Pollination services are mediated by bee functional diversity and landscape context

Kyle Teixeira-Martins

Faculty of Science, Department of Biology

McGill University
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Abstract

1. Wild bees, which exhibit multiple functional traits enabling pollination of apples (*Malus domestica* Borkh), potentially can compensate for recent declines in domesticated honey bees (*Apis mellifera* Linnaeus) that are conventionally employed to ensure apple fruit and seed set. Whether compensation is possible will depend on functional diversity in the wild bee community and on the distribution of habitat and resources within the landscape surrounding an orchard that affect wild bee abundance.
2. We studied pollination services and bee functional diversity in 20 apple orchards in southern Quebec, Canada. We evaluated pollinator efficacy by studying: apple visitation rates, approach (front or side-working), body size, foraging type (pollen or nectar foraging), sociality, temporal and climatic activity patterns, and pollen carrying habit. Pollination services were measured as apple fruit set and seed set. A distance-based measure of functional diversity, calibrated with bee traits and weighted by relative abundances in the wild bee community, was used to model pollination services. We correlated the landscape composition and configuration of surrounding natural (forest) and semi-natural (meadow) habitats with bee diversity and pollination services.
3. The incidence of fruit set and seed set in orchards increased with bee functional diversity. Complementarity between managed versus unmanaged bees in traits associated with foraging and resource use drove this relationship. Seed set was also negatively correlated with both the mean distance from surrounding meadows and the total area of surrounding orchards. Bee functional diversity was positively associated with surrounding meadow and forest area. These two land classes complement each other in their seasonal provision of foraging resources for bees.

4. *Synthesis and Application:* Our models can be used to prescribe management and conservation objectives for meadow and forest habitats that promote bee functional diversity and in turn pollination services. We identify useful wild bee pollinators and discuss their needs in terms of landscape composition and configuration.

Resumé

1. Les abeilles sauvages, qui possèdent plusieurs caractéristiques fonctionnelles contribuant à la pollinisation des pommiers (*Malus domestica* Borkh), ont le potentiel de compenser le déclin d'abeilles mellifères (*Apis mellifera* Linnaeus) qui sont normalement adoptées pour assurer la grenaison et la nouaison des fruits. Les facteurs qui rendront possible cette dite compensation dépendent de la diversité fonctionnelle de la communauté des abeilles sauvages, ainsi que de la répartition de leur habitat au sein du vaste paysage qui entoure les vergers.
2. Nous avons étudié les services écosystémiques et la diversité fonctionnelle des abeilles dans vingt vergers situés au sud du Québec. Nous avons ensuite évalué l'efficacité des abeilles pollinisatrices en étudiant les paramètres suivants : la vitesse de butinage, la fréquence des contacts avec les stigmates, la taille des abeilles, le type de récolte (pollen ou nectar), la sociabilité, le seuil d'activité en fonction des paramètres climatiques et temporaux, et finalement, le mode de transport du pollen. Les services de pollinisation ont été mesurés en fonction de la nouaison et de la grenaison des pommes. La diversité fonctionnelle a été estimée en tenant compte des caractéristiques fonctionnelles et des valeurs d'abondance relative des abeilles, et cette estimation a été utilisée pour modéliser les services écosystémiques. La composition et la configuration des habitats naturels (boisés) et semi-naturels (en friche) dans les paysages autour des vergers étudiés ont été corrélées avec la diversité et les services de pollinisation fournis localement.
3. L'incidence de la nouaison et la grenaison des pommes ont augmenté avec la diversité fonctionnelle des abeilles. La complémentarité dans l'utilisation des ressources entre les abeilles gérées et non gérées a été responsable de cette relation positive. La diversité fonctionnelle des abeilles a démontré une association positive avec la terre boisée et

avec la terre en friche. La phénologie des espèces fleurissantes dans ces deux types de terrain se complète mutuellement dans leur fourniture des ressources pour les abeilles.

