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Pollination of caged hybrid seed red rape with *Osmia cornuta* and *Apis mellifera* (Hymenoptera Megachilidae and Apidae)

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Abstract

Osmia cornuta was tested as a pollinator of caged hybrid red rape for seed production in comparison to *A. mellifera*. Nine screen cages were set up, each to enclose ten plants of red rape. Four cages were provided with 40 *O. cornuta* (10 females and 30 males). In each of other four cages a small nucleus (ca. 400 workers) of *A. mellifera* was introduced. To estimate seed yields without pollinators, no bees were introduced in the ninth cage. After bloom, seed yield, seed germination percentages after 5 and 7 days, and seed weight were measured. *Osmia cornuta* readily visited red rape flowers, and pollinated them successfully. The *O. cornuta* cages produced a 1.5-fold increase in seed yield compared to *A. mellifera* cages. Seed quality was similar in *O. cornuta* and *A. mellifera* cages. The "no pollinator" cage produced few seeds, and of low quality. Red rape seems to be a suitable plant on which to rear *O. cornuta*, since the *O. cornuta* population released was increased by 1.2-fold.

Key words: *Osmia cornuta*, *Apis mellifera*, pollination, seed yield, *Brassica rapa.*

Introduction

In the production of hybrid varieties, the standards for purity are extremely high. To prevent contamination by external pollen sources, plants of the parent lines are often isolated in cages (Brantjes, 2000). Small colonies of honey bees, *Apis mellifera* L., in three-frame-boxes (nuclei) containing 5000-6000 bees, are usually used for pollination. However, confinement in cages for more than one month causes severe bee worker losses. Confined honey bee workers often show aggressive behaviour, difficulting work within the seed production cages. Furthermore, in Northern Italy, red rape, *Brassica rapa* L. (Brassicaceae), blooms in early spring and honey bee pollination may be curtailed due to adverse climatic conditions. For these reasons, a mason bee, *Osmia cornuta* (Latreille), was tested as a red rape pollinator in cages (Ladurner *et al*., 2000). Because of its foraging behaviour and capacity to fly under less favourable weather conditions, *O. cornuta* has already been shown to be a more efficient pollinator than *A. mellifera* on almond and apple (Bosch and Blas, 1994; Vicens and Bosch, 2000a, 2000b).

Preliminary investigations, carried out in 1999, indicated that *O. cornuta* may be an interesting alternative pollinator for red rape (Ladurner *et al*., 2000). Pollen gathering activity was performed by *O. cornuta* females even under damp and cool weather conditions. As few as nine *O. cornuta* females provided better pollination than a nucleus colony of honey bees (Ladurner *et al*., 2000). Moreover, as with other solitary bee species tried in cages, *O. cornuta* require less maintenance than honey bees (Cox *et al*., 1996; Wilson *et al*., 1999).

In 2000, we pollinated caged red rape with *O. cornuta* and *A. mellifera*. The following questions were addressed:

1. How does *O. cornuta* perform on red rape, compared to *A. mellifera*, in terms of seed yield, seed weight and seed germination percentage?

2. Is it possible to maintain or increase *Osmia cornuta* populations on red rape in a confined environment?

Materials and methods

Nine anti-aphid screen cages (1.5 x 4.0 x 1.7 m) were set up, each to enclose ten plants of red rape. At the beginning of bloom (April 20), 40 *O. cornuta* (30 males and 10 females), still inside their cocoons, were released in each of four cages. Two of these cages contained one grooved-board nesting unit (Pinzauti, 1991; Krunic *et al*., 1995). Each nesting unit had 81 nesting holes. Holes were 15 cm long and 1 cm in diameter. The remaining two *O. cornuta* cages each contained two reed section bundles (ca. 50 reeds/bundle) of *Arundo donax* L. (Poaceae). Reed sections were 15-20 cm long and 0.8-1 cm in inside diameter. Other four cages were each provided with one small nucleus of honey bees (approximately 400 workers per nucleus). To estimate seed yields without pollinators, no bees were introduced in the ninth cage.

After bloom (May 24), the following parameters were measured in each cage:

1. Seed yield (weight in grams of total seed production): seeds were stored after harvest in a dry and ventilated room for exsiccation. To separate seed from straw, small electric threshers were used. The threshed seed was then transferred into a seed separator (WESTRUPP SLAGELSE, Denmark) that sorted seed by size. The resulting product was run by two seed cleaners (DELTA MACHINE, Benelux – Type BELAR and ROBER SAATREINIGER DHG, Minden-Westf., Germany), which further divided seeds, according to their specific weight. The resulting clean seed was weighed.

2. Seed germination percentage after 5 days: germination was tested by following the international rules for seed assays stipulated by the Association Internationale d'Essays de Semences (1999). For each cage, four sam

ples of 100 seeds were placed on filter paper in Petri dishes and kept at 20 °C for 5 days.

3. Seed germination percentage after 7 days: as above with seeds kept for 7 days.

4. Seed weight (weight in grams of one sample of 1000 seeds per replicate): seed weight was established by using the method, dictated by the Italian Ministerial Decree, issued on January 7, 1987 (Gazzetta Ufficiale della Repubblica Italiana, 1987). For each cage, four samples of 100 seeds were weighed. After having applied variation coefficients and standard formulas reported in Gazzetta Ufficiale della Repubblica Italiana (1987), the resulting mean weights were multiplied by 10, thus obtaining the weight of 1000 seeds per replicate.

