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Effects of pollen selection on progeny vigor in a *Cucurbita pepo* × *C. texana* hybrid

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Abstract We examined the effects of pollen selection for rapid pollen-tube growth on progeny vigor. First, we crossed a wild gourd (*Cucurbita texana*) to a cultivated zucchini (*Cucurbita pepo* cv 'Black Beauty') to produce an F_1 and then an F_2 generation. Half of the F_1 seeds were produced by depositing small loads of *C. texana* pollen onto the stigmas of *C. pepo*. These small pollen loads were insufficient to produce a full complement of seeds and, consequently, both the fast- and the slow-growing pollen tubes were permitted to achieve fertilization. An F_2 generation was then produced by depositing small loads of F_1 pollen onto stigmas of F_1 plants. The F_2 seeds resulting from two generations of small pollen loads are termed 'the non-selected line' because there was little or no selection for pollen-tube growth rate on these plants. The other half of the F_1 and F_2 seeds were produced by depositing large pollen loads (>10 000 pollen grains) onto stigmas and then allowing only the first 1% or so of the pollen tubes that entered the ovary to fertilize the ovules. We did this by excising the styles at the ovary at 12–15 h after pollination. The resulting F_2 seeds are termed 'the selected line' because they were produced by two generations of selection for only the fastest growing pollen tubes. Small pollen loads from the F_2 plants, both the selected and the non-selected lines, were then deposited onto stigmas of different *C. pepo* flowers, and the vigor of the resulting seeds was compared under greenhouse and field conditions. The results showed that the seeds fertilized by pollen from the selected line had greater vegetative vigor as seedlings and greater flower and fruit production as mature plants than the seeds fertilized by pollen from the non-selected line. This study demonstrates that selection for fast pollen-tube growth (selection on the microgametophyte) leads to a correlated increase in sporophyte (progeny) vigor.

Key words Microgametophyte selection · Pollen selection · Pollen competition · *Cucurbita texana* · *Cucurbita pepo*

Introduction

Over the past two decades, several studies have shown a relationship between the number of pollen grains that are deposited onto a stigma and the performance of the resulting progeny (see Mulcahy and Mulcahy 1975; Ottaviano and Mulcahy 1989; Walsh and Charlesworth 1992). In general, the progeny produced by large pollen loads have enhanced germination, more vigorous vegetative growth, and/or greater reproductive output (such as flower, fruit and seed production) than the progeny from smaller pollen loads (e.g., Mulcahy and Mulcahy 1975; Mulcahy et al. 1975, 1978; Fingerett 1979; Ottaviano et al. 1983; Stephenson et al. 1986; Davis et al. 1987; Winsor et al. 1987; Richardson and Stephenson 1992; Quesada et al. 1993, but see Snow 1990).

In 1979, Mulcahy (1979) proposed the pollen competition hypothesis to account for the effect of pollen load size on progeny performance. He hypothesized that under conditions of pollen competition (i.e., more pollen tubes than ovules; large pollen loads), only the fastest growing pollen tubes achieve fertilization, whereas both fast- and slow-growing pollen tubes achieve fertilization when there are fewer pollen tubes than ovules (small pollen loads). According to this hypothesis, the correlation between fast-growing pollen tubes and vigorous progeny is due to a large overlap in gene expression between the gametophytic and sporophytic stages of the life cycle.

Over the past 15 years it has been shown that a large portion of the genome of the microgametophyte is tran-

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scribed and translated and that the vast majority of the genes expressed by pollen are also expressed in the sporophyte (e.g., Tanksley et al. 1981; Willing and Mascarenhas 1984; Willing et al. 1988). When the pollen competition hypothesis was formulated, it was assumed that pollen competition could only act on heritable genetic variation in pollen performance among the microgametophytes deposited onto the stigma. However, genetically based pollen-pistil interactions leading to differences in pollen performance could also account for the effects of large pollen loads on progeny vigor, if the differences in pollen performance are determined by genetic similarity/dissimilarity between the maternal sporophyte and the microgametophyte (see Waser et al. 1987; Snow and Mazer 1988; Schlichting et al. 1990).

