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Bumble bee pollen use and preference across spatial scales in human-altered landscapes

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Abstract. 1. While the discussion of native pollinator decline has grown dramatically worldwide, information on how native pollinators utilise floral resources in natural and human-altered landscapes remains relatively limited. Specifically, little is known about the collection of pollen, an essential component of larval and adult bee food, and whether pollen collection patterns change across habitats, spatial scales, and/or native and non-native floral resource distributions.

2. In this study, the pollen collection patterns and preferences of the yellow-faced bumble bee, *Bombus vosnesenskii*, were examined across a gradient of natural and human-altered sites.

3. Three hypotheses were investigated: (i) landscape-scale (2 km) but not local-scale (250 m) floral and habitat composition factors are most predictive of bee-collected pollen composition; (ii) collection of native pollen is greater in sites with greater proportions of natural habitat at landscape scales; and (iii) bees exhibit greater 'preference' for native versus non-native pollen.

4. *Bombus vosnesenskii* collected more species of pollen when landscape-scale riparian forest cover was low but local- and landscape-scale floral species richness was high. Also, *B. vosnesenskii* collected pollen from a wide range of plant families and did not exhibit a significant preference for native versus non-native species.

5. Finally, preference analysis indicated that the only significant preference exhibited by *B. vosnesenskii* during the study period was for *Heteromeles arbutifolia*, a native shrub. Overall, results from this study reveal the importance of species-rich floral patches (native and non-native) for pollinator provisioning across natural and human-altered landscapes.

Key words. agriculture, ecosystem service, foraging behaviour, pollinator, resource dynamics.

Introduction

Pollination is a critical part of wild and cultivated plant reproduction and an invaluable ecosystem service in both natural and human-altered landscapes. Animal pollination, which is provided by both wild and managed pollinators, is valued at approximately US$200 billion worldwide (Gallai *et al.*, 2009) and contributes to the stability of food prices, food security, food diversity, and human nutrition (Steffan-Dewenter *et al.*, 2005; Eilers *et al.*, 2011). The urgent need to conserve native (wild) pollinators that provide crop pollination services has been exacerbated by recent catastrophic declines in managed honey bee populations (e.g. Neumann & Carreck, 2010), which illustrate that sole reliance on honey bees for crop pollination is risky. Further, recent work has shown that crop yields respond positively and far more consistently to wild pollinator richness and visitation than to honey bee visitation across many crop types, biomes, and continents (Garibaldi *et al.*, 2013). Although native pollinators are critical ecosystem service providers, their populations currently face many threats, most often associated with habitat destruction, resulting in the loss of nesting and foraging resources (Kearns *et al.*, 1998; Biesmeijer *et al.*, 2006; Winfree *et al.*, 2009).

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A number of studies have investigated the local- and landscape-scale factors influencing pollinator visitation of flowers in human-altered landscapes; however, much less is known about pollen collection patterns. For bees, one of the most important pollinator groups globally (e.g. Roubik, 1995), successfully obtaining pollen and nectar is an essential step in sustaining foraging adults and provisioning the brood (Michener, 2000). Because pollen is the primary food source for bee larvae, and can vary considerably in nutritive quality between species (Roulston & Cane, 2000; Tasei & Aupinel, 2008), it is hypothesised that bees may be more selective of pollen than of nectar resources (Wcislo & Cane, 1996; Cane & Sipes, 2006). Further, flower-visitation patterns may not serve as a proxy for pollen usage, as flower visitors often focus on nectar collection and thus neither actively collect nor transfer pollen (Herrera, 1987; Alarcon, 2010). Past studies have revealed that, for various insect species, habitat composition and floral abundance influence floral visitation patterns (e.g. Steffan-Dewenter *et al.*, 2002; Kremen *et al.*, 2004; Jha & Vandermeer, 2010), foraging distance and scale (Westphal *et al.*, 2006; Osborne *et al.*, 2008; Jha & Kremen, 2013a), gene flow patterns (Jha & Kremen, 2013b), and colony growth patterns (Williams & Kremen, 2007; Williams *et al.*, 2012), yet it remains unknown how these factors, across spatial scales, influence pollen collection.

In addition, although non-native plant species are often abundant in human-altered landscapes (e.g. Williams *et al.*, 2011; Morandin & Kremen, 2012), little is known about the collection of native versus non-native pollen by insect visitors, especially in relation to local- and landscape-scale floral and habitat composition. Some previous studies suggest that non-native plant species can disrupt native plant–pollinator interactions (Aizen *et al.*, 2008; Bartomeus *et al.*, 2008) or provide less attractive floral resources than native plants (e.g. Kearns *et al.*, 1998), while others suggest that non-native plants provide important floral resources (Ghazoul, 2002; Bjerknes *et al.*, 2007) and could even fill phenological gaps for native pollinators (Stout & Morales, 2009). Interestingly, 'preference' for a resource, defined as the statistical description of resource use relative to resource availability (Beyer *et al.*, 2010), has only recently been explored for pollen and flower visitors (Harmon-Threatt, 2011; Williams *et al.*, 2011; Davis *et al.*, 2012; Morandin & Kremen, 2012), despite its considerable relevance to pollinator restoration.

In this study, the pollen usage and preference patterns of the western yellow-faced bumble bee, *Bombus vosnesenskii*, were examined across a variety of natural and human-altered landscapes and varying floral resource distributions. Bumble bees are critical native pollinators for both wild and cultivated plants (reviewed in Kremen *et al.*, 2002; Goulson *et al.*, 2008a) and visit a broad range of flowering plant species, and thus are essential for the maintenance of many plant communities (Memmott *et al.*, 2004). Furthermore, bumble bees are ideal species for the study of pollen resource usage given their large colony sizes and high resource demands. Because of their sensitivity to floral resource levels (e.g. Carvell *et al.*, 2004; Pywell *et al.*, 2006; Williams *et al.*, 2012) and habitat composition (e.g. Knight *et al.*, 2009; Goulson *et al.*, 2010; Jha & Kremen, 2013a), among other factors, bumble bees are often the first bees to be extirpated in highly intensified human-altered landscapes (Larsen *et al.*, 2005), and a number of species have also exhibited declines across a wide range of geographic regions (Goulson *et al.*, 2008b; Cameron *et al.*, 2011); thus a better understanding of bumble bee pollen preference, even across single foraging trips, could be important for informing effective conservation practices.