4. *Résumé et application*: Nous voulons mettre en place des objectifs de conservation des habitats boisés et en friche afin de favoriser la diversité fonctionnelle des abeilles sauvages et les services de pollinisation. Une identification des espèces d'abeille sauvage les plus utiles à la pollinisation a été entamée, et nous avons discuté de leurs besoins en fonction de la composition et de la configuration du paysage.

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Research for this thesis involved the participation of 27 apple orchard owners across the 2011 and 2012 sampling campaigns. Additional field study was conducted in 2012, with over 100 property owners involved in the project altogether. I express gratitude towards the apple growers, small fruit farmers, garden owners, and others who have shared a concern for bee biodiversity in the Montérégie, Québec, Canada.

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Preface and Contribution of Authors

This thesis is in a manuscript-based format consisting of a single paper intended for publication in a peer-reviewed journal, and is accompanied by a set of appendices. The manuscript explores the interrelationship between bee functional diversity, landscape context and the pollination of apple orchards in the Montérégie region of southern Quebec, Canada.

I received valuable guidance and intellectual contributions from my co-supervisors, Dr. A. Gonzalez and Dr. M. Lechowicz. Both professors provided suggestions on sampling protocols and analyses as the project was at various stages of development and engaged in several rounds of editorial comments on the thesis. They helped situate my research within the broader context of biodiversity and ecological studies, as well as with relevance for stakeholders in the Montérégie Connection project. I was, however, the primary person designing the methodology, training research assistants, leading field sampling, identifying specimens, conceiving statistical analyses, and deriving meaning from results.

General Introduction

An ecosystem comprises a community of living organisms (e.g. plants, animals, microbes) as well as their abiotic environment (e.g. air, water, soil) (Chapin & Matson 2011). An "ecosystem service" is produced when we interact with the natural environment and derive a benefit (Fisher, Turner & Morling 2009). Humans derive services from ecosystems such as material goods (e.g. timber, water, food) and useful functions (e.g. nutrient and water recycling, pollination). In this thesis I focus on apple production and bee pollination as two key ecosystem services. We benefit from the production of apples, but also from the ecological processes that support the production of apples including the transfer of compatible pollen to the stigma of the apple flower that allows the fruit to be set. There is growing concern that biodiversity change is threatening the supply of ecosystem services such as pollination and crop production (Kremen et al. 2007).

The loss of a service provided by an ecosystem can pose a serious economic cost (Fitter 2013). For example, due primarily to shortages of pollinating bees in the Hindu Kush-Himalayan region of China, orchardists and their families have been forced to hand pollinate as much as 2000 ha of blooming apple trees. On average, it would be eight times cheaper for these farmers to rent honey bees to pollinate their crop (Partap, Partap & Yonghua 2000). Scaling from the farm level to that of an entire nation, it is estimated that the cost of replacing managed honey bee pollination in Canada would be US$0.66 billion per year (Winston & Scott 1984) and is between US$1.6 billion (Southwick & Southwick 1992) and $US14.6 billion per year (Morse & Calderone 2000) for the USA depending on how the service is evaluated (Allsopp De Lange & Veldtman 2008). These cost calculations and interventions involving honey bees beg the larger question of how best to manage "agroecosystems" without jeopardizing the ecosystem services we derive from them.

An agricultural landscape is a patchwork of crop fields threaded with hedgerows, scattered with forest fragments and intervening leas. In the study of fruit bearing crops, there are two key players that provide pollination services: managed honey bees (*Apis mellifera* Linnaeus) and various species of wild bees (Kremen et al. 2007). Honey bees are typically afforded at a cost and are introduced to orchards when the trees are in bloom (Free 1993). Wild bees are, for all intents and purposes, cost-free and primarily derived from the landscapes surrounding farms (Kennedy et al. 2013), including forest edges and meadows (Mandelik et al. 2012). While the depiction of bees in popular media is that of the honey bee or a bumble bee, there are in fact approximately 19,000 species of bees worldwide (Michner 2000). Unlike honey bees the majority of bees are not eusocial and nesting in hives but solitary, and all bees require specific nesting substrate, building materials and floral resources to survive (Packer Genaro & Sheffield 2007). Depending on the degree to which an agricultural landscape satisfies the requirements of wild bees, they can act as important pollinators to the benefit of farmers (Kennedy et al. 2013).

