AN ABSTRACT OF THE THESIS OF

Thomas R. Jahns for the degree of Doctor of Philosophy in Crop Science presented on July 31, 1990.

Title: Pollination Biology and Pollinator Alternatives in Mermaid Meadowfoam (Limnanthes alba Hartw. ex Benth.)

Abstract Approved: Redacted for Privacy
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Meadowfoam (cultivar Mermaid) is an entomophilous winter annual oilseed crop that has historically produced an average of only two of five seeds per flower. Reference to inadequate meadowfoam pollination exists in the literature, but quantitative evidence is lacking. Studies were undertaken to: 1) quantify meadowfoam pollination requirements and 2) evaluate the potential of an alternative pollinator. In vivo pollination biology studies tested pollen age, stigma age, stylar restriction, and pollen deposition rate effects on seed set. Yield efficacy of Osmia lignaria propinqua Cresson, a native wild bee pollinator, was compared in cages to a honey bee standard and a non-caged honey bee control. Osmia reproductive potential was also tested. Pollen 0-5 days old (postanthesis), stored at 3,

18, or 37^{0}C, did not appear to limit seed set. Stigma age was critical for seed set maximization. Seed set was not influenced by the number of stigmas pollinated per flower, but was limited by less than 25 pollen grains deposited per flower. Seed set and pollen deposition increased with increasing honey bee visits per flower. It was concluded that at least three honey bee colonies per acre should be used for commercial meadowfoam production. Osmia produced comparable individual plant yields to honey bees. Sixty Osmia produced similar solid stand yields to 4000 honey bees. Significantly greater solid stand yields per bee were obtained from Osmia when compared to the honey bee. Osmia survival and female production were negatively correlated with female density, while nest/male/total cell production was positively correlated with female density. Osmia demonstrated yield improvement potential as a meadowfoam pollinator.

Pollination Biology and Pollinator Alternatives in Mermaid Meadowfoam (*Limnanthes alba* Hartw. ex Benth.)

by

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PREFACE

This thesis was written in manuscript format. Dr. Gary D. Jolliff was co-author on all six manuscripts comprising Chapters I-V, and Appendix I of this thesis. Dr. Jolliff's monumental contributions to this thesis included: 1) recognizing initial pollination problems in meadowfoam, 2) establishing priority for pollination research, 3) generating research funding, 4) providing equipment and facilities, 5) recuiting me to identify pollination problems, 6) advising me on all phases of my research, and 7) critical review of all manuscripts.

Chapter I was written for Crop Science, and is currently in departmental review. Robert E. Franz is a co-author on this paper. Bobs' contributions to Chapter I included: 1) participation in hypothesis conception, 2) hand pollination and data taking on "Stigma Age" study, and 3) manuscript review.

Chapter II was published in Crop Science 30(4):850-853).

Chapter III was accepted by the Oregon State Agricultural Experiment Station, as Extension Circular #1360 and is scheduled for printing in early 1991. D. M. Burgett is co-author on this paper. Dr. Burgett's contributions to Chapter III included: 1) participation in hypothesis conception, 2) apicultural expertise and assistance, 3) equip-

ment and facility donations, 4) critical manuscript review, and 5) Extension Circular funding.

Chapter IV was tentatively accepted by Crop Science on July 24, 1990, and is currently being revised for return to associate editor.

Chapter V was submitted to the Journal of the Kansas Entomological Society on March 1, 1990.

The Appendix I paper was produced for the Department of Environmental Quality, Advisory Committee on Field Burning, as partial fulfillment of funded research conducted during the 1987 meadowfoam bloom period.

POLLINATION BIOLOGY AND POLLINATOR ALTERNATIVES IN MERMAID MEADOWFOAM (LIMNANTHES ALBA HARTW. ex BENTH.)

INTRODUCTION

This thesis is written in manuscript format. It reports the following five studies: 1) "Pollen Longevity, Stigmatal Receptivity, and Stylar Commonality Effects on Meadowfoam Seed Set", 2) "Pollen Deposition Rate Effects on Seed Set in Meadowfoam", 3) "Pollination and Seed Set of Meadowfoam", 4) "Osmia lignaria propinqua Cresson: an Alternative Pollinator for Meadowfoam in Cages", and 5) "Survival Rate and Reproductive Success of Osmia lignaria propinqua Cresson (Hymenoptera: Megachilidae) in Caged Meadowfoam, Limnanthes alba Benth. (Limnanthaceae)".

It has been speculated (Calhoun and Crane, 1978; Gentry and Miller, 1965; McGahuey, 1986; Franz and Jolliff, 1989) that inadequacies in pollination biology limit meadowfoam seed yield. Accelerated commercialization of this new crop is dependent on increased seed yield. These studies found that seed set was not influenced by the number of stigmas pollinated per flower or by pollen age, but was significantly influenced by all treatments which modified pollen deposition rate and timing. Therefore, it appears that the future of meadowfoam seed yield improvement will be highly dependent on the strength of pollinator populations and the precision of pollinator management.

CHAPTER I

POLLEN LONGEVITY, STIGMATAL RECEPTIVITY, AND STYLAR COMMONALITY EFFECTS ON MEADOWFOAM SEED SET

ABSTRACT

Limited quantitative information is available on the pollination limitations in meadowfoam (Limnanthes alba Hartw. ex Benth. cv. Mermaid). Three studies were conducted to determine whether pollen longevity, stigmatal receptivity, or stylar commonality were important seed set determinants. In vivo greenhouse hand-pollination studies tested: (i) pollen age (0-5 days postanthesis) on seed set production, under storage temperatures of 3, 18, and 37^{0}C; (ii) stigma age (0-120 hours postanthesis) on stigmatal receptivity; and (iii) pollination of 1, 3, and 5 stigmas per flower on seed set, as influenced by stylar commonality. Pollen age, between 0-5 days postanthesis, did not appear to reduce seed set under any of the three storage temperatures. Stigmas pollinated 24, 48, and 72 hours postanthesis produced 1.3, 3.3, and 0.9 seeds per flower, respectively. Hand pollination of 1 stigma per flower produced statistically similar seed set to 3 or 5 stigmas pollinated per flower. Pollen age and stylar structure offered insignificant limitations to meadowfoam pollination and seed set. Pollination at optimum stigmatal receptivity maximized seed set, indicating the importance of pollinator / peak bloom stigmatal receptivity synchrony for seed set maximization.

INTRODUCTION

Meadowfoam is an entomophilous, winter annual oilseed crop (Jolliff, 1989) that blooms in early May, when weather conditions are potentially unstable in Oregon's Willamette Valley. Cool (<15^{0}C) and wet weather conditions during meadowfoam bloom can inhibit flower opening (Kalin, 1971), thus delaying pollen dehiscence in new flowers and pollen removal (via honey bees) in older flowers. Hot (>25^{0}C) and dry weather conditions during bloom can stimulate pollen dehiscence and flower anthesis (opening), resulting in millions of newly opened flowers shedding pollen daily. If temperature extremes (<5^0 or >35^{0}C) and/or inadequate pollen foraging exists, tremendous pollen reserves may be subjected to potentially damaging temperatures (Huerta and Vasek, 1984), or may exhibit diminished prepollination viability with age (Morse, 1987), or both. The influence of pollen temperature exposure and pollen age on meadowfoam pollen viability has not been reported.

Stigmatal receptivity has been found to influence seed set in many agricultural, horticultural, and native plants (Chang and Struckmeyer, 1976; Schoper, et al., 1986; Morse, 1987). Preliminary studies revealed that stigma age, temperature exposure, and light quality may influence stigmatal receptivity in meadowfoam (Franz, 1990; Franz and Jahns -unpubl. data).

Little information is available in the meadowfoam literature on pollen viability or stigmatal receptivity. Ma-

son (1952) reported that 97.6% of L. alba var. alba pollen was viable, with protandry ranging from 1-3 days. Huynh (1971) found that L. douglasii pollen germinated easily in vitro when held at 25^{0}C for 12 hours. Devine and Johnson (1978) achieved up to 60% seed set (3 of 5 seeds per flower) when they applied hand pollination treatments to initially unreceptive stigmas of L. alba plants that were kept in a greenhouse at 30^{0}C. Kalin (1971) observed that L. alba var. alba took four days from anthesis to obtain stigmatal receptivity. Kesseli (1984) reported a similar protanrous duration in L. douglasii. Arroyo (1973) stated that a 2-3 day difference in the timing between anther dehiscence and stigmatal receptivity largely prevented self-pollination in L. alba. A 1-2 day delay between anther dehiscence and stigmatal receptivity was reported by Guerrant (1984) in L. alba.

In vivo studies, conducted to evaluate pollen age and stigmatal receptivity effects on seed production, have been a reliable method for substantiating fertilization and seed set capabilities (Huerta and Vasek, 1984; Schoper et al., 1986). To achieve maximum in vivo pollination in meadowfoam, at least 25 pollen grains per five stigmatal papillae (FSP) are required (Jahns and Jolliff, 1990). Mason (1952) reported that to achieve adequate hand-pollination, each of the five stigmatal papillae must be touched with an anther (containing viable pollen). In preliminary studies (Franz and Jahns-unpubl. data), we found that pollination to each

of the five stigmatal papillae may not be required to maximize seed set per flower.

A more thorough understanding of meadowfoam pollination requirements may enhance the development, management, and economic competitiveness of future cultivars. Consequently, we undertook in vivo greenhouse studies to establish the relative importance of (i) pollen age (under three storage temperatures) on seed production; (ii) stigma age on optimum pollination timing; and (iii) individual stigma pollinations on seed production, as influenced by stylar commonality.

MATERIALS AND METHODS

Greenhouse experiments were conducted at Oregon State University, Corvallis in 1987-88 (stigmatal receptivity study) and 1988-89 (pollen viability and stylar commonality studies). All plants used in these studies were grown from seed of Mermaid meadowfoam. Individual plant growing procedures and growth chamber environmental conditions, up to flower initiation (day-45), were identical to those reported by Jahns and Jolliff (1990). Fluorescent lights produced a photon flux density average of ca. 145 μmol m^{-2} s^{-1} throughout the testing periods.

Hand pollinations to all experimental units vectored a minimum of 25 pollen grains per FSP (Jahns and Jolliff, 1990) utilizing either a #000 camel's hair brush or a freshly dehiscing anther, depending upon treatment requirements. At harvest, seeds from each experimental assay were removed, sliced to measure embryo viability, and counted.

A randomized complete block design was used for each treatment study replicate (run). Data were analyzed using analysis of variance (ANOVA) procedures and treatment mean effects compared using Fisher's Protected LSD (FPLSD) at $P<0.05$. Homogeneous error terms between experimental replicates, as determined by an F-test of mean square error terms, allowed analysis pooling (Cochran and Cox, 1950).

Pollen Age and Seed Set

Pollen from 0-5 days postanthesis, was tested for seed production capability under three independent storage temperatures: (i) 3^0C (25-35% relative humidity (rh)), (ii) 18^0C (55-75% rh), and (iii) 37^0C (25-35% rh). The six post-anthesis storage periods included a 0-day control (18^0C 55-75% rh) through 5 days postanthesis, which is the ca. viability limit for an unpollinated flower (Franz and Jolliff, 1989). The three independent pollen storage temperatures represented: (i) extremely low, (ii) average, and (iii) extremely high temperatures encountered during meadowfoam bloom, over the past twelve years.

All assay-plants were kept in an 18^0C (55-78% rh) greenhouse from ca. 52 days postseeding through treatment application (ca. day-100) and harvest (ca. day-125). Anthers were excised from all open assay-plant flowers to inhibit pretreatment flower pollination by non-treatment pollen sources. A minimum of six flowers, at optimum stigmatal receptivity (Jahns and Jolliff, 1990), were required per assay plant. Six plants per experimental repetition met plant-assay requirements.

Treatment pollen was collected daily (0-5 days) from ca. 45-60 flowers that contained freshly dehiscing anthers, and placed into small storage vials (Jahns and Jolliff, 1990). Following pollen collection, the storage vial for each collection day was labeled and loosely capped to retain pollen and maintain gas exchange. Pollen (in vials)

was stored under one of three storage temperatures for five consecutive pollen collection days. On the sixth pollen collection day, the control (0 day) was obtained and the 0-5 day pollen-age treatments applied to all assay-plants per treatment replicate.

The six aged-pollen treatments were randomly assigned to six flowers per plant. Each treatment flower peduncle was labeled with the respective treatment coding. Pollen was vectored to the receptive treatment flower stigmas via a #000 camel's hair brush. Separate brushes were used for each treatment.

Plants were considered blocks, each receiving all aged-pollen treatments. Six plants (blocks) were used per experimental repetition. Aged-pollen studies were repeated twice under each of the three storage temperature regimes from 27 Nov. 1988 to 25 April 1989, for a total of twelve replicates per temperature study. The necessity to use the genotypically variable meadowfoam plants as blocks negated statistical comparisons between temperature treatments, because of the low flower production per plant.

Stigma Age and Seed Set

Hand pollinations were used in a greenhouse to evaluate whether an optimum stigmatal receptivity period exists in meadowfoam flowers. Stigma-age, measured at 24-hour intervals between 0-120 hours postanthesis, was bioassayed for seed setting ability under a 12-hour day/night temperature (% rh) cycle of: 23C(90)/18^{0}C(53% rh). As determined

from preliminary studies, the six stigma-age treatments represented an exaggerated stigmatal receptivity period range that was expected to encompass pre-, optimum, and poststigmatal receptivity periods.

Anthers were excised at anthesis from all flowers opening on each potential assay-plant to prevent accidental pollination of treatment flowers. A minimum of one new flower per plant per day was required to open for six consecutive days, before plants achieved assay status. Four out of fifteen plants per experimental repetition met the assay-plant criteria. The six treatment flowers per plant were labeled on peduncles with corresponding treatment codes prior to treatment application.

Each experimental repetition used eleven pollen-donor plants. Only stamens excised at anthesis, that possessed anthers containing freshly dehisced pollen, were used for treatment pollinations. Hand pollinations were applied to all six stigma-age treatments per assay-plant on 15 Nov., 28 Nov., and 23 Dec. 1987.

Plants were considered blocks, each receiving all six treatments per plant. Four plants (blocks) were used per experimental repetition. The entire experiment was repeated three times for a total of twelve replicates per treatment.

Viable stigma-age effects, as expressed by seed set, were compared using curvilinear regression analysis.

Stigma Pollination and Seed Set

Individual stigmas per flower were hand pollinated in a greenhouse to determine if a common style existed between stigmas and ovules, as measured by seed set. A stigma snipping (excising) technique was used to inhibit stigma pollination of non-treatment stigmas per flower. Assay-plants were grown in a greenhouse containing a 12-hour day/night temperature (% rh) regime of 27(80)/19^{0}C(50% rh).

Fifteen initial plants were selected per experimental repetition. All anthers were excised at anthesis from each flower per assay-plant to inhibit pollinations prior to treatment application. A minimum of four flowers per plant were required at optimum stigmatal receptivity on the day of treatment application. Four out of fifteen initial plants per repetition met assay-plant requirements.

One, 3, and 5 receptive stigmas per flower were pollinated with freshly dehisced pollen from excised stamens that had been removed from eleven pollen-donor plants per experimental repetition. A fourth treatment, the control, consisted of all five stigmatal papillae per flower being snipped, followed by the deposition of a minimum of 25 pollen grains to the five remaining stigmatal branch stubs.

Plants were considered blocks, each receiving four stigma-pollination treatments per plant. Four plants were used per experimental repetition. The entire experiment was repeated three times, from 28 April through 5 May 1989. A total of twelve replicates were used per treatment.

RESULTS AND DISCUSSION

Pollen Age and Seed Set

Pollen age, from 0-5 days postanthesis, had no significant ($P>0.39$) effect on seed set under the three storage temperatures used in this study (Table I.1). Coefficients of variation (CVs) ranged from 25-29% under the three temperature regimes, which compares favorably with CV levels encountered in previously reported field and greenhouse pollination studies (Jahns and Jolliff, 1990).

Control (0-day) values (Table I.1) demonstrated the importance of using meadowfoam plants as blocks for treatment comparisons. Further studies are required for temperature effect verification.

From these data, we conclude that pollen age does not appear to be limiting meadowfoam seed set. Further studies are needed to consider diurnal temperature, % rh, and light quality/intensity influences untested in this study.

Stigma Age and Seed Set

Stigma age had a highly significant ($P<0.001$) effect on seed set in meadowfoam. Stigmatal receptivity period treatments of 24, 48, and 72 hours postanthesis averaged 1.33, 3.33, and 0.87 seeds per flower (SE=0.3), respectively, under the greenhouse conditions used in this study. Pollinations of stigmas 0, 96, and 120 hours postanthesis produced no seed. Seed set comparisons ($FPLSD_{0.01} = 0.99$) between the five stigma-age treatments indicated that stig-

matal receptivity at 48 hours postanthesis produced significantly more seeds per flower than any other stigma age.

A highly significant ($P<0.001$) quadratic response was observed, as indicated from regression analysis, between stigma ages of 24, 48, and 72 hours postanthesis and seed set (Fig. I.1).

It appears from these data that stigma age is critical for maximizing seed set in Mermaid meadowfoam. Field observations (Franz and Jahns, unpubl. data) and preliminary growth chamber studies (Franz, 1990) have indicated that stigmatal receptivity may range from several hours to as long as five days postanthesis, depending upon environmental conditions. Field studies, to predict optimum stigmatal receptivity of peak bloom under variable environmental conditions, are needed for synchronization with adequate pollinator density for seed set maximization.

Stigma Pollination and Seed Set

Mermaid meadowfoam plants produced similar ($P=0.52$) seeds per flower, whether 1 (3.1), 3 (3.5), or 5 (3.6) stigmas per flower were hand pollinated (SE = 0.3). The control produced no seed from the twelve replicates used in this study, indicating that pollen must be applied directly to the papillae for seed set to take place. Experimental variation (CV=33.1%) was moderately high, though consistent with meadowfoam pollination studies.

Results from this study indicate that multiple stigma pollinations per flower are not critical for seed set in

meadowfoam, updating the findings of Mason (1952). While multiple stigma pollinations are not required, the importance of adequate pollen deposition onto receptive papillae surfaces (Jahns and Jolliff, 1990) must not be overlooked if seed set maximization is to be achieved.

Results from the three pollination studies reported in this paper indicate that stigmatal receptivity is of primary importance to meadowfoam seed set. Pollen age does not appear to influence seed set over a five day storage period. One receptive stigma pollinated per flower appears adequate for maximizing seed set when at least 25 pollen grains are deposited per stigma (Jahns and Jolliff, 1990), or in any combination between the five stigmas per flower. These studies should encourage further attention to optimum stigmatal receptivity periods which show potential for improving seed set via enhanced pollinator / flower synchrony at peak bloom.

Table I.1. Viable pollen longevity under three storage temperatures as measured by seed set per flower.

Control (18^0C) (day-0)	Pollen Storage Temp.	Pollen Age (days postanthesis) 1	2	3	4	5	SE[a]	CV[b]
		-- seeds per flower[c] --						
3.9	3^0C	3.4	3.5	3.4	3.7	3.2	0.3	25
3.3	18^0C	3.2	3.7	3.1	3.6	3.0	0.3	29
4.4	37^0C	4.4	4.2	4.0	3.6	4.1	0.3	26

[a]SE = standard error.
[b]CV = coefficient of variation.
[c]Seeds per flower values are the average from 12 replicates.