In this study, permutation tree modelling and preference analysis were used to investigate bumble bee pollen collection patterns and preference for native versus non-native plant species across land-use types and floral resource distributions. Past research has demonstrated that bumble bee foraging distances can frequently exceed 500 m (e.g. Darvill *et al.*, 2004; Knight *et al.*, 2005; Jha & Kremen, 2013a). Studies have also documented higher bumble bee abundances in areas with greater natural habitat cover (Steffan-Dewenter *et al.*, 2002; Greenleaf & Kremen, 2006), greater general flower cover (Williams *et al.*, 2012), and greater native flower cover (Carvell *et al.*, 2006; Pywell *et al.*, 2006). Therefore it was predicted that pollen collected by worker bumble bees per foraging trip will respond to habitat and daily resource levels at large (i.e. landscape) spatial scales, and that pollen loads will be comprised of predominantly native plant species. Specifically, three major hypotheses were tested: (i) landscape-scale (2 km) floral resource and habitat composition are more predictive of daily bumble bee worker pollen load composition than local-scale (250 m) floral resource and habitat composition; (ii) foraging bumble bee workers collect more native pollen species per day in areas dominated by natural habitat; and (iii) bumble bees exhibit greater 'preference' for native versus non-native pollen.

Materials and methods

Study sites and land classification

The research area extends from Sonoma to Sacramento County in northern California and includes a wide range of human-altered and natural habitat (e.g. Kremen *et al.*, 2002, 2004). Research was conducted from 1 June to 14 July of 2009 in eight independent study regions which varied in the proportion of human-altered habitat (cultivated crops, orchard, bare ground, and impervious areas) and natural habitat (grassland, oak woodland–chaparral, and riparian forest) within a 2 km radius (5–95% natural cover, mean = 51 ± 39%). Thus all cultivated study regions included at least 5% natural habitat, usually comprised of hedgerows or buffer strips inhabited by native plants. All study regions were separated by more than 3 km (Jha & Kremen, 2013a) (Fig. 1a).

In this study, 'local-scale habitat composition' is defined as land use within a 250 m radius and 'landscape-scale habitat composition' is defined as land use within a 2 km radius, the smallest and largest scales that have been consistently utilised for understanding land use in previous bumble bee studies (e.g. Steffan-Dewenter *et al.*, 2002; Goulson *et al.*, 2010). Using data from the National Agriculture Imagery Program (2009 NAIP, http://www.fsa.usda.gov/), land cover data was digitised and hand-classified within a 250 m and 2 km radius of each site

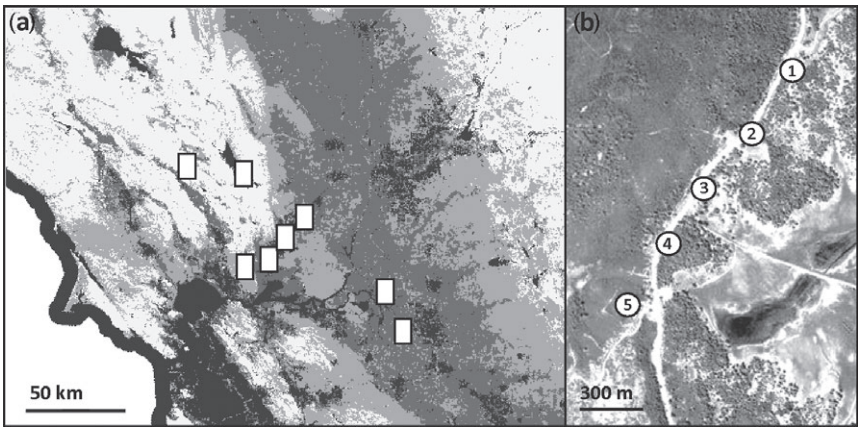

Fig. 1. (a) Map of the research area: white squares, study regions; lighter colours represent natural landscapes (grassland, oak woodland–chaparral, and riparian forest); darker colours represent water bodies and human-altered landscapes (cultivated crops and urban areas). (b) A close-up of the western-most study region showing the five sampling sites separated by 300 m.

using ArcGIS®. Habitat classifications were confirmed with ground-truthing surveys at every site. Habitat was classified into seven categories: crop, grassland, orchard, bare ground, paved ground, oak woodland–chaparral, and riparian forest; however only the three natural habitat cover types, grassland, oak woodland–chaparral, and riparian forest, were considered for the analyses because these were the only land types with flowering plants.

Study species and pollen measurements

Pollen loads of *Bombus vosnesenskii* Radoszkowski, the yellow-faced bumble bee, were examined across the eight study regions. This is the most common bumble bee in California and one of the most important native pollinators on the west coast (Kremen *et al.*, 2002). Like many other bumble bees, *B. vosnesenskii* is polylectic and nests primarily underground in large colonies of often 200–300 workers (Shelly *et al.*, 1991). To examine the pollen collection patterns of *B. vosnesenskii*, an average of 20.8 (± 2.18 SE) bees were net-collected in a single day across five sites located 300 m apart on a 1.2 km transect within each of the eight study regions (40 sample locations) (Fig. 1b). However, in three of these sites no workers were carrying pollen loads, leaving a total of 37 sample sites for the study. Individual bees were temporarily immobilised and pollen loads were collected using sterilised forceps to scoop pollen from the right corbicula. The pollen load was then immediately stored in 1 ml of 95% ethanol. Given that individual bumble bees often specialise on only a few plants (Kleijn & Raemakers, 2008), pollen loads were pooled from five individuals per site to estimate *B. vosnesenskii* pollen usage across multiple individuals. Five individuals were selected because this was the minimum number of individuals that carried pollen loads out of all individuals collected at any given site. Specifically, when more than five individuals carried pollen loads, five *B. vosnesenskii* individuals per site were randomly selected. Then 10 μl from each sample was mixed for a 50 μl total pooled sample per site. The 50 μl pooled sample was then vortexed for 1 min, and 10 μl was applied to a microscope slide containing 20 μl of a glycerol and fuschin stain (Kearns & Inouye, 1993). The stain and pollen microscope slide was allowed to rest for 24 h (to allow the pollen grains to rehydrate). The first 300 pollen grains were identified to species based on a reference collection (see details below); however, *Aster* species could not be reliably distinguished from one another, thus these species were pooled for analyses. As per other bumble bee diet breadth studies (e.g. Goulson & Darvill, 2004), Simpson's diversity index was calculated for each sample. To build the pollen reference collection, pollens from all flowering plant species encountered in the vegetation surveys (described below) were sampled by collecting and storing five anthers from each species in 95% ethanol. Following the staining protocol described above, the samples were mounted and used as a reference for pollen identification. More than 91% of pollen grains collected by bumble bees in this study were identified to species.