Not all bees are created equal. A recently published global synthesis of pollination systems worldwide has found that a diverse array of bees pollinates commercial crops more effectively than honey bees alone (Garibaldi et al. 2013). While honey bees are prized for being easily transportable, are loyal to the crop they visit, and can be maintained at high densities throughout the year (Gould & Gould 1988), wild bees exhibit various adaptations and 'functional traits' that can make them superior pollinators. For instance, undomesticated bumble bees can be more effective pollinators of commercial raspberries than honey bees in that they carry more pollen, deposit more pollen on stigmas, visit more flowers per minute and are active during unfavorable weather conditions (Wilmer, Bataw & Hughes 1994). Because of such specializations specific to particular bee species, bee diversity can ensure greater pollination through a "sampling

effect" -- a more diverse bee community is more likely to contain a species with a key functional trait that affects pollination in a particular crop (Hooper et al. 2005). In addition, a "complementarity effect" can occur when bee species differ in their contribution to pollination, and traits from several species combine to enhance overall functional performance (Blüthgen & Klein 2011). For example, Chagnon, Ingras and Oliveira (1993) found that while honey bees tend to pollinate stigma of the apical florets of strawberry inflorescences, smaller wild bees pollinate the basal florets such that a strawberry visited by both bees yields the largest fruit. This sort of positive relationship between functional diversity and pollination services has been validated both experimentally (Fontaine et al. 2005; Albrecht et al. 2012; Fründ et al. 2013) and in observational studies (Chagnon, Ingras & Oliveira 1993; Greenleaf & Kremen 2006; Hoehn et al. 2008; Klein et al. 2008; Brittain et al. 2013).

Since wild bees are sustained by the landscape surrounding farms, evaluating the contribution of bee diversity to pollination services hinges on understanding the distribution and spatial structure of habitat and resource requirements. Bees are central place foragers; they will travel only a certain maximum distance from their nests in search of floral rewards or nest building materials before returning to their point of origin. Bees thus experience the landscape within only a certain radial distance from their nests; this distance varies between species and is governed by physiology and body size (Greenleaf et al. 2007). Hence the influence of landscape structure on pollination services is often estimated from aerial images of farms and their surroundings. Concentric circles emanating from the center of a farm field are created with increasing radii, typically varying in length from 250 m to 3000 m; see Steffan-Dewenter et al. (2002) for an example. Variation across bee species in the landscape heterogeneity they experience is estimated by correlating a feature of the landscape inside a given concentric circle with the bee diversity found visiting the crop at its center.

For instance, the area of semi-natural land surrounding almond orchards in California positively correlates with flower visitation frequency by wild bees (Klein et al. 2012), and similar trends have been found across agroecosystems worldwide (Klein et al. 2007; Ricketts et al. 2008; Kennedy et al. 2013). This positive association between pollination services and wild bee diversity provides an incentive for farmers and municipal planners to conserve the diversity of habitats upon which wild bees depend (Tscharntke et al. 2005).

Our project focuses on pollination services provided by wild bees to apple growers in the Montérégie administrative region of southern Quebec, Canada. The region extends from the US-Canada border north to the St-Lawrence River, west to the Ontario border and east to the Appalachian highlands. The region is home to over 1.4 million residents (Statistics Canada 2011), encompassing the densely populated south shore of Montreal as well as rural towns to the south and east of the metropolis. This landscape outside the Montreal urban area is characterized by a mosaic of forest fragments, embedded in a sea of cash crop agriculture, and studded with suburban and exurban milieus.