Alternatively, because fruits produced from large pollen loads typically differ from those produced by small pollen loads in seed number, in the position of the seeds within the ovary, and in seed weight, it has been suggested that any differences in progeny vigor could be due to differences in the quantity or quality of the resources provided to the seeds by the maternal plant (Charlesworth 1988; Schlichting et al. 1990). Such environmental (non-genetic) maternal effects could be mediated by phytohormones produced or accumulated in the developing seeds (Lee 1984; Brenner 1987).

Finally, because most species regularly produce more ovules than mature seeds (see Lee 1988), differences in progeny vigor related to pollen load size could be due to non-random seed abortion. For example, it has been shown for several species that potentially viable seeds do abort (see Casper 1988; Lee 1988; Nakamura 1988; Rocha and Stephenson 1991), and two of these studies revealed that under conditions that foster the survival of these viable but normally abortive seeds, they produce less vigorous progeny than the normally non-abortive seeds (Casper 1988; Rocha and Stephenson 1991). Consequently, if differences in vigor among the developing seeds within a fruit influence which seeds mature, as suggested by several authors (Charnov 1979; Westoby and Rice 1982; Bawa and Webb 1984; Stearns 1987), then fruits produced by large pollen loads would have more fertilized ovules than could be developed to maturity and would have many opportunities for non-random seed abortion. In contrast, fruits produced by small pollen loads would not have a surplus of fertilized ovules and would have less opportunity for non-random abortion.

In this study, we tested the pollen competition hypothesis by subjecting *Cucurbita* pollen to two generations of selection for fast pollen-tube growth. We then compared, via greenhouse and field studies, the vigor of seeds fertilized by pollen from plants selected for fast pollen-tube growth with that of seeds fertilized by pollen from plants that have not been selected for fast pollen-tube growth. These experiments were designed to eliminate maternal effects, non-random seed abortion and pollen-pistil interactions as possible explanations for the data.

Materials and methods

Pollen selection treatment: first generation

In the summer of 1989, 10 *Cucurbita texana* plants (grown from seeds taken from a natural population in Texas and probably containing some genetic variability) were crossed to 10 'Black Beauty Bush' (an inbred cultivar of *C. pepo*) zucchini plants that were also grown from seeds. *C. texana* (a wild gourd that is native to the southwestern U.S. and northern Mexico) is either the wild progenitor of *C. pepo* (cultivated squashes) or an early escape from cultivation (Decker and Wilson 1987; Decker-Walters 1990). Moreover, *C. texana* is completely cross-fertile with *C. pepo* and, consequently, they could be considered one biological species (Quesada et al. 1993). Both species are monoecious vines with indeterminate growth and reproduction, but 'Black Beauty' has short internodes that give it a bushy appearance. Seeds of both parental species were sown into an experimental garden at the Pennsylvania State University Agricultural Experiment Station at Rock Springs, Pa. Two flowers were pollinated on each 'Black Beauty' plant using the pollen of *C. texana*. One flower received a small pollen load, while another flower on the same plant received a large pollen load (see Winsor et al. 1987; Quesada et al. 1991 for pollination technique). A small pollen load consisted of approximately 460±30 pollen grains (mean±SE; n=10), and a large pollen load consisted of saturating the stigma with excess pollen (>10 000 pollen grains). To prevent visitation by bees, both staminate (donor) and pistillate (maternal) flowers were covered with cheesecloth bags 1 day before anthesis. On the day of anthesis, pollen from staminate flowers from a minimum of 5 different plants was removed with a small brush, placed into a small plastic container, and thoroughly mixed. Following the controlled pollinations the cheesecloth bags were again placed over the pistillate flowers.

In order to separate the effects of differences in the intensity of pollen competition from the effects of differences in seed number, seed weight, and seed position within the ovary, we excised the style (at the ovary) of the flowers receiving large pollen loads 12–15 h after pollination (depending on the air temperature). A previous study revealed that 12 h is sufficient time (at 30°C) for only the fastest pollen tubes to enter the ovary (Stephenson et al. 1988b). We also excised the styles from flowers receiving small pollen loads at 24 h after pollination (after both fast- and slow-growing pollen tubes had entered the ovary). It was necessary to excise the styles because previous studies of *C. pepo* had shown that fruits resulting from large pollen loads contain more but smaller seeds than fruits from small pollen loads (Stephenson et al. 1988c) and that seed size has an effect on seedling growth (Winsor et al. 1987, Stephenson et al. 1988a). By excising the style in this study, we obtained similar seed numbers and seed weights in the fruits from the large and the small pollen load treatments (Table 1). The mature fruits were harvested in late autumn, and seeds were removed, washed, air-dried, counted, and stored in paper envelopes at room temperature.