Vegetation surveys and pollen reference collection

While satellite imagery could be used for local- and landscape-scale habitat classification and quantification, vegetation surveys were required for floral species identification and quantification. Vegetation surveys were conducted on the same day as the pollen collection, and comprised of 12 randomly placed 1 × 1 m quadrats that were surveyed at every site along each transect (five patches per transect) and between sites, 300 m perpendicular to the transect (four patches per transect) for a total of 108 1 × 1 m quadrats in nine patches surveyed on each transect (i.e. per study region). Flowering species richness, number of flowering inflorescences, and petal area per inflorescence were recorded for each quadrat. Floral cover was then calculated based on inflorescence count multiplied by the petal area per inflorescence. Inflorescence counts were found to be good predictors of overall pollen availability, as documented for similar plant species within the same bioregion (Harmon-Threatt, 2011). In this study, 'local-scale

floral' resource factors are those that were measured within each site (the mean, CV, and total species richness of the 12 vegetation quadrats *within* a patch), and 'landscape-scale floral' resource factors are those measured across all sites in the study region (the mean, CV, and total species richness *among* all 108 quadrats in all nine patches).

Data analysis

Role of local and landscape variables. Non-parametric regression trees were used to determine the most important factors predictive of the four pollen response variables: (i) pollen load species richness; (ii) pollen load diversity (Simpson's diversity index); (iii) pollen load native species richness; and (iv) proportion of native pollen. Regression trees are commonly used to examine patterns in ecological data, and are especially useful for developing habitat models because they do not assume linearities in response variables and allow factors to interact in a hierarchical fashion. In this study, non-parametric regression trees are especially useful, given their ability to resolve critical values (i.e. thresholds) for relevant explanatory variables, which can then be useful for developing guidelines for specific conservation management practices (reviewed in De'ath & Fabricius, 2000). Specifically, vegetation restoration protocols can be designed by directly using the thresholds derived for each of the explanatory vegetation variables. These thresholds represent a valuable new contribution to pollinator restoration ecology, given that most current pollinator restoration practices are based on visitation rather than pollen usage data (Frankie *et al.*, 2005; Menz *et al.*, 2011).

Non-parametric regression trees were built utilising the 'party' package in R (R Development Core Team, 2013; Strobl *et al.*, 2009). The non-parametric regression trees estimate a regression relationship by utilising a binary recursive data-partitioning algorithm. The trees function by first testing whether input variables are independent of one another and independent of the response variable. If this hypothesis is rejected, then the input variable with the greatest association to the response variable is selected, as measured by a P-value for the test of the partial null hypothesis of the single input variable and the response variable. The data are then split by the response variable into two sections (nodes) and the search for the input variable with the next greatest association to the response variable is repeated. This process continues until the criterion (0.95) does not exceed the minimum established in the analysis.

Because habitat composition was correlated across local and landscape scales, these two scales were examined in two separate sets of non-parametric regression trees. In the first set of trees, all floral resource factors were included: (1) local (within patch) floral cover, (2) local coefficient of variation (CV) of floral cover, (3) local floral species richness, (4) local CV of floral species richness, (5) landscape (among patches) floral cover, (6) landscape CV of floral cover, and (7) landscape total floral species richness. Then, for this first set of trees, only local-scale habitat composition factors were included: (8) the proportion of grassland within 250 m, (9) the proportion of oak woodland–chaparral within 250 m, and (10) the proportion of riparian forest in a 250 m radius of each site. In the second set of trees, all the floral resource factors (1–7) were included, along with the landscape-scale habitat composition factors: (8) the proportion of grassland within 2 km, (9) the proportion of oak woodland–chaparral within 2 km, and (10) the proportion of riparian forest within 2 km of each site. Site locations per study region exhibited substantial variation in floral and habitat composition at the 250 m scale, therefore the five sample sites per study region were treated as independent for the local-scale analyses. However, given the proximity of sampling sites within a study region, which could be interpreted as 'repeated sampling' and potentially bias tree-building (De'ath & Fabricius, 2000), the package 'coin' (Hothorn *et al.*, 2008) was used to examine the significance of local-scale explanatory variables, given stratification within a study region (these P-values are reported as 'study region-stratified'). Native plant cover and species richness was not analysed because native plant cover and species richness were significantly correlated with total plant cover and total plant species richness (Pearson correlation coefficient, $r = 0.536$, $P = 0.0006$, and $r = 0.645$, $P < 0.0001$, respectively).

Species usage and preference. Classical compositional analysis (Aebischer *et al.*, 1993) was used to identify species selection for all species comprising more than 0.1% of the pollen load. Compositional analysis can be used to identify pollen species that are collected significantly more than expected relative to their availability (e.g. Davis *et al.*, 2012). Compositional analysis of pollen was implemented using the 'adehabitatHS' package (Calenge, 2006) developed in R. In these analyses, matrices of floral species availability (proportion of floral cover) and floral species use (proportion of pollen load) per site were first transformed using log-ratio analysis, which is one of the most appropriate transformations for proportional values (Aitchison, 1982). Second, overall pollen selection was examined by testing if pollen species use is significantly non-random relative to availability per site, using a randomisation test (10 000 repetitions) and assuming Wilks' lambda (λ). Species were also classified into non-native and native groups and overall preference for either group was tested. Finally, a preference ranking matrix was built representing the mean difference between the proportion of pollen use and proportion of floral cover for each species per site, and a randomisation test (10 000 repetitions) was used to assess the significance of preference for one species over each other species (Aebischer *et al.*, 1993; Calenge, 2006).

Results

Role of local and landscape variables

A total of 30 plant species were found flowering within the sites (13 native and 17 non-native) and a total of 22 plant species were found in the pollen loads (eight native and 14 non-native) (Fig. 2). Native species comprised 63.4% of the