The Montérégie constitutes the largest apple-producing region of Quebec, with 66% of the total area of apple orchards in the province (MAPAQ 2011). Approximately 30% of the orchards in the region (MRNF 2003) are associated with the Montérégian Hills (Feininger and Goodacre 1995). The Montérégian Hills have been favored for apple growing since the earliest European settlement of the region. In 1650, Montreal's first orchard was planted on the flanks of Mont-Royal and subsequently many orchards were established on the other Monteregian Hills (Roy 1978). Given southern Quebec's insalubrious climate (Charrette & Krueger 1992), settlers favored locating orchards on slopes above the floor of the Saint Lawrence River

Valley to ensure cold air drainage, thus mitigating the risk of frost damage during the spring flowering of apples (Khanizadeh et al. 1998).

Early settlers presumably depended at least in part on wild bees to help ensure pollination of the apple crop, but in modern times orchardists have often had to rent honey bee hives for the blooming period to ensure that apple flowers develop into fruit. Over the past ten years, however, honey bee winter mortality rates in Quebec have been as high as 35% above historical levels (Currie, Pernal, & Guzmán-Novoa 2010), thus increasing the need for alternative, wild pollinators. In comparison to domesticated honey bees, wild bee species have multiple traits enabling apple pollination: they carry more pollen (Kendall & Solomon 1973), carry more compatible fruit pollen (Kendall 1973), transfer fruit pollen at a higher rate (Jacob-Remacle 1989; Thomson and Goodell 2001), show a stronger preference for *Malus* flowers (Johnson 1984, Kendall & Solomon 1973, Vicens & Bosch 2000) and can compensate for honey bees under adverse environmental conditions (Free 1960; Chansigaud 1975; Boyle-Makowski & Philogene 1985; Batra 1994). In the course of my thesis research, farmers have asked me again and again whether they in fact could depend on pollination by wild bees derived from landscapes surrounding their farms in lieu of renting honey bees.

Although honey bee declines are a concern for apple production in the Montérégie, the potential role of bee diversity and the surrounding landscape in pollination services remains poorly understood. The only published work we could find surveying wild bee diversity in a southern Quebec orchard was conducted by Oliveira, Pion and Paradis (1980) who reported a surprising diversity of bees visiting apple blooms, comprising at least 25 species. However, domesticated honey bees were dominant and wild bees encompassed only 19.5-29.1% of the bee community involved in apple pollination. Recent research on the value of wild bees in apple pollination includes that conducted by Watson, Wolf and Ascher (2011) in Wisconsin,

USA. They focused on the interrelation between bee diversity in orchards and the surrounding landscape and found that both wild bee species richness and abundance were positively predicted by the amount of forest area surrounding farms. However, neither in the historical literature (Hutson 1926; Brittain 1933, 1935; Phillips 1933; Loken 1958; Free 1964, 1966; Oliveira & Paradis 1980; Boyle & Philogene 1983, 1985; Scott-Dupree & Winston 1987; Jacob-Remacle 1989) nor in recent work (Gardner & Ascher 2006; Watson, Wolf & Ascher 2011; Adamson et al. 2012) has there been a study to quantitatively interrelate apple pollination services to individual pollinator efficacies, to bee diversity, and in turn to the landscapes from which wild bees are sourced.

This thesis therefore investigates the interplay between pollination services, bee species diversity and landscape context in apple orchards. The research is supported by and is part of the "Montérégie Connection Project", which seeks to understand the linkages between biodiversity, ecosystem services and landscape configuration in the Montérégie; to build landscape models used to inform decision makers about current and future provisions of ecosystem services; and to engage in active communication with corporations, government, and non-government organizations to ensure the utilization of research findings.

