-incompatibility and the breeding system of *Clarkia unguiculata* (Onagraceae). *Am. J. Bot.*, 74: 471-476.

Brewbaker, J. L., and S. K. Majumder. 1961. Cultural studies of the pollen population effect and the self-incompatibility inhibition. *Am. J. Bot.*, 48: 457-464.

Brink, R. A., and J. H. MacGillivray. 1924. Segregation for the waxy character in maize pollen and differential development of the male gametophyte. *Am. J. Bot.*, 11: 465-469.

Brown, S. W., and M. Cave. 1954. Detection and nature of dominant lethals in Lilium. I. Effects of X rays on the development of the post-fertilization ovule. *Am. J. Bot.*, 41: 451.

Buchholz, J. T. 1922. Developmental selection in vascular plants. *Bot. Gaz.*, 73: 249-286.

Buchholz, J. T., and A. F. Blakeslee. 1927. Abnormality in pollen tube growth in *Datura* due to the gene "Tricarpel." *Proc. Natl. Acad. Sci. USA*, 13: 242-249.

Carroll, S. B., and D. L. Mulcahy. 1990. Unexpected progeny sex ratios in dioecious *Silene latifolia*. *Am. J. Bot.*, 77 (Suppl.): 51.

Charlesworth, B. 1978. Model for the evolution of Y chromosomes and dosage compensation. *Proc. Natl. Acad. Sci. USA*, 75: 5618-5622.

———. 1987. The heritability of fitness. In J. W. Bradbury and M. B. Andersson (eds.), *Sexual Selection: Testing the Alternatives*, pp. 21-40. John Wiley & Sons, New York.

Charlesworth, D. 1987. Self-incompatibility systems in the flowering plants. *Perspect. Biol. Med.*, 30: 263-277.

———. 1988. Evidence for pollen competition in plants and its relationship to progeny fitness: a comment. *Am. Nat.*, 132: 298-302.

Charlesworth, D., and B. Charlesworth. In press. The effects of selection in the gametophyte stage on mutational load. *Evolution*.

Charlesworth, D., D. W. Schemske, and V. L. Sork. 1987. The evolution of plant reproductive characters: sexual versus natural selection. In S. Stearns (ed.), *The Evolution of Sex and its Consequences*, pp. 317-335. Birkhauser-Verlag, Basel.

Cleland, R. E. 1972. *Oenothera: Cytogenetics and Evolution*. Academic Press, New York.

Conn, J. S., and U. Blum. 1981. Sex ratio of *Rumex hastatulus*: the effect of environmental factors and certation. *Evolution*, 35: 1108-1116.

Correns, C. 1928. Bestimmung, Vererbung und Verteilung des Geschlechtes bei den höheren Pflanzen. *Handb. Vererbungswissenschaft*, 2c: 1-138.

Cruzan, M. B. 1986. Pollen tube distributions in *Nicotiana glauca*: evidence for density dependent growth. *Am. J. Bot.*, 73: 902-907.

———. 1989. Pollen tube attrition in *Erythronium grandiflorum*. *Am. J. Bot.*, 76: 562-570.

Darwin, C. R. 1876. *The Effects of Cross and Self fertilization in the Vegetable Kingdom*. John Murray, London.

Davis, L. E., A. G. Stephenson, and J. A. Winsor. 1987. Pollen competition improves performance and reproductive output of the common zucchini squash under field conditions. *J. Am. Soc. Hortic. Sci.*, 112: 712-716.

de Nettancourt, D. 1977. *Incompatibility in Angiosperms*. Springer Verlag, New York.

Dellaert, L. M. W. 1980. Segregation frequencies of radiation-induced viable mutants in *Arabidopsis thaliana* (L.) Heydh. *Theor. Appl. Genet.*, 57: 137-143.

Demerec, M. 1924. A case of pollen dimorphism in maize. *Am. J. Bot.*, 11: 461-464.

den Nijs, A. P. M., and A. van den Boom. 1983. Competition between irradiated and fresh pollen in *Cucumis sativis* L. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 397-404. Elsevier, New York.

Falconer, D. S. 1981. *Introduction to Quantitative Genetics*. 2nd ed. Longman, London.