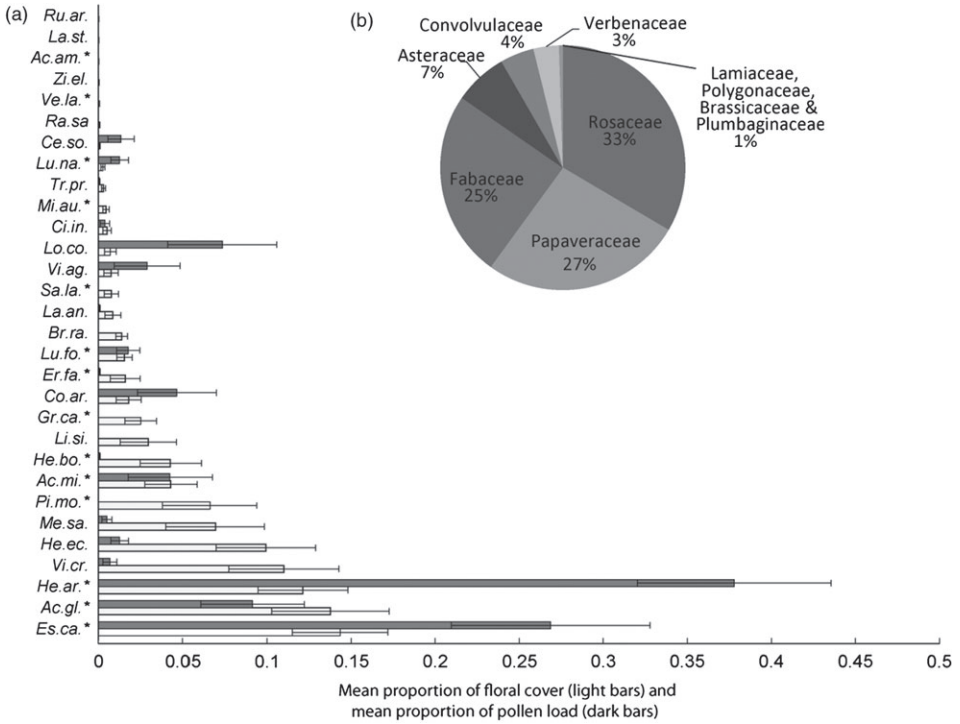

Fig. 2. (a) Mean area cover (m^{-2}) of each flowering plant species across all quadrats ($n = 08$) (light bars) and mean proportion of pollen load ($n = 37$) comprised by each flowering plant species (dark bars). Native species are marked with an asterisk; error bars represent standard error. (b) Mean proportion of pollen load comprised by each flowering plant family. Species codes: *Ac.mi., Achillea millefolium; Ac.am., Acmispon americanus; Ac.gl., Acmispon glaber; Br.ra., Brassica rapa; Ce.so., Centaurea solstitialis; Ci.in., Cichorium intybus; Co.ar., Convolvulus arvensis; Er.fa., Eriogonum fasciculatum; Es.ca., Eschscholzia californica; Gr.ca., Grindelia camporum; He.bo., Helianthus bolanderi; He.ec., Helminthotheca echioides; He.ar., Heteromeles arbutifolia; La.va., Lavandula angustifolia; La.st., Lavandula stoechas; Li.si., Limonium sinuatum; Lo.co., Lotus corniculatus; Lu.fo., Lupinus formosus; Lu.na., Lupinus nanus; Me.sa., Medicago sativa; Mi.au., Mimulus aurantiacus; Pi.mo., Pickeringia montana; Ra.sa., Raphanus sativus; Ru.ar., Rubus armeniacus; Sa.la., Salix laevigata; Tr.pr., Trifolium pretense; Ve.la., Verbena lasiostachys; Vi.cr., Vicia cracca; Vi.ag., Vitex agnus-castus; Zi.el., Zinnia elegans.*

floral cover and 81.1% of pollen loads collected by bumble bees. The largest fraction of pollen loads were comprised of three native species: *Heteromeles arbutifolia* (37.8%) (Rosaceae), *Eschscholzia californica* (26.8%) (Papaveraceae), and *Acmispon glaber* (9.12%) (Fabaceae) (Fig. 2). Of the identified pollen grains, 10 different families were represented, while the vast majority of pollen (> 85%) was collected from plant species in the Rosaceae, Fabaceae, and Papaveraceae (Fig. 2b).

Pollen load species richness and pollen load diversity were not significantly explained by any floral or habitat composition variable at either the local or landscape scale ($P > 0.15$ for all variables). The proportion of native pollens per load was significantly higher when mean local-scale floral cover was less than $0.192\,m^2$ ($P < 0.001$; study region-stratified, $P = 0.048$; Fig. 3a). The number of native species per pollen load was significantly higher when, jointly, the proportion of landscape-scale riparian forest cover was < 0.208 ($P = 0.001$), local-scale species richness was > 1 ($P = 0.012$; study region-stratified, $P = 0.049$), and landscape-scale floral species richness was > 4 ($P = 0.044$) (Fig. 3b).

Species usage and preference

The randomisation test for overall species selection was not significant ($\lambda = 0.205$, $P = 0.323$), nor was the test for native versus non-native species ($\lambda = 0.981$, $P = 0.867$). However, *B. vosnesenskii* significantly selected the native species, *H. arbutifolia*, in greater amounts relative to its abundance and significantly more than every other species except the non-native species, *Lotus corniculatus* (Table 1). Despite making up large portions of bumble bee pollen loads (Table 1, Fig. 1), two native species, *E. californica* and *A. glaber*, were not significantly selected in greater proportion than their abundances.

Discussion

This study examined the role of plant origin (native versus non-native) and multiple scales of floral resource and habitat composition on *B. vosnesenskii* pollen collection and preference per foraging trip. While no factors were found to predict total pollen richness or diversity collected per

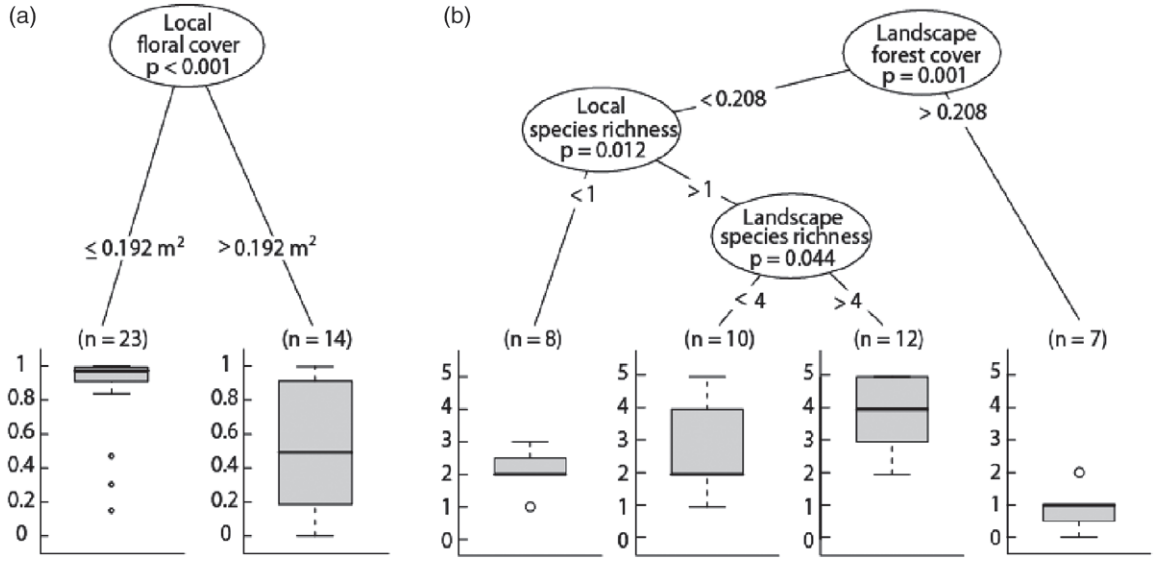

Fig. 3. Conditional inference tree for (a) proportion of native pollen; (b) number of native species in the pollen load (*y*-axes). Circles indicate explanatory variables showing the strongest association to the response variable (local-scale floral cover, and landscape-scale floral cover and local-scale floral species richness, respectively). The *P*-values listed at each node represent the test of independence between the explanatory variable and the response variable. Box plots at the terminal nodes show the distribution of (a) proportion of native pollen collected and (b) number of native species in the pollen load. Boxes represent the inner-quartile range of the data; dark horizontal lines within boxes represent the median; whiskers represent the extent of data within the 1.5× inner-quartile range. Circles above and below whiskers represent data points outside of this range. The number of sampling sites (*n*) that fall within each branch are listed above the box plots.