This part of the Montérégie Connection Project aims in particular to generate scientifically sound recommendations founded in field-based observations and ecological theory that can be implemented by our stakeholders – not only apple growers in the region, but also all those whose decisions influence the growing of apples. As such, we adopted the following four objectives:

1. To document the biodiversity of bee fauna visiting apple orchards in the Montérégie

2. To quantify the relationship between pollination services and bee functional diversity
3. To establish the bee functional traits implicated in the relationship between bee diversity and pollination services
4. To produce models predicting bee diversity in apple orchards as a function of the surrounding landscape

Our assessment spans two years of field sampling, beginning with a pilot study in 2011 on apple pollination by wild bumble bees (Appendix A) in relation to landscape context. In 2012, we then sampled the full gamut of wild bee visitors to orchards, observing their pollinator behavior, assessing fruit set and seed set in apples as a measure of pollination services, and drawing relationships with landscape metrics.

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CHAPTER 1: Pollination services are mediated by bee functional diversity and landscape context

Kyle Teixeira- Martins, Andrew Gonzalez, and Martin J. Lechowicz

McGill University, Department of Biology
1205 Dr. Penfield Avenue, Montreal, Quebec
Canada H3A 1B1

Introduction

Wild bees can pollinate crops more effectively than honey bees (*Apis mellifera* Linnaeus) alone. Recent meta-analysis shows that wild bees are positively associated with fruit set in agroecosystems worldwide (Garibaldi et al. 2013). Whereas wild bees are sourced from mixed habitat landscapes surrounding farms (Kennedy et al. 2013), domesticated honey bees are artificially introduced for the period during which the crop is in bloom. Research pertaining to alternative pollinators and their habitat requirements has been stimulated by recent declines in managed honey bees (Johnson et al. 2009; Currie, Pernal & Guzmán-Novoa 2010; Neumann & Carreck 2010) and the concomitant increase in demand for traditionally honey bee pollinated crops (Aizen & Harder 2009). Our study focuses on the effect of the wild bee community on pollination services and the role played by landscape composition and configuration surrounding farms on the availability of wild bees.

In a diverse assemblage of bees there is an inherent structure with species characterized by particular functional traits that facilitate pollination services to a greater or lesser degree (Blüthgen & Klein 2011). For instance, bees can exhibit interspecific variation in the number of flowers visited per unit time (Jacob-Remacle 1989; Willmer et al. 1994; Javorek et al. 2002), in their efficacy at depositing or removing compatible pollen on the stigma per visit (Thomson & Goodell 2001; Kremen, Williams & Thorp 2002, Kremen et al. 2004; Winfree et al. 2007), and in physiological restrictions that limit their activity to certain abiotic conditions (Rader et al. 2013). When contributions to ecosystem functioning differ among species, traits from several species taken together can enhance overall functional performance levels (Hooper et al. 2005; Blüthgen & Klein 2011). Hence functional diversity in the bee community can be anticipated to enhance pollination services, an expectation that has been validated both experimentally (Fontaine et al. 2005; Albrecht et al. 2012; Fründ et al. 2013) and in observational studies (Chagnon, Ingras &

Oliveira 1993; Greenleaf & Kremen 2006; Hoehn et al. 2008; Klein et al. 2008; Brittain et al. 2013).

The potential enhancement of pollination services by functional diversity arises in 1) the relative abundance of bee species in a community and 2) the degree to which species-specific traits influencing pollination vary in the community (Petchey & Gatson 2006). The greater the relative abundance of a species, the more its traits favoring pollination will enhance pollination service by the bee community. Provided that bees in the community do not exhibit 'functional redundancy' (Rosenfeld 2002), there is the potential for additional enhancement through complementarity effects as well (Hooper et al. 2005; Blüthgen & Klein 2011). Since assessing the effect on pollination services of every combination of bees exhibiting different traits at varying relative abundances in natural settings is logistically impractical, functional diversity typically has been treated through a simple index approach (Hoehn et al. 2008; Klein et al. 2008). Such indices of functional diversity, however, have yet to explicitly and simultaneously consider the relative abundance of many species weighted by multiple functional traits.

