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Pollination biology of *Prunus mahaleb* L.: deferred consequences of gender variation for fecundity and seed size

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This study describes the reproductive biology of *Prunus mahaleb*, a rosaceous treelet, in a southeastern Spanish population. The species is gynodioecious with 55.4% of the plants being male-fertile and 44.6% presenting non-functional, shrunken anthers with no pollen, and behaving as functional females. Individual trees produced the same flower morph in four consecutive study years. Fruit set in bagged inflorescences was absent in male-sterile trees and was very reduced in male-fertiles. Open-pollinated flowers of male-fertiles showed greater fruit set ($\bar{x} =$ 29.1%) than male-steriles ($\bar{x} =$ 25.3%). Average fruit set in three experimental treatments (control, selfed, and crossed) were 29.05%, 41.9% and 38.6%, respectively, for male-fertile trees; those for male-steriles were 25.3%, 0% and 39.2%, respectively. Seeds from male-sterile plants were heavier (71.33 mg) than those of male-fertile plants (66.05 mg) but did not differ in germination ability. Male-fertile and male-sterile trees differ significantly in average nectar concentration (73.5% and 55.9%, respectively) and nectar volume secreted/flower and day (0.142 μl and 0.171 μl, respectively). A total of 41 species of insect flower visitors were recorded. Calliphorid and tachinid flies (41.97% of total visits), and andrenid bees (30.30%), were the most frequent visitors. The diversity of insect visitors was greater in male-fertile trees. Bees and flies accounted for 50.5% and 49.5%, respectively of total visits to male-fertile trees. Flies were far more frequent than bees at male-sterile flowers (76.9% and 23.1%, respectively). Male-sterile trees received higher average visitation (47 insects/census) than male-fertile trees (32 insects/census). Individual trees showed a relative constancy of the fecundity rankings between years. Fruit production was significantly higher in male-sterile trees, with a four-year average of 6558 fruits, in contrast with male-fertile trees which yielded 4670 fruits. This 1.77-fold difference in favour of male-sterile trees over male-fertiles was not compensated by the greater fruit set of the latter. This is attributable to the lower outcrossing rate of male-fertiles, estimated as 52.68% from seed mass data. In addition, greater seed mass of female progeny, and higher visitation rate by insects might explain the maintenance of this polymorphism. These results have far-reaching implications for the demography and seed-dispersal ecology of this endozoochorus species.

KEY WORDS:— Cryptic breeding systems – fruit set – gynodioecy – Mediterranean – pollination – Rosaceae.

CONTENTS

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INTRODUCTION

It is widely accepted that diclinous plant breeding systems illustrate a gradient of evolutionary steps from hermaphroditism to dioecy, involving intermediate types of populations with hermaphrodite and either male-sterile or female-sterile individuals (Darwin, 1877). Gynodioecious populations frequently have some male-sterile plants together with hermaphrodite individuals and exemplify the usual path to dioecy from hermaphroditism. Thus, an initial evolution of the gynodioecious condition from hermaphroditism could give rise to dioecy or subdioecy by gradual reduction of seed fertility among hermaphrodites (Lloyd, 1976; Ross, 1978). Central to this idea is the proposition that some selective advantage must accrue to the female, male-sterile, individuals that contribute no genes to the population via pollen and seem to be at a selective disadvantage relative to perfect-flowered plants. However, few field studies have actually estimated this selection differential in gynodioecious species (e.g., Stevens, 1988; see also Kevan *et al.*, 1990; Ågren & Willson, 1991; Lloyd, 1976, and references therein).

When both nuclear and cytoplasmic factors are involved in the inheritance of male sterility (apparently a common situation in wild populations; see Lewis, 1941) a two-fold fecundity advantage of the females is required for their establishment in populations. However, as pointed out by Lewis (1941), the equilibrium frequencies of the two sexual phenotypes in the population would depend not only on fitness differences, but on the attainment of a 'hybridity optimum' (i.e. the seed set of females declines if they become too abundant and pollen-donor hermaphrodites are scarce). Therefore, aspects of the pollination ecology (insect visitation patterns, nectar secretion, phenology) are central to understanding the evolution of gynodioecy among entomophilous species. Information on these ecological factors among gynodioecious species is extremely scarce (Dommeé, Assouad & Valdeyvan, 1978; Atsatt & Rundel, 1982; Sun & Ganders, 1986; Ågren & Willson, 1991; Ashman & Stanton, 1991).

Recent studies have revealed subtle sexual differentiation within populations of apparently perfect-flowered species. Cryptically diclinous breeding systems appear as 'hidden' within the morphologically hermaphrodite condition. In these systems, one or both sexual forms are difficult to detect because they produce apparently perfect flowers, often with androecia or gynoecia similar in size and shape to the functional ones (Lloyd, 1972, 1973; Bawa & Webb, 1983; Stevens & Richards, 1985; Kevan *et al.*, 1990; Mayer & Charlesworth, 1991). Cases of male sterility have been frequently reported among cultivated varieties of rosaceous species (e.g. Vargas & Romero, 1976; also see Campbell, Greene & Dickinson, 1991; Dickinson & Campbell, 1991). These types of 'cryptic' polymorphisms might prove to be much more common than previously recognized (Young, 1972; Uno, 1982a; Primack & McCall, 1986; Stevens, 1988;

Steyn & Robbertse, 1988; Liston, Rieseberg & Elias, 1990; Ågren & Willson, 1991; see review in Mayer & Charlesworth, 1991).

The formal terminology based on flower morphology thus seems to hide a rich variety of angiosperm sexual systems which appear in the form of cryptic functional variation. Study and documentation of this type of variation are relevant to our understanding of breeding system evolution, but also have important implications for genetics and demography of plant populations. The objectives of this paper are: (1) to describe the diclinous breeding system of *Prunus mahaleb* L. (Rosaceae) in Southern Spanish wild populations exhibiting functional gynodioecy; (2) to examine some of the implications of this breeding system for individual differences in fecundity and seed size.

I examined the form of flowers on different individuals and the correlates of floral variation with fecundity, fruit and seed morphology, seed set, seed size, and differential nectar production and visitation by pollinators. A central objective was to document the deferred consequences of flower variation and gender expression for different fitness components in this species. Following Lewis (1941), I will refer to functional females as 'male-steriles' and to hermaphrodites as 'male-fertiles', to emphasize the differences with other gynodioecious breeding systems with both morphological and functional separation of the two gender types.

MATERIALS AND METHODS

Study area and plant characteristics

The main study site was located in Nava de las Correhuelas (1615 m elevation), a site in the highlands of the Parque Natural de las Sierras de Cazorla, Segura y las Villas (Jaén province, southeastern Spain). It extends over ca. 100 ha including both poldje deep soils and rocky slopes. Deciduous vegetation dominated by *Crataegus monogyna*, *Prunus mahaleb*, *Lonicera arborea*, *Berberis hispanica* and *Acer granatense* occupies the deep soils, while adjacent rocky slopes are dominated by open pine forest (*Pinus nigra* ssp. *salzmannii*) with an understory *Juniperus communis*, *J. sabina*, *J. phoenicea* and scattered *Taxus baccata* (Valle *et al.*, 1989).

Prunus mahaleb, the Saint Lucy's or Mahoma's cherry, is a widely distributed deciduous treelet in central and south Europe, extending to the Ukraine, south Belgium, Morocco, Syria, and west-central Asia (Webb, 1968). In the southeastern Iberian Peninsula it grows scattered in mesic and cool sites in limestone mountains, occupying both deep soils of the 'navas' (poldjes) and rocky slopes. Populations at Sierras de Cazorla, Segura y las Villas are found at elevations ranging from 1200 to 1800 m.

Plants at the main study site leaf by early May and flower between mid-May and mid-June. Ripe fruits can be found as early as the end of June at elevations under 1300 m. However, the main ripening and seed dispersal season at the denser populations at 1550–1650 m takes place between the last week of July and the end of August, extending in certain years to early September. Leaves are shed by late November.