Pollen selection treatment: second generation

In the summer of 1990, 210 F_1 seedlings obtained from the previous pollen competition experiment were transplanted into an experimental garden, 160 served as pollen donors and 50 as maternal plants (5 plants from each of 10 maternal plants used to produce the F_1). Of the 160 F_1 pollen donors, 80 were obtained from the large pollen load treatment and 80 from the small pollen load treatment. With the same pollination technique described above, two flowers were pollinated on each F_1 maternal plant. One flower received a small pollen load with pollen obtained from a minimum of 5 donors produced by a small pollen load from the previous generation. The other flower of the same recipient plant received a large pollen load with pollen obtained from a minimum of 5 donors produced by a large pollen load from the previous generation. Similar to the previous year, we were able to control for differences in seed number and seed weight in the progeny produced by the small and the large pollen loads by excising the style (at the ovary) of the flowers receiving large pollen loads

12–15 h after pollination while excising the style of the flowers receiving small pollen loads 24 h after pollination (Table 1). The style excision technique allowed us to again select for only the fastest pollen tubes in the large pollen load treatment and for both fast- and slow-growing pollen tubes in the flowers receiving small pollen loads. The mature fruits were harvested in late autumn and seeds were removed, washed, air-dried, counted, and stored in paper envelopes at room temperature.

Effects of two generations of pollen selection on progeny vigor

In the summer of 1991, 100 F_2 seedlings were transplanted into an experimental garden and these served as pollen donors. Fifty of these F_2 plants were obtained from the lines selected for fast pollen-tube growth (two generations of large pollen load treatments: denoted as F_1L-F_2L), and the other 50 F_2 plants were obtained from the lines that were not selected for fast pollen-tube growth (two generations of small pollen load treatments: denoted as F_1S-F_2S). Ten *C. pepo* cv 'Black Beauty Bush' plants were also sown into the experimental garden and these served as maternal plants. Two flowers were pollinated on each maternal plant using a small pollen load. One of the two flowers received pollen obtained from a minimum of 5 donors from the selected line (F_1L-F_2L). The other flower on the same maternal plant received pollen obtained from a minimum of 5 donors from the non-selected line (F_1S-F_2S). Pollen was gathered and applied in the same manner as the small pollen loads in the previous two generations. Similar seed numbers and seed weights were obtained from fruits resulting from the small pollen loads from the selected (102±10 seed number; 160±3 mg seed weight) and non-selected lines (103±11 seed number; 160±3 mg seed weight). The mature fruits were harvested in the autumn and seeds were removed, washed, air-dried, counted, and stored in paper envelopes at room temperature.

We conducted a greenhouse and a field experiment to determine the effects of two generations of selection for fast pollen-tube growth on progeny vigor. In March 1991, we chose two fruits (one produced by F_1L-F_2L pollen and the other one by F_1S-F_2S pollen) from each of the 10 maternal plants. Ten seeds from each fruit were randomly chosen, weighed, assigned to 4 l pots (containing equal parts of peat, perlite, vermiculite, and potting soil) and randomly assigned to positions on a greenhouse bench. We recorded the days to emergence, leaf area at 14, 21, and 28 days after emergence, and the dry weight of the above-ground biomass at 28 days after emergence. Leaf area was determined using a sizing tool described by Schlichting et al. (1990).

Table 1 Seed number per fruit and seed weight of the F_1 and F_2 progeny. The styles of flowers receiving large pollen loads were excised 12–15 h after pollination and the styles of flowers receiving small pollen loads were excised 24 h after pollination. Mean ± standard error

Generation	Seed number		Seed weight (mg)	
	Large pollen load	Small pollen load	Large pollen load	Small pollen load
F_1	54 ± 23	63 ± 23	200 ± 6	192 ± 6
F_2	146 ± 13	166 ± 12	100 ± 1	100 ± 1