Fenster, C. B., and V. L. Sork. 1988. Effects of crossing distance and male parent on *in vivo*

pollen tube growth in *Chamaecrista fasciculata*. *Am. J. Bot.*, 75: 1989–1903.

Fisher, R. A. 1930. *The Genetical Theory of Natural Selection*. 2nd rev. ed., 1958. Dover Publications, New York.

Ganeshaiah, K. N., R. U. Shaankar, and G. Shivashankar. 1986. Stigmatic inhibition of pollen grain germination—its implication for frequency distribution of seed number in pods of *Leucaena leucocephala* (Lam) de Wit. *Oecologia*, 70: 568–572.

Gastony, G. J., and L. D. Gottlieb. 1982. Evidence for genetic heterozygosity in a homosporous fern. *Am. J. Bot.*, 69: 634–637.

Gawel, N. J., and C. D. Robacker. 1986. Effect of pollen-style interaction on the pollen tube growth of *Gossypium hirsutum*. *Theor. Appl. Genet.*, 72: 84–87.

Gliddon, C., and C. Strobeck. 1975. Necessary and sufficient conditions for multiple-niche polymorphism in haploids. *Am. Nat.*, 109: 233–235.

Gross, K. L., and M. L. Kromer. 1986. Seed weight effects on growth and reproduction in *Oenothera biennis* L. *Bull. Torrey Bot. Club*, 113: 252–258.

Haldane, J. B. S. 1932. *The Causes of Evolution*. Harper, New York.

———. 1933. The part played by recurrent mutation in evolution. *Am. Nat.*, 42: 5–19.

Haldane, J. B. S., and S. D. Jayakar. 1963. Polymorphism due to selection of varying direction. *J. Genet.*, 58: 237–242.

Harris, P. J., K. Freed, M. A. Anderson, J. A. Weinhandl, and A. E. Clarke. 1990. An enzyme-linked immunosorbent assay (ELISA) for in vitro pollen growth based on binding of a monoclonal antibody to the pollen surface. *Plant Physiol.*, 84: 851–855.

Heribert-Nielsson, N. 1923. Zertationsversuche mit durchtrennung des griffes bei *Oenothera lamarckiana*. *Hereditas*, 4: 177–190.

Herrero, M., and A. Arbeloa. 1989. Influence of the pistil on pollen tube kinetics in peach (*Prunus persica*). *Am. J. Bot.*, 76: 1441–1447.

Heslop-Harrison, J., and Y. Heslop-Harrison. 1989. Actomyosin and movement in the angiosperm pollen tube: an interpretation of some recent results. *Sex. Plant Reprod.*, 2: 199–207.

Hessing, M. B. 1986. Pollen growth following self- and cross-pollination in *Geranium caespitosum* James. In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 467–472. Springer-Verlag, New York.

Hoekstra, F. A. 1983. Physiological evolution in angiosperm pollen: possible role of pollen vigor. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 35–41. Elsevier, New York

Hornby, C. A., and S.-C. Li. 1975. Some effects of multiparental pollination in tomato plants. *Can. J. Plant Sci.*, 55: 127–131.

Hutchinson, E. S. 1984. Seed size and quantitative characters in *Avena barbata*. *Heredity*, 52: 25–33.

Jennings, D. L., and P. B. Topham. 1971. Some consequences of raspberry pollen dilution for its germination and for fruit development. *New Phytol.*, 70: 371–380.

Johnson, C. M., and D. L. Mulcahy. 1978. Male gametophyte in maize: II. Pollen vigor in inbred plants. *Theor. Appl. Genet.*, 51: 211–215.

Jones, D. F. 1928. *Selective Fertilization*. The Univ. of Chicago Press, Chicago.

Kavetskaya, A. A., and L. O. Tokar. 1963. Negative effect of large quantities of pollen in pollinations of walnut trees. *Bot. Zh.*, 48: 580–585.

Kempthorne, O. 1957. *Introduction to Genetic Statistics*. John Wiley, New York.

Khush, G. S., and C. M. Rick. 1967. Studies on the linkage map of chromosome 4 of the tomato and on the transmission of induced deficiencies. *Genetica* (*The Hague*), 38: 74–94.