Small white flowers are produced in corymbs of 3–10, secrete very small amounts of nectar, and are visited and pollinated by bees

(Hymenoptera:Andrenidae, Apidae) and flies (Diptera: Calliphoridae, Syrphidae). Unripe fruits are green and ripe drupes are glossy black, 8.0 ± 4.4 mm long and 8.3 ± 0.5 mm wide ($n = 20$) (Herrera & Jordano, 1981). Chemical analyses of the dry pulp reported by these authors yielded 3.2% crude fat, 2.8% crude protein, 6.3% ash, and 5.7% fibre, with soluble carbohydrates amounting to 82.0%. Fruit crop sizes usually range between 700 and 25000. Ripe fruits are consumed, and the seeds dispersed, by frugivorous vertebrates (mostly birds and carnivorous mammals; Herrera & Jordano, 1981; Herrera, 1989; P. Jordano & E.W. Schupp, pers. obs.). At least 28 species of frugivorous birds, four mammals, and one lizard have been recorded feeding on the fruits.

Methods

A general survey of 102 trees (almost all the reproductive individuals) was carried out to obtain an estimate of the percentages of the two floral morphs in the population. A population of 52 marked plants was monitored for 4 years (1988–1991). Plant characteristics (height, number of trunks, trunk diameter at breast height, and the area of canopy projection) were measured for each tree. Intensive observations on phenology, breeding system, seed set, fruit production and seed dispersal were carried out during 1988 and 1989 in a subsample of 32 trees. Five individual branches were marked in these trees, and regular counts from bud differentiation to ripe fruit removal (every 4 days during flowering, or 5–7 days during fruit ripening) were carried out on marked branches to assess phenological patterns, fruit set, and fruit removal rates. Approximately 25 000–30 000 flowers were monitored in this way during 1988 and 1989. Fifteen flower buds in four plants were individually tagged to determine flower longevity. These flowers were monitored daily in order to determine the timing and sequence of occurrence of anthesis, anther dehiscence, stigma receptivity, corolla changes, and corolla abscission. Fresh flowers were collected for measurement of anther size and determination of pollen presence using a dissecting microscope.

Seed set under experimental field conditions was examined in the sample of 32 trees. Experimental treatments included: bagged flowers with insects excluded; bagged flowers with emasculation prior to anther dehiscence; hand cross pollinations between flowers of different trees (xenogamous crosses); hand cross pollinations between flowers of the same tree (geitonogamous crosses); flowers left open to natural pollination, serving as a control. Whenever possible, different groups of flowers of the same branch were bagged and a single treatment was applied in each bag. Three or four branches of the same tree were used for these experimental pollinations and the procedure was replicated in seven male-fertile trees (totalling 82 treatment bags with 4023 flowers in the two study years) and five male-sterile trees (80 bags, 4098 flowers).

I used the bag treatment to test for the effect of absence of insect intervention on fruit and seed set. In addition, I combined this treatment with flower emasculation to test for apomixis. I emasculated the flowers by gently separating the petals of flower buds ready to open and cutting the stamen filaments with forceps.

To pollinate the flowers, stamens were removed with forceps from bagged donor flowers and immediately transferred to receptive stigmata in a different flower of the same or other tree. The dehisced stamens were gently brushed against the stigma. Fruit set was recorded two weeks after pollination and again when fruits reached final size. Fruits were collected when ripe and the viable seeds in each treatment bag counted and weighed individually (dry mass after oven desiccation at 30°C). Samples of ripe fruits were also collected from non-marked branches before depletion of fruit crops by frugivores. Maximum length, cross diameter, fresh mass, dry mass, and dry mass of pulp and seed were determined for these fruits.

I carried out germination tests directly in the field. Seeds from individually-identified trees of the two morphs ($n = 60$ seeds) were sown in 16 replicate plots in different microhabitats and checked weekly for appearing seedlings. Values of percent seeds germinated were averaged over plots for each gender type. Results of germination response to frugivore treatment and microhabitat type, and its effects on seedling size and survival, will be reported elsewhere (P. Jordano, E. W. Schupp & L. López-Soria, in prep.).

Nectar production was determined for individual flowers that had been bagged (fine bridal veil) for the preceding 24 h. Measurements were obtained for flowers of different ages (semi-open buds, 1st day, 2nd day and ≥ 3 days). I bagged 5–10 branches on each tree, including several inflorescences, and then sampled for measurement from the enclosed flowers. The nectar volume secreted was collected using microcapillary tubes and determined from the column length of the capillary. Dissacharide sugar concentration was obtained with a Bellingham pocket refractometer specially modified for readings of small amounts of nectar. In order to determine simultaneously both nectar volume and concentration for as many flowers as possible, the nectar obtained from flowers with very small amounts was diluted in a known volume of distilled water and the resulting refractometer reading corrected accordingly. This procedure was used in $< 15\%$ of the readings. Mass of sugar produced per flower and day was estimated by the procedure outlined by Prys-Jones & Corbet (1978). The standing crop of nectar was sampled at different hours of the day in flowers open to visitation by pollinators. Only nectar volume was determined in these samples.

Insect visitors to the flowers were monitored during short censuses (10 min) on individual trees. A census involved the count and identification of all flower visitors in a canopy area of *c.* 2 m^2. Censuses were carried out during the whole flowering period, especially in days with fine weather. I carried out non-systematic observations at dusk and soon after nightfall both of the study population and at Roblehondo (1300 m), but these watches failed to reveal activity of crepuscular or nocturnal visitors. During censuses, the species of flower visitors were identified whenever possible or, alternatively, classified as distinct morphotypes; voucher specimens were collected for later identification of these morphotypes. These specimens are deposited within the insect collection at the Unidad de Ecología Evolutiva, Estación Biológica de Doñana, Seville, Spain. The behaviour of insects at flowers was also monitored to determine pollen or nectar foraging.

For statistical analyses I used parametric tests whenever their requirements were met. I used logarithmic (for linear or mass measurements) and angular transformations (for percentages) prior to ANOVA tests or regression analyses. In the case of extremely skewed distributions (e.g. crop sizes), I provide the median and 25–75 percentiles and used non-parametric tests; mean and standard errors of the mean are given in the other cases, unless otherwise stated. For the comparison of fruit-set differences resulting from experimental pollinations, as well as for the tests involving comparisons between the two gender types (male-fertiles and male-steriles) I used general linear models (SAS, 1986; Gagnon *et al.*, 1989). I performed orthogonal comparisons to test specific hypotheses comparing only the means of selected levels of a factor or combination of factors (see Snedecor & Cochran, 1989). Contrasts were used to perform planned comparisons of: (1) fruit set in male-fertile and male-sterile trees; (2) fruit set resulting from cross-vs. self-pollinations in each gender type; (3) fruit set resulting from experimental pollen addition (crossed or selfed) and from the controls; (4) all the above tests incorporating simultaneously the information of the two study years, since significant between-year differences were detected. Contrasts were used as an alternative to multiple comparison tests, or tests with data subsets involving only the desired effect levels, to avoid sacrificing statistical power. For unplanned, a posteriori, comparisons I used the Scheffé test to control the experimentwise error rate at 5%. Variance components were estimated with VARCOMP and NESTED procedures in SAS (1986).

RESULTS

Plant characteristics and variation in flower morphology

Two distinct gender types were present in the study population, as well as in other populations of the study area. I found no intermediate trees, although variation in anther size, anther number, and pollen grains/anther occurred in male-fertile individuals (see below). I recorded 59 plants (57.84%) that were male-fertile, presenting functional anthers with bright orange-yellow colour and globose shape, and filled with pollen. Male-sterile plants ($n = 43$; 42.16%) presented non-functional, shrunken, light-pink anthers, with no pollen. A census in 1990 at a nearby population (Torcal del Cerecino, 3 km away) yielded similar proportions out of $n = 76$ trees: 52.26% male-fertiles and 44.74% male-steriles. The relative frequencies of the two gender types do not differ significantly from 1:1 (for both populations, $\chi^2 < 2.51$, $P > 0.10$).