We employ Functional Dispersion (FDis; Laliberté & Legendre 2010) as a quantitative metric that combines both the relative abundance of bees and trait diversity elements of functional diversity, and use FDis to predict fruit and seed set in apple (*Malus domestica* Borkh). Functional dispersion is essentially a distance-based index that estimates functional diversity in terms of the relative abundances of bee taxa in a multidimensional trait space. Having conducted detailed observations of bee pollinator behavior, we are able to calibrate FDis with those functional traits most correlated with fruit and seed set in our system. In turn, we can identify target pollinator groups for conservation by understanding pollinator characteristics that enable fruit production.

The quantification of functional diversity in terms of bee relative abundance and the diversity of functional traits is especially important for tree crops such as apple. Apple cultivars mostly are self-incompatible and require insect mediated cross-pollination to set seeds and produce fruit (McGregor 1976). Poor pollination can reduce seed set, which leads to asymmetrical pomes (Brault & Oliveira 1995; Keulemans et al. 1995) that fail to meet market expectations for fruit quality. The receptive period of the apple stigma, however, is very brief (Oliveira, Pion & Paradis 1980; Sheffield, Smith & Kevan 2005) and in temperate settings occurs in tempestuous spring climates that can hamper pollinator activity (Gould 1939). Moreover, some bee genera, such as bumble bees, have not yet established prolific colonies in the spring when apples flower (Gardner & Ascher 2006). Orchardists therefore typically rent domesticated honey bees to ensure apple pollination (Free 1993). As indicated by insect surveys over the past century, a surprising diversity of wild, non-*Apis* bees (hereafter referred to only as 'wild bees') can be found in apple orchards (Hutson 1926; Brittain 1933, 1935; Phillips 1933; Loken 1958; Free 1964, 1966; Oliveira & Paradis 1980; Boyle & Philogene 1983, 1985; Scott-Dupree & Winston 1987; Jacob-Remacle 1989; Gardner & Ascher 2006; Park et al. 2010; Watson, Wolf & Ascher 2011; Adamson et al. 2012). In comparison to domesticated honey bees, wild bee species have been shown to exhibit multiple traits enabling apple pollination: they carry more pollen (Kendall & Solomon 1973), carry more compatible fruit pollen (Kendall 1973), transfer fruit pollen at a higher rate (Jacob-Remacle 1989; Thomson and Goodell 2001), show a stronger preference for *Malus* flowers (Johnson 1984, Kendall & Solomon 1973, Vicens & Bosch 2000) and compensate for honey bees under adverse environmental conditions (Free 1960; Chansigaud 1975; Boyle-Makowski & Philogene 1985; Batra 1994). A noteworthy inadequacy in the pollination of apples by honey bees is their tendency to "sidework", that is, drawing nectar from apple flowers without contacting and thus fecundating the stigma (Schneider *et al.*

2002, 2004). Pollination services delivered to apples thus depend upon not only domesticated honey bees but also wild bee visitors that differ in their relative abundance, seasonal phenology, temporal and climatic activity patterns, and diverse functional traits.

The degree to which a farmer may choose to import honey bees or depend on wild bees will largely be determined by the quality of the landscape surrounding farms from which wild bees are sourced. Habitats surrounding orchards support wild bees by provisioning nesting and foraging resources at different times of the year (Parish & Bazzazz 1979; Westrich 1996; Heinrich 2004). The composition and configuration of bee-frequented habitats in agroecosystems is strongly associated with the frequency and diversity of bee visitors to a given agricultural field (Klein et al. 2007; Ricketts 2008; Kennedy et al. 2013). Surrounding forest cover has been found to predict the abundance and richness of wild bees visiting apple orchards (Watson, Wolf & Ascher 2011). In a pilot study we established that bumble bee abundances in orchards were positively correlated with the surrounding area of open field habit but not with forest area (Appendix A). Similarly, forest cover and distance to nearest forest patch did not correlate with pollination services in either watermelon or vegetable crops (Winfree et al. 2007, 2008; Lonsdorf et al 2009). Mandelik et al (2012) report that wild bees respond to differences in flowering phenology, tracking resource availability from forested, agricultural, and open field habitats. We contribute to this literature by sampling the diversity of all *Apis* and non-*Apis* bees in apple orchards situated along a gradient of forest cover and open field habitat in a temperate environmental setting. By quantifying the relationship between bee diversity and landscape metrics, we provide clear objectives for land use managers regarding how best to ensure the security of pollination services (Kremen et al. 2007).