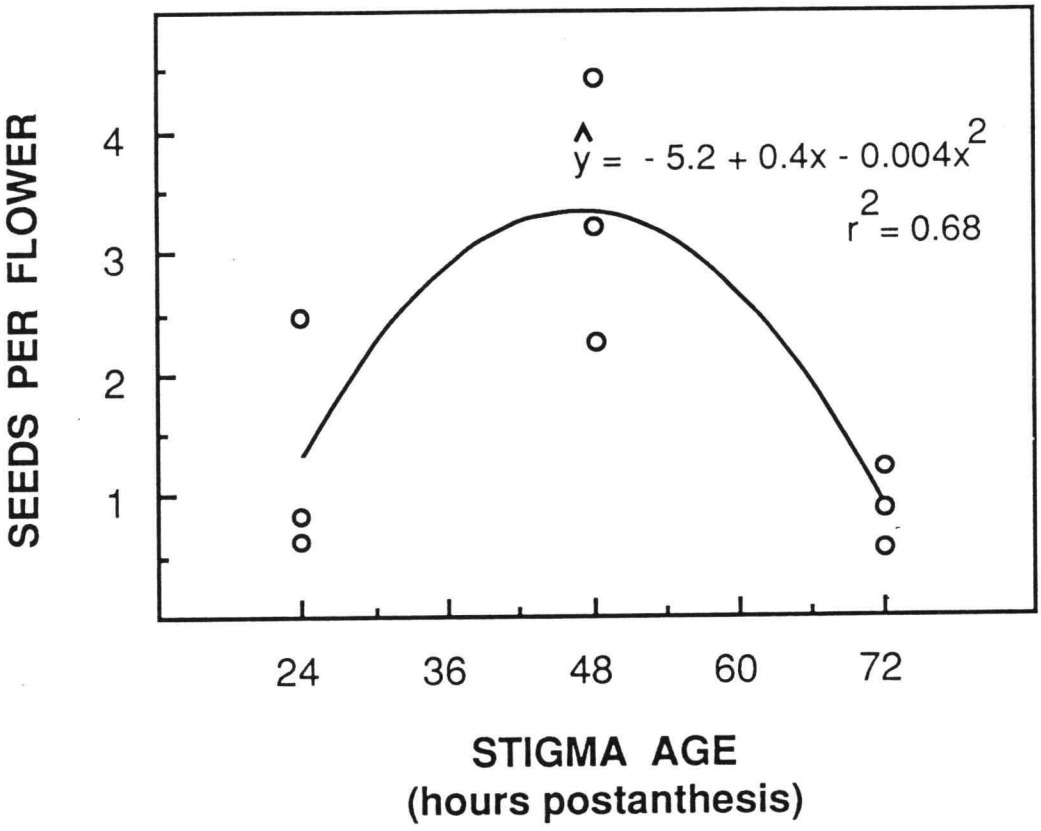

Figure I.1 Meadowfoam seeds per flower response to stigma age from three experimental replicates conducted in a greenhouse under a 12 hour day/night temperature (% rh) cycle of 23(90)/18^{0}C (53% rh) with a photon flux density average of ca. 145 µmol m^{-2} s^{-1}.

CHAPTER II

POLLEN DEPOSITION RATE EFFECTS ON SEED SET IN MEADOWFOAM

ABSTRACT

Meadowfoam (*Limnanthes alba* Hartweg ex Benth. cv. Mermaid) is an entomophilous, winter-annual oilseed crop which typically produces an average of only two out of five potential seeds per flower. This study was conducted to determine whether improved pollination will enhance seed set of this new crop. In the field, 1, 6, and 11 honey bee (*Apis mellifera* L.) visits per flower resulted in 1.6, 2.3, and 3.3 seeds per flower, respectively. In later pollen deposition studies, one and six honey bee visits per flower deposited an average of 15 and 43 pollen grains per five stigmatic papillae (FSP) per flower, respectively, in 1988, and 22 and 47 pollen grains per FSP, respectively, in 1989. Hand-applied pollen deposition treatments were studied in a greenhouse to investigate the cause of these flower visitation responses. Seeds per flower increased linearly (b= 0.086) in the range of 5 to 25 pollen grains deposited per receptive FSP. Seed set was 4.1 seeds (out of five potential seeds per flower) with 25 pollen grains per FSP. Although the greenhouse results cannot be directly extrapolated to the field, these data suggest that multiple honey bee visits to meadowfoam flowers are required for maximum pollination and seed set to occur.

INTRODUCTION

Meadowfoam is an entomophilous, winter-annual oilseed crop being domesticated in Oregon (Calhoun, 1975; Jolliff et al., 1981). Meadowfoam plants are herbaceous and multi-stemmed, with an indeterminate flowering habit. Meadowfoam flowers profusely. A 1982 study by Pearson and Jolliff (1986) revealed that in control plots, an average of 5.5 million flowers ha^{-1} were produced daily over a 16-day sampling period from Mermaid meadowfoam plants. Individual flowers contain ten anthers, five stigmas apically tipped with a papilla, and five ovaries. Flowers develop acropetally and are protandrous by 1 to 3 days (Mason, 1952). Although protandrous, flowers are highly self-compatible (Mason, 1952; Devine and Johnson, 1978) and receive many geitonogamous pollinations (Kalin, 1971) in native stands. Arroyo (1973) found that individual L. alba flowers contained copious pollen (467,000 to 933,000 pollen grains) and Kalin (1971) reported that the pollen was mostly removed on the day of anther dehiscence. Pollen viability does not appear to limit seed set, as Mason (1952) found over 97% of meadowfoam pollen to be viable at anthesis.

Honey bees are the primary pollinators of cultivated meadowfoam (Kalin, 1971). Honey bees forage on meadowfoam flowers for pollen and nectar and, when the FSP are receptive both nectar and pollen foragers are capable of successful pollination.

Less than optimum yields obtained in previous field studies have led to speculation that inadequate pollination may be limiting meadowfoam seed set (Gentry and Miller, 1965; Calhoun and Crane, 1978; Pearson and Jolliff, 1986). Despite abundant flowering, Mermaid meadowfoam rarely sets more than two out of five potential seeds per flower when grown in solid stands (Franz and Jolliff, 1989). Embryo abortion studies on seeds of Mermaid meadowfoam indicate that seed set may be reduced at least 20% by some form of temperature related embryo abortion (Franz and Jolliff, 1989). The difference between an average of two seeds per flower in the field and three to four seeds per flower in growth chamber (Alba, 1986) and greenhouse hand-pollination studies (T. Jahns, unpublished data) indicates that pollination may be limiting seed set in honey bee pollinated Mermaid meadowfoam flowers.

Results from pollinator visitation and pollen deposition studies over a variety of entomophilous plants suggest that the number of pollinator visits and the number of pollen grains deposited per receptive stigma could influence seed set in flowers of Mermaid meadowfoam. Bader and Anderson (1962) reported that seed set in birdsfoot trefoil (Lotus corniculatus L. cv. Viking) was positively correlated with pollinator visits. Shore and Barrett (1984) found that two to seven pollen grains are required to set a single seed in holley-rose (yellow alder) (Turnera ulmifolia L.), while $\geq$95 pollen grains were required to achieve

maximum seed set. Hand pollinations in trumpet creeper (Campsis radicans L.) showed that deposition of 200 to 800 pollen grains produced mature fruit on 20% of the pollinated flowers, while 100% of the pollinated flowers produced fruit when >800 pollen grains were deposited (Bertin, 1982). Individual flower pollination requirements for maximization of seed are not necessarily indicative of whole plant responses (Free, 1971), but they are an integral part of the pollination biology of a given crop. The effects of pollinator visitations and pollen deposition rates have not been reported in meadowfoam.

This study was undertaken to investigate the effects of hand-applied pollen deposition rates on seed set and to determine the number of honey bee visits needed for adequate pollen deposition to maximize seed set.

MATERIALS AND METHODS

Field experiments were conducted at the Oregon State University Hyslop Crop Science Field Laboratory near Corvallis, OR in 1986 and 1987 (seed-set study) and 1988 and 1989 (pollen-deposition study). All plants used in this study were grown from seed of Mermaid meadowfoam, established in the preceding year on an 18 cm row spacing planted at 22 kg ha^{-1}.

Honey Bee Visitations and Seed Set

Flower access was controlled to allow 1, 6, and 11 honey bee visits per flower from either pollen or nectar foragers. The three visitation rate treatments represented a daily low, medium, and high visitation spectrum, determined from preliminary studies; and the equal visitation rate spacing allowed statistical simplification via orthogonal polynomial contrasting. No other pollinators were allowed to visit a treatment flower, which was accomplished by physically tapping away the few non-honey bee pollinators present, using a flat wooden stick. Only a flower containing five receptive stigmatic papillae (i.e., the five stigmas spread, with each papilla bulbous) was used. Pretreatment honey bee visitations were allowed to remove dehiscing pollen from protandrous treatment flowers $\geq$1 day prior to stigmatal receptivity.

Wire meshed globes (common tea infusers) of 6.3-cm-diam. were used to prevent pollinator visitations. A 2.5-cm-diam. hole was cut in the bottom of each globe to allow

flower insertion, while restricting pollinator entry with the flower enclosed. Globes were glued to 0.9-m-long welding rods that were bent into 7-cm-diam. circles at one end for globe attachment. Welding rods were bent 90^0 to vertical, 10 cm below the attached globe so that the straight end of the welding rod could be pushed into the soil for support and the globe slipped over a flower without disturbing the flower position. Observation stations were placed within 1.5 m of all randomly selected treatment flowers, to expedite removal of globes, visual visitation counting, non-honey bee visitation protection, and reinsertion of flowers into the wire globes after the visitation-rate treatment. Treatment flowers were labeled with pressure-sensitive tape on each respective peduncle.

Visitation treatments were applied by removing flowers from within the globes, allowing the designated visitations to take place and returning the flowers to the globes. Following visitation exposure, flowers were retained in the globes 1 to 3 days, until permanently closed. Treated flowers were harvested at maturity and evaluated for seeds per flower. This pollination control method was derived from Walker (1943), who used cages made of cloth and wire to limit pollinator visits to flowers of watermelon [*Citrullus lanatus* (Thunb.) Matsum. & Nakai var. *lanatus*].

In both 1986 and 1987, we used the equivalent of eight colonies of honey bees per ha, a 60% increase over the recommended colony number (Karow et al., 1986). Treatment

flowers were randomly chosen throughout the 0.24-ha solid stand, using a completely randomized design with six replicates in 1986 and 24, 14, and 9 replications for 1, 6, and 11 visitations, respectively, in 1987. One observer was used in 1986 and two observers in 1987.

The time required to obtain 1 honey bee visit averaged 18 minutes, while 6 and 11 honey bee visits required an average of 2 and 5.5 hours, respectively. The disparity between the time required to complete visitation treatments, the observation capacity of the observers, and the need to maximize the number of replicates resulted in an unbalanced data set. Observations were made daily between 1100 and 1700 hours, at peak bloom in 1986 and one day post-peak in 1987.

The data from both years were combined into a 2 x 3 factorial arrangement, with treatment and interaction effects based on unweighted means. Preliminary analysis revealed that the variance was proportional to the mean, with zeros common for one visitation. Therefore, we added 0.5 to all response variables and performed a square-root transformation; we used 95% confidence limits in lieu of standard errors (Sokal and Rohlf, 1969). The analysis of variance (ANOVA) was performed on the 2 x 3 factorial and significance of the main factors and interactions determined.

Honey Bee Flower Visitations and Pollen Deposition Rates

Flower access was controlled to allow one and six honey bee visits per flower from either pollen or nectar foragers. Environmental conditions precluded an 11-visit treatment (see discussion, below). Otherwise, procedures were as described above for the seed-set study.

As in the seed-set study, the equivalent of eight honey bee colonies per ha^{-1} was used, and daily observations were made between 1100 and 1700 hours. Three sampling days were chosen per year, with day-one at or near peak bloom, followed by two post-peak-bloom treatment days. Sampling days were 21, 24, and 27 May 1988 and 14, 15, and 19 May 1989.

Immediately following visitation exposure, peduncles were cut 1 cm below the receptacle and flowers were placed into plastic collection boxes containing a 2 cm layer of 2% agar gel. Peduncles were inserted into the agar gel, up to the receptacle, to keep flowers isolated and stationary. Boxed flowers were stored at 3 to $5^{0}C$ for 24 to 48 hours prior to examination.

Floral stigmas were prepared for pollen counting by emasculating the flower, removing the calyx and corolla, and sticking the peduncle into a 1-cm-diam. plastic vial cap, 1-cm deep, filled with 2% agar gel. The agar-supported pistil was examined under 140X, using a stereozoom microscope with fiber-optic illumination, to record the number of pollen grains adhering to the stigmatic papillae.

Treatments were arranged in a 2 x 2 factorial, using a completely randomized design with six replicates. Years were compared by arranging treatments as a 2 x 2 x 2 factorial. Data were analyzed by ANOVA procedures and means compared using Fisher's protected LSD at $P<0.05$.

Pollen Deposition Rates and Seed Set

Greenhouse experiments were conducted at Oregon State University, Corvallis, OR in 1988 and 1989. Individual seed from Mermaid meadowfoam was sown into perlite-filled trays and placed in the dark at 10^0C for 7 days. On Day-7, trays were removed from the dark and placed into a growth chamber at 15^0C with an 8/16-hour light/dark cycle for vegetative growth. A photon flux density of ca. 140 µmol m^{-2} s^{-1} was maintained during the light cycle. At 21 days, seedlings were transplanted into 10 by 10 cm fiber pots containing a peat, sand, clay-loam mixture, and potted plants were returned to the growth chamber. At 45 days, plants were staked for support and moved into a greenhouse with a 16/8-hour light/dark cycle for photoinduction. Photon flux densities averaged ca. 145 µmol m^{-2} s^{-1}, under fluorescent lights, from Jan. through May 1989. Greenhouse temperatures for the 16/8-hour day/night regime averaged $21.3/13.8^0$C, respectively. Soil fertility was maintained using 12 weekly applications of 50 ml complete Hoagland's solution. At Day-90, 45 days post-photoinduction, plants began to flower, and pollen deposition treatments were administered.

Pollen deposition treatments consisted of 5, 15, 25, and ≥45 pollen grains deposited per FSP. We consider the hand-applied, ≥45 pollen deposition treatment as a standard for securing 100% pollination; this is supported by the findings of Devine and Johnson (1978) and has been successfully used in meadowfoam research for the last 12 years. Because a common style exists between stigmatic papillae and ovules (T.R. Jahns, 1989, unpublished data), deposition treatments did not require an even application of pollen grain numbers to each stigmatal papilla, but did require that each pollen grain be in direct contact with a papilla. To facilitate counting, a minimum of one pollen grain was applied per papilla.

Approximately 25 pollen-donor flowers, within 1 day postanthesis, were selected from 12 plants the day of treatment application. Plants receiving deposition treatments were excluded as pollen sources. Each pollen-donor flower was emasculated and the ten anthers tapped against the inside wall of a 1-cm-diam. plastic 5-ml vial cap. Repeated tappings released dehisced pollen into the cap. Following pollen collection the cap was covered with tape until deposition application.

Each plant receiving deposition treatments possessed a minimum of four receptive flowers the day of treatment application. Pre-treatment pollen contamination was kept to a minimum by emasculating potential treatment flowers at anthesis. Many more flowers per treatment plant were emas-

culated than used. Peduncles of treatment flowers were taped to plant-support stakes to reduce vibration during pollen application.

An eyelash glued to a small wooden dowel was used to vector the treatments of 5, 15, and 25 pollen grains to the five receptive stigmatic papillae. The eyelash was passed through the pollen stored in the vial cap, then touched to the stigmatic papillae. As for the field study, a stereo-zoom microscope was used at 140X to count pollen grains. Excess grains were removed from treatment stigmas, using a pollen-free eyelash or a #000 camelhair's brush. The $\geq$45 treatment consisted of touching a whole, freshly dehiscing anther (1 day post-anthesis) to the receptive stigmas of the $\geq$45 treatment flower. A single anther-donor flower was selected on the day of treatment and individual anthers removed as required to apply the $\geq$45 treatment to all plants within each experimental replication. Treatment flowers were labeled on peduncles with respective treatment numbers and allowed to mature in the greenhouse for ca. five weeks. At harvest, seeds were removed, sliced to verify viable embryo presence, and counted.

Plants were considered blocks, having all four deposition treatments applied simultaneously to each plant. Six plants were used per experimental replication. The entire experiment was repeated three times from 14 Jan. to 2 Apr. 1989, for a total of 18 replications per treatment. A randomized complete block design was used in a 3 x 4 factorial

arrangement. Data were analyzed by ANOVA and treatment effects compared using orthogonal polynomial contrasts at $P<0.05$.

RESULTS AND DISCUSSION

Honey Bee Flower Visitations and Seed Set

The number of honey bee visits to receptive flower stigmas had a significant (P=0.011) linear effect on seed set. When averaged across both years, flowers receiving 1, 6, and 11 honey bee visits set 1.6, 2.3, and 3.3 seeds per flower, respectively. This response is best described by the transformed $[(X + 0.5)^{1/2}]$ regression equation $\hat{y} = 1.270 + 0.05x$ ($r = 0.41$). Transformed means, with 95% confidence levels, for seed set from 1, 6, and 11 honey bee visits per flower were 1.39 ± 0.188, 1.63 ± 0.230, and 1.95 ± 0.266, respectively.

In general, a large variance is common in pollination studies, and this study was no exception, as demonstrated by the CV of 33%. The use of a larger sample size may help buffer the added variability encountered between plant and pollinator (Free 1970, p.6,7).

Honey Bee Flower Visitations and Pollen Deposition Rates

The number of honey bee visits had a highly significant ($P<0.01$) effect on the number of pollen grains deposited per stigma. From an average of 18 observations, in 1988, one honey bee visit deposited 15.2 pollen grains per FSP, 32% less than the 22.4 pollen grains per FSP in 1989. Six visits in 1988 resulted in 43.3 pollen grains per FSP, or 7.5% less than the 46.8 grains in 1989. Variation in deposition rates was greater in 1988 (SE=2.9) than in 1989 (SE= 1.3), and the CV was larger in 1988 (42.4 vs. 15.9).

In 1988, winds in excess of 9 m s^{-1} were prevalent from peak to late bloom, hindering honey bee pollination activity (Park, 1923). Originally, honey bee visitation treatments were to consist of 1, 6, and 11 visits per flower, but no more than 7 to 8 visits were observed per six hour sampling day from 21 to 27 May 1988. In 1989, warm, dry, and calm weather conditions appeared favorable for honey bee pollination, as an average of only 104 minutes were required to obtain the six honey bee visitation treatment. These results indicate that the environment plays a role in pollen deposition as well as honey bee visitations. The year effect was significant ($P<0.05$), although the trend was similar, and the year x treatment interaction was not significant.

Pollen Deposition Rates On Seed Set

The linear effect of pollen grain numbers per stigma on seed set was highly significant ($P<0.01$)(Table II.1). Five pollen grains per FSP set an average of 2.4 seeds per flower, while 25 pollen grains set an average of 4.1 seeds per flower. The $\geq$45 pollen grain deposition treatment produced identical seed set to the 25 pollen-grain treatment, indicating that 25 pollen grains are adequate to maximize seed set under the controlled conditions used in this experiment.

The linear relationship between pollen deposition number and seed set is best described by the regression equation $\hat{y} = 2.04 + 0.086x$ ($r = 0.51$), i.e., seed set increased

by ca. 0.09 seed for each additional pollen grain added, across the range used in this study.

Considerable variation was present in the experiment, as indicated by the CV of 31%. Plants and hand-pollination techniques used in this study were considered optimum for pollination and subsequent seed set. This excellent seed set from the 25 and ≥45 hand-pollination treatments (4.1 out of a possible 5 seeds per flower) indicates that pollination is limiting seed set in field grown plants of Mermaid meadowfoam.

Except for poor honey bee foraging conditions that may limit rates of pollen deposition, limitations to pollination and seed set in the field have not yet been identified. They may be linked to factors limiting seed yield, such as water stress (Pearson and Jolliff, 1986), plant variability (Alba, 1986), seed-fill limitations (Krebs and Jain, 1985), and embryo abortion (Franz and Jolliff, 1989). Even though other factors may affect seed yield, adequate pollination is essential to achieve maximum seed set per flower, especially under field conditions where pollination is widely variable.

Increased honey bee visitations per flower enhanced both pollen deposition rates and seed set. These data illustrate the importance of having an adequate number of pollinators available to maximize pollen deposition under less than optimum foraging conditions. Twenty-five pollen grains per stigma appeared adequate for maximum seed set

under greenhouse conditions. The field studies conducted in 1986 through 1989 indicate that 25 pollen grains per FSP may be inadequate to maximize seed set in field grown plants, but further studies are required for verification. Although pollination requirements of individual flowers for maximum seed set are not necessarily indicative of whole plant responses (Free, 1971), these data illustrate the importance of pollen deposition rates both from hand and honey bee pollinations on seed set in flowers of Mermaid meadowfoam.

Table II.1. Number of pollen grains deposited per five stigmatic papillae (FSP) and mean number of seeds per flower observed and expected in plants of Mermaid meadowfoam.

Number of pollen grains per FSP	Seeds per flower[a]	
	Observed	Expected[b]
5	2.4	2.5
15	3.5	3.3
25	4.1	4.2
≥45 (standard)	4.1	
SE	0.3	
CV	32.2	

[a]Seeds per flower = average of 18 observations.
[b]Expected = fitted linear equation $\hat{y} = 2.04 + 0.086x$ ($r^2 = 0.26$).