Production of pollen or lack of it was a constant character, expressed consistently by each tree in the four study years. Microscopic examination of the anthers revealed total absence of pollen grains in male-sterile plants. In contrast, those of male-fertile trees liberated large quantities of pollen. Additional evidence of male-sterility comes from failure to set seed in experimentally self-pollinated male-sterile trees (see below).

Anther size differed significantly between the two gender types. Anther length and width of male-fertiles (0.71 ± 0.01 mm and 0.77 ± 0.01 mm, respectively, $n = 36$) were significantly greater than those of male-steriles (0.57 ± 0.01 and 0.46 ± 0.02 mm, respectively, $n = 42$) ($F \geq 114.0$, $P < 0.001$, d.f. = 1,76, for the two possible comparisons).

Nectar production

Mean daily nectar production/flower, including data from first-and second-day flowers, was 0.100 ± 0.005 μl ($n = 356$; male-fertile trees) and 0.098 ±

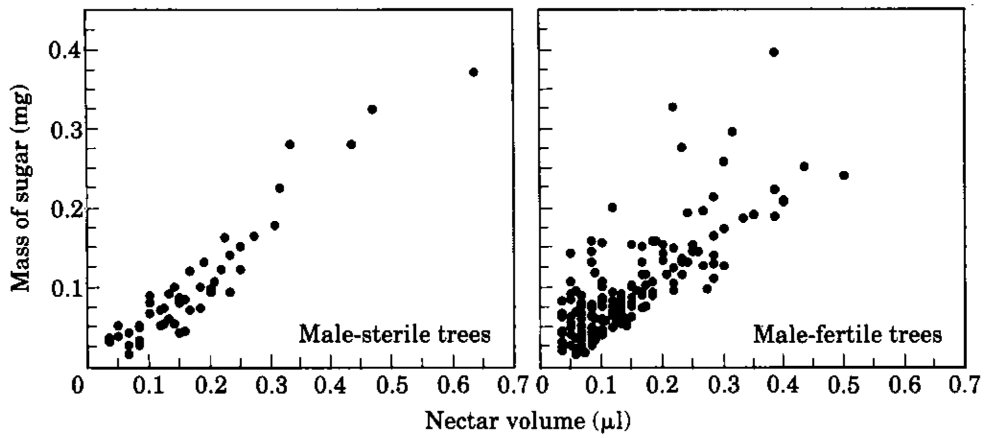

Figure 1. Relationship between the mass of sugar/flower and the nectar volume secreted/24h (μl) by individual flowers of *Prunus mahaleb* of the two gender types. Regression equations (log-transformed data): $y = 0.678x-0.457$; $F = 197.30$, d.f. = 1,169; $R^2 = 0.539$, for male-fertile trees. $y = 0.951x-0.294$; $F = 159.31$, d.f. = 1,51; $R^2 = 0.758$, for male-sterile trees.

0.004 μl ($n = 224$; male-sterile trees). The large buds, which are sometimes visited by insects the day before anthesis, secrete 0.077 ± 0.018 μl ($n = 18$); secretion reaches a maximum during the first and second day, and then declines in flowers = 3d old (0.003 ± 0.002 μl; $n = 22$). All these figures are for cumulative, 24h secretion. Average standing crops of flowers open to pollinator visitation (only 1st and 2nd day flowers included) was 0.006 ± 0.003 μl ($n = 30$), with 86.7% of them being empty.

Male-fertile and male-sterile trees differ significantly in the concentration of the nectar ($F = 8.53$, d.f. = 1, 222, $P < 0.004$), and only marginally in the volume secreted ($F = 3.41$, d.f. = 1, 222, $P = 0.066$). Mean values of these variables were: 73.5 ± 3.3% and 0.142 ± 0.007 μl for male-fertile trees ($n = 171$ flowers); and 55.9 ± 2.5% and 0.171 ± 0.015 μl ($n = 53$ flowers) for male-sterile trees.

To test for differences in nectar production, I compared the total amount of sugar produced/24h. Nectar volume and mass of sugar produced were significantly related for both male-fertile trees ($F = 264.3$, d.f. = 1, 169, $P < 0.0001$; $R^2 = 0.610$) and male-sterile trees ($F = 456.8$, d.f. = 1, 51, $P < 0.0001$; $R^2 = 0.899$) (Fig. 1). An ANCOVA showed statistical differences in the slope of the regressions; male-fertile trees secreted 0.633 mg/μl while male-sterile trees yielded 0.547 mg/μl ($F = 172.5$, d.f. = 3, 220, $P < 0.0001$). This difference was largely due to the higher concentration of the nectar of male-fertile trees and the slight trend for more dilute nectar in male-sterile flowers.

Insect visitors

At least 41 species of insects were recorded visiting *P. mahaleb* flowers (Table 1). The flower visitors assemblage included a diverse assortment of bees and flies, with 38 taxa identified to species, one asilid, one empid, and two unidentified flies. All these insect visitors, with the probable exception of the two lepidopterans can be considered as pollinators of the flowers since contact with anthers and/or stigmata was recorded in all instances. Hymenopterans

TABLE 1. Insect visitors, number of visits (*n*), and percentage of total visits for trees of different functional gender

	Functional gender					
	Male-fertile		Male-sterile		Total	
	n	%	*n*	%	*n*	%
Lepidoptera						
*Hemaris tityus**	0	–	0	–	1	0.05
*Iphiclides podalirius**	0	–	0	–	2	0.11
Hymenoptera						
Psithyrus vestalis obenbergeri	1	0.16	0	–	4	0.22
Bombus pratorum (m)	1	0.16	0	–	2	0.11
Bombus pratorum (wk)	6	0.94	0	–	15	0.82
Bombus pascuorum (wk)	1	0.16	0	–	2	0.11
Bombus pascuorum (q)	1	0.16	1	0.11	2	0.11
Bombus terrestris (wk)	27	4.24	11	1.16	41	2.25
Bombus terrestris (q)	2	0.32	1	0.11	3	0.16
Xylocopa violacea	1	0.16	1	0.11	6	0.33
Apis mellifera	4	0.63	6	0.63	19	1.04
Osmia rufa	1	0.16	0	–	6	0.33
Andrena fulva	245	38.45	166	17.57	482	26.37
Andrena synadelpha/nigroaenea†	4	0.63	1	0.11	8	0.44
Andrena trimmerana	0	–	2	0.21	4	0.22
Andrena congruens	20	3.14	18	1.90	52	2.85
Andrena tibialis corvina	0	–	0	–	1	0.05
Andrena albopunctata/morio†	1	0.16	4	0.42	6	0.33
Andrena assimilis	3	0.47	0	–	3	0.16
Halictus quadricinctus	0	–	5	0.53	9	0.49
Halictus tridivisus	0	–	0	–	1	0.05
Halictus scabiosae	5	0.78	2	0.21	8	0.44
Lasioglossum xanthopum	0	–	0	–	1	0.05
Sphecodes gibbus	0	–	0	–	4	0.22
Megachile centuncularis	0	–	0	–	1	0.05
Ancistrocerus nigricornis	0	–	0	–	1	0.05
Diptera						
Eristalis tenax	50	7.85	45	4.76	112	6.14
Eristalis pratorum	0	–	0	–	7	0.38
Eristalis arbustorum	4	0.63	6	0.63	17	0.93
Cheilosia glossa	17	2.67	3	0.32	26	1.42
Mallota fuciformis	5	0.78	0	–	5	0.27
Dasysyrphus albostriatus	1	0.16	2	0.21	4	0.22
Syrphus vitripennis	15	2.35	11	1.16	28	1.53
Syrphus ribessi	1	0.16	1	0.11	2	0.11
Criorhina berberina	0	–	0	–	3	0.16
Calliphora vomitoria	143	22.44	399	42.22	577	31.56
Gonia ornata	31	4.87	83	8.78	133	7.29
Pollenia rudis	13	2.04	33	3.49	49	2.68
Tachina corsicana	6	0.94	0	–	7	0.38
Asilidae n.i.	23	3.61	33	3.49	56	3.06
Empididae n.i.	2	0.31	109	11.53	113	6.19
Diptera n.i.	3	0.47	2	0.21	5	0.27
Total records	637		945		1828	