Table 1. Species-ranking matrix where values represent preferences for row species compared with column species.

	*He.ar.**	*Lu.fo.**	*Vi.ag.*	*Lo.co.*	*Co.ar.*	*Ce.so.*	*Es.ca.**	*Ac.mi.**	*Ac.gl.**	Pollen load	Floral availability
*He.ar.**	0	+++	+++	+	+++	+++	+++	+++	+++	1	3
*Lu.fo.**	–	0	–	+	+	+	+	+	+	7	7
Vi.ag.	–	+	0	–	+	–	+	+	+	8	5
Lo.co.	–	–	+	0	–	+	+	+	+	3	8
Co.ar.	–	–	–	+	0	–	+	+	+	6	6
Ce.so.	–	–	+	–	+	0	+	+	–	9	9
*Es.ca.**	–	–	–	–	–	–	0	+	+	2	2
*Ac.mi.**	–	–	–	–	–	–	–	0	+	5	4
*Ac.gl.**	–	–	–	–	–	+	–	–	0	4	1

Thus at the intersection of row *i* and column *j*, a positive symbol (+) indicates that species *i* is used more than species *j*, and a negative symbol (−) indicates otherwise. Significant differences are indicated by tripled signs. Pollen load indicates ranking according to abundance in pollen load; Floral availability indicates overall floral availability across sites, where 1 = highest rank. Native species are marked with an asterisk.

day, native pollen collection was predicted by both local-scale (within patch) and landscape-scale (among patches) floral cover and species richness. There was no support for the second hypothesis that greater natural habitat cover at the landscape scale would result in higher pollen richness. Natural habitat cover was a significant predictor for only one pollen response variable (number of native pollens per sample), but in the opposite direction to our prediction. There was also no support for the third hypothesis; while bumble bees collect more native than non-native pollen (absolutely and relative to abundance), this relationship was not significant. However, analyses indicate that *B. vosnesenskii* did exhibit a significant preference for the native rosaceous species *H. arbutifolia*.

Role of local and landscape variables

Regression tree analyses indicate that *B. vosnesenskii* responds to floral resource levels across both local (within patch) and landscape (among patch) scales. Past studies indicate that bumble bees can forage long distances (Darvill *et al.*, 2004; Knight *et al.*, 2005; Hagen *et al.*, 2011; Jha & Kremen, 2013a) and that their visitation is often related to resource availability at large spatial scales (750–3000 m; Steffan-Dewenter *et al.*, 2002; Westphal *et al.*, 2006, 2009). Results from this study, though gathered across a single foraging day, also support the importance of both local and landscape scales, and additionally suggest that bumble bees gather pollen from a greater number of native plant species

when both landscape and local floral species richness is higher. Bumble bees are generalists (Memmott, 1999) and often visit several floral species for pollen and nectar within a single foraging bout (Goulson *et al.*, 2008a). The act of collecting multiple pollen species could be advantageous to bumble bees because the nutrition content of pollen and nectar varies considerably between plant species (Gardener & Gillman, 2001; Kitaoka & Nieh, 2009) and because multiple pollen species are important for bee larval growth (Genissel *et al.*, 2002; Tasei & Aupinel, 2008).

Habitat composition was a significant predictor of native species pollen richness per sample, although native species richness was significantly lower in landscapes with greater riparian forest cover. One potential explanation for this pattern could be greater bumble bee specialisation (collection of fewer native plant species) in landscapes with more extensive riparian forest cover. Foraging studies using experimental arrays indicate that within a foraging bout, bumble bees will continue foraging on a single species as long as rewarding conspecific flowers are available within close distance (e.g. Chittka *et al.*, 1997), thus if greater riparian forest availability provides more consistent access to a preferred native species, a reduction in native species richness of the collected pollen may be evident. Furthermore, if riparian forest habitats provide higher resource levels than other habitat types, as documented within the study region for a different native bee species (Williams & Kremen, 2007), then *B. vosnesenskii* may forage 'optimally', by increasing diet breadth in low resource areas, and decreasing diet breadth in high resource areas (Macarthur & Pianka, 1966), such as the riparian forest habitat. Experimental work on individual pollinators has found strong support for optimal foraging (e.g. Kunin & Iwasa, 1996), especially for bumble bees (Pyke, 1978; Fontaine *et al.*, 2008), although other studies suggest that bumble bees may actually increase foraging distance in order to visit patches with high flowering plant species richness (Jha & Kremen, 2013a).

Two previous studies have found that habitat composition alone can be predictive of pollen load composition for some bee species (Goulson *et al.*, 2002; Williams & Kremen, 2007). In this study, while habitat was predictive for native pollen species richness, the dominant variables influencing pollen collection were related to floral composition within and across patches. Although pollen collection patterns were assessed across foraging trips, research conducted across multiple time periods within the same study system has found that floral resource availability, calculated over a landscape scale, was also strongly related to growth of experimental bumble bee colonies (Williams *et al.*, 2012). Overall, findings from this study support the importance of floral resource availability in determining native pollen collection patterns at local and landscape scales.

Species use and preference

The three largest portions of *B. vosnesenskii* pollen loads were comprised of the native species *H. arbutifolia*, *E. californica*, and *A. glaber* (Rosaceae, Papaveraceae, and Fabaceae, respectively). Bumble bee pollen loads were not dominated by the Fabaceae, as seen in many past studies (Goulson & Darvill, 2004; Goulson *et al.*, 2008b), but included substantial amounts of pollen from the Papaveraceae and Rosaceae (as seen in Carvell *et al.*, 2006). While this study examined pollen loads per foraging trip, unlike these past studies, findings from this study are congruent with the findings of Carvell and colleagues in indicating an important role for plants within the Fabaceae, but an equally strong role for other plant families.