CHAPTER III

POLLINATION AND SEED SET IN MEADOWFOAM

INTRODUCTION

Meadowfoam (Limnanthes alba Benth., Limnanthaceae) is an oilseed crop that requires insect (primarily honey bee) pollination to set seed. Effective honey bee management will increase meadowfoam yields, improving the economic competitiveness of this new resource for Oregon. The purpose of this extension circular is to review the flowering characteristics and seed set requirements of meadowfoam; and to offer suggestions for increasing honey bee management effectiveness for improving meadowfoam pollination and subsequent yield.

The first recorded farm-scale planting of meadowfoam (cultivar Foamore) in Oregon took place in 1975-1976. A second cultivar, Mermaid was released exclusively to the Oregon Meadowfoam Growers Association in 1985. Both Mermaid and Foamore are open-pollinated cultivars that require bee pollination to set seed. Mermaid meadowfoam seed yields have ranged from 702 to 1567 lb/A in research plots while commercial yields have averaged 770 lb/A. Greenhouse pollination studies using hand pollination on selected Mermaid meadowfoam plants have repeatedly produced greater than 4 seeds per flower, while honey bee pollinated field populations have rarely averaged greater than 2.5 seeds per flower. The seed set disparity observed between greenhouse and field pollination, as well as yearly yield fluctuations in field research plots, have aroused speculation that inadequate pollination may be limiting seed set. Effective

bee management is essential to maximize meadowfoam pollination and subsequent seed set.

FLOWERING CHARACTERISTICS

Meadowfoam is a short, fleshy plant that produces 1-12 flowers per stem and 1-10 stems per plant under solid stand field production. The flowers on each stem develop in a sequential order from bottom to top. Each flower is attached to a stem via a peduncle. Peduncles elongate prior to flower opening. Each flower has ten stamens, each stamen containing a pollen-producing anther, the male reproductive structure (Fig. III.1). The ten stamens surround the female reproductive structure (pistil). The pistil is made up of 5 stigmas (female receptors of pollen), a common style, and five ovaries, each containing one ovule (Fig. III.1). The five ovaries are located at the base of the style. Each ovary has the potential to produce one seed, technically known as a nutlet. Prior to initial flower opening, five petals and five sepals cover the stamen and pistil. Once flower opening commences, flower sepals and petals open during the day, exposing the stamens and pistil to potential bee pollinators, and close at night. Bee visitations, apparently for nectar, are required to initiate the nightly petal and sepal closing mechanism. Flowers on caged plants lacking pollinator exposure do not close at night. Flowers continue to open and close until successful pollination and subsequent fertilization permanently close the petals and sepals around the reproductive structures.

Meadowfoam pollen is sufficiently heavy and sticky to inhibit wind pollination, thus requiring insects, primarily

honey bees, for pollen transport. Within each flower, pollen is shed before the stigmas are receptive (Fig. III. 2A &B). This non-synchronous development of male pollen before the female stigma becomes receptive is called protandry. Because of protandry, pollen for seed set usually comes from another flower, either on the same or a different plant. Pollen produced within any given flower is capable of setting seed within the same flower (self-pollination); however, in the field, self pollination is not likely because: 1) honey bees remove the majority of the flower pollen prior to stigmatal receptivity; and 2) the remaining pollen adheres to stamens that are physically located below the receptive stigmas (Fig. III.2C).

POLLINATION

The pollination period for meadowfoam ranges from 2 to 4 weeks during May and early June. Individual flowers open for 1 to 4 days during bloom, depending on the temperature. Pollination occurs when pollen is inadvertently transferred from the anthers of one or more flowers, via honey bees, onto the stigma or stigmas of a receptive flower. Pollen germination on a stigma is followed by pollen tube growth down the style into the ovule where the union of sperm and egg cell results in embryo fertilization and subsequent seed production.

When pollen is abundant, either honey bee pollen foragers or honey bee nectar foragers can successfully accomplish pollination. Pollen collecting honey bees are most

prevalent from approximately 11 am to 2 pm, when the majority of available pollen is collected. Honey bees are found foraging primarily for nectar between 2 and 7 pm. The daily foraging periods of the pollen and nectar collecting honey bees overlap from late morning to early afternoon.

FACTORS AFFECTING POLLINATION AND SEED SET

Plant

Genetic Variability. Individual meadowfoam plants in greenhouse and field experiments have averaged from 0 to 5 seeds per flower under adequate pollination. Meadowfoam plants are genetically variable, resulting in a range of individual plant yield responses within a solid stand.

Flower Position and Timing. Flower production starts at the bottom and proceeds to the top of each stem over time. Flowers located on the bottom two-thirds of the stems produce more seeds per flower than flowers located on the top one-third of the stems.

Embryo Abortion. Embryo abortion has been observed in individual Mermaid plants. The risk of reduced seed development, by as much as one seed per flower over an entire plant, increases with temperatures above 80^{0}F.

Stigmatal Receptivity. The stigmas of individual meadowfoam flowers become receptive to deposited pollen at specific times. Pollen deposition before (Fig. III.2A&B) or after prime stigmatal receptivity is less likely to maximize seeds per flower than at optimum stigmatal receptivity (Fig. III.2C). The greater the number of pollinator

visitations per flower during the stigmatal receptivity period, the greater the assurance that pollen will be deposited at or near peak stigmatal receptivity, maximizing seed set.

Pollen Deposition. The deposition of approximately 25 viable pollen grains to any or all of the five stigmas per flower is required to maximize seed set in Mermaid flowers grown in the greenhouse. In a two year field study it was found that one honey bee visitation per flower deposited between 15 and 22 pollen grains per stigma, while six honey bee visitations deposited between 43 and 47 pollen grains per stigma. While the rate of pollen deposition required to maximize seed set in the field has not been determined, multiple honey bee visits per flower increase the likelihood of adequate pollen deposition for maximum seed set.

Honey bee

Nutritional Requirements. Honey bees require both nectar and pollen for maintenance and expansion of individual colonies. The low nectar availability of Mermaid flowers inhibits honey production, limiting colony maintenance and growth. Pollen for colony growth does not appear limiting in meadowfoam.

Canopy Penetration. Meadowfoam plants in solid stand grow together, entwining the stems and flowers of adjacent plants. Production practices such as seeding rate, row spacing, and nitrogen fertilization influence the distribution of flower opening over time and flower location in the

canopy. Secondary meadowfoam stem and flower growth may produce a number of flowers below the canopy that have the potential to set seed and increase yield. The majority of foraging bees do not actively seek out flowers in the depths of the canopy because their movement is restricted by the entwined plants. By increasing the number of foraging honey bees, pollination and subsequent seed set of these secondary flowers should increase.

Weather

Temperature. Plant pollination requirements and honey bee activity are strongly affected by temperature. Temperatures below 55^0F will hinder flower opening as well as honey bee flight. On warm days following inhibited flowering, millions of flowers may open per acre, resulting in an enormous pollination requirement. As temperatures rise, there is a tremendous demand for pollen deposition onto the receptive stigma(s) of individual flowers. The prime stigmatal receptivity period may be as long as 24 hours under temperatures below 70^0F and as short as one hour with temperatures above 90^0F.

Humidity. High humidity, like low temperatures, may inhibit flower opening creating increased pollination demand when a "backlog" of flowers open.

Wind. At wind speeds greater than 15 to 20 mph, honey bee flight and subsequent meadowfoam pollination are restricted to areas adjacent to hives.

Honey bee management

Colony Strength. One strong colony will pollinate more flowers than 2 or 3 weak colonies. Section 55-005 (Chapter 603) of the Oregon Administrative Rules sets the "minimum" standard for pollination colony strength at 25,000 adult honey bees per colony, resulting in approximately 12,000 total foragers. Larger colonies are preferred, but may not be available. Colony strength is often difficult to assess, so the use of reputable beekeepers is highly recommended (The Pacific Northwest Cooperative Extension Bulletin #245 is an available guide for evaluating honey bee colonies for pollination).

Honey Reserves. Without adequate honey reserves stored in the hive, pollination deficiencies and colony loss may result. At least 30% of the available frames per hive should contain honey reserves for colony maintenance and pollination strength.

Colony Spacing. Proper hive placement has been shown to increase yields in some crops. During periods unfavorable to foraging, bees tend to work areas close to the hive. On meadowfoam acreages greater than ten acres, a minimum colony spacing of 30 hives every ten acres should increase pollination and subsequent yields, although no data are available to verify this.

POLLINATION RECOMMENDATIONS

Colony number

Three hives per acre are recommended to insure adequate pollination. During long bloom periods with prevalent cool, wet conditions, more than three hives per acre may be required to fulfill peak pollination demands.

Timing

Hives should be moved into meadowfoam fields when 5-10% of the flowers are in bloom. Introduction at 10% bloom will help to discourage colonies from initially foraging on competing plants, but if honey bee hive introduction is delayed past 10% bloom, severe yield reductions may result. In 1987, caged field studies revealed that two days after 10% Mermaid meadowfoam bloom was established, peak bloom occurred. A one week delay in honey bee colony introduction would have left 83% of the available flowers opened from one to seven days without pollination. As a result of the female stigma aging process, 50% of the previously unpollinated flowers may not have been able to set seed, dramatically reducing seed yield.

Removal of honey bee hives from the field can begin when less than 5% of the bloom remains.

Competing bloom

Meadowfoam should not be planted within two miles of commercial acreages of competing crops such as crimson clover or a late blooming rapeseed variety. Superior nectar

resources are present in these crops, attracting honey bees away from meadowfoam, drastically reducing pollination.

Weeds commonly in bloom during meadowfoam flowering include the mustards and wild radish. These plants, if present in large numbers, can directly compete for colony pollination visitations. Cultural or chemical control of these weeds will increase meadowfoam pollination.

Studies in 1981 and 1982 showed that meadowfoam pollen preference within any given honey bee hive dropped off after 2-5 days. Meadowfoam pollen collection per colony was increased by replacing existing hives with fresh hives every five days. This would be expected to increase meadowfoam pollination and seed set, although it has not been tested.

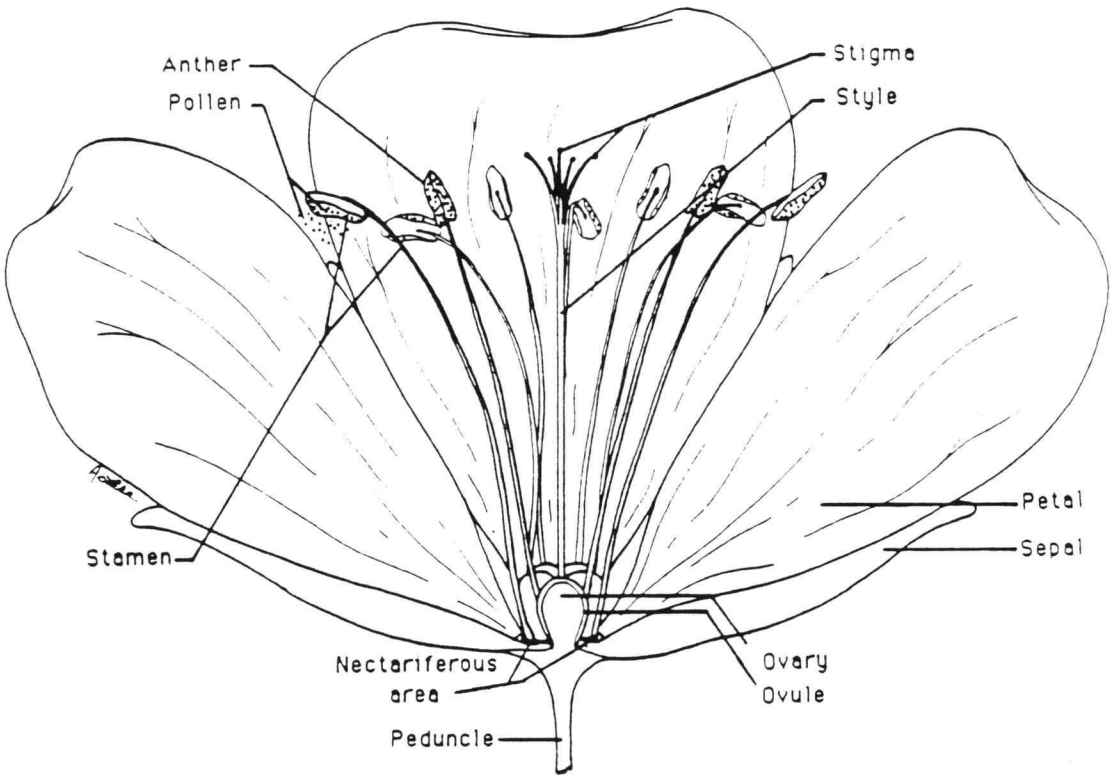

Figure III.1 Longitudinal section of a Mermaid meadowfoam flower with one stamen and two petals removed, X 7.

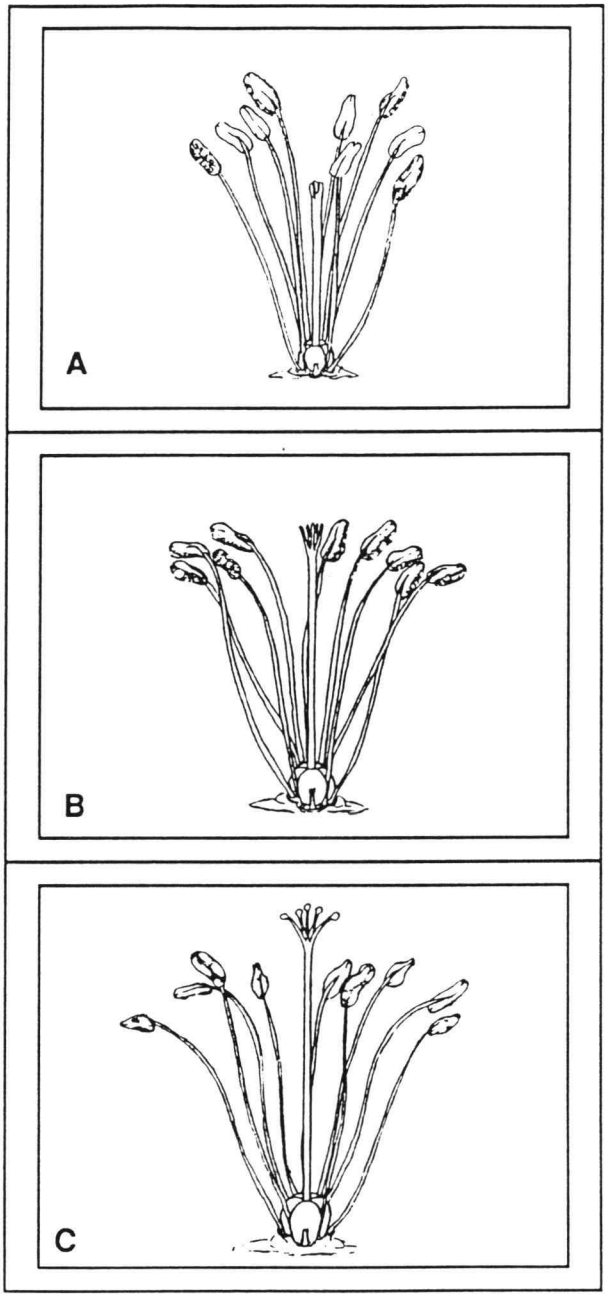

Figure III.2 Sequential reproductive development of a Mermaid meadowfoam flower from: (A) flower opening with initial pollen availability (dehiscence) and unreceptive stigmas, (B) maximum pollen shed with unreceptive stigmas, and (C) maximum stigmatal receptivity with reduced pollen availability.

CHAPTER IV

OSMIA LIGNARIA PROPINQUA CRESSON: AN ALTERNATIVE POLLINATOR FOR MEADOWFOAM IN CAGES

ABSTRACT

Meadowfoam (Limnanthes alba Hartw. ex Benth. cv. Mermaid) is an entomophilous winter annual oilseed crop with an unstable seed yield history. A two-year study was undertaken to test the efficacy of Osmia lignaria propinqua Cresson bees as meadowfoam pollinators, based upon the following criteria: (i) whole-plant seeds per flower yield; (ii) solid stand yield, seed weight, and seed-oil production; and (iii) seed yield per bee. Three O. l. propinqua density treatments (10, 35, and 60 adult females) were compared to a 4000 honey bee (Apis mellifera L.) density treatment standard, and a 4000 honey bee + 35 O. l. propinqua bee density treatment. Cages were used to isolate treatments. A non-caged honey bee treatment, equivalent to 8 honey bee colonies ha^{-1}, served as the control. No significant differences were found in whole plant seeds per flower produced between any of the caged-pollinator treatments in either year, although there were significantly more seeds per flower in 1986 (2.9) than in 1987 (2.2). No significant solid stand yield differences were found between the three O. l propinqua density treatments and the two caged honey bee treatments in 1986. Sixty female O. l. propinqua bees per cage produced similar solid stand yields to all honey bee treatments (inside or outside cages) in 1987. A positive linear relationship (b= 4.5x) between O. l. propinqua density and yield suggests that greater than 60 female O. l. propinqua bees per cage may be required to

maximize solid stand yields. There were no significant differences found in seed weights between any of the pollinator treatments tested in 1986. In 1987, the 10 and 35 O. l. propinqua density treatments produced significantly higher seed weights than all honey bee treatments, indicating, via yield component compensation, potential pollination inadequacies. No significant differences in seed-oil content existed between any of the pollinator treatments in either 1986 or 1987. All O. l. propinqua treatments produced significantly greater yields per bee than either of the caged honey bee treatments in 1987. A similar trend existed in 1986, although only the 10 O. l. propinqua density treatment produced significantly higher yields per bee. While regression analysis indicated that yield per bee declined with increasing O. l. propinqua density, average yield per bee was at least 1.7x greater than any of the caged honey bee treatments. Meadowfoam seed yield produced from 60 O. l. propinqua bees per cage was as high as the best caged honey bee treatment in 1986 or 1987. Although further studies are required to determine optimum O. l. propinqua density levels for yield maximization, O. l. propinqua showed potential as an alternative pollinator of meadowfoam.

INTRODUCTION

Meadowfoam is an entomophilous, winter annual oilseed crop being developed at Oregon State University, Corvallis (Calhoun, 1975; Jolliff, 1989). Historically (1977 to 1989), meadowfoam yields have fluctuated from 788 to 1760 kg ha^{-1} in replicated yield trials (Jolliff, unpublished data). Commercially grown meadowfoam (1983 to 1987) has produced considerably lower yields, ranging from 570 to 989 kg ha^{-1} (M. Ringsdorf, President, Meadowfoam Growers Association, 28781 Bodenhamer Rd., Eugene, OR-unpublished data). It has been speculated that inadequate pollination is contributing to both the annual yield fluctuations (Calhoun and Crane, 1978; Franz and Jolliff, 1989) and the yield disparity between research and commercial meadowfoam production (Ringsdorf-pers. comm.). If honey bee pollination is limiting seed set, then an alternative meadowfoam pollinator, that could consistently produce higher yields, would improve the economic competitiveness of this new resource.

The honey bee is the primary pollinator of cultivated meadowfoam in Oregon (Karow et al., 1986). During the May bloom period, cool ($<16^{0}C$), wet, and windy (>24 km hr^{-1}) weather conditions may occur, limiting honey bee foraging activity (Lundie, 1925). These weather conditions may also inhibit flower opening (Kalin, 1971), delay anthesis (Pearson and Jolliff, 1986), and intensify the need for timely pollination to maximize seed set.

The problems associated with honey bee pollination of meadowfoam flowers are similar to those reported by Torchio (1976) in early-season orchard crops. Inclement weather is prevalent at this time of year, bloom is of short duration, and the amount of bloom is enormous. We selected O. l. propinqua (the orchard mason bee-Klostermeyer, 1979; the blue orchard bee-Yarris, 1983) as a potential meadowfoam pollinator because this bee: (i) has out-performed the honey bee in pollination efficacy of selected, early-season crops (Torchio, 1979, 1985; Kuhn and Ambrose, 1984); (ii) has early season flight activity (Torchio, 1976) which can be synchronized with meadowfoam bloom; and (iii) has populations that can be managed in open and caged environments (Torchio, 1976, 1979, 1985).

Successful evaluation of pollinator yield potential is complex (Free, 1971), and the importance of pollinator foraging density has too often been overlooked (Kehr and LaBerge, 1966; Brewer, 1974; Koelling et al., 1981) when assessing pollinator performance. While pollinator efficacy offers a baseline for pollinator assessment, the use of pollinator foraging density can also provide yield per bee evaluations which have proven invaluable for assessing pollinator benefits (Waller et al., 1985; Berger et al., 1988).

This study evaluates O. l. propinqua pollination efficacy in caged meadowfoam plots on: (i) whole-plant seeds per flower yield; (ii) solid stand, seed weight, and seed-

oil yields; and (iii) yield per bee based on results of caged O. l. propinqua compared to results of caged or non-caged honey bee controls.