*The species was recorded outside census time in trees of both gender types

†Includes records for the two species, which are difficult to identify during field censuses

Data for bumblebees (*Bombus* spp.) includes separate entries for females (q), males (m), and workers (wk)

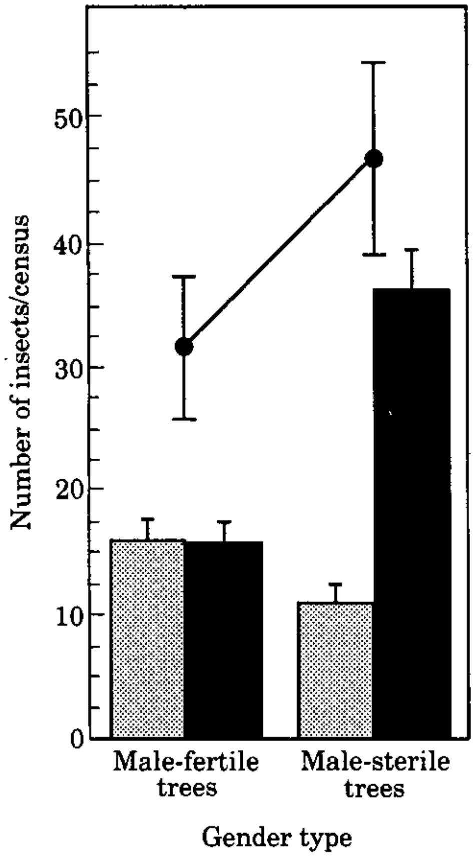

Figure 2. Differences between male-fertile and male-sterile trees in the mean total number of insect visits per census (± one SE) (dots and vertical lines). For each gender type, dotted bars indicate the mean ± one SE overall visit rate by Hymenoptera; black bars represent mean ± one SE overall visit rate by Diptera.

accounted for 37.16% of total visits, dipterans for 62.28%, and lepidopterans for 0.16%. The more frequent visitors were calliphorid and tachinid flies (41.97% of records; mostly *Calliphora vomitoria*), and andrenid bees (30.30%, mostly *Andrena fulva*).

Insects visited most frequently flowers in the first and second day of bloom, when nectar secretion was maximum and anther dehiscence occurred. Out of 218 visits where flower age was recorded, 90.8% were to 1st or 2nd day flowers. Semi-opened buds in the day prior to anthesis received only 3.7% of visits and flowers >2d, 5.5%.

Bees and flies accounted for similar proportions of total visits to male-fertile trees (50.5% and 49.5%, respectively). However, flies were far more frequent than bees at male-sterile flowers (76.9% and 23.1%, respectively). Insect visitation rates differed significantly between the two gender types (Fig. 2). Male-sterile trees (47 ± 4 insects/census) received higher visitation than male-fertile trees (32 ± 3 insects/census) ($F = 7.76$, d.f. = 1, 38, $P < 0.008$). The difference was largely due to the striking difference in visitation rate by flies, which favoured male-sterile (36 ± 3 insects/census) over male-fertile trees (16 ±

2 insects/census) ($F = 30.5$, d.f. = 1, 38, $P < 0.0001$). In contrast, visitation rate by bees was greater for male-fertile trees (16 ± 2 insects/census v. 11 ± 1 insects/census in male-steriles) ($F = 5.16$, d.f. = 1, 38, $P = 0.0029$) (Fig. 2). Most bee species were collecting pollen in addition to nectar (e.g. *Bombus* spp., *Andrena* spp.) and this might explain the difference in visitation patterns.

The two gender types differed significantly in both the diversity of the flower visitor assemblage and the relative proportions of the major taxa (dipterans and hymenopterans, Table 1). The diversity of insect visitors was highest for male-fertile trees ($H' = 3.02$, Shannon-Wiener index) than for male-sterile trees ($H' = 2.72$). This was largely an effect of the greater diversity of the fly assemblage at male-fertile trees; the diversities of the bee assemblage at the two types of trees were very similar (see Table 1).

Experimental pollinations and seed set

P. mahaleb is partially self-compatible in the study area and requires cross-fertilization for maximum fruit set. In addition, differences in seed set between the two sexual forms are readily apparent, since male-steriles effectively behaved as functional females (Table 2, see below).

Fruit set was absent in bagged inflorescences of male-sterile individuals and was very reduced in male-fertiles (11.6% for the two years pooled) (Table 2). Fruit set was totally absent in emasculated-bagged inflorescences. Bagged inflorescences yielded significantly reduced fruit set relative to the other treatments, suggesting that insect intervention is necessary for successful pollination and maximum seed set ($P < 0.007$, for the possible Scheffé post-hoc comparisons between the bagged and other treatments, only male-fertile trees).

TABLE 2. Percentage of flower setting fruits from controlled pollinations

Functional gender and year	Pollination treatment	Number of flowers	% fruit set (*n*)*
Male-fertile			
1988	Control	1208	59.1 ± 2.2 (5)
	Bagged	–†	–†
	Self-pollination	32	69.9 ± 4.3 (4)
	Cross-pollination	–†	–†
1989	Control	1724	23.3 ± 1.8 (26)
	Bagged	535	11.5 ± 1.6 (23)
	Self-pollination	340	33.9 ± 3.5 (14)
	Cross-pollination	184	24.5 ± 6.8 (10)
Male-sterile			
1988	Control	2315	40.8 ± 4.5 (26)
	Bagged	211	0.2 ± 0.1 (11)‡
	Self-pollination	51	1.5 ± 1.5 (5)‡
	Cross-pollination	69	66.1 ± 7.7 (6)
1989	Control	712	12.3 ± 3.1 (13)
	Bagged	229	0 (10)
	Self-pollination	350	0 (13)
	Cross-pollination	161	17.4 ± 3.7 (11)

*Mean, standard error, and number of experimental branches

†No data available

‡One experimental bag partially open; fruit set attributable to accidental pollination within the bag

A three-way ANOVA testing simultaneously for the effects of gender type, pollination treatment, and year showed highly significant differences associated with each effect and their interactions ($F = 42.45$, d.f. = 13, 148, $P < 0.0001$), accounting for 85.2% of total variance in fruit set. Gender expression explained only 6.55%, while the treatment and year effects accounted for 78.62% (37.54% and 41.08%, respectively).

For the two years pooled, control branches of male-fertile trees showed greater fruit set ($\bar{x} = 29.1 \pm 2.8$, $n = 31$) than male-steriles ($\bar{x} = 25.3 \pm 3.8$, $n = 23$) ($F = 15.80$, d.f. = 1, 50, $P < 0.0001$). Between-year differences were also significant ($F = 91.43$, d.f. = 1, 50, $P < 0.0001$), but did not interact significantly with the gender type ($F = 1.64$, d.f. = 1, 50, $P = 0.21$). Thus, fruit set of male-fertile controls decreased from 59.1% in 1988 to 23.3% in 1989. Male-sterile controls showed a similar trend, from 40.8% to 12.3%. The difference can be explained by the persistent fine weather during the spring of 1988, favouring insect activity; this contrasted with the more rainy weather during the flowering period of 1989.

When compared to the open-pollinated controls, the cross-pollination treatment significantly enhanced fruit set only in the male-sterile trees ($F = 18.67$, $P = 0.0001$; Table 2). In male-fertile trees, fruit set resulting from cross-pollinations was significantly greater than in selfed crosses ($F = 7.51$, $P = 0.008$) but did not differ significantly from controls ($F = 0.33$, $P = 0.57$) (Table 2). Finally, selfed crosses in male-fertile trees significantly enhanced fruit set over controls ($F = 6.22$, $P = 0.02$).