For the field experiment conducted in the summer of 1992, we randomly selected 5 seeds from each of the same two fruits (one fruit produced by F_1L-F_2L pollen and the other one by F_1S-F_2S pollen) obtained from each of the same 10 maternal plants used in the greenhouse experiment. We weighed each seed, assigned it to a 125-ml vermiculite-filled peat pot and placed these 125-ml pots side by side in flats in a greenhouse. After emergence, the seedlings were transplanted to a field at The Pennsylvania State University Agricultural Field Station at Rock Springs, Pa. The transplanted seedlings were arranged at 2-m intervals in a random block design in the field, with two seedlings from each maternal plant represented in each of five different blocks (one seedling fertilized by F_1L-F_2L pollen and the other one by F_1S-F_2S pollen). Staminate and pistillate flower production and fruit production were recorded daily for each plant throughout the growing season.

Results

In the greenhouse experiment, 175 of the 200 seeds germinated and grew for the duration of the experiment. A multivariate analysis of variance (which treats all five of our measures of progeny vigor as if they were a single trait, such as total vigor, as well as accounting for potential non-independence of the five measures of vigor) indicated a highly significant overall effect of pollen selection on progeny vigor (Wilks' Lambda=0.91 $F(5,150)=2.97$ $P<0.01$) (GLM MANOVA SAS 1990). An analysis of covariance (GLM SAS 1990) was conducted to independently evaluate each measure of progeny vigor produced with F_1L-F_2L or F_1S-F_2S pollen. This analysis showed a significant pollen selection effect on the number of days to seedling emergence and a marginal effect ($0.05<P<0.10$) on the leaf area at 14 days after emergence (Table 2). The progeny obtained from small pollen loads from F_1L-F_2L plants (selected donors) germinated faster and had a greater leaf area at 14 days after emergence than the progeny from the unselected pollen donors (F_1S-F_2S) (Table 3). By 21 and 28 days post-emergence there were no differences in leaf area. We suspect that the progeny of the selected donors became pot-bound after 14 days, whereas the progeny of the non-selected donors became pot-bound only after 21 days. In addition to the effects of the pollen selection treatment, the maternal plant and the interaction of the maternal plant and the pollen selection treatment also signif-

Table 2 Mean squares and significance levels from the analysis of covariance (seed weight as covariate) of the greenhouse experiment

Source of variance	*df*	Days to emergence	Leaf area (cm^2)			Biomass (g)
			14 days	21 days	28 days	
Maternal plant (MP)	9	9.93***	26 817**	78 434	108 015	9.44
Pollen selection treatment (PST)	1	28.5***	35 954*	93	481	1.36
MP by PST	9	10.4***	12 918	32 288	54 792	3.67
Seed weight	1	8.08	13 893	13 911	1 399	0.06
Error	154	3.24	13 752	49 642	68 799	6.17

*, **, *** Significant at the 0.10, 0.05, and 0.01 levels, respectively

Table 3 Effects of two generations of pollen selection on progeny vigor in the greenhouse. Least square mean ± standard error

Pollen selection treatment	Days to emergence	Leaf area (cm^2)			Biomass (g)
		14 days	21 days	28 days	
Selected	5.6 ± 0.2	352 ± 14	651 ± 26	760 ± 31	7.7 ± 0.3
Non-selected	6.6 ± 0.2	319 ± 13	652 ± 25	756 ± 30	7.5 ± 0.3

Table 4 Mean squares and significance levels from the analysis of covariance (seed weight as covariate) of the field experiment

Source of variance	*df*	Staminate flowers per plant	Pistillate flowers per plant	Fruits per plant
Maternal plant (MP)	9	58.8	4.47	0.76
Pollen selection treatment (PST)	1	101.8*	2.27	5.63***
MP by PST	9	78.9***	6.93	1.52
Block	8	33.4	9.58**	1.19
Seed weight	1	350.8***	8.02	0.93
Error	65	33.7	4.94	0.97

*, **, *** Significant at the 0.10, 0.05, and 0.01 levels, respectively

Table 5 Effects of two generations of pollen selection on progeny reproductive performance in the field experiment. Least square means ± standard error

Pollen selection treatment	Staminate flowers per plant	Pistillate flowers per plant	Fruits per plant
Selected	20 ± 0.9	5.2 ± 0.3	2.4 ± 0.1
Non-selected	18 ± 0.8	4.8 ± 0.3	1.9 ± 0.1

icantly influenced the days to seedling emergence (Table 2), while the maternal plant influenced the leaf area at 14 days after emergence (Table 2).