MATERIALS AND METHODS

Plot Preparation

Caged field experiments were conducted in 1986 and 1987 at the Oregon State University Hyslop Crop Science Field Laboratory near Corvallis, Oregon. A 0.24 ha field was planted with Mermaid meadowfoam seed at a 22 kg ha^{-1} seeding rate on an 18-cm row spacing on 3 Oct. 1985 and 9 Oct. 1986. Plot perimeter dimensions were 3.7 by 3.7 m. Prior to bloom each plot was hand-trimmed, leaving a 3.7 by 2.4 m (8.9 m^2) solid stand down the center (north to south) of each plot. In the remaining 0.65 by 3.7 m spaces on each side of the solid stand, all but four plants per side were removed. These four plants were isolated 0.74-m apart (north to south) and staked for support. At ca. 1% bloom all plots received an insecticide application of 'Mavrik' ([(RS)-alpha-cyano-3-phenoxybenzyl(R)-2-[2-chloro-4-(trifluoro-methyl)anilino]-3-methylbutanoate), at 0.1 kg ha^{-1} (a.i.), on the morning of plot-caging to reduce incidental pollination by non-treatment insects (Free, 1970). The plots were covered with natural-colored polyvinylidene chloride (Saran) cages, of 20 by 20 mesh, measuring 3.7 by 3.7 by 1.8 m. In 1987 the center section of each cage was supported with a 5-cm diam. polyvinyl chloride (PVC) tube ca. 3-m long, to prevent precipitation from pooling on cage tops.

Pollinator Preparation

Honey bees

Treatment one consisted of honey bee nuclei colonies manipulated to obtain a density level of ca. 4000 adult bees per pollination unit. Colony populations were measured by estimating the number of adult honey bees occupying combs within the colonies (Burgett and Burikam, 1985). Colonies were assembled between 0530 hours and 0630 hours on the morning of introduction into cages. Colonies were introduced into cages at 10% meadowfoam bloom (17 May 1986 and 5 May 1987). One frame of capped honey in 1986 and at least two full frames in 1987 were inserted into each colony prior to cage introduction. In 1986 supplemental sugar syrup was provided to the colonies throughout bloom. In 1987 no sugar syrup was furnished. Water was supplied throughout bloom. Colonies were removed and densities estimated (postpollination) 2 June, 1986 and 21 May 1987.

Treatment two was the uncaged "standard" treatment, which utilized the equivalent of eight honey bee colonies ha^{-1}, a 60% increase over the colony number recommended (Karow et al., 1986) for commercial meadowfoam pollination.

Osmia lignaria propinqua

Treatments three, four, and five consisted of 10, 35, and 60 adult female O. l. propinqua bees per caged plot, respectively. A corresponding number of adult males per plot were introduced at the same time for mating purposes. Treatment density selections were slightly different, but

patterned after O. l. propinqua bee levels used by Torchio (1979) for caged almond tree pollination, which consisted of 10, 20, 30, and 40 introduced O. l. propinqua females per caged tree.

In 1985, a population of O. l. propinqua was obtained from the USDA-ARS Bee Biology Lab in Logan, Utah for release in 1986. In 1986, 30% of the O. l. propinqua bee population for release in 1987 was obtained from USDA-ARS, 60% from Dr. Ray Lynn (Star Rt., Mendon, UT), and 10% from 1986 offspring produced in caged meadowfoam plots. Adults were shipped and over-wintered in 7-mm (inside diam.) straws ca. 15-cm long. All straws were X-rayed upon receipt to identify and remove parasites and parasitoids (Stephen and Undurraga, 1976). Straws were stored at 3-5^{0}C from the time they were acquired (9 Oct 1985 and 3-8 Oct 1986) until the time of emergence-incubation (13-15 May 1986 and 2-4 May 1987).

Two days prior to emergence-incubation, O. l. propinqua cocoons were removed from their straws and individually placed into #000 gelatin capsules. Capsules (containing cocoons) were placed in 13 to 30^{0}C fluctuating incubators set at 12-hour intervals. Emerged adults (within capsules) were sorted by sex and stored at 3-5^{0}C. These incubation procedures were repeated for three days, until surplus numbers of bees emerged.

O. l. propinqua bees were grouped by sex into densities of 10, 35, and 60 males or females. Each sexed den-

sity group was then placed into a cardboard release box measuring ca. 30 by 15 by 10 cm. A small flap cut into the top of each release box facilitated O. l. propinqua release into cages.

One wood nesting unit was placed (facing SE) in each cage assigned to the O. l. propinqua 10 and 35 bee density treatments, and two nesting units were placed (facing SE and SW) in the 60 bee density treatment cages. All nesting units were supported on two stakes, 1 m above soil level. Each nesting unit measured 17.8 by 15.2 by 13.3 cm and contained 54 paper straws. Each straw was 15 cm long by 7 mm inside diam.

O. l. propinqua males and females were released into cages 16 May 1986 and 5 May 1987. Nesting units were inspected every other night throughout the flowering period with the aid of a small pen-light directed into nesting holes to facilitate identification of bees by sex. Additional females were added to the cages 23 and 24 May 1986 and 11 and 19 May 1987 to re-establish original female densities released. Mud and water were made available in tubs placed in each treatment cage.

Honey bees + Osmia lignaria propinqua bees

A sixth treatment, consisting of 4000 honey bees and 35 O. l. propinqua female bees, was included for comparison. Thirty-five male O. l. propinqua bees were also introduced for mating purposes. Preparation of the two bee

species used in this treatment was the same as described above.

Bloom Phenology and Environmental Monitoring

Bloom phenology

Bloom phenology was monitored daily between 1600 and 1700 hours, from the solid stand area of three caged plots (without pollinators), by removing and counting all opened flowers within a 0.1 m^2 frame placed randomly within each plot. The caged-bloom periods were from 12 May to 1 June 1986 and 2 to 21 May 1987. The bloom phenology sampling period started on the day of pollinator introduction (10% bloom) and ended on the day of pollinator removal (>99% bloom), from 16 May to 1 June 1986 and 5 to 21 May 1987.

Environmental monitoring

Hourly temperatures were recorded inside and outside of the caged meadowfoam plots using thermographs placed under white vented wood covers.

Daily photon flux density was measured parallel to the top of the meadowfoam flower canopy inside and outside of the caged plots at 15 to 30 minute intervals using a line quantum sensor (Model 191S, Line Quantum Sensor, LI-COR, Inc., Lincoln, NE 68304).

Daily precipitation was recorded 300 m away from the treatment plots at the Hyslop Crop Science Field Laboratory Weather Station.

Pollinator-Treatment Yield Efficacy

The six pollinator treatments were assigned to plots and analyzed as a completely randomized design (CRD), with three replicates per treatment, unless otherwise stated. All data were analyzed using analysis of variance (ANOVA) procedures and treatment mean comparisons made using Fisher's Protected Least Significant Differences (FPLSD) at $P< 0.05$ or $P<0.01$. Data are reported as individual year responses unless otherwise noted.

Individual plants

Following pollinator exposure, four of eight isolated plants (subsamples) per plot were harvested for seed set evaluation on 20 June 1986 and 12 June 1987. Harvesting was accomplished by cutting plants at soil level and placing each in a paper bag. Bagged plants were stored at room temperature for ca. 3 weeks to facilitate seed fill prior to flower position and whole-plant seeds per flower evaluations.

Pollinator treatment efficacy on whole-plant seeds per flower yield was measured by averaging seeds per flower yield from all flowers per sampled plant within each respective pollinator treatment.

The effect of flower position on seeds per flower yield was evaluated across all six pollinator treatments. Flower position was determined by dividing the total number of flowers per stem into thirds (bottom, middle, and top). Seeds per flower response to flower position was then mea-

sured by averaging the number of seeds produced per flower within the bottom, middle, and top positions per stem, followed by the averaging of each position response across all stems per plant.

Pollinator and flower position treatments were arranged in a nested classification (Snedecor and Cochran, 1980) using a CRD with three replicates and four subsamples per replicate. Data were analyzed with SAS (SAS Institute, Inc., 1985).

Solid stand

Following pollinator treatment exposure and plant maturation, a 3.7 by 0.9 m swath (3.3 m^2) was harvested from the solid stand of each plot using a flail-chopper (Carter Manufacturing, Brookston, IN 47923) on 25 June 1986 and 15 June 1987. Plant material from each plot was placed in a burlap bag and allowed to air-dry. Materials were then threshed, seed was cleaned, and seed yield (kg ha^{-1}), seed-weight (mg), and seed-oil content (Comstock and Culbertson, 1958) evaluated per treatment. Simple linear regression procedures were used to assess O.l. propinqua density responses to seed yield (kg ha^{-1}) and seed weight (mg) where applicable. Yields from caged plots devoid of pollinators were included as a non-pollinator check (Free, 1970), but these data were withheld from pollinator treatment analysis as a method to increase treatment mean separation sensitivity.

Foraging-Bee Density

Foraging-bee densities per treatment were monitored at 0.5 hour intervals, from 1100 to 1700 hours daily, throughout the 1986 and 1987 bloom periods. Four bamboo stakes (ca. 1 cm diam. by 1 m in length) were pushed into the soil to demarcate a 0.25 m^2 observation area, which was randomly placed in the solid stand area of each plot. The 13 daily instantaneous bee counts, taken from each 0.25 m^2 area per plot, were averaged over the entire pollination period to estimate the number of pollinators actively foraging per treatment (Burgett-pers. comm.). Treatments were arranged in a CRD, with three replicates per treatment. Foraging-bee densities per treatment were analyzed using ANOVA procedures and treatment means compared using FPLSD at $P<0.01$.

Individual Bee Yield Efficacy

A second order polynomial regression analysis was used to evaluate the relationship between yield (kg ha^{-1}) and bees m^{-2} per plot, over the five caged-pollinator treatments. Yield per bee was determined over the same five treatments, using the average yield per plot divided by the average number of foraging bees per plot (8.9 m^2). A second order polynomial regression analysis was then used to evaluate the relationship between the number of female *O. l. propinqua* bees maintained per treatment and yield per bee within each treatment.

RESULTS AND DISCUSSION

Bloom Phenology and Environmental Monitoring

Bloom phenology

The bloom phenology sampling periods were from ca. 10% initial bloom to >99% bloom. In 1986 and 1987 a daily average of 5477 and 4736 flowers were produced per cage (8.9 m^2), respectively (Table IV.1).

Peak bloom in 1986 took place on the tenth foraging day, while peak bloom in 1987 took place on foraging day three (Fig. IV.1).

Environmental monitoring

Hourly temperature values were found to be identical inside and outside of the cages with daily temperatures during peak pollinator activity (1000-1900 hours) averaging 21.8^{0}C and 21.5^{0}C in 1986 and 1987, respectively (Fig. IV. 1). In general, average daily temperatures progressed from cool to hot in 1986, while in 1987 a hot to cool temperature trend occurred during the potential foraging period.

Regression analysis between average number of daily flowers produced per cage (Table IV.1) and daily average temperatures observed per foraging day (Fig. IV.1) indicated no significant relationship ($P=0.347$; d.f.=1,13; $F=0.95$) in 1986, except between peak bloom and peak temperature. In 1987, a highly significant ($P<0.01$; d.f.=1,15; $F=80.76$) linear relationship existed ($r^2=0.84$; $\hat{y}=-14010 + 873.2x$, where $\hat{y}$ = average number of flowers per cage and x = temperature ^{0}C) between bloom phenology and temperature.

There was a 31% ($P<0.001$; inside/outside correlation $r^2=0.96$; $n=45$) and 38% ($P<0.001$; inside/outside correlation $r^2=0.93$; $n=135$) average reduction in photon flux density inside (versus outside) cages in 1986 and 1987, respectively. These differences may be attributed in part to a darkening of the cage materials, both from dirt accumulation in the handling and storage of the cages as well as long-term sun exposure (from use on other research plots) during the summer months, between years.

Twenty-four hour monitoring of precipitation indicated that at least trace amounts were recorded on 8 of 17 potential foraging days in 1986, while in 1987 5 of 17 foraging days had a trace or more of precipitation.

Flowers failed to open on foraging day five and six in 1986 (Table IV.1). Both precipitation (Table IV.1) and cool temperatures (Fig. IV.1) occurred on these two days, supporting Kalin's (1971) observation that Limnanthes flowers may not open on wet and cool days. Flowers opened every day during the 1987 foraging period.

Pollinator Treatment Yield-Efficacy

Individual plants

Individual plant data from both years were pooled for analysis and reporting, because of no year interactions and a non-significant ratio of mean square error terms between years (Snedecor and Cochran, 1980). The variation among sub-sampled plants within a treatment was also found to be non-significant ($P=0.53$).

There were no significant differences found in whole-plant seeds per flower yield between any of the caged pollinator treatments. In 1986 whole-plant seeds per flower yields varied from 2.6 to 3.1 (SE=0.3), with an overall average of 2.9. The 1987 data ranged from 2.0 to 2.4 (SE= 0.25), with a significantly ($P<0.001$) lower overall whole-plant seeds per flower average of 2.2. Average seeds per flower rankings (highest to lowest) among the five caged-pollinator treatments were inconsistent between years.

The non-caged pollinator treatment produced greater numbers of average seeds per flower in both 1986 (3.4) and 1987 (2.5) than any of the caged pollinator treatments. Although the non-caged pollinator treatment was significantly greater than some of the caged pollinator treatments, the significant treatment differences were inconsistent between years.

On the basis of the non-significant yield responses obtained between caged pollinator treatments, we conclude that 10, 35, or 60 female O. l. propinqua bees per cage are capable of pollinating individually isolated meadowfoam plants as effectively as ca. 4000 honey bees per cage.

Flower position had a highly significant ($P<0.001$) effect on seeds per flower across all six pollinator treatments tested in 1986 and 1987. The bottom, middle, and top flower positions per plant averaged 2.8, 2.7, and 2.2 seeds per flower, respectively, with a treatment mean standard

error (SE) of 0.3. The top (last) one-third of the flowers produced per stem set significantly ($FPLSD_{0.01}$ = 0.2) fewer seeds per flower than either the bottom or middle flower positions.

Solid stand

O. l. propinqua density treatments produced slightly lower yields (kg ha^{-1}) than the caged honey bee treatments in 1986 and 1987. There were no significant differences (P=0.30) found in yield production between O. l. propinqua and caged-honey bee treatments in 1986, although O. l. propinqua produced a 21% lower average yield (Table IV.2). All five caged-pollinator treatments averaged only 524.6 kg ha^{-1}, while the non-caged honey bee treatment produced a significantly (P=0.001) greater 1076 kg ha^{-1} average (Table IV.2). The disparity in yield between the caged and non-caged treatments in 1986 was a direct result of 9.5 mm of rainfall (Table IV.1) which passed through the sagging tops of the cage material as large, heavy droplets, lodging the solid stand area of all caged plots. In 1987, there were no significant differences found between any of the honey bee treatments (inside or outside cages) and the 60 O. l. propinqua treatment (Table IV.2), although 60 O. l. propinqua bees produced a 17% lower yield than the two caged-honey bee treatments. While the 35 and 10 O. l. propinqua treatments produced significantly lower yields than any of the honey bee treatments, they were not significantly lower than the 60 O. l. propinqua treatment. A positive linear

relationship (P= 0.11; r^2=0.3) existed between O. l. propinqua density and yield which is best described by the regression equation $\hat{y}$ = 611.7 + 4.5x. Overall seed yield (kg ha^{-1}) was significantly (P<0.001) lower in 1986 than 1987, averaging 616.5 and 946.7 kg ha^{-1} across all six pollinator treatments, respectively. O. l. propinqua did not significantly increase yield when added to 4000 honey bees per plot in either 1986 or 1987. Caged treatments without pollinators produced 63 and 133 kg ha^{-1} in 1986 and 1987, respectively (Table IV.2), thus substantiating meadowfoam requirements for insect pollination. The cage support tubes used in 1987 (see Materials and Methods above) appeared to eliminate lodging and subsequent yield loss from precipitation that fell on caged plot treatments.

There were no significant differences (P=0.15) found in seed weights between any of the pollinator treatments in 1986. Average pollinator treatment seed-weights varied from 6.3 to 7.1 mg $seed^{-1}$ (SE=0.20). Lodging of caged-plots may have influenced these results. In 1987, 10 and 35 female O. l. propinqua bees per cage produced average seed-weights of 10.3 and 10.2 mg, respectively. These weights were significantly (P=0.002; $FPLSD_{0.01}$=0.74; SE= 0.17) heavier than the caged-honey bee (9.2 mg), caged-honey bee + O. l. propinqua (9.4 mg), or non-caged-honey bee (9.3 mg) treatments. Sixty O. l. propinqua females per cage produced an intermediate seed-weight of 9.7 mg which was not significantly different from the seed-weights pro-

duced from either O.l. propinqua or honey bee pollinator treatments. A negative linear relationship existed between O. l. propinqua density and seed weight ($P=0.12$; $r^2=0.3$), which is best described by the regression equation $\hat{y} = 10.5 - 0.01x$. Overall seed weight was significantly ($P<0.0001$) lower in 1986 compared to 1987, averaging 6.6 and 9.7 mg $seed^{-1}$ ($SE=0.11$), respectively. If seed set efficiency and seed-weight are powerful predictors of yield in meadowfoam (Krebs and Jain, 1985) then seed-weight produced between pollinator treatments may also be an indicator of pollination efficacy. While the yield (Table IV.2) and seed-weight responses to O. l. propinqua density support this hypothesis, further studies are required for verification.

Seed-oil content was not significantly ($P=0.15$) different between any of the caged pollinator treatments tested in 1986 or 1987. Seed-oil content ranged from 24.8 to 27.5% ($SE=0.66$).

Although the results obtained from solid stand experiments indicate that 10 and 35 O. l. propinqua bees may be less efficient meadowfoam pollinators than 4000 honey bees, the disparity in the total number of pollinators per treatment must not be overlooked when assessing pollinator potential. By monitoring the actual number of foraging bees per treatment we were able to assess individual bee performance on a seed yield per bee basis, which we feel is a more accurate measure of pollinator potential in Mermaid.

Foraging-Bee Density

In 1986 and 1987, both caged honey bee treatments had significantly ($P<0.01$) more bees foraging on flowers than either O. l. propinqua or non-caged honey bee treatments (Table IV.2). While this was predicted, the magnitude of foraging honey bees to foraging O. l. propinqua bees in cages (across all treatments) was unexpectedly low, averaging 10.5:1 and 5.4:1 in 1986 and 1987, respectively (Table IV.2). From these results we conclude that roughly 5% of the Osmia population and <0.5% of the honey bee population were constantly foraging in the caged-meadowfoam plots.

All honey bee treatments in 1987 experienced an average bee m^{-2} reduction of at least 47% below 1986 levels, while O. l. propinqua increased average foraging bees m^{-2} by ca. 47% (Table IV.2). The lack of sugar syrup supplementation, which reduced colony populations near the end of bloom (Jahns, unpublished data), undoubtedly lowered caged-honey bee foraging rates in 1987.

A significant ($P<0.05$) curvilinear relationship existed between yield (kg ha^{-1}) and bees m^{-2} over the five caged-pollinator treatments in both 1986 and 1987 (Fig. IV.2). These results indicate that bees m^{-2} is a more sensitive measure of caged-pollinator activity in relation to yield than is pollinator treatment density (see linear relationship between O. l. propinqua density and yield, under Solid stand, above).

Impact of Bee Efficacy on Yield

Ten O. l. propinqua bees per cage in 1986 and all O. l. propinqua bee density treatments in 1987 produced a significantly ($P<0.01$) higher yield per bee than either the caged honey bee or the caged honey bee + O. l. propinqua bee treatments tested (Table IV.2). While the 35 and 60 female O. l. propinqua treatments did not produce a significantly higher yield per bee than the honey bee treatments in 1986, a similar yield-per-bee trend was evident between the two years. The exceptionally high amount of experimental variation (CV= 78) present in 1986, undoubtedly influenced these results. While significant (1986 = $P<0.05$; 1987 = $P<0.01$) regression analysis indicated that yield per bee declined with increasing O. l. propinqua density (Fig. IV.3), the average yield-per-bee values were 1.7x greater than any of the caged honey bee treatments tested.

We conclude from this study that 60 O. l. propinqua bees are as efficient at pollinating caged meadowfoam plots as are 4000 honey bees. Despite significantly lower foraging pollinator density levels, O. l. propinqua produced similar individual plant seed yields and significantly higher solid stand seed yields per bee. While further studies are needed to determine optimum O. l. propinqua density levels for meadowfoam yield maximization, O. l. propinqua demonstrated potential as an alternative pollinator of caged meadowfoam.