The between-gender contrasts are summarized in Table 3. As in the above set of comparisons, to test for the difference between the two gender types in treatment effects, I contrasted simultaneously the 1988 and 1989 values of each level. The pooled means for the three treatments (control, selfed, and crossed) were 29.05%, 41.9%, and 38.6%, respectively, for male-fertile trees; those for male-steriles were 25.3%, 0.43% (due to accidental breakage of a bag), and 39.2%, respectively. Male-fertile trees showed greater fruit set than male-steriles in the control treatment only (in addition, of course, to the self-pollination, as expected from the functional femaleness of male-steriles) (Table 3). Fruit set did not differ, however, between the cross-pollinations in the two gender types.

TABLE 3. Summary of the contrasts in fruit set between different pollination treatments in *P. mahaleb* trees of two gender types (see Table 2). The direction of the difference in mean fruit set (two years pooled) is indicated. MF, male-fertile; MSt, male-sterile; MS, Mean Square

	Male-fertile trees		
Male-sterile trees*	Control	Self-pollination	Cross-pollination
Control	MS = 0.263 $F = 15.80^{***}$ MF > MSt	MS = 0.641 $F = 38.55^{***}$ MF > MSt	MSt = 0.145 $F = 8.75^{*}$ MF > MSt
Cross-pollination	MS = 0.03 $F = 1.88^{\mathrm{NS}}$ –	MS = 0.02 $F = 1.30^{\mathrm{NS}}$ –	MS = 0.04 $F = 2.50^{\mathrm{NS}}$ –

*Only results for the control and cross-pollination are reported since male-steriles behaved as functional females, with absence of pollen production (see text).

Arcsine transformed data prior to analysis. d.f. = 1,107.

$^{***}P < 0.0001$; $^{*}P < 0.05$; NS, non-significant.

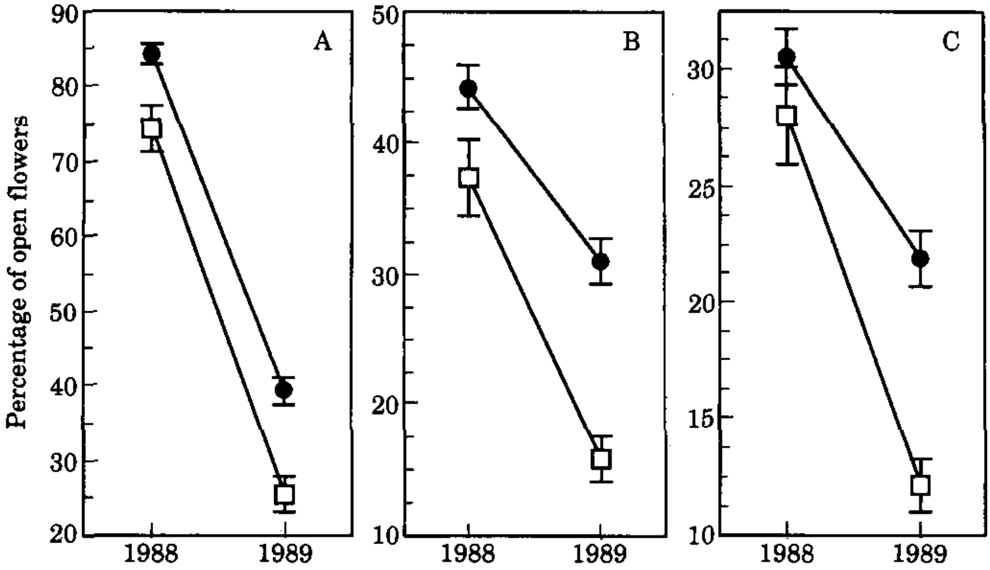

Figure 3. Differences between gender type and year in fruit set (relative to the number of open flowers) at different stages of fruit development. A, fruits initiated (2 mm diameter). B, fruits developed to 4 mm (approximately 50% full-size). C, fruits developed to final size. ●, Male-fertile trees; □, male-sterile trees; data represented as mean ± 1 SE for the two gender types.

The results of the counts in marked branches confirmed the above differences between the two gender types. The proportion of fully developed fruits (fruits at final size) relative to open flowers was significantly greater among male-fertile ($\bar{x} = 26.2 \pm 0.9\%$, $n = 123$) than male-sterile ($\bar{x} = 20.1 \pm 1.5\%$, $n = 80$) ($F = 11.73$, d.f. = 1, 161, $P = 0.003$) trees (Fig. 3C). In 1988, differences in fruit set between the two gender types were not significant ($F = 1.53$, $P = 0.22$), but in 1989 male-fertile trees showed a significantly greater mean fruit set ($F = 32.10$, $P < 0.0001$; Fig. 3). Male-fertile trees also showed a fecundity advantage over male-steriles when considering, in addition to the greater proportion of full-size fruits set, the proportion of flowers initiating fruit growth (Fig. 3A), and the proportion setting enlarged fruits (those with fruits reaching at least 3–4 mm diameter) (Fig. 3B). This difference was maintained both in 1988 and 1989.

Seed size and germination

Dry mass of seeds resulting from artificial pollination differed significantly both between gender types ($F = 8.46$, d.f. = 1, 385, $P < 0.005$) and between treatments ($F = 7.99$, d.f. = 2, 385, $P < 0.0001$), with no significant interaction

TABLE 4. Variation in dry seed mass (mg) of fruits resulting from experimental hand pollination treatments

Functional gender	Control	Self-pollination	Cross-pollination
Male-fertile	65.66 ± 0.77 $n = 174$	62.28 ± 1.57 $n = 4$	71.75 ± 1.99 $n = 39$
Male-sterile	74.33 ± 1.30 $n = 72$	–	77.01 ± 3.68 $n = 20$

($F = 0.18$, $P = 0.67$). This model, however, explained a relatively small fraction of total variance ($R^2 = 0.140$). Gender accounted for 8.06% of the variance, and the treatment effect for 1.58%. When I incorporated a plant effect nested within the gender type effect, it explained 19.10% of total variation. Thus, most variance in seed mass in the pollination experiments was related to within-plant variation and was independent of gender effects. Table 4 summarizes the mean seed dry masses obtained in each combination of the two effects.

As regards germination in natural conditions for seeds from the two morphs ($F = 0.18$, d.f. = 1, 136), average percent germination for seeds from male-sterile trees ($\bar{x} = 28.5 \pm 2.8\%$; $n = 64$) did not differ significantly from that of male-fertile trees ($\bar{x} = 29.7 \pm 2.4\%$; $n = 80$). A significant effect of microhabitat type on germination percentage, but no significant interaction with gender type has been observed (L. López-Soria, E.W. Schupp & P. Jordano, pers. obs.).

Differences in fecundity

The overall difference in fruit crop size between the two gender types is significant (Table 5; $F = 4.90$, d.f. = 1 and 93, $P = 0.03$), especially in the years of greatest fruit production (1988 and 1991). Male-sterile trees produced a four year average of 6558 fruits, while male-fertile ones yielded 4670 fruits. However, these differences in total fecundity vanished after accounting for variation in canopy size by means of ANCOVA ($F = 0.451$; d.f. = 1, 93; $P = 0.503$; least-squares fruit crop-size means for male-sterile and male-fertile trees were 5334 and 5423 fruits, respectively; $P = 0.904$). Male-fertile trees and male-sterile trees, therefore, are similar in fecundity relative to size.

Male-steriles had a fecundity advantage only in 1988 and 1991 (Table 5). Fruit crop sizes were more similar between the two types of trees during 1989 and 1990, when persistent rainy weather during the pollination period (late May-early June) occurred. In the two years with better climatic conditions for pollination (1988 and 1991), male-steriles showed a notorious fecundity advantage.