This is the first study of its kind to interrelate multiple functional traits, interspecific bee abundances, pollination services and the landscapes upon which bees depend. We adopt a "production function" approach (Tallis & Pollasky 2009) to trace a mathematical relationship between pollination services and bee FDis (Laliberté & Legendre 2010), as well as landscape metrics and environmental covariates. We identify the pollination service point of delivery in apples at the incidence of seed and fruit set (see Liss et al. 2013). These measures are the most direct results of pollination (Dennis 2003) that benefit orchardists without being confounded by local management decisions that influence crop yield (Dale & Polasky 2007). Our findings can be used to inform orchard and land use managers on the relevance and requirements for fostering wild bee diversity in this system. We organize our investigation around two specific questions: 1) when weighted by bee relative abundance in the local community are multiple functional traits correlated with apple fruit set and seed set? and 2) which landscape characteristics best predict bee diversity and pollination services in apple orchards?

Methods

Study Area and Site Context

The study took place in 20 apple orchards on the lower slopes of the Monteregian Hills (Feininger & Goodacre 1995) and the surrounding floor of the St. Lawrence River Valley to the east of Montreal in southern Quebec, Canada (Figure 1). The Montérégie constitutes the largest apple-producing region of Quebec, with 66% of the total apple orchard area and 50% of all farms in the province (MAPAQ 2011). The landscape is characterized by a mosaic of forest fragments, embedded in a sea of cash crop agriculture, and studded with suburban and exurban milieus.