Table IV.1. Climatic and bloom phenology data during pollinator exposure to caged meadowfoam in 1986 and 1987.

	1986			1987		
Forage Day	Date	Flowers Per Cage[a]	Light[b]	Prec. (mm)[c]	Date	Flowers Per Cage	Light	Prec. (mm)
1	5-16	4599	622	0	5-5	5547	626	0
2	5-17	3322	706	0	5-6	7565	636	0
3	5-18	3055	450	0	5-7	15664	630	0
4	5-19	9078	586	T[d]	5-8	13824	593	0
5	5-20	0	337	4.87	5-9	9612	636	0
6	5-21	0	501	2.31	5-10	7387	596	0
7	5-22	10384	721	2.31	5-11	7358	552	0
8	5-23	5756	407	0	5-12	3026	274	7.69
9	5-24	9256	629	T	5-13	5222	486	0.26
10	5-25	14774	726	T	5-14	2254	327	0.51
11	5-26	12756	413	0	5-15	445	267	0.26
12	5-27	9968	488	T	5-16	1246	606	0.51
13	5-28	4599	681	0	5-17	534	471	0
14	5-29	3531	714	0	5-18	327	589	0
15	5-30	1691	684	0	5-19	118	596	0
16	5-31	227	692	T	5-20	207	652	0
17	6-1	105	707	0	5-21	178	604	0
Averages:		5476.5	592	0.6		4736	538	0.5

[a]Flowers per cage- average of 3 replicates from 8.9 m^2 area per cage.
[b]Light- Average daily photon flux density in μ mol m^{-2} sec^{-1} inside cages.
[c]Prec.-is daily, 24-hour precipitation (mm) recorded outside of cages.
[d]T-Precipitation measured as a trace amount.

Table IV.2. Pollinator treatment seed yield, foraging pollinators m^{-2}, and seed yield-per-bee within caged and non-caged meadowfoam plots, in 1986 and 1987.

Pollinator treatment	1986[a] Yield kg/ha	1986[a] Bees m^{-2}	1986[a] Yield / bee[b]	1987[a] Yield kg/ha	1987[a] Bees m^{-2}	1987[a] Yield / bee
10 Osmia	456	0.5	110.5	673	0.8	94.7
35 Osmia	492	1.3	48.9	736	1.6	53.8
60 Osmia	477	3.6	15.3	897	2.3	44.6
4000 Apis (Std)	593	20.4	3.3	1052	7.6	16.2
4000 Apis +35 Osmia	605	17.5	3.9	1119	9.2	14.2
LSD	NS	5.7**	73.1**	261*	4.2**	25.7**
CV	19	25	78	16	22	22
Non-caged (Control)[c]	1076	8.4	14.4	1203	4.3	31.4
LSD	326**	5.0**		329**	3.8**	
CV	21	23		14	36	
No Pollinators[d]	63	0		133	0	

*,** LSD performed at 0.05 and 0.01 level of significance, respectively.

[a]Values presented are means from three replicates.

[b]Yield / Bee = yield equivalent (kg ha^{-1}) per plot / the average number of bees per 8.9 m^2 foraging area per plot. The actual values used differ slightly from potential values calculated from the table, the result of rounding and averaging table values.

[c]Non-caged (Control) = non-caged plots with pollinator densities equivalent to eight honey bee colonies ha^{-1}.

[d]No Pollinators = caged plots without pollinators.

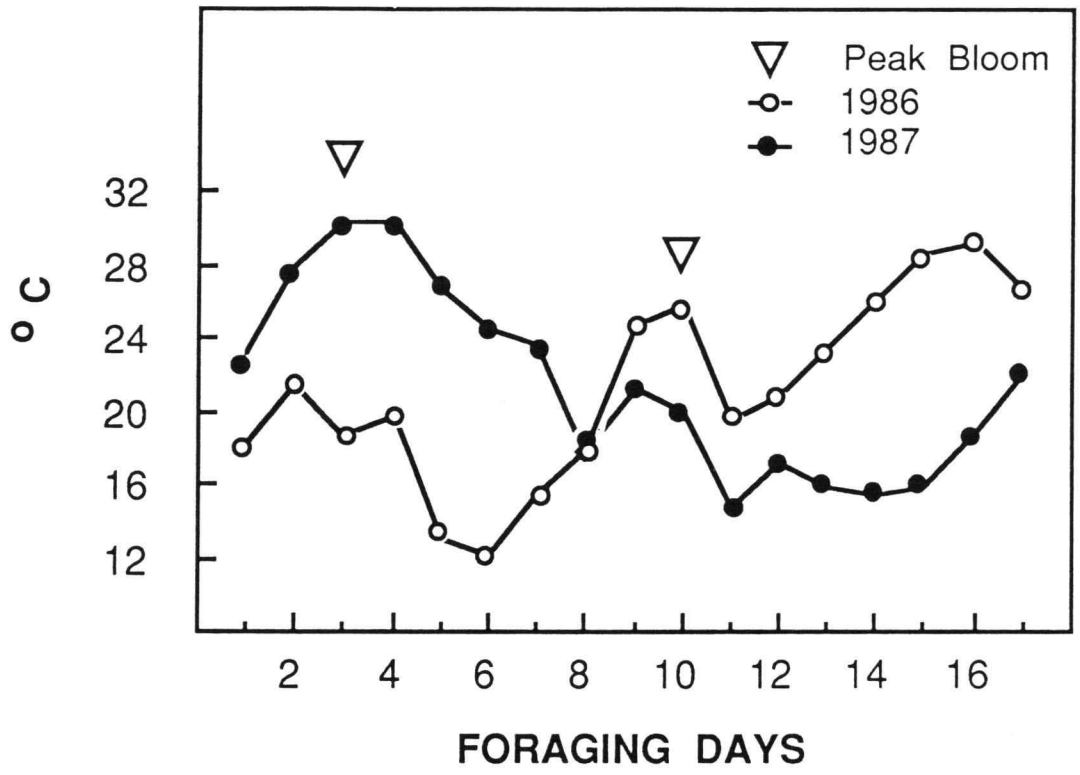

Figure IV.1 Daily average temperatures during pollinator foraging periods (1000 to 1900 hours) and days of peak bloom for 1986 and 1987.

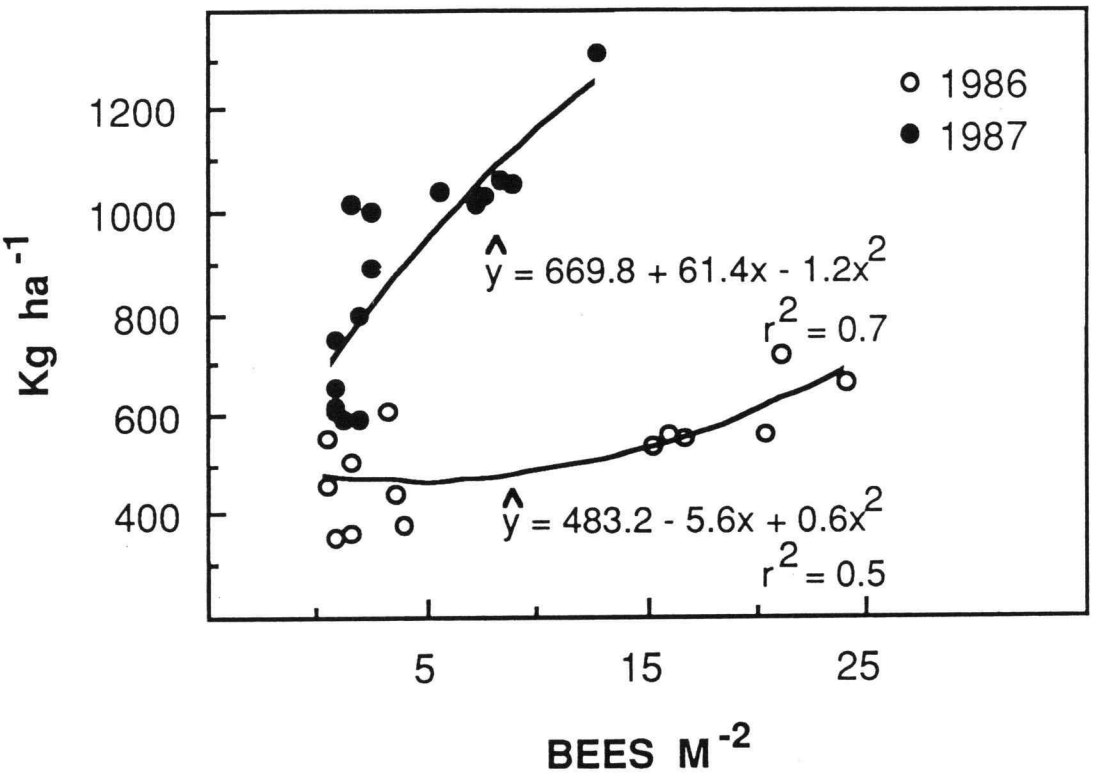

Figure IV.2 Relationship between foraging-bee pollinators m^{-2} and yield (kg ha^{-1}) over the five caged-pollinator treatments used in 1986 and 1987.

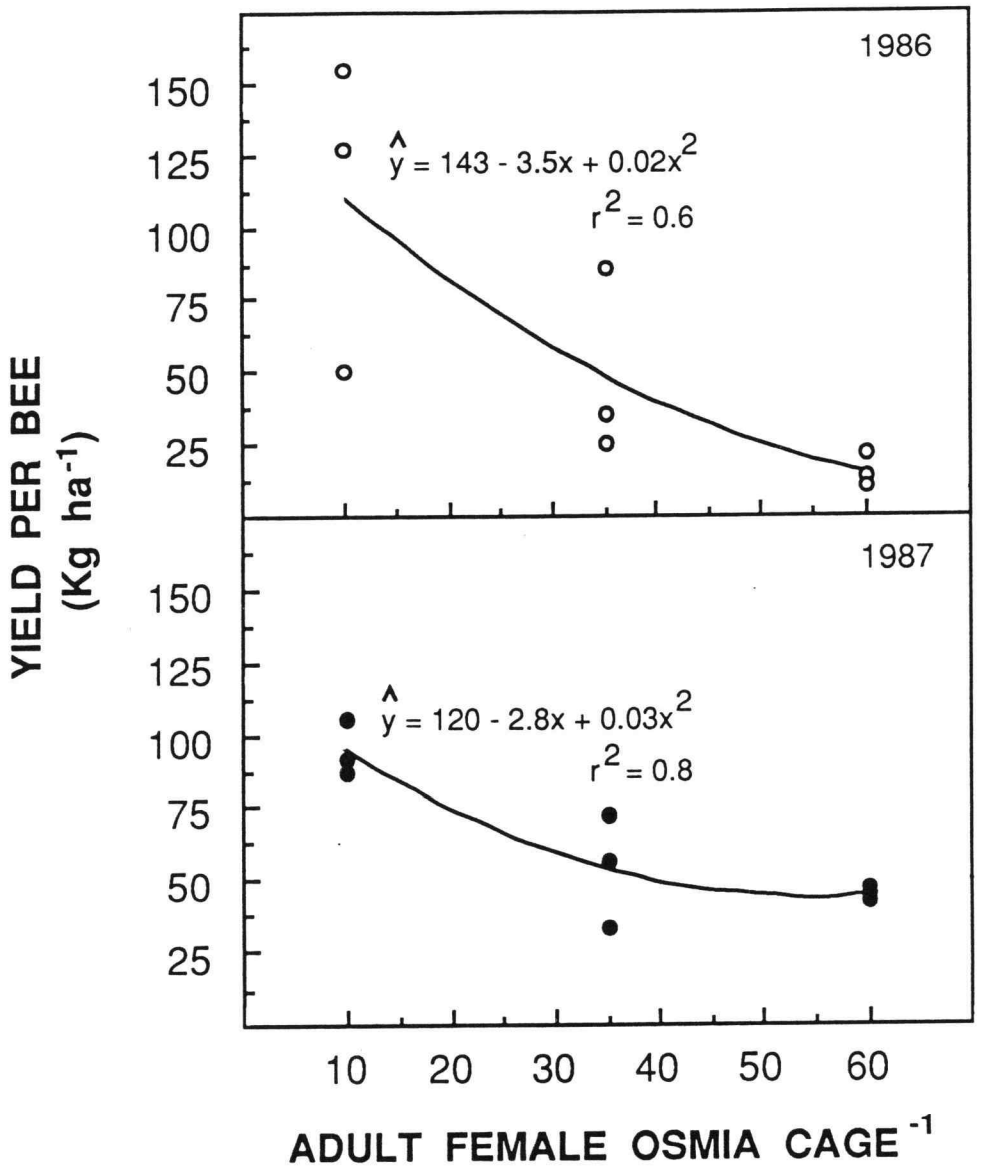

Figure IV.3 Change in meadowfoam yield-per-bee in relation to an increase in adult female O. l. propinqua density (between 10 and 60 bees per cage), in 1986 and 1987.

CHAPTER V

SURVIVAL RATE AND REPRODUCTIVE SUCCESS OF *OSMIA LIGNARIA PROPINQUA* CRESSON (HYMENOPTERA: MEGACHILIDAE) IN CAGED MEADOWFOAM, *LIMNANTHES ALBA* BENTH. (LIMNANTHACEAE)

ABSTRACT

Meadowfoam (Limnanthes alba Benth.: Limnanthaceae) is an entomophilous, winter annual oilseed crop with an unstable seed yield history that may be attributed to inadequate honey bee pollination. Osmia lignaria propinqua Cresson, a wild bee pollinator, may improve meadowfoam pollination, but its availability is somewhat restricted. A two-year survival and reproductive efficacy study was undertaken, within caged plots, to evaluate the potential of O. l. propinqua for sustaining future meadowfoam pollination populations. Pollinator density treatments of 10, 35, and 60 female bees per cage were maintained during the 1986 and 1987 meadowfoam bloom periods. Percent survival was negatively correlated with female density, producing ca. a 50% average survival rate from 18 female bees. Nest, male, and total cell production increased linearly with increasing female O. l. propinqua densities. Female progeny production in 1986 and 1987 averaged 2.1, 0.6, and 0.3 females per bee from the 10, 35, and 60 female bee density treatments, respectively. The male:female ratio from 10, 35, and 60 female bees per cage averaged 2.33:1, 2.63:1, and 5.62:1, respectively, indicating resources (ie. pollen or nectar) may become limited as bee densities increase. While adult female survival and nesting success were density dependent, O. l. propinqua demonstrated the potential to survive and reproduce from resources obtained solely from foraging on Mermaid meadowfoam flowers.

INTRODUCTION

Meadowfoam (Limnanthes alba Benth. cv. Mermaid) is an entomophilous, winter annual oilseed crop being developed at Oregon State University, Corvallis (Calhoun, 1975; Jolliff, 1989). The oil extracted from meadowfoam seed has generated industrial interests for uses ranging from high temperature lubrication to cosmetics (Miwa and Wolff, 1962; Gentry and Miller, 1965; and Purdy and Craig, 1987). From 1977 to 1988, Mermaid meadowfoam seed yield fluctuated from 788 to 1760 kg ha^{-1} in replicated yield trials (Jolliff, 1988). It has been speculated that inadequate pollination may be one of several possible causes for these yearly yield fluctuations (Calhoun and Crane, 1978; Franz and Jolliff, 1989). Consistently higher yields would improve the economic competitiveness of this new resource.

The honey bee, Apis mellifera L., is the primary pollinator of cultivated meadowfoam in Oregon (Karow et al., 1986). During the May bloom period, cool ($<16^{0}C$), wet, and windy (>24 km hr^{-1}) weather conditions may occur, inhibiting honey bee foraging activity (Lundie, 1925). These weather conditions may also inhibit flower opening (Kalin, 1971), delay anthesis (Pearson and Jolliff, 1986), and intensify the need for timely pollination to maximize seed set.

The problems associated with honey bee pollination of meadowfoam flowers are similar to those reported by Torchio (1976) in early-season orchard crops. Inclement weather is

prevalent at this time of year, bloom is of short duration, and the amount of bloom is enormous. Based on the success of Osmia lignaria propinqua Cresson as an early-season orchard crop pollinator (Torchio, 1979,1981,1985; Kuhn and Ambrose, 1984), we selected O. l. propinqua as a potential meadowfoam pollinator because O. l. propinqua: (1) is a species which is gregarious and develops a reasonably large local population which can be managed (Torchio, 1976); (2) engages in early season activity (Torchio, 1976) which can be synchronized with meadowfoam bloom; and (3) has a tendency to specialize on abundant crop resources (Torchio 1976,1981; Torchio and Asensio, 1985).

The objectives of this study were to examine the effects of three O. l. propinqua densities in caged meadowfoam plots on: (1) the rate of survival and (2) the reproductive potential for sustaining future pollination populations.

MATERIALS AND METHODS

Plot Preparation

Caged field experiments were conducted in 1986 and 1987 at the Oregon State University Hyslop Crop Science Field Laboratory near Corvallis, Oregon. A 0.24 ha field was planted with seed from the meadowfoam cultivar Mermaid at a 22 kg ha^{-1} seeding rate on an 18 cm row spacing on 3 Oct. 1985 and 9 Oct. 1986. Plot perimeter dimensions were 3.7 by 3.7 m. Prior to bloom each plot was hand-trimmed, leaving a 3.7 by 2.4 m solid stand down the center (north to south) of each plot. In the remaining 0.65 by 3.7 m spaces on each side of the solid stand, all but four plants per side were removed. These four plants were isolated 0.74 m apart (north to south) and staked for support. At ca. 1% bloom all plots received an insecticide application of 'Mavrik' ([(RS)-alpha-cyano-3-phenoxybenzyl(R)-2-[2-chloro-4-(tri-fluoro-methyl)anilino]-3- methylbutanoate]), at 0.1 kg/ha (ai), the morning of plot caging to reduce incidental pollination by non-treatment insects (Free, 1970). The plots were covered with natural colored poly-vinylidene chloride (Saran) cages, of 20 by 20 mesh, measuring 3.7 by 3.7 by 1.8 m.

Bloom Phenology and Environmental Monitoring

Bloom phenology was monitored daily between 1600 and 1700 hours, from the solid stand area of three caged plots (without pollinators), by removing and counting all opened flowers within a 0.1 m^2 frame placed randomly within each

plot. The caged bloom periods were from 12 May to 1 June 1986 and 2 to 21 May 1987. The bloom phenology sampling period started on the day of pollinator introduction (10% bloom) and ended on the day of pollinator removal (>99% bloom), from 16 May to 1 June 1986 and 5 to 21 May 1987.

Hourly temperatures were recorded inside and outside of the caged meadowfoam plots using thermographs placed under white vented wood covers. Daily photon flux density was measured parallel to the top of the meadowfoam flower canopy inside and outside of the caged plots at 15-30 minute intervals using a line quantum sensor (Model 191S, Line Quantum Sensor, LI-COR, Inc., Lincoln, NE 68304). Daily precipitation was recorded 300 m away from the treatment plots at the Hyslop Crop Science Field Laboratory Weather Station.

Pollinator Preparation

In 1985, a population of O. l. propinqua was obtained from the USDA-ARS Bee Biology Lab in Logan, Utah for release in 1986. In 1986, 30% of the O. l. propinqua bee population released in 1987 was obtained from USDA-ARS, 60% from Dr. Ray Lynn (Star Rt., Mendon, UT), and 10% from 1986 offspring produced in caged meadowfoam plots. Adults were shipped and over-wintered in 7 mm straws ca. 15 cm long. All straws were X-rayed upon receipt to identify and remove any parasites and parasitoids present (Stephen and Undurraga, 1976). Straws were kept at 3-5^{0}C from the time they

were received (9 Oct 1985 and 3-8 Oct 1986) until they were incubated for emergence (13-15 May 1986 and 2-4 May 1987).

Density treatments of 10, 35, and 60 adult females per caged plot were used. A corresponding number of adult males per plot were also introduced at the same time for mating purposes. Two days before incubation, O. l. propinqua cocoons were removed from their straws and individually placed into #000 gelatin capsules. Capsules (containing cocoons) were stuck to the sticky side of a 27.9 x 21.6 cm label sheet that was stapled (for support) to an identically sized sheet of cardboard. These sheets were then placed in 30^0C incubators for 12 hours followed by another 12 hour treatment at 13^0C. Sheets were inspected for adult O. l. propinqua cocoon emergence at 10 minute intervals. Emerged adults (within capsules) were sorted by sex and placed into the $3\text{-}5^0$C cooler for storage. These incubation procedures were repeated for 3 days, until a surplus of females was collected.