Kimura, M. 1958. On the change of population fitness by natural selection. *Heredity*, 12. 145–167.

Klekowski, E. J. 1984. Mutational load in clonal plants: a study of two fern species. *Evolution*. 38: 417–426.

———. 1988. *Mutation, Developmental Delection, and Plant Evolution*. Columbia Univ. Press, New York.

Lande, R. 1976. The maintenance of genetic variability by mutation in a polygenic character with linked loci. *Genet. Res. Camb.*, 26: 221–235.

———. 1982. A quantitative genetic theory of life history evolution. *Ecology*, 63: 607–615.

———. 1988. Quantitative genetics and evolutionary theory. In B. S. Weir, E. J. Eisen, M. M. Goodman, and G. Namkoong (eds.), *Proceedings of the Second International Conference on Quantitative Genetics*, pp. 71–84. Sinauer, Sunderland.

Lawrence, C. W. 1963. Genetic studies on wild populations of *Melandrium*. *Heredity*, 18: 149–163.

Lee, T. D., and A. P. Hartgerink. 1986. Pollination intensity, fruit maturation pattern, and offspring quality in *Cassia fasciculata* (Leguminosae). In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 417–422. Springer-Verlag, New York.

Levin, D. A. 1975. Gametophytic selection in *Phlox*. In D. L. Mulcahy (ed.), *Gamete Competition in Plants and Animals*, pp. 207–218. North Holland, Amsterdam.

Levin, D. A. 1990. Sizes of natural microgameto-

phyte populations in pistils of *Phlox drummondii*. *Am. J. Bot.*, 77: 356–363.

Lewontin, R. C. 1964. The genetics of selection and linkage. I. General considerations; heterotic models. *Genetics*, 49: 49–67.

Lloyd, D. G. 1974. Female-predominant sex ratios in angiosperms. *Heredity*, 32: 35–44.

Malti and K. R. Shivanna. 1985. The role of the pistil in screening compatible pollen. *Theor. Appl. Genet.*, 70: 684–686.

Marshall, D. L., and N. C. Ellstrand. 1986. Sexual selection in *Raphanus sativus*: experimental data on non-random fertilization, maternal choice, and consequences of multiple paternity. *Am. Nat.*, 127: 446–461.

Marshall, D. L., and K. L. Whittaker. 1989. Effects of pollen donor identity on offspring quality in wild radish, *Raphanus sativus*. *Am. J. Bot.*, 76: 1081–1088.

Mascarenhas, J. P. 1966. Pollen tube growth and ribonucleic acid synthesis by vegetative and generative nuclei of *Tradescantia*. *Am. J. Bot.*, 53: 563–569.

———. 1975. The biochemistry of angiosperm pollen development. *Bot. Rev.*, 41: 259–314.

———. 1989. The male gametophyte of flowering plants. *Plant Cell*, 1: 657–664.

Mascarenhas, N. T., D. Bashe, A. Eisenberg, R. P. Willing, C.-M. Xiao, and J. P. Mascarenhas. 1984. Messenger RNAs in corn pollen and protein synthesis during germination and pollen tube growth. *Theor. Appl. Genet.*, 68: 323–326.

Mazer, S. J. 1987. Parental effects on seed development and seed yield in *Raphanus raphanistrum*: implications for natural and sexual selection. *Evolution*, 41: 355–371.

Maynard Smith, J. 1978. *The Evolution of Sex*. Cambridge Univ. Press, Cambridge.

McKenna, M. A. 1986. Heterostyly and microgametophytic selection: the effect of pollen competition on sporophytic vigor in two distylous species. In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 443–448. Springer-Verlag, New York.

McKenna, M. A., and D. L. Mulcahy. 1983. Ecological aspects of gametophytic competition in *Dianthus chinensis*. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 419–424. Elsevier, New York.

Meinke, D. W. 1982. Embryo-lethal mutants of *Arabidopsis thaliana*: evidence for gametophytic expression of the mutant genes. *Theor. Appl. Genet.*, 63: 381–386.

