



The intertwined effects of natural vegetation, local flower community, and pollinator diversity on the production of almond trees

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ARTICLE INFO

Keywords:

Agro-systems
Crop pollination
Ecosystem service
Fruit set
Honeybees
Landscape characteristics
Local flower community
Pollinator-species richness
Percentage of natural habitat
Prunus dulcis

ABSTRACT

Wild pollinators are a valuable natural resource for crops, as they often increase their production and quality. For this reason, there is currently a great interest in the development of management and conservation tools that help to maintain a wide variety of wild pollinators in agro-systems. To achieve this, it becomes a priority to study the diversity of wild pollinators in relevant crops as well as the local and landscape characteristics that benefit them. The almond tree (*Prunus dulcis*) is a crop of high economic interest, with a large dependence on pollinators due to the self-incompatibility of most of its varieties and, thus, it is very vulnerable to pollinator losses. By using field data and habitat characterization of 18 almond fields in Mallorca Island (Spain), we assessed how the abundance and diversity of pollinators varied with local and landscape characteristics (at 1 and 2 km buffer zones) of the fields, and how those affected almond production (fruit set). Almond trees were mostly pollinated by honeybees, but they were also visited by a large number of wild pollinators. The percentage of natural area in the 2 km buffer zones increased both pollinator-species richness and honeybee visits. At the field level, the flower community in the ground positively influenced almond production, both directly and indirectly by increasing the diversity of wild pollinators. Pollinator-species diversity directly increased fruit production but was negatively affected by honeybee abundance, which suggests that a high density of honeybees might result in negative effects on almond production through competition with wild pollinators. Management strategies to improve almond production might include favoring wild pollinators through the maintenance of natural habitats surrounding crop fields, and preserving the flowering herb community that occurs spontaneously in the ground-cover of almond fields in Mediterranean areas.

1. Introduction

Pollination is an essential ecosystem service (Klein et al., 2007) currently threatened by the increasing disappearance of both wild pollinators and honeybees (Biesmeijer et al., 2006; Winfree et al., 2007; Burkle et al., 2013). This pollinator loss is causing great concerns in agriculture production, mainly because two-thirds of the plant species cultivated by humans are pollinated by insects, and 35% of world food production depends on animal pollination (Gallai et al., 2009). Although the use of honeybee hives for crop pollination is common practice (Winfree et al., 2007), it is known that wild pollinators are an important and valuable natural resource for crops, as they usually increase their production (Klein et al., 2007; Garibaldi et al., 2013) and quality (Brittain et al., 2014); and therefore also the net profits earned by farmers (Morandin and Winston, 2005). In addition, wild bees are often more effective crop pollinators than honeybees (Sadeh et al., 2007; Garibaldi et al., 2013; Zhang et al., 2015). For this reason, there is currently a great interest in the development of management and

conservation tools that help to maintain a wide variety of wild pollinators in agro-systems (Aizen and Harder, 2009; Potts et al., 2010; Garibaldi et al., 2013). However, the actual role that wild pollinators play in crops of some important productive areas of the world is still unknown. Further research is also needed on the relationship between managed honeybees and wild pollinators. While high local densities of managed honeybees could lead to competition between them and wild pollinators (Goulson, 2003, 2004), a more balanced relationship between wild and managed bees might be beneficial, given that an increase in wild pollinators might enhance honeybee movement (Carvalho et al., 2011) and positively influence honeybee effectiveness per visit (Brittain et al., 2013a).

Almond trees (*Prunus dulcis* Mill., F. Rosaceae) are very appreciated world-wide for their nuts and flowers. Currently, Spain has become the second largest almond producer of the world after United States of America, producing the 11.9% (2.31 millions of tons) of world production (FAO, 2010), and the Balearic Islands is one of the main regions of almond production in this country (FAO, 2010). Despite this, there is

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scarce information either on their pollinator assemblage or the impact that wild pollinators have on almond production in these islands. This crop is very vulnerable to eventual pollinator losses (Garibaldi et al., 2013), as most varieties are self-incompatible (Cortal et al., 2002), and it flowers early in the season, when wild bee populations are just starting to emerge from diapause. For this reason, many almond fields depend on rented honeybee hives for their pollination (Vargas and Romero, 1987; Delaplane and Mayer, 2000), but fruit production in commercial almond fields is still usually limited, with fruit sets of ca. 30% (Gary et al., 1976; Tombesi et al., 2016). To solve this problem, several studies have focused on testing the efficiency of other managed bees, such as *Osmia cornuta* (Bosch and Blas, 1994; Márquez et al., 1994), *Osmia lignaria* (Artz et al., 2013), and *Bombus terrestris* (Dag et al., 2006) on the pollination of almond flowers. Some works have also described the diversity of almond trees' wild pollinators (Ortiz-Sánchez and Tianut, 1993; Mandelik and Roll, 2009; Klein et al., 2012; Brittain et al., 2013b), and the local or/and landscape characteristics that benefit them in some areas where it is cultivated, such as Israel (Mandelik and Roll, 2009), California (Klein et al., 2012), Australia (Saunders et al., 2013), and Egypt (Norfolk et al., 2016). These studies show that the increase in local flowering resources and/or the percentage of surrounding natural habitat favor both wild pollinators (Mandelik and Roll, 2009; Klein et al., 2012; Saunders et al., 2013; Norfolk et al., 2016) and fruit production (Klein et al., 2012; Norfolk et al., 2016). However, the relative importance of local versus landscape context on almond production is still little explored, and nothing is known about almond production in productive insular habitats that are characterized by depauperated pollinator communities (e.g. Barrett, 1996; Anderson et al., 2001).