Emerged O. l. propinqua bees retained at $3\text{-}5^0$C were removed from gelatin capsules, grouped into densities of 10, 35, and 60 males or females, and placed into cardboard release boxes measuring ca. 30 by 15 by 10 cm. A small flap, cut into the top of each release box, was taped shut until the boxes were introduced into treatment cages. The flaps of these boxes were then opened to facilitate O. l. propinqua emergence.

One wood nesting unit was placed (facing SE) in each cage assigned to the O. l. propinqua 10 and 35 bee density treatments, and two nesting units were placed (facing SE and SW) in the 60 bee density treatment cages. All nesting units were supported on two stakes, 1 m above soil level. Nesting units measured 17.8 by 15.2 by 13.3 cm and contained 54 paper straws 15 cm long with a 7 mm diameter.

O. l. propinqua males and females were released into cages 16 May 1986 and 5 May 1987. Nesting units were inspected every other night throughout the flowering period with the aid of a small pen-light directed into the nesting holes of the nesting medium to facilitate identification and counting of male and female bees. Additional females were added to the cages on 23 and 24 May 1986 and 11 and 19 May 1987 to re-establish the same number of females originally released. Mud and water were made available in tubs placed in each treatment cage.

Pollinator Survival

Adult female O. l. propinqua survival was calculated from the number of females remaining per treatment divided by the total number of females introduced (initial and supplemented) per cage over the two bloom periods.

The density-treatment data were analyzed as a completely randomized design (CRD) with three replications. The addition of years created a 2 x 3 (year x density) factorial and the combined data were used for testing year main effects. Data were analyzed by analysis of variance

(ANOVA) and treatment effects compared using orthogonal polynomial contrasts and regression analysis at $P<0.05$, unless otherwise stated.

Reproductive Success

O. l. propinqua was evaluated under caged meadowfoam plots for nest production, live bee production (male/female), and total cell production. The evaluation process began with O. l. propinqua nest completion, which was culminated by a mud cap at the entrance of the straw nest. Capped straws were collected every other night, following the counting of adult bees. At least five straws were retained per nesting unit to stimulate foraging (Torchio, personal communication). Removed straws were labeled and kept at room temperature (18^0C-24^0C) until late October. These straws were then taped to cardboard sheets and X-rayed for cell counting (number of progeny development chambers), developmental stage (larvae or adult), and parasite identification. The number of straw nests containing cells were recorded and all nest associates removed. Adult progeny retained in the straw nests were then placed in paper bags and stored at 3-5^0C until the following spring. In April, emergence procedures were again followed to determine male, female, and total cell production. 'Other' production was also recorded, which grouped all cells in which larvae or adults were visually observed (X-ray analysis) prior to the 3-5^0C incubation period, but from which bees emerge.

The 10, 35, and 60 female O. l. propinqua density treatments were arranged and the data analyzed as a CRD with three replications. The addition of years created a 2 x 3 (year x density) factorial. Data were analyzed by ANOVA and treatment effects compared using orthogonal polynomial contrasts and regression analysis at $P<0.05$, unless otherwise stated.

RESULTS AND DISCUSSION

Bloom Phenology and Environmental Monitoring

The bloom phenology sampling periods were from ca. 10% initial bloom to >99% bloom. In 1986 and 1987 a daily average of 5477 and 4736 flowers were produced per cage, respectively (Table V.1). Peak bloom occurred ten days after pollinator introduction in 1986, while in 1987 it occurred on day three (Table V.1).

Hourly temperature values were found to be identical inside and outside of the cages with daily temperatures during peak pollinator activity (1000-1900 hours) averaging $21.8^{0}C$ and $21.5^{0}C$ in 1986 and 1987, respectively (Fig. V.1). In general, average daily temperatures progressed from cool to hot in 1986, while 1987 experienced a hot to cool temperature trend during the potential foraging period (Fig. V.1).

Peak bloom in 1986 took place on the tenth foraging day, which contained the fifth highest temperature during the 1986 foraging period. Peak bloom in 1987 took place on the day (foraging day three) on which the maximum daily average temperature was reached (Fig. V.1). Regression analysis between the average number of daily flowers produced per cage (Table V.1) and the daily average temperatures observed per foraging day (Fig. V.1) indicated no significant relationship ($P=0.347$; d.f.=1,13; $F=0.95$) in 1986, except between peak bloom and peak temperature. In 1987, a highly significant ($P<0.001$; d.f.=1,15; $F=80.76$) linear relation-

ship existed (r^2=0.84; $\hat{y}$=-14010 + 873.2x, where $\hat{y}$ = average number of flowers per cage and x = temperature ^{0}C) between bloom phenology and temperature.

There was a 31% (P<0.001; inside/outside correlation r^2=0.96; n=45) and 38% (P<0.001; inside/outside correlation r^2=0.93; n=135) average reduction in photon flux density inside (versus outside) cages in 1986 and 1987, respectively. These differences may be attributed in part to a darkening of the cage materials, both from dirt accumulation during the handling and storage of the cages as well as long-term sun exposure (from use in other research) during the summer months, between years.

Twenty-four hour monitoring of precipitation indicated that at least trace amounts were recorded on 8 of 17 potential foraging-days in 1986, while 1987 had 5 of 17 foraging-days with a trace or more of precipitation. Flowers failed to open on foraging days five and six of the 1986 bloom period (Table V.1). Both precipitation (Table V.1) and cool temperatures (Fig. V.1) occurred on these two days, supporting Kalin's (1971) observation that Limnanthes flowers may not open on wet and cool days. Flowers opened every day during the 1987 foraging period.

Pollinator Survival

The effects of foraging in flowers of the meadowfoam cv. Mermaid (under cages) on O. l. propinqua survival was influenced by female density levels in both 1986 and 1987 (Fig. V.2). In 1986, a quadratic relationship (P=0.070)

existed between the number of O. l. propinqua females and percent survival. Ten females per cage averaged a 299% and 195% greater survival rate than the 35 and 60 female density treatments, respectively. In 1987 a similar quadratic relationship (P=0.047) was found. Ten females per cage averaged a 156% and 186% greater survival rate than the 35 and 60 female density treatments, respectively. Under the caged plot configuration used in this study, it appears that to obtain at least a 50% survival rate, no more than 15 to 18 female O. l. propinqua bees should be maintained per cage (Fig. V.2).

While the error term disparity between the two years prohibited a combining of the data, the similarity (P=0.52; d.f.=1,12; F=0.45) in O. l. propinqua survival rates between 1986 and 1987 is of importance when considering that 1987 had an average of 4.7 times more supplemented females introduced per cage than in 1986 (Table V.2) and a 13.5% reduction in average bloom per cage (Table V.1). These data indicate that percent survival of O. l. propinqua females is density-dependent and surprisingly stable under the caged conditions of this study. Although quantification of mortality and escapement was not attempted, the conclusion can be drawn that an increased death rate (at higher density levels) resulting from a reduction in available resources (ie. pollen or nectar) would offer survival-response stability, allowing for density-dependent factors to be controlled by available plant resources.

Nesting Success

Results obtained from dissections of treatment nest materials are summarized in Table V.3. O. l. propinqua density treatment analysis in 1986 and 1987 indicated that a positive linear response existed between adult female bee density and the number of nests produced, the number of males produced, and the number of total cells produced (Fig. V.3). Nest production was significantly affected by the number of adult females per cage in 1986 ($P<0.001$; $r^2=0.91$) and 1987 ($P=0.08$; $r^2=0.37$) (Fig. V.3A). Sixty female O. l. propinqua bees produced 58% and 140% more nests in 1986 and 25% and 55% more nests in 1987 than the 35 and 10 female density treatments, respectively (Table V.3). Male production in 1986 ($P<0.001$; $r^2=0.90$) and 1987 ($P=0.02$; $r^2=0.57$) increased with increasing numbers of females per density treatment (Fig. V.3B). In 1986, the 60 female density treatment produced 60% and 151% more males, and in 1987, 77% and 115% more males than the 35 and 10 density treatments, respectively (Table V.3). Total cell production was also greatest in the 60 female density treatment in both 1986 ($P<0.001$; $r^2=0.87$) and 1987 ($P=0.05$; $r^2=0.43$) (Fig. V.3C). A 37% and 103% greater number of total cells in 1986 and a 55% and 77% greater number of total cells in 1987 were produced in the 60 compared to the 35 and 10 female density treatments, respectively (Table V.3). From a total of 385 females (and 315 males) introduced into the caged Mermaid meadowfoam plots in 1986 (Table V.2) a total

of 1340 cells were produced in 398 nests. Out of the 1340 cells produced, 1170 progeny survived (913 males and 257 females) resulting in a total population increase of 204% (Table V.3). In 1987, 712 females (and 315 males) (Table V.2) produced a total of 1137 cells in 333 nests. Out of 1137 cells produced, 1006 progeny survived (764 males and 242 females) resulting in a total population increase of 41% (Table V.3).

Total female production in both 1986 and 1987 was highest in the 35 female density treatment (Table V.3) which was best described by a quadratic response in 1986 ($\hat{y} = 14.40 + 1.28x - 0.02x^2$; $r^2=0.52$; $P=0.04$) and by no significant ($P= 0.98$) response differences between the three density treatments in 1987. The lack of response between the three density treatments in 1987 was undoubtedly influenced by the early and late supplementation of 468% more females than in 1986 (Table V.2), although further studies are required for verification. While total female production is of value for assessing total progeny production, the results are misleading, especially with respect to female progeny production per bee.

In 1986 a total average of 3.5 cells were produced per female, with the 10, 35, and 60 female density treatments producing a per-bee average of 2.5, 0.9, and 0.3 female progeny, respectively (Fig. V.4). From 385 females released in 1986, 257 living females were produced, a 33% reduction in the future female pollinator population. A 54%

lower average cell number (1.60) was produced per female in 1987 compared to 1986, with the 10, 35, and 60 female density treatments producing 1.7, 0.3, and 0.2 female progeny per bee, respectively (Fig. V.4). From 712 females released in 1987, 242 female progeny were produced resulting in a 66% reduction in the potential female pollinator population. The results of these data indicate that to sustain or increase future female O. l. propinqua bee populations for caged meadowfoam pollination, a limit of between 18 to 32 female bees per cage should be maintained (Fig. V.4).

The male:female sex ratio response to female bee density was best described by combining the density treatment data from 1986 and 1987. Orthogonal polynomial contrasting revealed a highly significant (P=0.003) quadratic response with 10, 35, and 60 female O. l. propinqua density treatments averaging 2.3:1, 2.6:1, and 5.6:1 (male:female) sex ratios, respectively (Fig. V.5). These findings are supported inpart by Torchio (1985) who found that a reduced number of bees (along with a longer bloom period and nesting season) lowered the male-biased sex ratio of progeny. A potential saturation of bees within cages (as females per cage increased) may have resulted in Mermaid meadowfoam resources (ie. nectar and pollen) becoming limited. Resource limitations, found by Torchio (1985) in apple pollination studies with O. l. propinqua, produced an extended period for individual cell construction, a large bias toward male progeny production (Fig. V.5), an increase in immature mor-

tality ("other" cell production - Table V.3), and a potential reduction in adult survival rates (Fig. V.2). While resource limitations cannot be verified from this study, nesting success results indicate that potential resource limitations may be occurring at the higher female bee density and supplementation levels (Table V.3). Further resource limitation assessment is needed for quantification.

Indoor storage (18-24^{0}C) of O. l. propinqua nests prior to overwintering (3-5^{0}C) and dissection (Stephen, personal communication) and pollinator supplementation (for meadowfoam yield maximization) may have compromised progeny production levels. Further research is required to assess the reproductive impact of these techniques. Regardless of experimental shortcomings, O. l. propinqua proved it can successfully survive and nest using pollen and nectar resources obtained solely from foraging in Mermaid meadowfoam plots under cages.

From the results obtained over the 1986 and 1987 bloom periods, we conclude that both survival and female reproduction are negatively correlated to female pollinator density, with densities of less than twenty adult female O. l. propinqua bees per cage appearing optimum for maintaining both adult survival and future bee populations.

Table V.1. Climatic and bloom phenology data during pollinator exposure to caged meadowfoam in 1986 and 1987.

	1986			1987		
Forage Day	Date	Flowers Per Cage[a]	Light[b]	Prec. (mm)[c]	Date	Flowers Per Cage	Light	Prec. (mm)
1	5-16	4599	622	0	5-5	5547	626	0
2	5-17	3322	706	0	5-6	7565	636	0
3	5-18	3055	450	0	5-7	15664	630	0
4	5-19	9078	586	T[d]	5-8	13824	593	0
5	5-20	0	337	4.87	5-9	9612	636	0
6	5-21	0	501	2.31	5-10	7387	596	0
7	5-22	10384	721	2.31	5-11	7358	552	0
8	5-23	5756	407	0	5-12	3026	274	7.69
9	5-24	9256	629	T	5-13	5222	486	0.26
10	5-25	14774	726	T	5-14	2254	327	0.51
11	5-26	12756	413	0	5-15	445	267	0.26
12	5-27	9968	488	T	5-16	1246	606	0.51
13	5-28	4599	681	0	5-17	534	471	0
14	5-29	3531	714	0	5-18	327	589	0
15	5-30	1691	684	0	5-19	118	596	0
16	5-31	227	692	T	5-20	207	652	0
17	6-1	105	707	0	5-21	178	604	0
Averages:		5476.5	592	0.6		4736	538	0.5

[a]Flowers per cage- averaged from three replications.
[b]Light- Average daily photon flux density in μ mol m^{-2} sec^{-1} inside cages.
[c]Prec.-is daily, 24-hour precipitation (mm) recorded outside of cages.
[d]T-Precipitation measured as a trace amount.

Table V.2. Female O. l. propinqua supplementation into treatment cages during the 1986 and 1987 caged meadowfoam bloom periods.

Year	Starting Density	Females Added[a]	Ending Density
1986	10	0.0	6.7
	35	5.3	6.7
	60	18.0	17.7
1987	10	6.3	11.0
	35	51.3	23.0
	60	74.7	32.7

[a]Females added= average number of female O. l. propinqua added per density treatment replication (X 3 for total number of females added per treatment).

Table V.3. Nesting results of O. l. propinqua in 1986 and 1987 at three density levels under caged plots containing plants of the meadowfoam cultivar Mermaid. Density response values are the totals produced from three replicates at each density level.

Den.	# holes	# nests	% util.[a]	Total cells	M	F	Other[b]	Sex ratio
				1986				
10	162	81	50	297	180	76	41	2.4:1
35	162	123	76	441	282	109	50	2.6:1
60	324	194	60	602	451	72	79	6.3:1
Total:	648	398	61	1340	913	257	170	3.6:1
				1987				
10	162	88	54	291	175b	81	35	2.2:1
35	162	109	67	332	213b	83	36	2.6:1
60	324	136	42	514	376a	78	60	4.8:1
Total:	648	333	51	1137	764	242	131	3.2:1

[a]% Util.= number of nests / number of holes
[b]Other includes all cells lacking adult emergence.

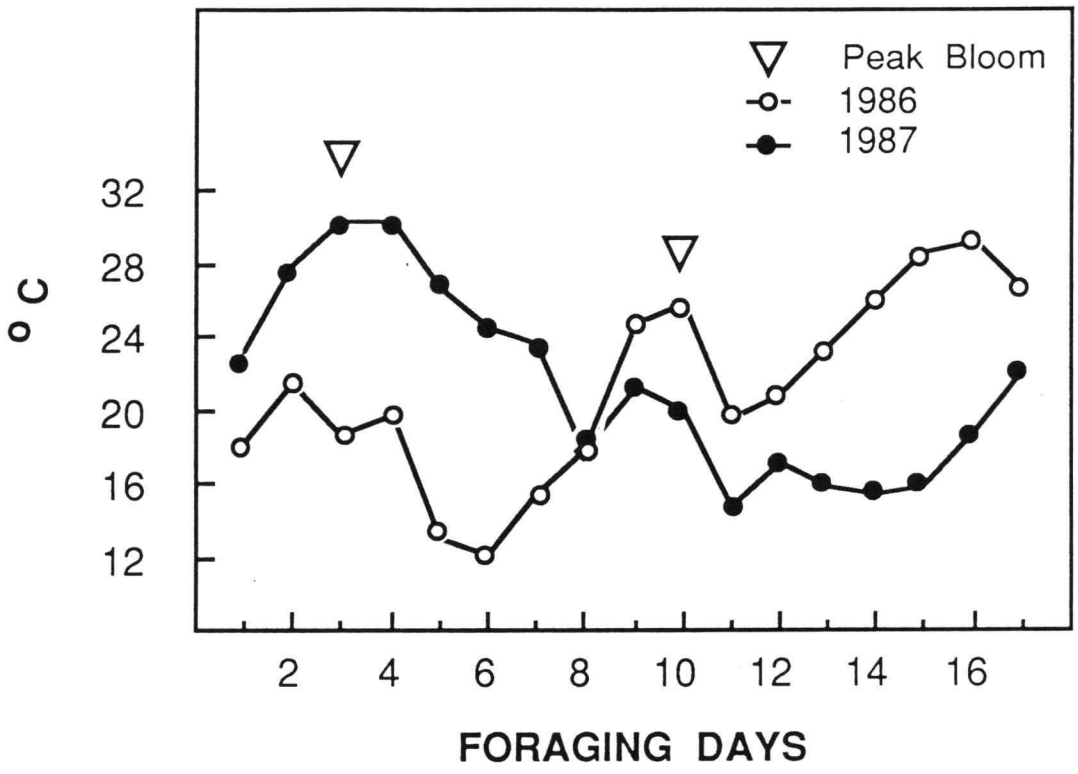

Figure V.1 Daily average temperatures (1000 to 1900 hours) during O. l. propinqua foraging; and days of peak Mermaid meadowfoam bloom for 1986 and 1987.

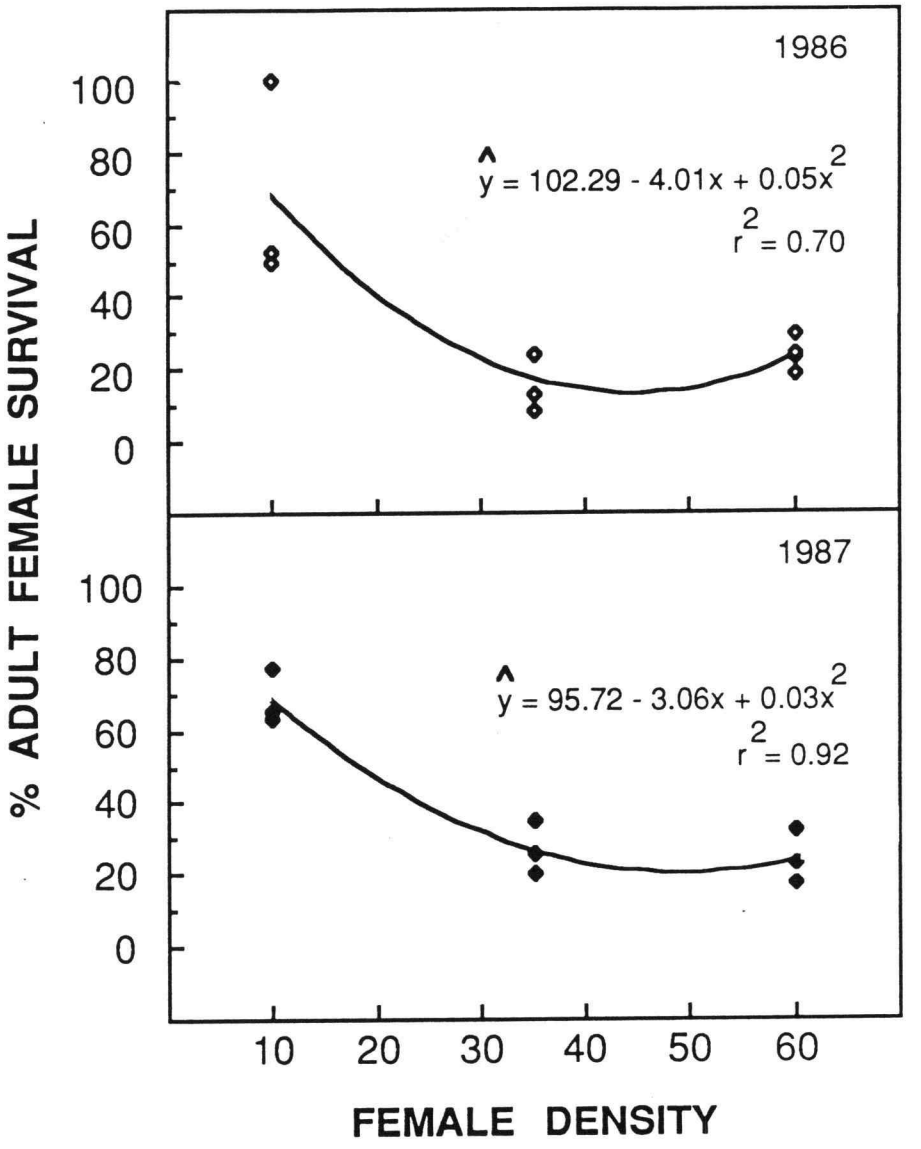

Figure V.2 Adult female survival response to female O. l. propinqua bee density levels in 1986 and 1987, when foraging in caged plots containing Mermaid meadowfoam plants.