In the field experiment, 94 of the 100 seeds germinated and grew to maturity. The following measures of reproductive performance were recorded on each plant: (1) total number of staminate flowers; (2) total number of pistillate flowers; (3) total number of fruits. A multivariate analysis of variance (MANOVA SAS 1990) revealed a marginal overall effect of pollen selection on our three measures of reproductive performance (Wilks' Lambda=0.90 $F(3,63)=2.30$ $P<0.08$). Again, it should be noted that this test treats all three measures as a single trait, reproductive performance, and it accounts for the potential non-independence among the measures. In addition, we performed an ANCOVA (GLM SAS 1990) in order to independently examine each measure of reproductive performance. The ANCOVA revealed a highly significant effect of pollen selection on total fruit production per plant and a marginal effect ($0.05<P<0.10$ on total staminate flower production per plant (Table 4). The progeny obtained from the F_1L-F_2L pollen (selected donors) produced more fruits and more staminate flowers per plant than the progeny obtained from the F_1S-F_2S pollen (unselected donors) (Table 5). In addition, both seed weight and the interaction of the maternal plant with the pollen selection treatment influenced the production of staminate flowers.

Discussion

Numerous studies of both cultivated (Mulcahy et al. 1975, 1978; Mulcahy and Mulcahy 1975; Ottaviano et al. 1983; Stephenson et al. 1986; Davis et al. 1987; Winsor et al. 1987) and non-cultivated (Fingerett 1979; McKenna and Mulcahy 1986; Ramstetter and Mulcahy 1988; Lee and Hartgerink 1986; Bertin 1990; Richardson and Stephenson 1992) species have shown a positive relationship between the size of the pollen load and progeny vigor. However, the factors responsible for this relationship have been subject of much debate (e.g., Mulcahy 1979; Charlesworth et al. 1987; Charlesworth 1988; Stephenson et al. 1988c; Snow and Mazer 1988; Schlichting et al. 1990; Charlesworth and Charlesworth 1992; Quesada et al. 1993). This study demonstrated that selection for fast pollen-tube growth leads to a correlated increase in progeny vigor as predicted by the pollen competition hypothesis (Mulcahy 1979). This study, however, differed from previous tests of the pollen competition hypothesis in that it excluded the alternative hypotheses that have been advanced to explain the relationship between pollen load size and progeny vigor (i.e., pollen-pistil interactions, environmental maternal effects, and non-random seed abortion).

The small loads used in this study were insufficient to produce a full complement of seeds (which is approximately 300 seeds). Consequently, the seeds resulting from small pollen loads were produced under conditions of little or no pollen competition and were the result of fertilization by both fast- and slow-growing pollen tubes. In short, there was no selection for microgametophyte performance (speed of germination and/or pollen-tube growth rate) on tubes from the plants produced by two generations of small pollen loads (F_1S-F_2S). It is not clear as to why only 15–50% of the pollen grains deposited onto the stigma actually produced a mature seed, but a recent survey revealed that similar pollen deposition to seed ratios are the norm among angiosperms (Stephenson et al. 1995). In contrast, the large pollen loads used in this study greatly exceeded that necessary to produce a full complement of seeds (Stephenson et al. 1988a) and, by excising the styles at 12–15 h after pollination, we permitted only the fastest growing pollen tubes (<1% of the pollen grains) to fertilize the ovules (Stephenson et al. 1988b). By using this procedure on both the parental and F_1 generations, we created F_1L-F_2L plants that were selected for vigorous microgametophytes (fast germination and/or fast pollen-tube growth). Because of the style excision technique, the selected and the non-selected lines were produced in fruits that did not differ in seed number (both less than a full complement), seed weight, or in the positions of the ovules that produce mature seeds (Table 1). Consequently, there were probably no differences in maternal environmental effects between the selected and non-selected plants, nor were there differences in the opportunity for non-random seed abortion.