The size of fruit crops sizes produced by individual plants in successive years (period 1988–1991) were strongly correlated ($r_s > 0.438$, $P < 0.03$), indicating a relative constancy of the fecundity rankings between years; in any given year, the best predictor of the number of fruits produced was plant size.

Data on flower production and seed set are available for 25 intensively-studied trees in 1988 and 1989 (Table 6). These data further support the greater fecundity of male-sterile trees. Male steriles produced, on average, a greater

TABLE 5. Differences between gender types in fruit production in the four study years. Figures are median crop sizes and 25%–75% quantiles

Year	Number of fruits	
	Male-sterile trees $n = 10$	Male-fertile trees $n = 15$
1988	9580 (7565–14 700)	4625 (2665–8008)
1989	1990 (1515–3655)	1525 (1024–4596)
1990	2005 (1200–3250)	1505 (961–3585)
1991	8540 (7623–13 559)	7630 (4092–9607)

TABLE 6. Production of flowers and fruits, proportion of flowers setting final-sized fruits (fruit set) and seed dry mass of plants differing in functional gender

Year and functional gender	Number of flowers*	Number of fruits*	Fruit set†	Seed mass‡ (mg)
1988				
Male-fertile	15960	4625	0.334	–
(n = 15 trees)	9141–26 373	2545–8155	0.294–0.375	
Male-sterile	21 676	9580	0.330	–
(n = 10 trees)	12 852–50 407	6758–14 939	0.253–0.407	
1989				
Male-fertile	8243	1525	0.232	65.50
(n = 15 trees)	4218–24 760	995–4920	0.180–0.285	0.70
				(n = 297)
Male-sterile	21062	1990	0.128	74.91
(n = 10 trees)	14 158–31 902	1499–3681	0.069–0.186	1.29
				(n = 92)

*Median, 25% and 75% quantiles

†Mean and 95% confidence limits

‡From a sample of ripe fruits of the 1989 season. Mean, standard error and sample size.

number of flowers than male-fertiles. During 1988 both types of trees showed similar fruit set (33%; Table 6). However, during 1989, fruit set decreased probably as a result of the adverse weather during the pollination period. Male-steriles experienced a greater decrease of fruit set (20.2% less), relative to the previous year, than male-fertiles (10.2% less) and this fact tended to obscure the differences in absolute fecundity.

DISCUSSION

Flower biology and gynodioecy

Individual *P. mahaleb* trees showed either hermaphroditism, producing perfect flowers, or functional femaleness, with production of pollen-sterile flowers, a situation of functional gynodioecy. Careful inspection of the flowers in the field readily revealed the gender of each tree, since the anthers differ strikingly in size, shape, and colour. In addition, experimental self-pollinations (both autogamous and geitonogamous crosses) always resulted in no seed-set in cryptic putative females. Male-sterile trees represented 42.4% of the reproductive population, and censuses at nearby populations indicated that this male-sterile proportion is common in the general study area. The gender expression of individuals was consistent during four consecutive study years. As far as I know, this is the first report of such reproductive behaviour in *P. mahaleb* and one of the few cases of gynodioecy in Rosaceae (see Jain, 1959). Richards (1986: 305; also see Krohne, Baker & Baker, 1980) reported a very similar situation in *Plantago lanceolata*, where females show well formed anthers which are yellow, rather than cream colour, because they lack pollen. Male-sterility has been reported previously for cultivated rosaceous species (Jain, 1959; Vargas & Romero, 1976; also see Campbell *et al.*, 1991; Dickinson & Campbell, 1991). The presence of these types of 'hidden' gender expression might be much more common in wild populations of rosaceous trees and shrubs than previously recognized.

The evidence in favour of a situation of stable gynodioecy in wild *P. mahaleb* populations is at best preliminary. In stable gynodioecy, male-sterility is frequently controlled by at least one nuclear factor unlinked with a second factor which is frequently cytoplasmic (Ross, 1978). Jain (1959) pointed out in his review that nuclear factors alone are responsible for male-sterility in other *Prunus* species and absence of plasm factors has been documented. The data obtained in the present study show that male-steriles have a fitness advantage over male-fertiles, but a two-fold fecundity advantage of male-steriles occurred only in certain reproductive episodes. This is the fecundity differential needed, with a model of nuclear-gene inheritance of male-sterility, for females to be maintained in the population (Lewis, 1941; Lloyd, 1974). In terms of total fecundity (number of full-size ripe fruits), male-sterile trees produced a four-year average 1.45 times greater than male-fertile trees. Annual mean crop sizes of male-sterile trees for the years 1988 to 1991 showed 2.1, 1.3, 1.3 and 1.1-fold advantages, respectively, relative to male-fertile averages. This fecundity advantage was greater during years with favourable conditions for pollination. Individual plants showed consistent between-year rankings in fruit crop sizes, suggesting that a slight fecundity differential associated with gender expression might have long-term consequences in the relative fitness of the two groups of individuals. These results emphasize the need for several years of monitoring of individually-identified plants to reveal a consistent fecundity advantage of male-steriles, or the existence of sporadic reproductive seasons when a large fecundity difference between the two morphs occurs.

Variation in fitness components associated with gender type

In addition to variation in total fecundity, differences in other fitness components that may influence directly the lifetime female fitness are less well documented in wild populations of gynodioecious species (Ågren & Willson, 1991). Previous studies have demonstrated that the fitness advantage of male-sterile plants is frequently brought about by differences in relative fecundity (e.g. fruit set), seed size, seedling survival, or vegetative growth, and not by an initial absolute fecundity differential (Richards, 1986; Gouyon & Couvet, 1987). Male-sterile *P. mahaleb* trees produced more flowers, fruits, and heavier seeds than male-fertiles. However, fruit-set/flower and the number of ripe fruits/flower were much lower in male-steriles. This fruit-set disadvantage did not compensate for the difference in absolute fecundity, and male-sterile trees produced a final crop size, on average, 1.45 times that of male-fertile trees. A number of researchers (e.g. van Damme, 1984; Stevens, 1988; Uno, 1982a) have documented reduced fruit set in male-sterile plants but better performance of this morph in other fitness components. The results of the present study concur with this. Differences with other studies (Assouad *et al.*, 1978; Ågren & Willson, 1991) probably originate from differences between the species in the effects of inbreeding depression.

The hypothesis of pollinator limitation of fruit set in *P. mahaleb*, especially in male-sterile trees, is supported by the results of experimental pollinations. First, male-fertile trees showed greater fruit set than male-steriles. Second, addition of cross pollen significantly enhanced fruit set over controls only in male-sterile trees, suggesting that fruit set was pollen-limited in this morph rather than resource-limited, most likely as a result of the impossibility for self-pollination. Third, reductions of fruit set in male-fertiles during unfavourable years were less severe than in male-steriles, as expected by the possibility for geitonogamous crosses.

The greater total fecundity of male-sterile plants is usually brought about by increased seed set/ovule and not by a difference in absolute fecundity (i.e. greater flower production or greater number of ovules/flowers) (see reviews in Richards, 1986; Gouyon & Couvet, 1987). Addition of cross pollen frequently increases the number of seeds/fruit in multi-seed fruits, supporting the idea that selective embryo abortions, rather than stylar incompatibility reactions, occur (Ågren & Willson, 1991). Results with *P. mahaleb*, however, suggest that the effect of cross-pollination would benefit male-sterile trees only if conditions for pollination are favourable. As stressed by Lewis (1941), the advantage of male-steriles due to outcrossing will depend on the balance between the advantage conferred by selfing in unfavourable 'pollination environments', and the negative effects of inbreeding depression on the selfed progeny.