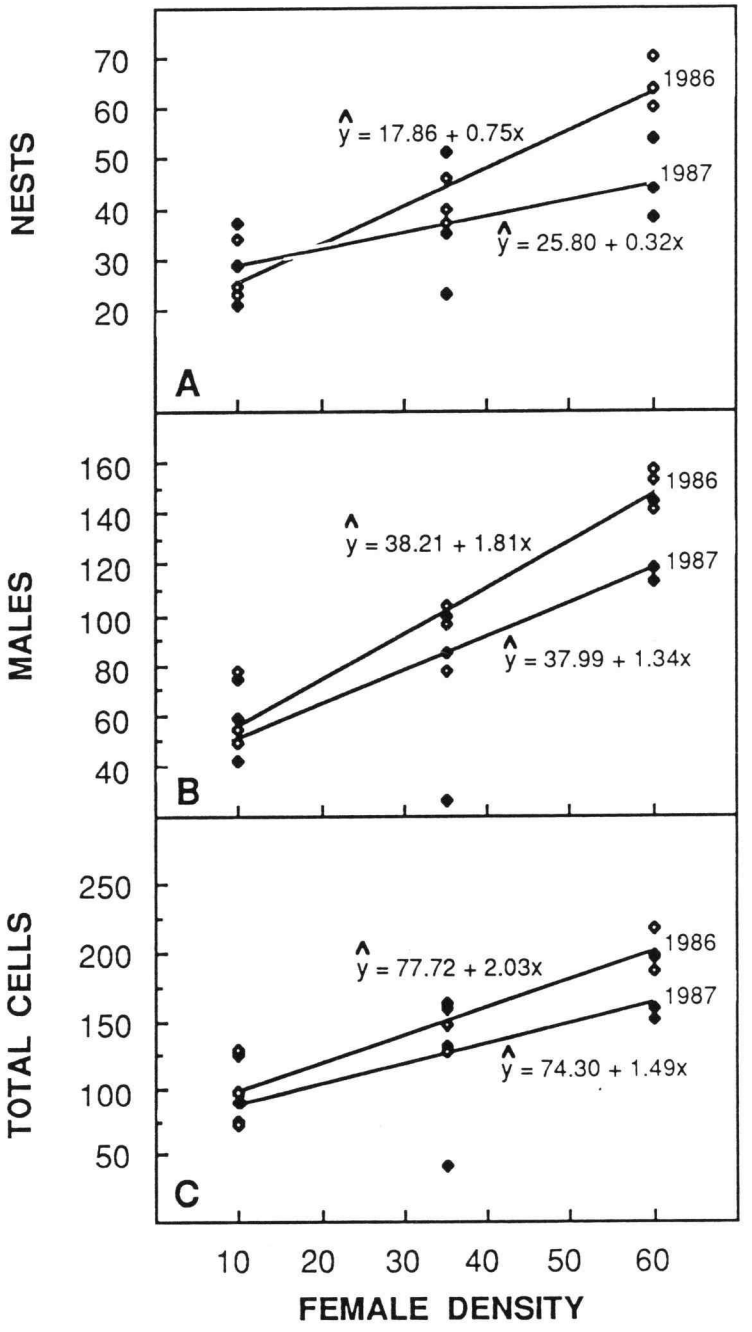

Figure V.3 Nest (A), male (B), and total cell (C) production in response to female O. l. propinqua bee density levels within caged plots containing Mermaid meadowfoam plants, in 1986 and 1987.

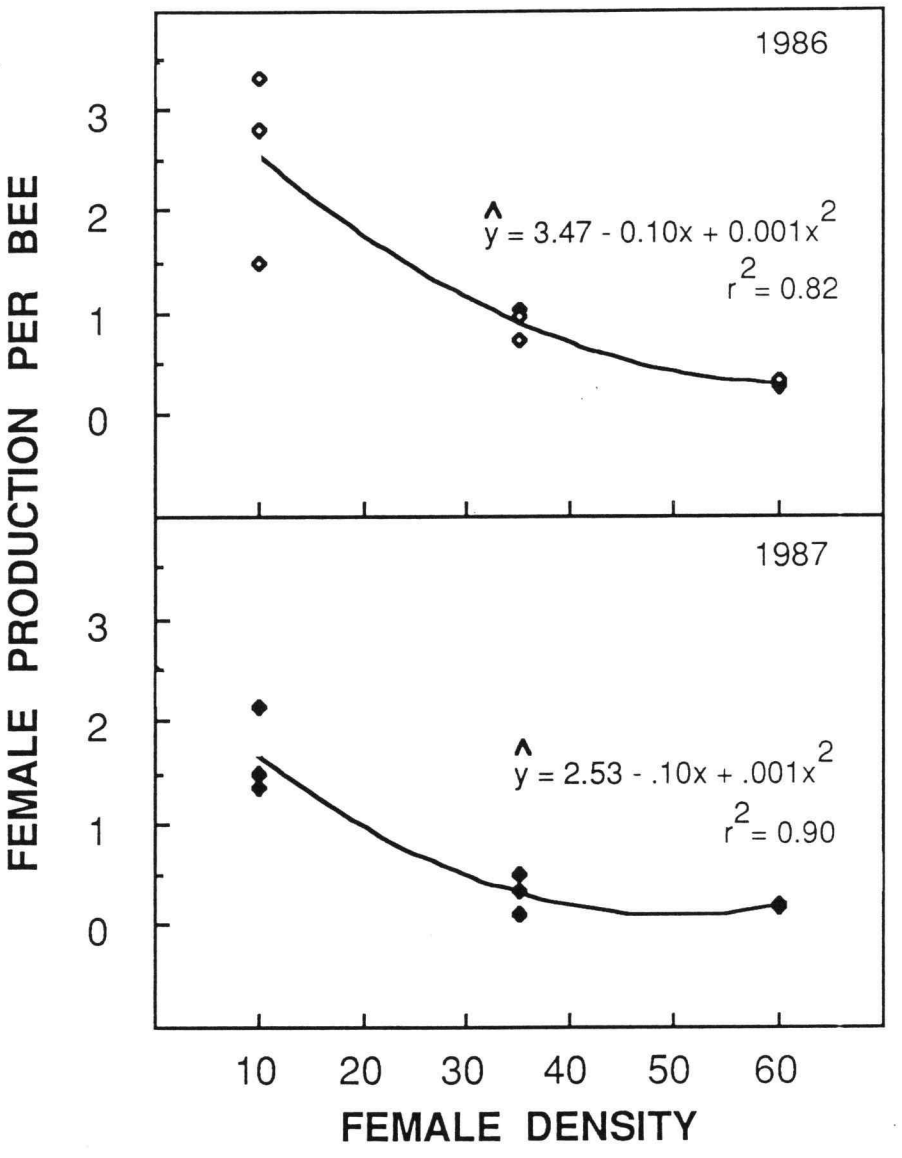

Figure V.4 Female density level effects on female progeny production per bee in caged plots containing Mermaid meadowfoam plants, in 1986 and 1987.

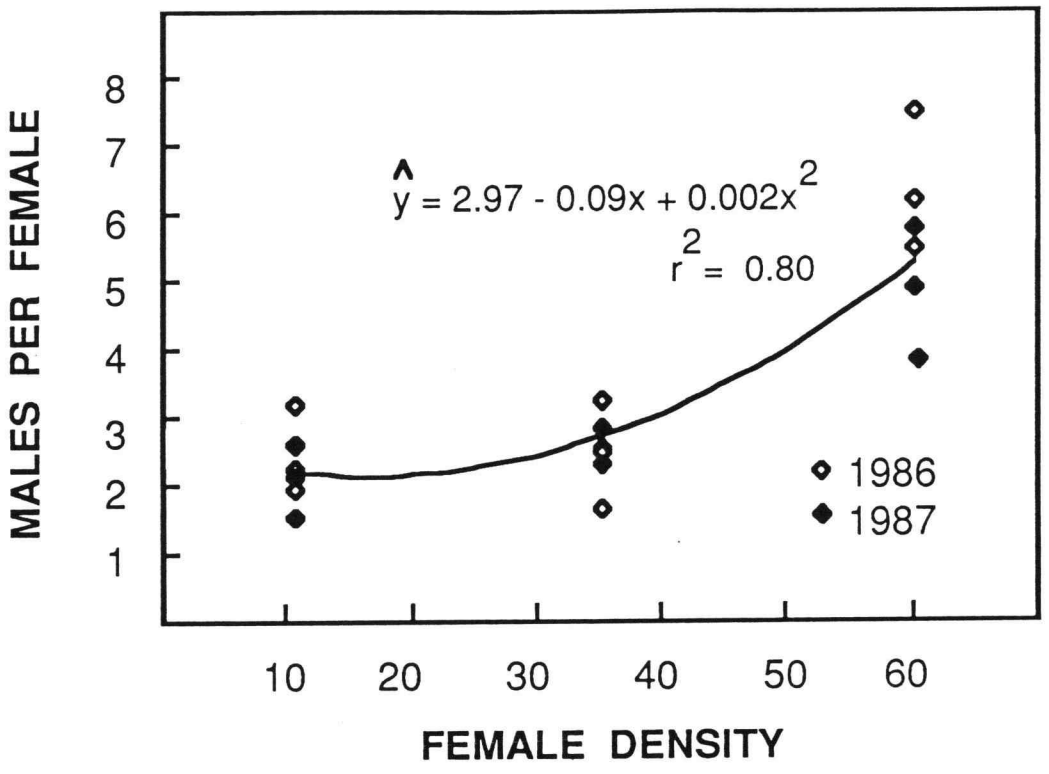

Figure V.5 Sex ratio (male:female) response to adult female O. l. propinqua density levels within caged plots containing Mermaid meadowfoam plants (combined 1986 and 1987 data).

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APPENDICES

APPENDIX I

EMULATION OF MEADOWFOAM SINGLE-PLANT PERFORMANCE WITHIN A SOLID STAND

This preliminary study was completed during the 1987 meadowfoam bloom period. It was initiated to determine if plant spacing, via our field plot design, would emulate morphological and reproductive solid stand plant development. This plant spacing design was used in the O. l. pro-pinqua studies reported in Chapters four and five.

ABSTRACT

Evaluation of individual meadowfoam (Limnanthes alba Benth.) plants of the cv. Mermaid from solid stand is complicated by the intertwining of numerous stems and flowers. This study tested a plant spacing field plot design which eliminated the need to disentangle stems and flowers. Individually spaced plants were compared to solid stand plants grown on 18 cm row spacings. A spaced-plant nursery treatment grown on a 91 cm row spacing was used to illustrate yield component plasticity within plants of the cv. Mermaid. No significant differences in morphological and reproductive plant responses were found between spaced and solid stand plants grown on the 18 cm row spacing. The plants in the 91 cm row spacing contained between 8-9 times more stems and flowers per plant than those planted on 18 cm rows. Spaced plants sown at the recommended 18 cm row spacing retained the competitive plant environment required for morphological and reproductive emulation of solid stand plant responses.

INTRODUCTION

Meadowfoam is a newly developed, winter annual oilseed crop well-adapted to the Mediterranean climate and poorly drained soils of Oregon's Willamette Valley (Calhoun, 1975; Jolliff et al., 1981). Diverse manufacturers, from areas such as the lubrication and cosmetic industry (Miwa and Wolff, 1962; Gentry and Miller, 1965; Higgins et al., 1971; Purdy and Craig, 1987), have been interested in the oil because of its unique composition.

Historically, meadowfoam yield has been measured on a unit area basis, averaging plant responses within a given area (Higgins et al., 1971; Calhoun and Crane 1978; Jolliff et al., 1981). The yield per area evaluation method often masks size and performance variation of individual plants (Ambrose and Hedley, 1984), which may contribute disproportionately to total yield (Obeid et al., 1967). Identification and selection of superior plant types within solid stands is critical for crop improvement.

Evaluation of widely spaced meadowfoam plants for correlation to solid stand performance also has its limitations (Krebs and Jain, 1985). Spaced plants are often much larger than plants in solid stand, with pronounced phenotypic differences. Spaced-plant evaluations may also be confounded by genotype x environment interactions, generating high yield within a "good" environment while accentuating sub-par performance under a "poor" environment (Hayward and Vivero, 1984)

Solid stand yield performance of many crops is dependent upon the ability of the individual plants within the solid stand to successfully compete. Plant competition studies have been used for evaluating single-plant performance within solid stands (Obeid et al., 1967; Edmeades and Daynard 1979; Ambrose and Hedley, 1984). The only meadowfoam competition study reported in the literature is a seeding rate study evaluated on a unit area basis by Calhoun and Crane (1978). No single-plant evaluation or inference from solid stand has been reported.

Individual plant assessment is complicated by the growth habit of solid stand meadowfoam plants. Meadowfoam is a short, herbaceous plant that produces multiple stems on which numerous flowers develop acropetally. As plant stems and peduncles elongate within a solid stand, they become entwined both with themselves and adjacent plants. In solid stands this growth habit complicates yield component analysis, because separation of individual plants is difficult and time consuming.

No techniques have been reported that facilitate harvest of individual meadowfoam plants grown in solid stand. The objective of this study was to evaluate a single-plant spacing field plot design for emulation of solid stand morphological and reproductive plant responses.

MATERIALS AND METHODS

A field study was conducted in 1987 at Oregon State University Hyslop Crop Science Field Laboratory near Corvallis, OR. to evaluate a single-plant spacing field plot design. On 6 Oct. 1986, 336 kg ha^{-1} of fertilizer (16-20-0) was applied (pre-plant) to a Woodburn silt loam (fine-silty, mixed, mesic Aquultic Agrixerolls) soil. Propachlor (2-chloro-N-isopropyl-acetanilide), a pre-emergence herbicide, was applied at 2.2 kg ha^{-1} active ingredient, 16 Oct. 1986. Plant emergence took place approximately 22 Oct. 1986. On 27 Feb. 1987 a spring N application of 40 kg ha^{-1} (40-0-0-6) was applied. Honey bees were used for pollination. Bee densities approaching twice the recommended number per hectare (Karow et al., 1986) were used to reduce the potential for pollination deficiencies. The bloom period began 4 May and ended 19 May 1987.

Emulation of solid stand plant responses was evaluated using two field plot plant spacing treatments. Experimental units for both treatment plots were planted on an 18 cm row spacing at a 22 kg ha^{-1} seeding rate on 9 Oct. 1986. Four replications of each treatment were assigned to two of twelve rows (Appendix Fig. I.1), with each treatment plot 7.5 m long. Treatment one was the conventional solid stand which was allowed to grow as planted. Treatment two, the single plant spacing field plot design, allowed plants to grow in solid stand until approximately five days pre-bloom (30 April 1987), then 12 in-row plants per plot were spaced

62.5 cm apart and staked for support. Spacing of individual plants was accomplished by careful hand thinning of adjacent in-row plants. A completely randomized design (CRD) was used. Two plants were randomly sampled per plot, resulting in a nested CRD.

A third treatment was included in this study to simulate yield component responses of plants of the cv. Mermaid to our spaced-plant breeding nursery configuration. Poor plant stands in some plots prevented design randomization of all three treatments (Appendix Fig. I.1), disqualifying direct statistical comparisons between treatment three and the other two treatments.

The spaced-plant nursery treatment was planted 10 Oct. 1986 adjacent to the twelve 18 cm spaced rows (Fig. 1). Two rows were planted 91 cm apart and contained four plots, 7.5 m long with an initial 15 cm in-row distance between plants. Twelve plants within each of the four 7.5 m long plots were thinned in-row to 60 cm between plants 45 days pre-bloom (21 March 1987). Two plants were sampled per plot.

On 11 and 12 June 1987 individual plants were harvested. Randomly selected plants from each of the three treatments were cut at soil level and bagged for drying. The two solid stand plants per plot from 18 cm row spacings were isolated from adjacent plants prior to bagging. Plants were allowed to dry until 1 July 1987. Individual plants from both row spacings were then dismantled and

stems per plant, flowers per stem, flowers per plant, and seeds per flower data were recorded.

Analysis of variance was used to assess solid stand and spaced treatment significance using the replication x treatment error term. Sub-sample significance was also tested. The spaced-plant nursery treatment was sampled for mean and standard error (SE) analysis. Yield component production averages, generated from the spaced-plant nursery configuration, were included to illustrate plant response to increased row spacing.

RESULTS AND DISCUSSION

Spacing of individual plants from the 18 cm row spacing showed no significant differences ($P<0.05$) in morphological and reproductive development compared to solid stand plants grown on the same row spacing (Appendix Table I.1). The average seed set per flower was significantly ($P<0.05$) different among sub-sampled plants. The variance in seeds per flower was also great, with a mean standard error (SE) of 21%. Historically the variability in seeds per flower has been high in L. alba (Brown et al., 1979; Alba, 1986), and is supported by these findings. Stems per plant, flowers per plant, and flowers per stem resulted in 7%, 12%, and 9% SE, respectively. Plant sub-samples for these three yield components were not significantly ($P<0.05$) different. The coefficient of variation (CV) for each yield component model was relatively high (Appendix Table I.1), although consistent with yield component analysis from plants of the meadowfoam cv. Mermaid (Norberg, 1989; Fiez, 1989). The 17% CV for flowers per stem was an exception, showing relatively good stability across all plants sampled.

Mean and SE analysis on the 91 cm row spacing showed a great amount of variability between plants sampled, with flowers per stem again showing the greatest stability (Appendix Table I.1). While no direct statistical comparison to solid stand plants can be made, plant response under a less competitive row and plant spacing demonstrates the

value of the single plant spacing for emulation of solid stand plant responses. By increasing row spacing 500%, plants produced approximately 900% more stems and flowers per plant than in solid stands, while seeds per flower and flowers per stem increased only 1.8% and 11%, respectively.

Stem and flower differences per plant have been positively correlated to yield in plants of the meadowfoam cv. Mermaid (Pearson, 1983 and McGahuey 1986), although not found exclusively throughout L. alba (Krebs and Jain, 1985). If a technique is to successfully emulate solid stand plant responses, stem and flower production between solid stand and treatment plants must be duplicated. Yearly environmental influences would be expected to alter the genotypic yield component responses from plants grown under either row spacing presented in this study (Hayward and Vivero, 1984). But, yearly yield component production should not vary significantly between plants grown at similar plant spacings up to five days prior to bloom, especially under the short cultivated meadowfoam plant bloom periods (Pearson, 1983; McGahuey, 1986).

Results of this investigation support all four yield component similarities between spaced and solid stand plants grown on 18 cm row spacings. This spacing technique successfully emulates plants of the meadowfoam cv. Mermaid grown in solid stand.

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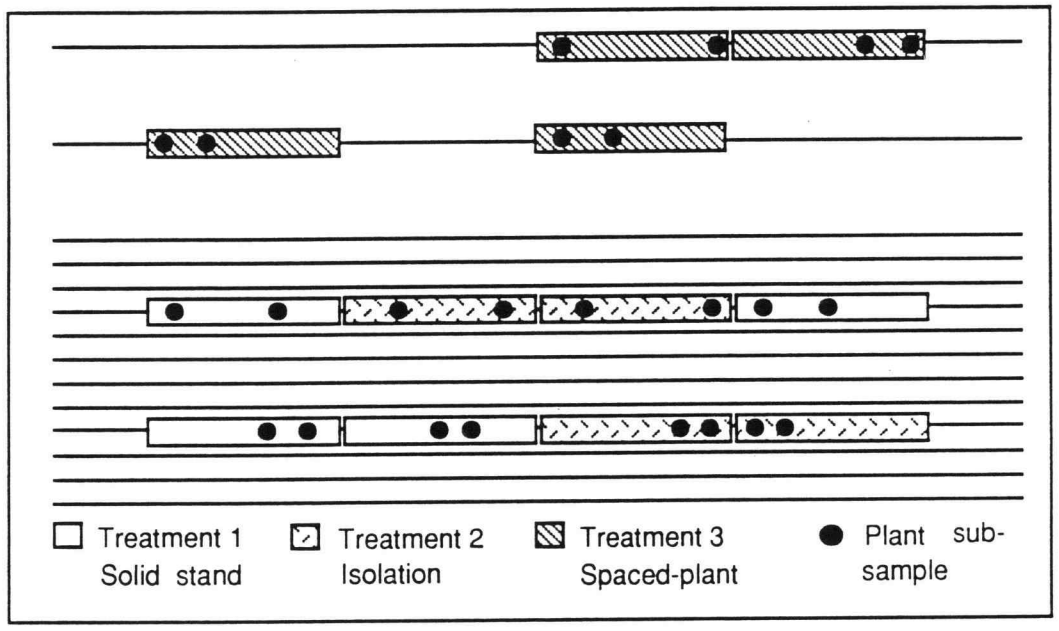
Appendix Table I.1. Means for vegetative and reproductive component production under spaced and solid stand treatments on an 18 cm row spacing, and from a spaced-plant nursery configuration treatment with a 91 cm row spacing.