Previously, we reported that F_2 plants produced by large pollen loads were more vigorous than F_2 plants produced by small pollen loads under greenhouse conditions (Quesada et al. 1993). The present study showed that the effects of selection for fast pollen-tube growth on progeny vigor extend across generations. Because we deposited only small pollen loads taken from both the selected (F_1L-F_2L) and the non-selected (F_1S-F_2S) plants onto separate flowers of the 'Black Beauty Bush' cultivar (inbred, genetically uniform) of *C. pepo*, there was no selection on the pollen that fertilized the seeds in the generation (third) that we examined for vigor in this study. Moreover, these small pollen loads produced mature fruits with similar seed numbers (less than a full complement), seed weights, and seed positions within the ovary. Consequently, the differences in progeny (sporophyte) performance that we found are unlikely to be due to maternal environmental effects or to non-random seed abortion. Rather, our findings (greater vegetative vigor and increased reproductive output on progeny resulting from fertilization by selected donors) are most likely the result of prior selection on heritable variation in pollen (microgametophyte) performance and a genetic correlation between pollen performance and progeny vigor.

Previous studies that have attempted to document the heritability of pollen performance and/or its correlated effect on progeny performance present a confusing and much debated picture (Snow and Mazer 1988; Schlichting et al. 1990; Charlesworth and Charlesworth 1992). In *Petunia hybrida*, the beneficial effect of large pollen loads were observed to extend into the next generation (Mulcahy et al. 1978), but this study used one clone as a pollen donor and one clone as a pollen recipient so that generalization may not be warranted. In *Raphanus raphanistrum*, two generations of weak versus intense pollen competition (pollen selection) produced plants whose pollen did not differ in performance (Snow and Mazer 1988), but the differences between the weak and the intense selection were surprisingly small (see Schlichting et al. 1990). Consequently, differences in pollen performance would be very difficult to detect in the Snow and Mazer (1988) study. In maize, *Zea mays*, four generations of selfing produced plants resulting from weak and strong pollen competition (selection). The pollen produced from the strong pollen selection line fertilized more seeds than the pollen from the weak selection line in mixtures of pollen from a reference line (Ottaviano et al. 1988) indicating that it is possible to select for pollen performance, at least within a given stylar genotype (see also Johnson and Mulcahy 1978). Ottaviano et al. (1988) also subjected a genetically variable population of maize to four generations of pollen selection and reported both improved micogametophyte performance and a correlated beneficial effect on one sporophytic trait (seed weight). Landi et al. (1986) selected for improved grain yield in maize and found a correlated response with pollen performance. Finally, Schlichting et al. (1990) found that additive genetic variance in the genes expressed during both stages of the life cycle played a significant but small role in the positive effect of large pollen loads on progeny vigor in the 'Black Beauty Bush' cultivar of zucchini (*C. pepo*). Because this is an inbred cultivar, pollen-pistil interactions, maternal effects, and/or non-random patterns of seed abortion played a more important role in altering progeny vigor.

In this study, the genetic variation (upon which we selected) was produced via hybridization of the common zucchini with a wild gourd. It should be noted, however, that the level of heterozygosity and the percentage of polymorphic loci in the F_1 of this study is similar to that typically reported for outcrossing herbaceous plants (Decker and Wilson 1987; Hamrick and Godt 1989). In natural populations, genetic variation for pollen performance can result from mutation, gene flow among populations, and environmental heterogeneity within populations (e.g., Snow and Mazer 1988; Young and Stanton 1990; Schlichting et al. 1990; Charlesworth and Charlesworth 1992).

The data from this study indicate that large pollen loads have the potential to improve the vigor of the resulting progeny via microgametophyte selection, especially in outcrossing highly heterozygous populations. However, with the exception of only one study (Snow and Mazer 1988), all the studies on the heritability of pollen performance have used cultivated species. Carefully controlled studies using non-cultivated species are now needed to determine the efficacy of pollen selection in nature.

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References

Bawa KS, Webb CJ (1984) Flower, fruit and seed abortion in tropical forest trees: Implications for evolution of paternal and maternal reproductive patterns. Am J Bot 71:736–751

Bertin RI (1990) Effects of pollination intensity in *Campsis radicans*. Am J Bot 77:178–187

Brenner ML (1987) The role of hormones in photosynthate partitioning and seed filling. In: Davies, PJ (ed) Plant hormones and their role in plant growth and development. M. N. Shoff, Minneapolis, MN, pp 474–493

Casper BB (1988) Evidence for selective embryo abortion in *Cryptantha flava*. Am Nat 132:318–326

Charlesworth B, Charlesworth D (1987) Inbreeding and its evolutionary consequences. Ann Rev Ecol Syst 18:237–268