Meinke, D. W., L. Franzmann, A. Baus, D. Patton, R. Weldon, J. D. Heath, and C. Monnot. 1985. Embryo-lethal mutants of *Arabidopsis thaliana*. In M. Freeling (ed.), *Plant Genetics*, pp. 129–146. Alan Liss, New York.

Modlibowska, I. 1942. Bimodality of crowded pollen tubes in *Primula obconica*. *J. Hered.*, 33: 187–190.

Moh, C. C., and R. A. Nilan. 1956. Reduced gene transmission in radiation-induced mutant barley. *J. Hered.*, 47: 129–131.

Mulcahy, D. L. 1967. Optimal sex ratio in *Silene alba*. *Heredity*, 22: 411–423.

———. 1971. A correlation between gametophytic and sporophytic characteristics in *Zea mays* L. *Science*, 171: 1155–1156.

———. 1974. Adaptive significance of gamete competition. In H. F. Liskens (ed.), *Fertilization in Higher Plants*, pp. 27–30. North Holland, Amsterdam.

———. 1979. The rise of the angiosperms: a genecological factor. *Science*, 206: 20–23.

Mulcahy, D. L., and G. B. Mulcahy. 1975. The influence of gametophytic competition on sporophytic quality in *Dianthus chinensis*. *Theor. Appl. Genet.*, 46: 277–280.

———, and ———. 1987. The effects of pollen competition. *Am. Sci.*, 75: 44–50.

Mulcahy, D. L., P. S. Curtis, and A. A. Snow. 1983. Pollen competition in a natural population. In C. E. Jones and R. L. Little (eds.), *Handbook of Experimental Pollination Biology*, pp. 330–337. Van Norstrand Reinhold, New York.

Murakami, K., and K. Yamada. 1972. Selective fertilization in maize, *Zea mays* L. I. Advantage of pollen from F_1 plant in selective fertilization. *Jpn. J. Pl. Breed.*, 22: 203–208.

Murdy, W. H., and M. E. B. Carter. 1987. Regulation of the timing of pollen germination by the pistil in *Talinum mengesii* (Portulacaceae). *Am. J. Bot.*, 74: 1888–1892.

Nakamura, R. R. 1988. Seed abortion and seed size variation within fruits of *Phaseolus vulgaris*: pollen donor and resource variation effects. *Am. J. Bot.*, 75: 1003–1010.

Namai, H., and R. Ohsawa. 1986. Variation of reproductive success rates of ovule and pollen deposited upon stigmas according to the different number of pollen on a stigma in angiosperm. In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 423–428. Springer-Verlag, New York.

Ottaviano, E., and D. L. Mulcahy. 1989. Genetics of angiosperm pollen. *Adv. Genet.*, 26: 1–64.

Ottaviano, E., D. Petroni, and M. E. Pe. 1988. Gametophytic expression of genes controlling development in maize. *Theor. Appl. Genet.*, 75: 252–258.

Ottaviano, E. M., M. Sari-Gorla, and I. Arenari. 1983. Male gametophyte competitive ability in maize: selection and implications with regard

to the breeding system. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 367–373. Elsevier, New York.

Ottaviano, E., M. Sari-Gorla, and D. L. Mulcahy. 1975. Genetic and intergametophytic influences on pollen tube growth. In D. L. Mulcahy (ed.), *Gamete Competition in Plants and Animals*, pp. 125–143. North Holland, Amsterdam.

Ottaviano, E., M. Sari-Gorla, and D. L. Mulcahy. 1980. Pollen tube growth in *Zea mays*: implication for genetic improvement of crops. *Science*, 210: 437–438.

Pederson, S., V. Simonsen, and V. Loeschche. 1988. Overlap of gametophytic and sporophytic gene expression in barley. *Theor. Appl. Genet.*, 75: 200–206.

Pfahler, P. L. 1965. Fertilization ability of maize pollen grains. I. Pollen sources. *Genetics*, 52: 513–520.

———. 1967. Fertilization ability of maize pollen grains. II. Pollen genotype, female sporophyte and pollen storage interactions. *Genetics*, 57: 513–521.

Pfahler, P. L., H. F. Liskens, and D. L. Mulcahy. 1981. Effect of pollen ultraviolet radiation on the abortion frequency and segregation patterns at various endosperm mutant loci in maize. *Environ. Exp. Bot.*, 21: 5–13.