Component Production	18 cm Row Spacing				91 cm Row Spacing	
	Spaced	Solid stand	SE[a]	CV[b]	Spaced nursery	SE
	-------	no.[c]	------	%	----	no.[c] ----
Stems per plant	17.4	16.5	(0.4)	33	145.6	(21.9)
Flowers per plant	100.5	98.6	(4.2)	34	861.8	(147.1)
Flowers per stem	5.8	5.7	(0.2)	17	5.8	(0.1)
Seeds per flower	2.0	1.8	(0.1)	29	2.0	(0.2)

[a]SE = standard error
[b]CV = coefficient of variation
[c]No.= means averaged from 8 plants



Appendix Figure I.1 Plot layout and sub-sample location within plots. Treatments 1 and 2 were planted on an 18 cm row spacing and treatment 3 was planted on a 91 cm row spacing.

APPENDIX II

TABLES OF MEANS

HONEY BEE: FLOWER VISITS PER MINUTE AND CONSECUTIVE VISITS OVER TIME

Appendix II consists of means for honey bee flower visits per minute and consecutive visits over time during the 1986 and 1987 meadowfoam bloom periods.

Appendix Table II.1. Means of honey bee flower visits per minute taken during the 1986 and 1987 meadowfoam bloom periods.

1986				1987			
Date	V/M[a]	n[b]	SE[c]	Date	V/M	n	SE
5-18	12.6	10	1.4	5-6	14.3	30	0.9
5-19	12.9	10	0.9	5-7	11.2	50	0.4
5-22	12.5	30	0.6	5-8	12.3	50	0.6
5-23	13.1	10	1.8	5-9	12.1	90	0.3
5-24	13.8	30	0.7	5-10	12.5	80	0.4
5-25	12.6	20	0.8	5-11	12.1	80	0.4
5-26	14.5	30	0.8	5-13	15.9	20	1.1
5-27	13.8	30	0.9	5-14	14.0	1	---
5-28	14.5	30	0.7	5-16	13.6	35	0.9
5-29	13.5	30	0.6	5-18	10.9	10	0.5
Grand Mean	13.4				12.9		

[a]V/M = average honey bee flower visits per minute.
[b]n = sample size
[c]SE = standard error

Appendix Table II.2. Number of consecutive flower visits, per pollen foraging honey bee, during the 1987 meadowfoam bloom period.

Date	Time	Visits	Minutes
5-11	1410	71	7.34
	1420	60	4.54
	1440	150	13.03
	1610	180	14.53
5-13	1445	40	3.49
	1505	181	16.04
	1640	319	25.10
5-14	1520	124	7.52
	1620	134	14.26
5-16	1050	84	5.23
	1210	38	3.55

APPENDIX III

TABLES OF MEANS

POLLEN COUNTS: FLOWERS, BEE BODIES, AND BEE LEGS (CORBICULA)

Appendix III consists of raw data and means for pollen counts taken from flowers and bees during the 1986 and 1987 meadowfoam bloom periods.

Appendix Table III.1. Raw data and means of flower pollen counts taken during the 1986 and 1987 meadowfoam bloom periods. Actual raw data and column mean values are X 10E4.

Pollen Grains Per Flower						
1986	1987					
5-29	5-8	5-9	5-11	5-13	5-14	5-16
52.6	73.2	65.4	69.4	36.2	54.0	30.4
52.4	56.8	60.8	79.8	49.4	41.0	42.8
68.0	48.6	94.0	63.8	61.6	62.8	42.4
78.6	62.2	46.4	65.2	17.4	57.2	39.6
68.4	40.8	79.8	62.4	39.6	50.2	45.4
74.0	44.4	105.4	58.4	16.2	54.4	59.6
64.4	48.2	67.0	57.2	29.0	64.2	65.0
88.0	64.4	83.2	50.4	59.4	47.0	40.2
54.4	50.0	74.0	67.6	49.2	43.6	70.8
81.6	79.2	75.6	81.4	30.4	45.4	56.0
56.2	65.4	66.4	66.6	26.8	45.0	48.6
87.4	69.0	55.4	71.6	66.8	45.6	41.6
59.8	73.8	57.0	50.0	25.8	34.4	41.6
68.0	54.4	61.0	55.6	39.1	46.4	26.8
56.0	61.8	74.6	53.0	(16.2)	42.2	35.0
76.6	65.4	62.8	40.0		25.4	47.0
38.3	62.8	55.6	44.6		44.2	63.8
59.4	51.6	27.2	39.0		59.2	47.6
46.8	56.4	41.8	76.2		47.2	29.2
55.6	62.6	63.2	46.0		60.8	30.2
54.4	80.0	68.4	42.0		41.2	31.0
51.2	65.6	68.6	49.6		29.8	20.8
64.8	49.6	73.6	55.8		21.8	25.8
56.6	44.2	78.8	95.8		44.2	32.8
59.8	49.4	68.8	59.6		47.0	36.6
66.6	59.2	67.0	60.0		46.2	42.0
57.6	s =(10.4)	(16.5)	(13.8)		(10.5)	(12.8)
58.4						
83.0						
75.0						
63.8						
s =(12.1)						

Grand Means

1986	1987
638,000	522,500

Appendix Table III.2. Raw data and means of foraging honey bee body pollen counts taken during the 1986 and 1987 meadowfoam bloom periods. Actual raw data and column mean values are X 10E4.

Pollen Grains Per Bee Body						
1986	1987					
5-29	5-8	5-9	5-11	5-13	5-14	5-16
16.1	14.0	15.3	9.9	11.8	9.7	13.4
9.3	23.3	14.4	9.4	12.1	15.9	10.9
12.7	25.7	29.9	9.5	32.7	11.0	10.2
13.4	21.1	16.9	20.4	21.7	16.1	8.8
21.6	28.8	17.2	7.7	10.3	4.8	12.5
7.6	29.5	25.8	6.3	13.8	14.9	12.2
13.8	19.8	11.8	13.2	18.3	11.8	9.3
12.3	29.6	15.7	8.4	20.6	12.7	8.2
29.2	34.3	13.6	15.9	23.7	11.1	10.2
9.2	26.2	19.3	10.1	12.4	6.7	8.4
14.2	25.2	18.0	11.1	17.7	11.5	10.4
4.8	s =(5.6)	(5.4)	(4.0)	(6.7)	(3.5)	(1.7)
5.8						
3.3						
11.2						
9.7						
11.6						
13.6						
5.6						
24.7						
19.8						
11.3						
19.8						
4.6						
9.2						
8.8						
8.5						
7.1						
10.0						
9.8						
12.0						
s =(6.0)						

Grand Means

1986	1987
119,500	156,500

Appendix Table III.3. Raw data and means of honey bee corbicula pollen counts taken from samples collected during the 1986 and 1987 meadowfoam bloom periods. Actual raw data and column mean values are X 10E4.

Pollen Grains Per Corbicula						
1986	1987					
5-29	5-8	5-9	5-11	5-13	5-14	5-16
66.0	183.2	215.2	204.0	365.6	124.0	224.4
89.4	118.4	90.4	200.4	240.8	114.4	225.6
258.0	187.2	134.4	192.8	162.8	112.8	200.0
124.8	191.6	134.4	137.2	156.8	116.8	83.2
175.6	142.4	74.0	195.2	247.6	132.8	122.4
51.8	204.0	138.0	153.2	158.8	98.4	128.8
53.8	130.8	159.6	96.4	153.6	122.8	112.4
262.0	169.2	87.6	210.8	178.8	95.2	81.6
217.0	63.2	104.4	155.6	117.2	115.2	116.4
156.0	125.6	200.0	99.2	202.0	36.4	201.2
290.0	155.2	120.8	127.2	289.2	141.6	66.0
83.0	145.2	110.4	110.8	174.4	200.8	116.0
108.2	204.4	160.8	147.2	177.6	87.6	181.2
98.6	52.4	64.4	128.0	184.0	170.4	159.6
87.0	106.4	130.8	166.8	211.2	123.2	195.6
90.6	121.6	60.4	90.8	106.4	128.0	190.0
87.8	155.6	154.8	100.0	55.2	134.0	203.2
77.2	165.2	111.2	92.8	106.0	157.6	182.0
101.8	121.2	63.2	57.2	138.8	150.4	118.0
123.0	172.0	178.4	78.8	122.4	164.0	192.4
112.2	145.7	124.7	137.2	177.5	126.3	155.0
62.4	s =(40.9)	(43.6)	(45.4)	(68.3)	(33.8)	(49.2)
174.0						
105.0						
191.0						
322.0						
224.0						
189.0						
274.0						
115.0						
145.7						
s =(76.1)						

Grand Means

1986	1987
1,457,000	1,444,000

APPENDIX IV

ANOVA TABLES

Appendix IV consists of ANOVA tables. These analysis were used in the writing of Chapters I through V and Appendix I.

Appendix Table IV.1. Pollen age response to seed set under 3^0C, 18^0C, and 37^0C storage temperature regimes. Data are combined from two replicated experiments at each temperature regime.

Source	df	Mean Squares	Sign.
		3C	
Runs (R)	1	2.72	
Reps (w/in R)	10	2.09	
TRT (T)	5	0.86	NS[a]
(R)x(T)	5	0.39	NS
Pooled Error	50	0.80	
		18C	
Runs (R)	1	1.13	
Reps (w/in R)	10	1.84	
TRT (T)	5	0.85	NS
(R)x(T)	5	1.23	NS
Pooled Error	50	0.96	
		37C	
Runs (R)	1	0.13	
Reps (w/in R)	10	0.53	
TRT (T)	5	1.19	NS
(R)x(T)	5	0.59	NS
Pooled Error	50	1.15	

[a]NS = non significant (P>0.05).

Appendix Table IV.2. Stigma age seed production from 0, 24, 48, 72, 96, and 120 hours postanthesis; and seed production from 24, 48, and 72 hours postanthesis. Data are combined from three replicated experiments.

Source	df	Mean Squares	Sign.
Run (R)	2	2.36	
Reps (w/in R)	9	0.62	
TRT (T)	5	22.65	**
(R)x(T)	10	0.40	NS[a]
Pooled Error	45	0.82	
Ages 24, 48, and 72 Hours Postanthesis Only			
Regression	2	5.13	**
Residual	6	0.82	

** significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix Table IV.3. Stylar commonality seed set from 1, 3, and 5 stigmas pollinated per flower. Data are combined from three replicated experiments.

Source	df	Mean Squares	Sign.
Run (R)	2	1.03	
Reps (w/in R)	9	3.31	
TRT (T)	2	0.86	NS[a]
(R)x(T)	4	1.57	NS
Pooled Error	18	1.26	

[a]NS = non significant (P>0.05).

Appendix Table IV.4. Honey bee flower visitation effects from 1, 6, and 11 visits on seeds per flower. Data are transformed $[(X + 0.5)^{1/2}]$ and combined from 1986 and 1987 observations.

Source	df	Mean Squares	Sign.
Yr (Y)	1	0.98	
TRT (T)	2	1.30	
linear	(1)	2.58	**
(Y)x(T)	2	0.13	NS[a]
Error	59	0.27	

** significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix Table IV.5. Pollen deposition rates from 1 and 6 honey bee visits sampled on three days during the 1988 and 1989 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1988		
Days (D)	2	208.00	
TRT (T)	1	7,084.00	**
(D)x(T)	2	437.40	
Error	30	154.10	
	1989		
Days (D)	2	968.70	
TRT (T)	1	5,329.00	**
(D)x(T)	2	157.80	
Error	30	30.22	

** Significant at P<0.01.

Appendix Table IV.6. Seed production from hand pollination, applying 5, 15, and 25 pollen grains per five stigmatal papillae (FSP) per flower. Experiments were replicated three times in a greenhouse between 1988 and 1989.

Source	df	Mean Squares	Sign.
Runs (R)	2	0.39	
Reps (w/in R)	15	2.17	
TRT (T)	2	13.72	**
linear	(1)	26.69	**
(R)x(T)	4	2.46	NS[a]
Pooled Error	30	1.05	

** Significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix Table IV.7. Environmental monitoring of: flower phenology vs daily average temperature; and photon flux density inside vs outside treatment cages.

Flower Phenology vs Daily Avg. Temp.

Source	df	Mean Squares	Sign.
	1986		
Regression	1	0.003	NS[a]
Residual	15	28.30	
	1987		
Regression	1	327,200,000.00	**
Residual	15	4,051,000.00	

Photon Flux Density Inside vs Outside Cages

Source	df	Mean Squares	Sign.
	1986		
Regression	1	4,439,000.00	**
Residual	43	4,692.00	
	1987		
Regression	1	10,030,000.00	**
Residual	133	6,013.0	

** Significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix Table IV.8. Individual plant seed set per flower and flower position yield effects (bottom, middle, and top locations per stem) under five caged pollinator treatments from combined data obtained during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
Yr	1	44.12	**
TRT	4	1.15	NS[a]
POS	2	11.54	**
YrxTRT	4	1.41	NS
YrxPOS	2	0.09	NS
TRTxPOS	8	0.59	NS
YrxTRTxPOS	8	0.29	NS
Error	330	0.68	
SUBS(YrxTRTx POS)	90	0.67	NS
Error	240	0.69	

** Significant ant P<0.01.
[a]NS = non significant (P>0.05).

Appendix Table IV.9. Solid stand yield (kg ha^{-1}) from caged pollinator treatments, and caged + non-caged pollinator treatments during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
1986 Caged Pollinators			
TRT	4	14,410.00	NS[a]
Error	10	10,260.00	
1986 Caged + Non-Caged Pollinators			
TRT	5	163,600.00	**
Error	12	17,050.00	
1987 Caged Pollinators			
TRT	4	112,000.00	*
Error	10	20,590.00	
1987 Caged + Non-Caged Pollinators			
TRT	5	137,100.00	**
Error	12	17,420.00	
1987 Caged Osmia-Only			
Regression	1	75,331.22	**
Residual	7	22,013.33	
1986 and 1987 Caged + Non-Caged Pollinators			
Yr	1	981,700.00	**
TRT	5	264,800.00	**
YrxTRT	5	35,940.00	NS
Error	24	17,233.33	

*, ** Significant at $P<0.05$ and $P<0.01$, respectively.
[a]NS = non significant ($P>0.05$).

Appendix IV.10. Seed weight produced from caged and non-caged pollinator treatments during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	5	0.25	NS[a]
Error	12	0.13	
	1987		
TRT	5	0.66	**
Error	12	0.09	
	1987 Caged Osmia-Only		
Regression	1	0.43	NS
Residual	7	0.14	
	1986 and 1987		
Yr	1	87.42	**
TRT	5	0.48	NS
YrxTRT	5	0.43	NS
Error	24	0.11	

** Significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix IV.11. Seed oil produced from caged pollinator treatments combined from the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
Yr	1	7.20	NS[a]
TRT	4	6.17	NS
YrxTRT	4	4.28	NS
Error	20	3.25	

[a]NS = non significant (P>0.05).

Appendix IV.12 Foraging-bee density (bees m^{-2}) within caged and non-caged pollinator treatments during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	5	216.50	**
Error	12	4.03	
	1987		
TRT	5	35.14	**
Error	12	2.34	

** Significant at P<0.01.

Appendix IV.13. Yield in relation to bees m^{-2} within caged pollinator treatments during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
		1986	
Regression	2	35,724.90	**
Error	12	7,397.56	
		1987	
Regression	2	228,461.02	**
Error	12	16,433.51	

** Significant at P<0.01.

Appendix IV.14. Yield-per-bee within caged pollinator treatments during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
Trt	4	6,181.00	**
Error	10	797.10	
	Caged Osmia-Only		
Regression	2	6,994.21	**
Error	6	1,328.53	
	1987		
TRT	4	3,245.00	**
Error	10	98.86	
	Caged Osmia-Only		
Regression	2	2,139.80	**
Error	6	157.21	

** Significant at $P<0.01$.

Appendix IV.15. O. l. propinqua survival at three density levels in caged meadowfoam plots during the 1986 and 1987 bloom periods.

Source	df	Mean Squares	Sign.
		1986	
TRT	2	2,238.00	**
linear	(1)	2,917.00	**
quadratic	(1)	1,559.00	NS[a]
Error	6	322.70	
		1987	
TRT	2	1,744.00	**
linear	(1)	2,953.00	**
quadratic	(1)	534.60	*
Error	6	86.33	
		1986 and 1987	
Yr	1	84.07	NS
TRT	2	4,052.00	**
YrxTRT	2	36.72	NS
Error	12	188.30	

*, ** Significant at P<0.05 and P<0.01, respectively.
[a]NS = non significant (P>0.05).

Appendix IV.16. Nest production from three O. l. propinqua densities within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	2	1,087.00	**
linear	(1)	2,128.00	**
Error	6	27.78	
	1987		
TRT	2	193.00	NS[a]
linear	(1)	384.00	NS
Error	6	109.00	

** Significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix IV.17. Male production from three O. l. propingua densities within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	2	6,245.00	**
linear	(1)	12,240.00	**
Error	6	175.10	
	1987		
TRT	2	3,801.00	*
linear	(1)	6,734.00	**
Error	6	694.20	

*, ** Significant at P<0.05 and P<0.01, respectively.

Appendix IV.18. Total cell production from three O. l. propinqua densities within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	2	7,760.00	**
linear	(1)	15,500.00	**
Error	6	383.40	
	1987		
TRT	2	4,696.00	*
linear	(1)	8,288.00	**
Error	6	1,651.00	

*, ** Significant at P<0.05 and P<0.01, respectively.

Appendix IV.19. Female production from three O. l. propinqua densities within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
TRT	2	137.40	*
linear	(1)	2.67	NS[a]
quadratic	(1)	272.20	**
Error	6	42.89	
	1987		
TRT	2	2.11	NS
Error	6	110.80	

*, ** Significant at P<0.05 and P<0.01, respectively.
[a]NS = non significant (P>0.05).

Appendix IV.20. Female production per bee under three O. l. propingua density treatments within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
	1986		
Regression	2	3.98	**
Residual	6	0.30	
	1987		
Regression	2	1.96	**
Residual	6	0.07	

** Significant at P<0.01.

Appendix IV.21. Male:female production from three O. l. propinqua densities within cages during the 1986 and 1987 meadowfoam bloom periods.

Source	df	Mean Squares	Sign.
Yr	1	1.70	NS[a]
TRT	2	19.84	**
linear	(1)	32.41	**
quadratic	(1)	7.27	**
YrxTRT	2	0.91	NS
Error	12	0.55	

** Significant at P<0.01.
[a]NS = non significant (P>0.05).

Appendix IV.22. Stem production (per plant) comparison between isolated and solid stand plants grown on an 18 cm row spacing.

Source	df	Mean Squares	Sign.
TRT	1	9,506.25	NS[d]
Rep(TRT)	6	3,837.79	NS
Error	8	1,387.25	

[d]NS = non significant (P>0.05).

Appendix IV.23. Flower production (per plant) comparison between isolated and solid stand plants grown on an 18 cm row spacing.

Source	df	Mean Squares	Sign.
TRT	1	382,851.56	NS[a]
Rep(TRT)	6	170,524.65	NS
Error	8	54,386.44	

[a]NS = non significant (P>0.05).

Appendix IV.24. Flower production (per stem) comparison between isolated and solid stand plants grown on an 18 cm row spacing.

Source	df	Mean Squares	Sign.
TRT	1	0.01	NS[a]
Rep(TRT)	6	0.56	NS
Error	8	0.75	

[a]NS = non significant (P>0.05).

Appendix IV.25. Seed production (per plant) comparison between isolated and solid stand plants grown on an 18 cm row spacing.

Source	df	Mean Squares	Sign.
TRT	1	0.73	NS[a]
Rep(TRT)	6	0.23	NS
Error	8	0.33	

[a]NS = non significant (P>0.05).