The progeny of females can be fitter either as a result of outbreeding or due to increased resource allocation to seeds relative to hermaphrodites (Ågren & Willson, 1991; Mitchell-Olds & Waller, 1985; Richards, 1986). Experimental pollinations with *P. mahaleb* support the idea that the increased fitness advantage of male-sterile trees was an effect of having all the seeds outcrossed. In self-compatible species that show inbreeding depression, females may benefit by having all flowers that set fruit outcrossed, while a fraction of the seeds of hermaphrodites results from selfing. In *P. mahaleb*, cross-pollination increased fruit set of male-fertiles over self-pollination, a result expected if difference between morphs in seed production is partly due to differences in outcrossing rates (Ågren & Willson, 1991). On the other hand, under a resource allocation hypothesis, male-steriles would be able to re-allocate more resources to seed production than male-fertiles. Consequently, the seed set of male-fertiles would be lower than in male-steriles, even when contrasting the cross-pollinations. The results do not support this hypothesis, since the fruit set in the cross-pollination treatments in the two gender types did not differ significantly.

Differences in seed mass between the two gender types of gynodioecious species, with females producing heavier seeds than hermaphrodites, have been repeatedly documented in the literature (see review in Richards, 1986; Ågren & Willson, 1991) and this seems a very general pattern associated with the gynodioecious breeding system or other outcrossing systems (Mitchell-Olds & Waller, 1985). Male-sterile *P. mahaleb* trees also differed significantly from male-fertiles in seed mass, producing, on average, heavier seeds. The hypothesis of the outcrossing advantage is supported again by the analysis of seed masses from the experimental pollinations. First, outcrossed seeds were heavier ($\bar{x} = 73.5$ mg) than both controls ($\bar{x} = 68.2$ mg) and those from self-pollinations ($\bar{x} = 62.3$ mg). Second, the outcrossed seeds of male-fertile trees were heavier ($\bar{x} = 71.7$ mg) than their selfed seeds ($\bar{x} = 62.2$ mg). Although these differences did not translate into different germination abilities, they may have deferred consequences for seedling vigour and survival (P. Jordano, E. W. Schupp & L. López-Soria, in prep.). Also, if seed-set and seed size of male-fertile trees are resource-limited, I would expect the outcrossed seeds of male-steriles to be heavier (all outcrossed). This is not supported by the data; the masses of outcrossed seeds of the two gender types were not significantly different.

Pollinator activity in the two floral morphs

To my knowledge, previous studies of entomophilous gynodioecious species have not documented differences among sexual morphs in pollinator activity. Differences in visitation by pollinator assemblages dominated by different insect taxa (e.g. bees and flies) would have important implications for pollen flow between hermaphrodites and females, and for selfing rates of hermaphrodites. For example, in general, bees have been found to carry more pollen grains than flies (Herrera, 1987, and references therein) and fly longer distances, although great variation within each of these taxa occurs and generalizations might not be valid in a biological context. Uno (1982a,b) found similar nectar flow patterns of gynodioecious *Iris douglasiana* flowers of the two morphs and stated that male-sterile flowers depend on the same pollinators that visit hermaphrodite flowers and could be thought of as mimics of them. Atsatt & Rundel (1982) reported that hermaphrodite *Fuchsia lycioides* produce up to six times more nectar than females, but apparently are visited with similar frequency by foraging hummingbirds. In *P. mahaleb*, male-sterile and male-fertile flowers secreted a similar amount of sugar per day, but the concentration of nectar was significantly higher in male-fertile flowers which secreted somewhat less nectar volume. This pattern has been reported previously in other cryptically-diclinous species (see Mayer & Charlesworth, 1991).

The differences in the pollinator assemblages visiting the two morphs could be caused by differences in nectar secretion patterns, in flower production (more showy displays of male-sterile trees), the presence of pollen as a reward itself in pollen-fertile flowers, or a combination of these. The result was a striking difference, both qualitative and quantitative, in the composition of insect visitor assemblages in the two *P. mahaleb* morphs. Visitation rates were much higher at male-sterile trees (a 1.5-fold difference with male-fertiles) and the assemblage was dominated by calliphorid flies. Male-fertile trees were visited in equal proportions by pollen-collecting solitary bees (mostly *Andrena* spp.) and flies, with the dipteran assemblage showing a greater proportion of syrphid flies. Greater proportion of syrphids in male-fertiles is expected from their known pollinivorous habit (Gilbert, 1981).

Outcrossing and effects on fitness via pollen and seeds

The extent to which pollination events limit fruit-set, particularly in partially self-compatible species, will depend strongly on the 'pollinator environment', as stressed some time ago by Lewis (1941). The estimates of outcrossing for gynodioecious hermaphrodites are extremely variable, ranging between 25%–83% (Richards, 1986, and references therein), but indicate substantial outcrossing. For example, with fine weather and high abundance of pollinators, male-steriles can obtain maximum fruit set whereas in situations of pollinator limitation, the ability of male-fertiles to self-pollinate may compensate the negative effects of infrequent outcrossing (Jain, 1976; Lloyd, 1979).

The lower natural levels of seed-set documented for male-sterile *P. mahaleb* relative to male-fertiles fit well with the relatively infrequent visits by bees, presumably responsible for most pollen transfer. I would expect a relatively high outcrossing rate for male-fertiles, given the importance of bees as visitors to this morph. A preliminary estimate of the outcrossing rates of male-fertile *P. mahaleb* trees can be gained from the results of the experimental pollinations; obviously, more direct methods such as assays of progeny arrays for polymorphic allozyme loci are needed to validate this estimate. If seed set is limited by the level of outcrossing, a rough estimate of selfing rate is obtained by the expression: $S = (P_{cross}\text{-}P_{open})/(P_{cross}\text{-}P_{self})$ (Charlesworth, 1988; Ågren & Willson, 1991), where S is selfing rate, and P_{cross}, P_{self}, and P_{open}, are the seed value (seed mass, in this case), after cross-, self-, and open-pollination, respectively. The estimate of selfing rate for *P. mahaleb* male-fertile trees yields $S = 0.4732$, a figure well below average selfing values of self-compatible species ($\bar{x} = 0.6010$, from Richards, 1986, Table 9.5). This result is similar to the figure obtained by Sun & Ganders (1986) with Hawaiian *Bidens* (Asteraceae), who found selfing rates of hermaphrodites ranging between 40–60% in populations with 30–40% female plants.

From a theoretical standpoint, a 1:1 sex ratio as documented here for *P. mahaleb* supports the contention that most gene transfer by male-fertile trees is via pollen (see Lloyd, 1976). Conservative estimates of the proportion of genes contributed via pollen for this morph, based on seed mass data (eqn. 8 in Lloyd, 1976) yield 0.69. This fits well with the low selfing rate estimated above and suggests that *P. mahaleb* is closer to the dioecious end of the hermaphroditism-dioecy continuum. However, extreme between-year variations in seed set and fecundity of hermaphrodites, together with similar germination ability indicate that the seed contribution of this morph has a great demographic relevance. Further study is needed, encompassing a broader geographical scale, to test the consistency of these patterns across *P. mahaleb* populations.

Concluding remarks

The gynodioecious breeding system described here for *P. mahaleb* has important implications beyond the pollination biology of the species. Gouyon & Couvet (1987) have outlined possible ecological, as well as genetic, consequences of the presence of functionally female individuals in these populations: these include increase of neighbourhood area by the decrease in selfing and a potential increased dispersal of the female progeny, and increased reproductive output of the population as a whole by the presence of more fecund females with longer life span. *P. mahaleb* seeds are dispersed predominantly by birds, but mammals also play a role (Herrera & Jordano, 1981; Herrera, 1989). The foraging modes of these frugivores within a population of trees is affected by the levels of fruit production of individual trees, and the characteristics of the fruits as well (seed size, fruit size, pulp/seed ratio, etc.; Jordano, 1992). From a demographic perspective, deferred consequences of differences in fecundity and seed size between morphs will have obviously a potential influence on the interactions of *P. mahaleb* with the frugivorous animals that disperse the seeds. For example, events during the dispersal phase (e.g. differential fruit removal from the two morphs) could compensate for fitness differences originating from the pollination phase. Further research is needed to elucidate the combined role of animal pollinators and seed dispersers in the demography and population genetics of diclinous species.