Pfahler, P. L., D. L. Mulcahy, and B. Barnabas. 1986. The effect of forced shedding on pollen traits, seedsetting, and transmission at various maize (Zea mays L.) endosperm mutant loci. *Acta Bot. Neerl.*, 35: 195–200.

Polito, V. S., and E. Pimienta. 1982. A rapid and accurate fluorescence method using ovule whole mounts to assess fertilization in plants. *Mikroskopie*, 39: 32–34.

Queller, D. C. 1987. Sexual selection in flowering plants. In J. W. Bradbury and M. B. Andersson (eds.), *Sexual Selection: Testing the Alternatives*, pp. 165–179. John Wiley and Sons, New York.

Rajora, O. P., and L. Zsuffa. 1986. Sporophytic and gametophytic gene expression in *Populus deltoides* Marsh, *P. Maximowiczii* Henry. *Can. J. Genet. Cytol.*, 28: 476–482.

Ramirez, N., and Y. Brito. 1990. Reproductive biology of a tropical palm swamp community in the Venezuelan Llanos. *Am. J. Bot.*, 77: 1260–1271.

Rieger, R. A., and A. A. Michaelis. 1958. *Genetisches und Cytogenetisches Wörterbuch.* Springer-Verlag, Berlin.

Rocha, O. J., and A. G. Stephenson. 1990. Effects of ovule position on seed production, seed weight, and progeny performance in *Phaseolus coccineus* L. (Leguminosae). *Am. J. Bot.*, 77: 1320–1329.

Rose, M. R. 1982. Antagonistic pleitropy, dominance and genetic variation. *Heredity*, 48: 63–78.

Rychlewski, J., and K. Zarzycki. 1975. Sex ratio in seeds of *Rumex acetosa* L. as a result of sparse or abundant pollination. *Acta. Biol. Cracov. Ser. Bot.*, 18: 101–114.

Sanders, L. C., and E. M. Lord. 1989. Directed movement of latex particles in the gynoecia of three species of flowering plants. *Science*, 243: 1606–1608.

Sari Gorla, M., C. Frova, and M. Villa. 1986. Extent of gene expression at the gametophytic phase in maize. In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 27–32. Springer-Verlag, New York.

Sari Gorla, M., E. Ottaviano, and D. Faini. 1975. Genetic variability of gametophytic growth rate in maize. *Theor. Appl. Genet.*, 46: 289–294.

Schaal, B. A. 1980. Reproductive capacity and seed size in *Lupinus texensis*. *Am. J. Bot.*, 67: 703–709.

Schemske, D. W., and C. Fenster. 1983. Pollen-grain interactions in a neotropical *Costus*: effects of clump size and competitors. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 405–410. Elsevier, New York.

Schemske, D. W., M. F. Willson, M. N. Melampy, L. J. Miller, L. Verner, K. M. Schemske, and L. B. Best. 1978. Flowering ecology of some spring woodland herbs. *Ecology*, 59: 351–366.

Schlichting, C. D., A. G. Stephenson, L. A. Small, and J. A. Winsor. 1990. Pollen loads and progeny vigor in *Cucurbita pepo*: the next generation. *Evolution*, 44: 1358–1372.

Schrauwen, J. A. M., M. W. M. Derks, P. F. M. Groot, W. H. Reijnen, M. M. A. van Herpen, and G. J. Wullems. 1988. Differential gene-expression during microsporogenesis with *Nicotiana tabacum*. In M. Cresti, P. Gori, and E. Pacini (eds.), *Sexual Reproduction in Higher Plants*, pp. 3–7. Springer-Verlag, New York.

Schwartz, D. 1971. Genetic control of alcohol dehydrogenase—a competition model for regulation of gene action. *Genetics*, 67: 411–425.

Searcy, K. B., and D. L. Mulcahy. 1985. Pollen tube competition and selection for metal tolerance in *Silene dioica* (Caryophyllaceae) and *Mimulus guttatus* (Scrophulariaceae). *Am. J. Bot.*, 72: 1695–1699.