Most importantly, a significant preference for the native species *H. arbutifolia* was documented in this study. *Heteromeles arbutifolia* is common across chaparral and riparian forest systems in the tall shrub communities of California (Hanes, 1974). This species provides structural diversity to landscapes often dominated by shorter-statured vegetation and thus is believed to be important for birds and small mammals in the form of nesting resources and predation protection (McMurray, 1990). Because of its critical role in bird and mammal ecology and its rapid growth and erosion control abilities, *H. arbutifolia* is often used in native plant restorations in California (Magill, 1974; Lowry, 1999; Morandin *et al.*, 2011). Findings from this study illustrate the ecological importance of *H. arbutifolia* as a major component of *B. vosnesenskii* pollen collection and thus highlight the potential overlap in species selection to restore both degraded habitats and native pollinator communities (Menz *et al.*, 2011).

Research examining historical and contemporary bumble bee pollen collection in the UK suggests that narrow diet breadth and inability to utilise non-native species may explain the rarity of some European bumble bee species (Kleijn & Raemakers, 2008). In the study system described, pollen use was examined over a much shorter time scale and for a species that does not appear to be in decline (Cameron *et al.*, 2011); here, a preference for non-native or native species was not documented. Rather, the utilisation of non-native or native plant species generally corresponded with abundance of those species in the community. Studies conducted within the same study region have similarly found that, while collection of non-native pollen by native bees is substantial, it is no greater than expected given the abundance of non-native plants in the community (Williams *et al.*, 2011). Interestingly, results from this study contrast a recent study conducted on restored native plant hedgerows in the same study region, where flower-visiting bee abundance, richness, and diversity were greater on native plants compared with exotic weedy species (Morandin & Kremen, 2012). These differences may occur because the current study focused on a single bee species rather than the entire bee community, and because pollen collection was measured rather than floral visitation, which assesses visits for both pollen and nectar.

Broader impacts

A critical step in pollinator conservation is determining what floral species are preferred by pollinators for nectar and pollen resources (Williams *et al.*, 2010; Winfree, 2010). While pollinator restoration projects, like the installation of flowering hedgerows, are gaining attention across the globe (Pywell *et al.*,

2005; Carvell *et al.*, 2006; Winfree, 2010), many restoration plant species lists are based primarily on visitation records, not on pollen and nectar usage (Frankie *et al.*, 2005; Menz *et al.*, 2011). Results from this study indicate that *B. vosnesenskii* utilise pollen from a wide range of plant families, including both native and non-native species. The results also indicate that a greater proportion of native pollen is collected when local floral cover is low, such as in agricultural landscapes with monoculture plantings. This is probably due to collection on the few native plants found within the hedgerows and buffer strips, and thus suggests that native plantings within agricultural landscapes can be important food resources for native bees (e.g. Morandin & Kremen, 2012). Additionally, results from this study suggest that maintaining high-flowering plant species richness across spatial scales may allow bumble bee colonies to collect greater numbers of pollen species.

Overall, these results are congruent with restoration practices which suggest that farmers and land managers do not need to rely solely on the availability of existing natural habitat to provide pollinator resources (e.g. Pywell *et al.*, 2006; Carvell *et al.*, 2007; Morandin & Kremen, 2013). Farmers and land managers can coordinate the creation of multiple diverse floral resource patches across their land to provide pollinators with floral resources at both local and landscape scales.

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References

Aebischer, N.J., Robertson, P.A. & Kenward, R.E. (1993) Compositional analysis of habitat use from animal radio-tracking data. *Ecology*, **74**, 1313–1325.

Aitchison, J. (1982) The statistical analysis of compositional data. *Journal of the Royal Statistical Society, Series B: Statistical Methodology*, **44**, 139–177.

Aizen, M.A., Morales, C.L. & Morales, J.M. (2008) Invasive mutualists erode native pollination webs. *PLoS Biology*, **6**, 396–403.

Alarcon, R. (2010) Congruence between visitation and pollen-transport networks in a California plant–pollinator community. *Oikos*, **119**, 35–44.

Bartomeus, I., Vila, M. & Santamaria, L. (2008) Contrasting effects of invasive plants in plant–pollinator networks. *Oecologia*, **155**, 761–770.

Beyer, H.L., Haydon, D.T., Morales, J.M., Frair, J.L., Hebblewhite, M., Mitchell, M. *et al.* (2010) The interpretation of habitat preference metrics under use-availability designs. *Philosophical Transactions of the Royal Society of London Series B: Biological Sciences*, **365**, 2245–2254.

Biesmeijer, J.C., Roberts, S.P.M., Reemer, M., Ohlemüller, R., Edwards, M., Peeters, T. *et al.* (2006) Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science*, **313**, 351–354.

Bjerknes, A.L., Totland, O., Hegland, S.J. & Nielsen, A. (2007) Do alien plant invasions really affect pollination success in native plant species? *Biological Conservation*, **138**, 1–12.

Calenge, C. (2006) The package "adehabitat" for the R software, a tool for the analysis of space and habitat use by animals. *Ecological Modelling*, **197**, 516–519.

Cameron, S.A., Lozier, J.D., Strange, J.P., Koch, J.B., Cordes, N., Solter, L.F. *et al.* (2011) Patterns of widespread decline in North American bumble bees. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 662–667.

Cane, J. & Sipes, S. (2006) Characterizing floral specialization by bees, analytical methods and a revised lexicon for oligolecty. *Plant–Pollinator Interactions, From Specialization to Generalization* (ed. by N. Waser and J. Ollerton), pp. 99–122. University of Chicago Press, Chicago, Illinois.

Carvell, C., Meek, W.R., Pywell, R.F. & Nowakowski, M. (2004) The response of foraging bumblebees to successional change in newly created arable field margins. *Biological Conservation*, **118**, 327–339.

Carvell, C., Westrich, P., Meek, W.R., Pywell, R.F. & Nowakowski, M. (2006) Assessing the value of annual and perennial forage mixtures for bumblebees by direct observation and pollen analysis. *Apidologie*, **37**, 326–340.

Carvell, C., Meek, W.R., Pywell, R.F., Goulson, D. & Nowakowski, M. (2007) Comparing the efficacy of agri-environment schemes to enhance bumble bee abundance and diversity on arable field margins. *Journal of Applied Ecology*, **44**, 29–40.

Chittka, L., Gumbert, A. & Kunze, J. (1997) Foraging dynamics of bumble bees, correlates of movements within and between plant species. *Behavioral Ecology*, **8**, 239–249.

Darvill, B., Knight, M.E. & Goulson, D. (2004) Use of genetic markers to quantify bumblebee foraging range and nest density. *Oikos*, **107**, 471–478.