Charlesworth D (1988) A comment on the evidence for pollen competition in plants and its relationship to progeny fitness. Am Nat 132:298–302

Charlesworth D, Charlesworth B (1992) The effects of selection in the gametophyte stage on mutational load. Evolution 46:703–720

Charlesworth D, Schemske DW, Sork VK (1987) The evolution of plant reproductive characters, sexual vs. natural selection. In: Stearns SC (ed) The evolution of sex and its consequences. Birkhauser Verlag, Basel, pp 317–335

Charnov EL (1979) Simultaneous hermaphroditism and sexual selection. Proc Natl Acad Sci USA 76:2480–2484

Davis LE, Stephenson AG, Winsor JA (1987) Pollen competition improves performance and reproductive output of the common zucchini squash under field conditions. J Am Soc Hortic Sci 112:711–716

Decker-Walters DS (1990) Evidence for multiple domestication of *Cucurbita pepo*. In: Bates DM, Robinson RW, Jeffrey C (eds) Biology and utilization of the Cucurbitaceae. Cornell University Press, New York, pp 96–101

Decker DS, Wilson HD (1987) Allozyme variation in *Cucurbita pepo* complex. *C. pepo* var. *ovifera* vs. *C. texana*. Syst Bot 12:263–273

Fingerett ER (1979) Pollen competition in a species of evening primerose, *Oenothera organensis* Munz. MSc thesis, Washington State University, Pullman, Wash.

Hamrick JL, Godt MJW (1989) Allozyme diversity in plant species. In: Brown AHD, Clegg MT, Kahler AL, Weir BS (eds) Plant population genetics, breeding and genetic resources. Sinauer Associates, Sunderland, Mass., pp 43–63

Johnson CM, Mulcahy DL (1978) Male gametophyte in maize: II Pollen vigor in inbred plants. Theor Appl Genet 51:211–215

Landi P, Frascaroli E, Speranza A (1986) Pollen characteristics of maize (*Zea mays* L.) inbred lines selected for grain yield. Genet Agric 40:397–404

Lee TD (1984) Patterns of fruit maturation: a gametophytic competition hypothesis. Am Nat 123:427–432

Lee TD (1988) Patterns of fruit and seed production. In: Lovett Doust J, Lovett Doust L (eds) Plant reproductive ecology: patterns and strategies. Oxford University Press, N.Y., pp 179–202

Lee TD, Hartgerinck AP (1986) Pollination intensity, fruit maturation pattern and offspring quality in *Cassia fasciculata* (Leguminosae). In: Mulcahy DL, Ottaviano EM (eds) Pollen: biology and implications for plant breeding. Elsevier Biomedical Press, New York, pp 417–422

McKenna M, Mulcahy DL (1983) Ecological aspects of gametophytic competition in *Dianthus chinensis*. In: Mulcahy DL, Ottaviano EM (eds) Pollen: biology and implications for plant breeding. Elsevier Biomedical Press, New York, pp 443–448

Mulcahy DL (1979) The rise of angiosperms: a genecological factor. Science 206:20–23

Mulcahy DL, Mulcahy GB (1975) The influence of gametophytic competition on sporophytic quality in *Dianthus chinensis*. Theor Appl Genet 46:277–280

Mulcahy DL, Mulcahy GB, Ottaviano E (1975) Sporophytic expression of gametophytic competition in *Petunia hybrida*. In: Mulcahy DL (ed) Gamete competition in plants and animals. North Holland Publishing Company, Amsterdam, pp 227–232

Mulcahy DL, Mulcahy GB, Ottaviano E (1978) Further evidence that gametophytic selection modifies the genetic quality of the sporophyte. Soc Bot Fr Actual Bot 1–2:57–60

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

Ottaviano EM, Mulcahy DL (1989) Genetics of angiosperm pollen. Adv Genet 26:1–64

Ottaviano EM, Sari-Gorla M, Arenari I (1983) Male gametophytic competition in maize: selection and implications with regard to breeding systems. In: Mulcahy DL, Ottaviano EM (eds) Pollen: biology and implications for plant breeding. Elsevier Biomedical Press, New York, pp 367–374

Ottaviano E, Sari-Gorla M, Villa M (1988) Pollen competitive ability in maize: within population variability and response to selection. Theor Appl Genet 76:601–608