Shivanna, K. R., and B. M. Johri. 1985. *The Angiosperm Pollen.* Wiley Eastern Limited, Daryaganj.

Singh, M. B., and R. B. Knox. 1986. Expression of β-galactosidase gene in pollen of *Brassica campestris*. In D. L. Mulcahy, G. B. Mulcahy,

and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 3-8. Springer-Verlag, New York.

Singleton, W. R., and P. C. Mangelsdorf. 1940. Gametic lethals on the fourth chromosome of maize. *Genetics*, 25: 366-390.

Smith, B. W. 1963. The mechanism of sex determination in *Rumex hastatulus*. *Genetics*, 48: 1265-1288.

Snow, A. A. 1986. Pollination dynamics in *Epilobium canum* (Onagraceae): consequences for gametophytic selection. *Am. J. Bot.*, 73: 139-151.

———. 1990a. Effects of pollen load size and number of donors on sporophyte fitness in wild radish (*Raphanus raphinistrum*). *Am. Nat.*, 136: 742-758.

———. 1991. Effects of pollen load size on sporophyte competitive ability in two *Epilobium* species. *Am. Midl. Nat.*, 125: 348-355.

Snow, A. A., and S. J. Mazer. 1988. Gametophytic selection in *Raphanus raphanistrum*: a test for heritable variation in pollen competitive ability. *Evolution*, 42: 1065-1075.

Snow, A. A., and T. P. Spira. 1991. Pollen vigour and the potential for sexual selection in plants. *Nature*, 352: 796-797.

Soltis, D. E., and P. S. Soltis. 1986. Electrophoretic evidence for inbreeding in the fern *Botrychium virginianum* (Ophioglossaceae). *Am. J. Bot.*, 73: 588-592.

Sprague, G. F. 1939. Pollen tube establishment and the deficiency of waxy seeds in maize. *Proc. Natl. Acad. Sci. USA*, 19: 838-841.

Stadler, L. J., and H. Roman. 1948. The effect of X-rays upon mutation of the gene A in maize. *Genetics*, 33: 273-303.

Stanton, M. L. 1984a. Developmental and genetic sources of seed weight variation in *Raphanus raphanistrum* L. (Brassicaceae). *Am. J. Bot.*, 71: 1090-1098.

———. 1984b. Seed variation in wild radish: effect of seed size on components of seedling and adult fitness. *Ecology*, 65: 1105-1112.

Stebbins, G. L. 1950. *Variation and Evolution in Plants*. Columbia Univ. Press, New York.

Stephenson, A. G., and R. I. Bertin. 1983. Mate competition, female choice and sexual selection in plants. In L. Real (ed.), *Pollination Biology*, pp. 109-149. Academic Press, Orlando.

Stephenson, A. G., J. A. Winsor, and L. E. Davis. 1986. Effects of pollen load size on fruit maturation and sporophyte quality in zucchini. In D. L. Mulcahy, G. B. Mulcahy, and E. Ottaviano (eds.), *Biotechnology and Ecology of Pollen*, pp. 429-434. Springer-Verlag, New York.

Stephenson, A. G., J. A. Winsor, C. D. Schlichting, and L. E. Davis. 1988. Pollen competition, nonrandom fertilization, and progeny fitness: a reply to Charlesworth. *Am. Nat.*, 132: 303-308.

Stinson, J. R., A. J. Eisenberg, R. P. Willing, M. E. Pe, D. D. Hanson, and J. P. Mascarenhas. 1987. Genes expressed in the male gametophyte of flowering plants and their isolation. *Plant Physiol.*, 83: 442-447.

Tanksley, S. D., D. Zamir, and C. M. Rick. 1981. Evidence for extensive overlap of sporophytic and gametophytic gene expression in *Lycopersicon esculentum*. *Science*, 213: 453-455.

Ter-Avanesian, D. V. 1978a. The effect of varying the number of pollen grains used in fertilization. *Theor. Appl. Genet.*, 52: 77-79.

———. 1978b. Significance of pollen amount for fertilization. *Bull. Torrey Bot. Club*, 105: 2-8.

Thomson, J. D. 1989. Germination schedules of pollen grains: implications for pollen selection. *Evolution*, 43: 220-223.