A one-way ANOVA was used to compare seed yields, seed germination percentages after 5 and 7 days, and seed weight in *O. cornuta* versus *A. mellifera* cages.

At the end of bloom, *O. cornuta* nesting materials were taken to the laboratory and the total number of brood cells was determined.

Results

Seed yields, germination percentages and seed weights in cages with *O. cornuta*, *A. mellifera* and no pollinators, respectively, are reported in table 1.

Climatic conditions during bloom were favourable to both *A. mellifera* and *O. cornuta* activity (Free, 1993; Vicens and Bosch, 2000b): daily wind speed was low (range: 0.6 - 2.3 m/s); daily solar radiation ranged between 102 and 512 cal/cm², relative humidity between 51 and 91%, and mean daily temperature between 15.2°C and 21.3°C. Nevertheless, cages pollinated by *O. cornuta* yielded 32.5% more seed than those pollinated by *A. mellifera* (ANOVA: $F_{(1, 6)} = 8.5217$, $p = 0.0267$) (table 1). For all the other parameters analysed, i.e. percentage of germination after 5 and 7 days and seed weight, no significant differences emerged: seed quality on plants pollinated by mason bees was comparable to that on plants pollinated by honey bees (germination percentage after 5 days: $F_{(1, 6)} = 4.9167$, $p = 0.0689$; germination percentage after 7 days: $F_{(1, 6)} = 0.5493$, $p = 0.5370$; seed weight: $F_{(1, 6)} = 0.5494$, $p = 0.4866$) (table 1).

The contribution of self- and/or wind pollination (cage with no pollinators) was very low (table 1). The control cage also yielded the lowest germination percentages and seed weight of all nine cages.

The *O. cornuta* nesting materials recovered contained 195 cells with live adults and 84 cells with dead immatures. Most mortality occurred in pre-defecating larval stages.

Discussion

Our results show that *O. cornuta* is a more efficient pollinator than *A. mellifera* on red rape in confined environment. The significant increase in seed yield in *O. cornuta* cages is not only due to *O. cornuta*'s greater tolerance to inclement weather (Vicens and Bosch, 2000b), as might be concluded from previous results (Ladurner *et al*., 2000), but also to *O. cornuta*'s foraging behaviour. As on almond and apple (Bosch and Blas, 1994, Vicens and Bosch, 2000a), *O. cornuta* females systematically landed on the reproductive organs of red rape flowers. They introduced their head through the stamens to reach the nectaries with simultaneous scrabbling movements of the legs against the anthers to detach the pollen. Honey bees, conversely, visited red rape flowers mostly for nectar, reaching the nectaries from the side by introducing their proboscis through the basis of the petals, without contacting the stigmas or getting dusted with pollen (side-working behaviour) (Robinson, 1979). Furthermore, *O. cornuta* females were observed visiting fewer flowers per red rape plant, and changed rows more often than honey bees, thus enhancing cross-pollination. Similar results have been observed on fruit trees (Bosch and Blas, 1994, Vicens and Bosch, 2000a).

In previous cage studies (Ladurner *et al*., 2000), *O. cornuta* population increase on red rape was 2.9-fold. In 2000, all adult progeny obtained survived the winter. Thus, a 1.2-fold population increase was obtained, despite the high larval progeny mortality recorded (30.1%). A possible reason for this high larval mortality are pesticides, sprayed against aphids and fungi on May 4. All caged plants were treated with Klartan (taufluvalinate) and copper. Some insecticides are known to be toxic to immatures of other Megachilid bee species (Guirguis and Brindley, 1974; Johansen *et al.*, 1983; Torchio, 1983; Tasei and Carré, 1985). In a previous study, in which no pesticides were applied, immature mortality was only 0.02%.

Our results show that one *O. cornuta* female and 3 males per plant are enough to achieve significant red rape seed yield increase compared to honey bee pollination, while maintaining high seed quality standards.

Table 1. Seed yield, percentage of germination after 5 and 7 days, and seed weight in red rape cages with various pollinators*.

Pollination treatment	Seed yield (g) (mean±sd)	Germination after 5 days (%) (mean±sd)	Germination after 7 days (%) (mean±sd)	Seed weight (g) (mean±sd)
A. mellifera	290.0±87.6 (a)	96.3±1.0 (a)	97.5±0.6 (a)	4.3±0.1 (a)
O. cornuta	430.0±39.2 (b)	94.8±1.0 (a)	97.8±0.5 (a)	4.3±0.1 (a)
No insects	30.0	78.0	81.0	3.9

* Different letters indicate significant statistical differences (one-way ANOVA: $p<0.05$).

The required rate of *O. cornuta* per red rape plant to achieve adequate pollination might actually be lower. Red rape in our study produced 2,000-3,000 flowers per plant. It has been estimated that *O. cornuta* females visit 9,000-23,000 flowers during their nesting period in almond and apple orchards (Bosch, 1994; Vicens and Bosch, 2000a).

Further studies should determine the number of *O. cornuta* visits per flower necessary to achieve maximum seed set on red rape. Red rape appears to be a suitable plant on which to rear *O. cornuta*. Red rape produces abundant pollen and nectar, and blooms for longer (over one month in our study) than most commercial fruit tree orchards. *Osmia cornuta* larvae of this and our previous study (Ladurner *et al*., 2000) developed into adults on provisions containing exclusively red rape pollen. By being able to maintain or increase *O. cornuta* populations on caged red rape, seed producers could minimize the risk of sting incidents, frequent when honey bees are used in confinement, as *O. cornuta* is not aggressive to humans.

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