Quesada M, Schlichting CD, Winsor JA, Stephenson AG (1991) Effects of genotype on pollen performance in *Cucurbita pepo*. Sex Plant Reprod 4:208–214

Quesada M, Winsor JA, Stephenson AG (1993) Effects of pollen competition on progeny performance in a heterozygous cucurbit. Am Nat 142:694–706

Ramstetter J, Mulcahy DL (1988) Consequences of pollen competition for *Aureolaria flava* seedlings. Bull Ecol Soc Am Suppl 69:269–270

Richardson TE, Stephenson AG (1992) Effects of parentage and size of the pollen load on progeny performance in *Campanula americana*. Evolution 46:1731–1739

Rocha OJ, Stephenson AG (1991) Effects of nonrandom seed abortion on progeny performance in *Phaseolus coccineus* L. Evolution 45:1198–1208

SAS Institute (1990) SAS user's guide: statistics. SAS Institute, Cary, N.C.

Schlichting CD, Small L, Stephenson AG, Winsor JA (1990) Pollen loads and progeny vigor: the next generation. Evolution 44:1358–1372

Snow AA (1990) Effects of pollen-load size and number of donors on sporophyte fitness in wild radish (*Raphanus raphanistrum*). Am Nat 136:742–758

Snow AA, Mazer SJ (1988) Gametophytic selection in *Raphanus raphanistrum*: a test for heritable variation in pollen competitive ability. Evolution 42:1065–1075

Stearns SC (1987) The selection arena hypothesis. In: Stearns SC (ed) The evolution of sex and its consequences. Birkhauser Verlag, Basel, pp 337–349

Stephenson AG, Winsor JA, Davis LE (1986) Effects of pollen load size on fruit maturation and sporophyte quality in zucchini. In: Mulcahy DL, Ottaviano EM (eds) Biotechnology and ecology of pollen. Springer, Berlin Heidelberg New York, pp 429–431

Stephenson AG, Devlin B, Horton JB (1988a) The effects of seed number and prior fruit dominance on the pattern of fruit production in *Cucurbita pepo* (zucchini squash). Ann Bot 62:653–661

Stephenson AG, Winsor JA, Schlichting CD (1988b) Evidence for non-random fertilization in the common zucchini, *Cucurbita pepo*. In: Cresti M, Gori P, Pacini E (eds) Sexual reproduction in higher plants. Springer, Berlin Heidelberg New York, pp 333–338

Stephenson AG, Winsor JA, Schlichting CD, Davis LE (1988c) Pollen competition, non-random fertilization, and progeny fitness: a reply to Charlesworth. Am Nat 132:303–308

Stephenson AG, Quesada MR, Schlichting CD, Winsor JA (1995) Consequences of variation in pollen load size. In: Hoch PC, Stephenson AG (eds) Experimental and molecular approaches to plant systematics. Monogr Syst Bot 53:233–244

Tanksley S, Zamir D, Rick CM (1981) Evidence for the extensive overlap of sporophytic and gametophytic gene expression in *Lycopersicon esculentum*. Science 213:453–455

Walsh NE, Charlesworth, D (1992) Evolutionary interpretations of differences in pollen-tube growth rates. Q Rev Biol 67:19–36

Waser NM, Price MV, Montalvo AM, Gray RN (1987) Female choice in a perennial herbaceous wildflower. *Delphinium nelsonii*. Evol Trends Plants 1:29–33

Westoby M, Rice B (1982) Evolution of the seed plants and inclusive fitness of plant tisues. Evolution 36:713–724

Willing RP, Mascarenhas JP (1984) Analysis of the complexity and diversity of mRNAs from pollen and shoots of *Tradescantia*. Plant Physiol 75:865–868

Willing RP, Bashe B, Mascarenhas JP (1988) Analysis of quantity and diversity of messenger RNAs from pollen and shoots of *Zea mays*. Theor Appl Genet 75:751–753

Winsor JA, Davis LE, Stephenson AG (1987) The relationship between pollen load and fruit maturation and the effect of pollen load on offspring vigor in *Cucurbita pepo*. Am Nat 129:643–656

Young HJ, Stanton ML (1990) Influence of environmental quality on pollen competitive ability in wild radish. Science 248:1631–1633