Tupy, J. 1982. Alterations in polyadenylated RNA during pollen maturation and germination. *Biol. Plant.*, 24: 331-340.

Van Breukelen, E. W. M. 1982. Competition between 2X and X pollen in styles of *Solanum tuberosum* determined by a quick in vivo method. *Euphytica*, 31: 585-590.

Waser, N. M. 1978. Competition for hummingbird pollination and sequential flowering in two Colorado wildflowers. *Ecology*, 59: 934-944.

Waser, N. M., M. V. Price, A. M. Montalvo, and R. N. Gray. 1987. Female mate choice in a perennial herbaceous wildflower, Delphinium nelsonii. *Evol. Trends Plants*, 1: 29-33.

Weeden, N. C. 1983. Identification of duplicate loci and evidence for post-meitoic gene expression in pollen. In D. L. Mulcahy and E. Ottaviano (eds.), *Pollen: Biology and Implications for Plant Breeding*, pp. 9-14. Elsevier, New York.

Weeden, N. F., and L. D. Gottlieb. 1979. Distinguishing allozymes and isozymes of phosphoglucoseisomerases by electrophoretic comparisons of pollen and somatic tissues. *Biochem. Genet.*, 17: 287-296.

Weis, I. M. 1982. The effects of propagule size on germination and seedling growth in *Mirabilis hirsuta*. *Can. J. Bot.*, 60: 1868-1874.

Weller, S. G., and R. Ornduff. 1989. Incompatibility in Amesinckia grandiflora (Boraginaceae): distribution of callose plugs and pollen tubes following inter- and intramorph crosses. *Am. J. Bot.*, 76: 277-282.

Wendel, J. F., M. D. Edwards, and C. W. Stuber. 1987. Evidence for multilocus genetic control of preferential fertilisation in maize. *Heredity*, 58: 297-301.

Williams, E. G., and J. L. Rouse. 1990. Relationships of pollen size, pistil length and pollen tube growth rates in Rhododendron and their influ-

ence on hybridization. *Sex. Plant Reprod.*, 3: 7–17.

Winn, A. A. 1985. Effects of seed size and microsite on seedling emergence of *Prunella vulgaris* in four habitats. *J. Ecol.*, 73: 831–840.

Willing, R. P., and J. P. Mascarenhas. 1984. Analysis of the complexity and diversity of mRNAs from pollen and shoots of *Tradescantia*. *Plant Physiol.*, 75: 865–868.

Willing, R. P., D. Bashe, and J. P. Mascarenhas. 1988. Analysis of the quantity and diversity of messenger RNAs from pollen and shoots of *Zea mays*. *Theor. Appl. Genet.*, 75: 751–753.

Willson, M. F., and N. Burley. 1983. *Mate Choice in Plants: Tactics, Mechanisms, and Consequences*. Princeton Univ. Press, Princeton.

Willson, M. F., L. J. Miller, and B. J. Rathcke. 1979. Floral display in *Phlox* and *Geranium*: adaptive aspects. *Evolution*, 33: 52–63.

Winsor, J. A., L. E. Davis, A. G. Stephenson. 1987. The relationship between pollen load and fruit maturation and its effects on offspring vigor in *Cucurbita pepo*. *Am. Nat.*, 129: 643–656.

Wyatt, R., I. J. Odrzykoski, and A. Stoneburner. 1989. High levels of genetic variability in the haploid moss *Plagiomnium ciliare*. *Evolution*, 43: 1085–1096.

Young, H. J., and M. L. Stanton. 1990. Influence of environmental quality on pollen competitive ability in wild radish. *Science*, 248: 1631–1633.

Zamir, D., S. D. Tanksley, and R. A. Jones. 1982. Haploid selection for low temperature tolerance of tomato pollen. *Genetics*, 101: 129–137.

Zimmerman, J. K., and I. M. Weis. 1982. Fruit size variation and its effects on germination and seedling growth in *Xanthium strumarium*. *Can. J. Bot.*, 61: 2309–2315.

Zuk, J. 1969. Autoradiographic studies in Rumex with special reference to sex chromosomes. *Chromosomes Today*, 2: 183–188.