Davis, E.S., Reid, N. & Paxton, R.J. (2012) Quantifying forage specialisation in polyphagic insects, the polylectic and rare solitary bee, *Colletes floralis* (Hymenoptera, Colletidae). *Insect Conservation and Diversity*, **5**, 289–297.

De'ath, G. & Fabricius, K.E. (2000) Classification and regression trees, a powerful yet simple technique for ecological data analysis. *Ecology*, **81**, 3178–3192.

Eilers, E.J., Kremen, C., Greenleaf, S.S., Garber, A.K. & Klein, A.M. (2011) Contribution of pollinator-mediated crops to nutrients in the human food supply. *PLoS ONE*, **6**, e21363.

Fontaine, C., Collin, C.L. & Dajoz, I. (2008) Generalist foraging of pollinators, diet expansion at high density. *Journal of Ecology*, **96**, 1002–1010.

Frankie, G.W., Thorp, R.W., Schindler, M., Hernandez, J., Ertter, B. & Rizzardi, M. (2005) Ecological patterns of bees and their host ornamental flowers in two northern California cities. *Journal of the Kansas Entomological Society*, **78**, 227–246.

Gallai, N., Salles, J.M., Settele, J. & Vaissiere, B.E. (2009) Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. *Ecological Economics*, **68**, 810–821.

Garibaldi, L.A., Steffan-Dewenter, I., Winfree, R., Aizen, M.A., Bommarco, R., Cunningham, S.A. *et al.* (2013) Wild pollinators enhance fruit set of crops regardless of honey bee abundance. *Science*, **339**, 1608–1611.

Gardener, M.C. & Gillman, M.R. (2001) Analyzing variability in nectar amino acids, composition is less variable than concentration. *Journal of Chemical Ecology*, **27**, 2545–2558.

Genissel, A., Aupinel, P., Bressac, C., Tasei, J.N. & Chevrier, C. (2002) Influence of pollen origin on performance of *Bombus terrestris* micro-colonies. *Entomologia Experimentalis et Applicata*, **104**, 329–336.

Ghazoul, J. (2002) Flowers at the front line of invasion? *Ecological Entomology*, **27**, 639–640.

Goulson, D. & Darvill, B. (2004) Niche overlap and diet breadth in bumblebees; are rare species more specialized in their choice of flowers? *Apidologie*, **35**, 55–63.

Goulson, D., Hughes, W.O.H., Derwent, L.C. & Stout, J.C. (2002) Colony growth of the bumblebee, *Bombus terrestris*, in improved and conventional agricultural and suburban habitats. *Oecologia*, **130**, 267–273.

Goulson, D., Lye, G.C. & Darvill, B. (2008a) Decline and conservation of bumble bees. *Annual Review of Entomology*, **53**, 191–208.

Goulson, D., Lye, G.C. & Darvill, B. (2008b) Diet breadth, coexistence and rarity in bumblebees. *Biodiversity and Conservation*, **17**, 3269–3288.

Goulson, D., Lepais, O., O'Connor, S., Osborne, J.L., Sanderson, R.A., Cussans, J. *et al.* (2010) Effects of land use at a landscape scale on bumblebee nest density and survival. *Journal of Applied Ecology*, **47**, 1207–1215.

Greenleaf, S.S. & Kremen, C. (2006) Wild bee species increase tomato production and respond differently to surrounding land use in Northern California. *Biological Conservation*, **133**, 81–87.

Hagen, M., Wikelski, M. & Kissling, W.D. (2011) Space use of bumblebees (*Bombus* spp.) revealed by radio-tracking. *PLoS ONE*, **6**, e19997.

Hanes, T. (1974) The vegetation called chaparral. *Symposium on Living with the Chaparral: Proceedings* (ed. by M. Rosenthal), pp. 1–5. The Sierra Club, San Francisco, California.

Harmon-Threatt, A. (2011) Pollen preference by native bumble bees. PhD Thesis dissertation, University of California, Berkeley, California.

Herrera, C.M. (1987) Components of pollinator quality–comparative analysis of a diverse insect assemblage. *Oikos*, **50**, 79–90.

Hothorn, T., Hornik, K., van de Wiel, M.A.V. & Zeileis, A. (2008) Implementing a class of permutation tests, the coin package. *Journal of Statistical Software*, **28**, 1–23.

Jha, S. & Kremen, C. (2013a) Bumble bee foraging in response to landscape heterogeneity. *Proceedings of the National Academy of Sciences of the United States of America*, **8**, 555–558.

Jha, S. & Kremen, C. (2013b) Urban land use limits regional bumble bee gene flow. *Molecular Ecology*, **22**, 2483–2495.

Jha, S. & Vandermeer, J.H. (2010) Impacts of coffee agroforestry management on tropical bee communities. *Biological Conservation*, **143**, 1423–1431.

Kearns, C.A. & Inouye, D.W. (1993) *Techniques for Pollination Biologists*. University Press of Colorado, Niwot, Colorado.

Kearns, C.A., Inouye, D.W. & Waser, N.M. (1998) Endangered mutualisms, the conservation of plant–pollinator interactions. *Annual Review of Ecology and Systematics*, **29**, 83–112.

Kitaoka, T.K. & Nieh, J.C. (2009) Bumble bee pollen foraging regulation, role of pollen quality, storage levels, and odor. *Behavioral Ecology and Sociobiology*, **63**, 501–510.

Kleijn, D. & Raemakers, I. (2008) A retrospective analysis of pollen host plant use by stable and declining bumble bee species. *Ecology*, **89**, 1811–1823.

Knight, M., Martin, A.P., Bishop, S., Osborne, J.L., Hale, R.J., Sanderson, R.A. *et al.* (2005) An interspecific comparison of foraging range and nest density of four bumblebee (*Bombus*) species. *Molecular Ecology*, **14**, 1811–1820.

Knight, M.E., Osborne, J.L., Sanderson, R.A., Hale, R.J., Martin, A.P. & Goulson, D. (2009) Bumblebee nest density and the scale of available forage in arable landscapes. *Insect Conservation and Diversity*, **2**, 116–124.

Kremen, C., Williams, N.M. & Thorp, R.W. (2002) Crop pollination from native bees at risk from agricultural intensification. *Proceedings of the National Academy of Sciences of the United States of America*, **99**, 16812–16816.

Kremen, C., Williams, N.M., Bugg, R.L., Fay, J.P. & Thorp, R.W. (2004) The area requirements of an ecosystem service, crop pollination by native bee communities in California. *Ecology Letters*, **7**, 1109–1119.