In this paper, we study the role of wild pollinators on almond tree pollination in Mallorca (Balearic Islands, Spain), by analyzing flower visitation frequencies (as a proxy for pollination) and fruit production. Specifically, we determined the abundance and identity of flower visitors and assessed their effect on fruit set in 18 crop fields across Mallorca Island during two years. Furthermore, we evaluated the effect of local (flower abundance and diversity in the groundcover of almond fields) and landscape (percentage of natural habitat surrounding the fields, and field size) characteristics on pollinator visits and fruit production on this crop. Particularly, we asked: (1) Does the frequency of wild pollinator visits to almond trees increase with natural resources for pollinators (natural habitat, floral resources) at the local and landscape levels?; (2) Does the diversity (richness and Simpson's diversity index) of pollinators increase with the availability of natural resources at the local and landscape levels?; (3) Does the presence of managed honeybees affect the abundance and diversity of wild pollinators visiting almond trees? (4) Does almond production increase with the visits of wild pollinators?; and, (5) Does almond production increase with the natural resources for them at the local and landscape levels? We expected almond pollination and fruit production to be positively related to the amount of natural habitat for pollinators both at the local and landscape level. Moreover, we predicted that a large abundance of honeybees might have a negative influence on wild-pollinator visitation.

2. Material and methods

2.1. Study species and sites

The deciduous tree *Prunus dulcis* (Mill.) belongs to the Rosaceae family and is one of the main fruit crops that requires pollination by insects (Garibaldi et al., 2013). Its flowers are open, whitish, and 3 to 5 cm in diameter; they normally appear solitary or in groups of 2 or 4. The fruits, almonds, take 5 to 6 months to mature and are used as food for their nutritional properties (fatty acids, vitamin E, fiber, riboflavin and minerals) and also to make oil and emollients.

We selected 18 almond fields across Mallorca Island, Balearic Islands, Spain (Fig. 1). Sites were distributed across all the area where

almond trees are cultivated in Mallorca, with a minimum distance of 850 m between sites. Sites were chosen to differ in size, visual differences in surrounding landscape, and presence of managed honeybee hives (Table A1, Appendix). Almond trees within the fields were of similar size and were planted in rows with each tree being separated by its closer neighbor by 5–10 m. Each study field included several varieties of almond trees, sometimes unknown by the farmers. The mix of varieties also differs among fields, with those flowering earlier having traditional Mallorcan varieties whereas those flowering later having foreign varieties (see Table A1 for varieties in each study site; all the varieties included in this study were auto-incompatible).

2.2. Landscape and local characteristics

To determine landscape characteristics, we calculated the size of each study almond field by means of orto-photos (year 2006). In addition, we estimated the percentages of both natural area (different types of forest and shrublands) and cultivated (mainly trees with dry fruit – such as almond and carob trees –, but also some olive groves and citrus) area in the 1 km- and 2 km- radius buffer zone surrounding the sampling area in each field, as different pollinators may respond to the landscape at different scales (e.g., Steffan-Dewenter et al., 2002; Kennedy et al., 2013). For this, we used ArcMap 10.3 (Environmental Systems Research Institute, Redlands, CA) and maps of land use coverage (Instituto Geográfico Nacional, 2010).

To estimate floral abundance and diversity in the groundcover of almond fields, we used 20 sampling squares (50 × 50 cm), randomly placed across the field each day the pollinator censuses were conducted (see below). Within each square we recorded the abundance and identity of the different plant species with open flowers. With these data we calculated: 1) flower abundance, as the total number of open flowers found in the sampled area; 2) flower richness, as the total number of flowering species found in the sampled area; and 3) flower diversity, as the inverse Simpson (1949) calculated as: $1/\sum_i p_i^2$, and where p_i was the proportional number of flowers of the species i , and S was the flowering species richness. This index varies from 0 (lowest diversity) to a maximum of $[1 - 1/S]$.

2.3. Pollinator visitation

We observed flower visits to almond tree flowers during two flowering seasons (2015 and 2016), from late January to late March, covering the whole flowering period of this species in Mallorca. To observe flower visitors, we haphazardly selected and marked 20 individual trees located approximately in the middle of each of the 18 orchards. Each sampling day, we performed focal observations of flower-visitors to each of the marked individual trees, using 5 min observation periods (5 min × 20 trees = 100 min observation per site and sampling day each year). Censuses were conducted always between 09:30 and 18.00 h, on days with weather conditions that allowed pollinator activity. The observation protocol was optimized during preliminary observations. Each study year, every site was visited between 3 and 5 days during its flowering peak (always including morning, midday and afternoon), except for 'Sa Marineta' which was observed only one day the first year, and 'Son Blai' which could only be studied the first year (4 sampling days) because almond trees were cut the second sampling year. Excluding from the analyses 'Sa Marineta' the first year, did not change the results (results not shown) and, therefore, we kept this sampling day in the analyses. Table A1 (Appendix) shows the number of sampling days in each site each year.

Observations of flower visitors were conducted on selected branches or areas of the canopy, where we counted the number of flowers (those branches contained a mean of 414 ± 12.76 flowers). During each census period, we recorded the number and identity of flower visitors and the number of flowers contacted by them. A pollinator visit was considered only when the visitor's body contacted the flower

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