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REFERENCES

Ågren J, Willson M F. 1991. Gender variation and sexual differences in reproductive characters and seed production in gynodioecious *Geranium maculatum*. *American Journal of Botany* **78:** 470–480.

Ashman T, Stanton M. 1991. Seasonal variation in pollination dynamics of sexually dimorphic *Sidalcea oregana* ssp. *spicata* (Malvaceae). *Ecology* **72:** 993–1003.

Assouad MW, Dommée B, Lumaret R, Valdeyron G. 1978. Reproductive capacities in the sexual forms of the gynodioecious species *Thymus vulgaris*. *Botanical Journal of the Linnean Society* **77:** 29–39.

Atsatt PR, Rundel PW. 1982. Pollinator maintenance vs. fruit production: partitioned reproductive effort in subdioecious *Fuchsia lycioides*. *Annals of the Missouri Botanical Garden* **69:** 199–208.

Bawa KS, Webb CJ. 1983. Floral variation and sexual differentiation in *Muntingia calabura* (Elaeocarpaceae), a species with hermaphrodite flowers. *Evolution* **37:** 1271–1282.

Campbell CS, Greene CW, Dickinson TA. 1991. Reproductive biology in subfam. Maloideae (Rosaceae). *Systematic Botany* **16:** 333–349.

Charlesworth D. 1988. A method of estimating outcrossing rates in natural populations of plants. *Heredity* **61:** 469–471.

Darwin C. 1877. *The different forms of flowers on plants of the same species*. London: Murray.

Dickinson TA, Campbell CS. 1991. Population structure and reproductive ecology in the Maloideae (Rosaceae). *Systematic Botany* **16:** 350–362.

Dommeé B, Assouad MW, Valdeyron G. 1978. Natural selection and gynodioecy in *Thymus vulgaris* L. *Botanical Journal of the Linnean Society* **77:** 17–28.

Gagnon J, Roth JM, Carroll M, Hofmann R, Haycock KA, Plamondon J, Feldman Jr, DS, Simpson J. 1989. *SuperAnova. Accessible general linear modeling.* California: Abacus Concepts.

Gilbert FS. 1981. Foraging ecology of hoverflies: morphology of the mouthparts in relation to feeding on nectar and pollen in some common species. *Ecological Entomology* **6:** 245–262.

Gouyon PH, Couvet D. 1987. A conflict between two sexes, females and hermaphrodites. In: Stearns SC, ed. *The evolution of sex and its consequences:* Basel: Birkhäuser, 245–261.

Herrera CM. 1987. Components of pollinator "quality": comparative analysis of a diverse insect assemblage. *Oikos* **50:** 79–90.

Herrera CM. 1989. Frugivory and seed dispersal by carnivorous mammals, and associated fruit characteristics, in undisturbed mediterranean habitats. *Oikos* **55:** 250–262.

Herrera CM, Jordano P. 1981. *Prunus mahaleb* and birds: the high efficiency seed dispersal system of a temperate fruiting tree. *Ecological Monographs* **51:** 203–21.

Jain SK. 1959. Male sterility in flowering plants. *Bibliographia Genetica* **18:** 101–166.

Jain SK. 1976. Evolution of inbreeding in plants. *Annual Review of Ecology and Systematics* **7:** 469–495.

Jordano P. 1992. Fruits and frugivory. In: Fenner M ed. *Seeds: the ecology of regeneration in plant communities.* London: C.A.B. International. 105–156.

Kevan PG, Eisikowitch D, Ambrose JD, Kemp JR. 1990. Cryptic dioecy and insect pollination in *Rosa setigera* Michx. (Rosaceae), a rare plant of Carolinian Canada. *Biological Journal of the Linnean Society* **40:** 229–243.

Krohne DT, Baker I, Baker HG. 1980. The maintenance of the gynodioecious breeding system in *Plantago lanceolata* L. *American Midland Naturalist* **103:** 269–279.

Lewis D. 1941. Male-sterility in natural populations of hermaphrodite plants. *New Phytologist* **40:** 56–63.

Liston A, Rieseberg LH, Elias TS. 1990. Functional androdioecy in the flowering plant *Datisca glomerata*. *Nature* **343:** 641–642.

Lloyd DG. 1972. Breeding systems in *Cotula* L. (Compositae, Anthemideae) I. The array of monoclinous and diclinous systems. *New Phytologist* **71:** 1181–1194.

Lloyd DG. 1973. Sex ratios in sexually dimorphic Umbelliferae. *Heredity* **31:** 239–249.

Lloyd DG. 1974. The genetic contributions of individual males and females in dioecious and gynodioecious angiosperms. *Heredity* **32:** 45–51.

Lloyd DG. 1976. The transmission of genes via pollen and ovules in gynodioecious angiosperms. *Theoretical Population Biology* **9:** 299–316.

Lloyd DG. 1979. Some reproductive factors affecting the selection of self-fertilization in plants. *American Naturalist* **113:** 67–79.

Mayer SS, Charlesworth D. 1991. Cryptic dioecy in flowering plants. *Trends in Ecology and Evolution* **6:** 320–325.

Mitchell-Olds T, Waller DM. 1985. Relative performance of selfed and outcrossed progeny in *Impatiens capensis*. *Evolution* **39:** 533–544.

Primack RB, McCall C. 1986. Gender variation in a red maple population (*Acer rubrum*; Aceraceae): a seven-year study of a "polygamodioecious" species. *American Journal of Botany* **73:** 1239–1248.

Prys-Jones O, Corbet SA. 1978. *Bumblebees*. Cambridge: Cambridge Univ. Press.

Richards AJ. 1986. *Plant breeding systems*. London: Allen & Unwin, Ltd. **Ross MD. 1978.** The evolution of gynodioecy and subdioecy. *Evolution* **32:** 174–188.

SAS 1986. *User's guide: Statistics, v. 6.0*. Cary, North Carolina: SAS Institute.

Snedecor GW, Cochran WG. 1989. *Statistical methods*. Ames, Iowa: Iowa State Univ. Press.

Stevens DP. 1988. On the gynodioecious polymorphism in *Saxifraga granulata* L. (Saxifragaceae). *Biological Journal of the Linnean Society* **35:** 15–28.

Stevens DP, Richards AJ. 1985. Gynodioecy in *Saxifraga granulata* L. (Saxifragaceae). *Plant Systematics and Evolution* **151:** 43–54.

Steyn EMA, Robbertse PJ. 1988. Flower production and sexuality in *Bequaertiodendron magalismontanum*. *South African Journal of Botany* **54:** 481–486.

Sun M, Ganders FR. 1986. Female frequencies in gynodioecious populations correlated with selfing rates in hermaphrodites. *American Journal of Botany* **73:** 1645–1648.

Uno GE. 1982a. Comparative reproductive biology of hermaphroditic and male-sterile *Iris douglasiana* Herb. (Iridaceae). *American Journal of Botany* **69:** 818–823.

Uno GE. 1982b. The influence of pollinators on the breeding system of *Iris douglasiana*. *American Midland Naturalist* **108:** 149–158.

Valle F, Gómez F, Mota F, Díaz C. 1989. *Parque Natural de Cazorla, Segura y Las Villas. Guía botánico-ecológica*. Madrid: Editorial Rueda.

Van Damme JMM. 1984. Gynodioecy in *Plantago lanceolata* L. III. Sexual reproduction and the maintenance of male steriles. *Heredity* **52:** 77–93.

Vargas FJ, Romero M. 1976. Androesterilidad en la variedad de almendro "Rof". I. Cong. Int. de Almendra y Avellana, Reus, pp. 523–525.

Webb DA. 1968. *Prunus* L. In: Tutin TG *et al.*, eds. *Flora Europaea, vol. 2*, Cambridge: Cambridge Univ. Press.

Young DA. 1972. The reproductive biology of *Rhus intergrifolia* and *Rhus ovata* (Anacardiaceae). *Evolution* **26:** 406–414.