Kunin, W. & Iwasa, Y. (1996) Pollinator foraging strategies in mixed floral arrays, density effects and floral constancy. *Theoretical Population Biology*, **49**, 232–263.

Larsen, T.H., Williams, N. & Kremen, C. (2005) Extinction order and altered community structure rapidly disrupt ecosystem functioning. *Ecology Letters*, **8**, 538–547.

Lowry, J. (1999) Gardening with a Wild Heart, Restoring California Native Landscapes at Home. UC Press, Berkeley, California.

Macarthur, R.H. & Pianka, E.R. (1966) On optimal use of a patchy environment. *American Naturalist*, **100**, 603–609.

Magill, A. (1974) *Photinia arbutifolia* Lindl., christmasberry. *Seeds of Woody Plants in the United States* (ed. by C. Schopmeyer), Agriculture Handbook No. 450, pp. 582–583. U. S. Department of Agriculture, Forest Service, Washington, DC.

McMurray, N. (1990) *Heteromeles arbutifolia* Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fort Collins, Colorado.

Memmott, J. (1999) The structure of a plant–pollinator food web. *Ecology Letters*, **2**, 276–280.

Memmott, J., Waser, N.M. & Price, M.V. (2004) Tolerance of pollination networks to species extinctions. *Proceedings of the Royal Society of London Series B: Biological Sciences*, **271**, 2605–2611.

Menz, M.H.M., Phillips, R.D., Winfree, R., Kremen, C., Aizen, M.A., Johnson, S.D. *et al.* (2011) Reconnecting plants and pollinators, challenges in the restoration of pollination mutualisms. *Trends in Plant Science*, **16**, 4–12.

Michener, C.D. (2000) *Bees of the World*. Johns Hopkins University Press, Baltimore, Maryland.

Morandin, L. & Kremen, C. (2012) Bee preference for native versus exotic plants in restored agricultural hedgerows. *Restoration Ecology*, **21**, 27–32.

Morandin, L. & Kremen, C. (2013) Hedgerow restoration promotes pollinator populations and exports native bees to adjacent fields. *Ecological Applications*, in press.

Morandin, L., Long, R., Pease, C. & Kremen, C. (2011) Hedgerows enhance beneficial insects on farms in California's Central Valley. *California Agriculture*, **65**, 197–201.

Neumann, P. & Carreck, N. (2010) Honey bee colony losses. *Journal of Apicultural Research*, **49**, 1–6.

Osborne, J.L., Martin, A.P., Carreck, N.L., Swain, J.L., Knight, M.N., Goulson, D. *et al.* (2008) Bumblebee flight distances in relation to the forage landscape. *Journal of Animal Ecology*, **77**, 406–415.

Pyke, G.H. (1978) Optimal foraging–movement patterns of bumblebees between inflorescences. *Theoretical Population Biology*, **13**, 72–98.

Pywell, R.F., Warman, E.A., Carvell, C., Sparks, T.H., Dicks, L.V., Bennett, D. *et al.* (2005) Providing foraging resources for bumblebees in intensively farmed landscapes. *Biological Conservation*, **121**, 479–494.

Pywell, R.F., Warman, E.A., Hulmes, L., Hulmes, S., Nuttall, P., Sparks, T.H. *et al.* (2006) Effectiveness of new agri-environment schemes in providing foraging resources for bumblebees in intensively farmed landscapes. *Biological Conservation*, **129**, 192–206.

R Development Core Team (2013) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria.

Roubik, D. (1995) *Pollination of Cultivated Plants in the Tropics*. Food and Agriculture Organization of the United Nations, Rome, Italy.

Roulston, T.H. & Cane, J.H. (2000) Pollen nutritional content and digestibility for animals. *Plant Systematics and Evolution*, **222**, 187–209.

Shelly, T.E., Buchmann, S.L., Villalobos, E.M. & Orourke, M.K. (1991) Colony ergonomics for a desert-dwelling bumble bee species (Hymenoptera, Apidae). *Ecological Entomology*, **16**, 361–370.

Steffan-Dewenter, I., Munzenberg, U., Burger, C., Thies, C. & Tscharntke, T. (2002) Scale-dependent effects of landscape context on three pollinator guilds. *Ecology*, **83**, 1421–1432.

Steffan-Dewenter, I., Potts, S.G. & Packer, L. (2005) Pollinator diversity and crop pollination services are at risk. *Trends in Ecology & Evolution*, **20**, 651–652.

Stout, J.C. & Morales, C.L. (2009) Ecological impacts of invasive alien species on bees. *Apidologie*, **40**, 388–409.

Strobl, C., Hothorn, T. & Zeileis, A. (2009) Party on!. *The R Journal*, **1**, 14–17.

Tasei, J.N. & Aupinel, P. (2008) Nutritive value of 15 single pollens and pollen mixes tested on larvae produced by bumblebee workers (*Bombus terrestris*, Hymenoptera, Apidae). *Apidologie*, **39**, 397–409.

Wcislo, W. & Cane, J. (1996) Floral resource utilization by solitary bees and exploitation of their stored foods by natural enemies. *Annual Review of Entomology*, **41**, 257–286.

Westphal, C., Steffan-Dewenter, I. & Tscharntke, T. (2006) Foraging trip duration of bumblebees in relation to landscape-wide resource availability. *Ecological Entomology*, **31**, 389–394.

Westphal, C., Steffan-Dewenter, I. & Tscharntke, T. (2009) Mass flowering oilseed rape improves early colony growth but not sexual reproduction of bumblebees. *Journal of Applied Ecology*, **46**, 187–193.

Williams, N.M. & Kremen, C. (2007) Resource distributions among habitats determine solitary bee offspring production in a mosaic landscape. *Ecological Applications*, **17**, 910–921.

Williams, N.M., Crone, E.E., Roulston, T.H., Minckley, R.L., Packer, L. & Potts, S.G. (2010) Ecological and life-history traits predict bee species responses to environmental disturbances. *Biological Conservation*, **143**, 2280–2291.

Williams, N.M., Cariveau, D., Winfree, R. & Kremen, C. (2011) Bees in disturbed habitats use, but do not prefer, alien plants. *Basic and Applied Ecology*, **12**, 332–341.

Williams, N., Regetz, J. & Kremen, C. (2012) Landscape-scale resources promote colony growth but not reproductive performance of bumble bees. *Ecology*, **93**, 1049–1058.

Winfree, R. (2010) The conservation and restoration of wild bees. *Year in Ecology and Conservation Biology 2010*, Vol. 1195, pp. 169–197. Blackwell Publishing, Oxford, U.K..

Winfree, R., Aguilar, R., Vazquez, D.P., LeBuhn, G. & Aizen, M.A. (2009) A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology*, **90**, 2068–2076.

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