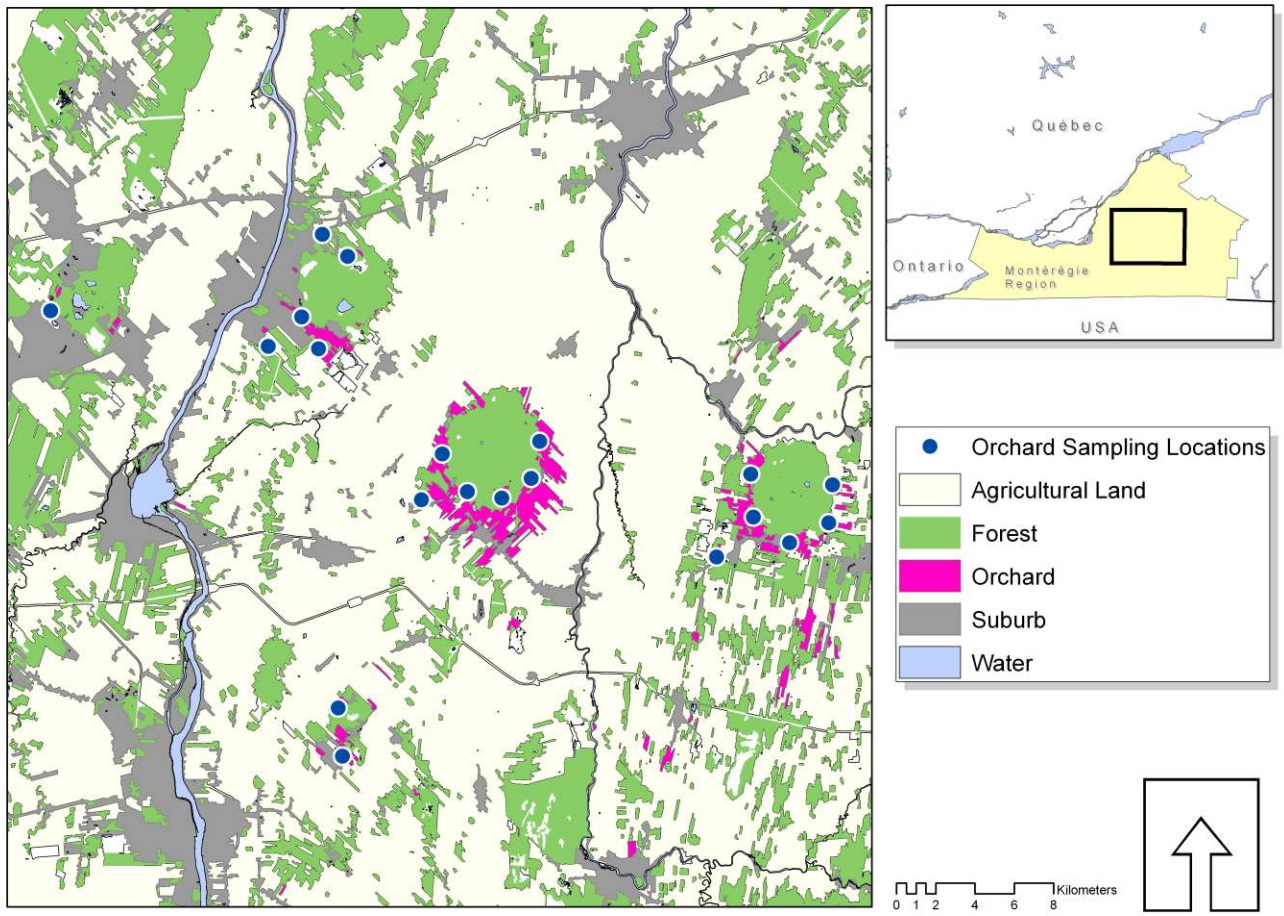

Figure 1: Map of Study Orchards

Locations of study orchards and landscape characteristics in the Montérégie administrative region of southern Quebec, Canada. The map was produced using with ArcGIS 10.1.

We studied commercial orchards at sites with good cold air drainage in spring that minimized the risk of frost damage to apple flowers (Khanizadeh et al. 1998). In the process of site selection, we ensured that 1) forest area and meadow area were uncorrelated ($r<0.50$) within a radial distance of 1000 m from sites, and 2) increased in percent cover in the surrounding landscape by 8-fold and 12-fold, respectively. Ten of the study orchards had standard apple tree rootstocks, and ten dwarf to semi-dwarf rootstocks. The minimum distance between sites was 1766 m with a mean nearest neighbor distance of 2702 m. All the farmers practiced integrative pest management (Bloomers 1994; Bourgeault 2009) in consultation with a provincial agronomist. To control for between-cultivar variation in fruit and seed set (Dennis 1979) we focused our study on McIntosh apples, which constitute 64% of the Quebec market (MAPAQ 2011) and are the leading cultivar in the north-eastern United States and eastern Canada (Hampson & Kemp 2003). McIntosh apples are altogether self-incompatible and cannot be wind-pollinated (Oliveira, Pion & Paradis 1980), so Courtland, Paula Red and other varieties are interplanted either between or within rows to ensure cross-pollination. Fourteen of the 20 orchards rented honey bees during the period of our observations, and all sites were within the flight distance of an orchard that did rent honey bees.

Site Construction and Characterization

Our sampling sites within each orchard were 100 m in length and 50 m in width (Figure 2), with placement largely determined by the availability of McIntosh trees. We measured a suite of covariates describing orchard management strategies including tree basal trunk diameter (cm), canopy radius (m), tree height (m) and density (trees/m^2). Ambient light and

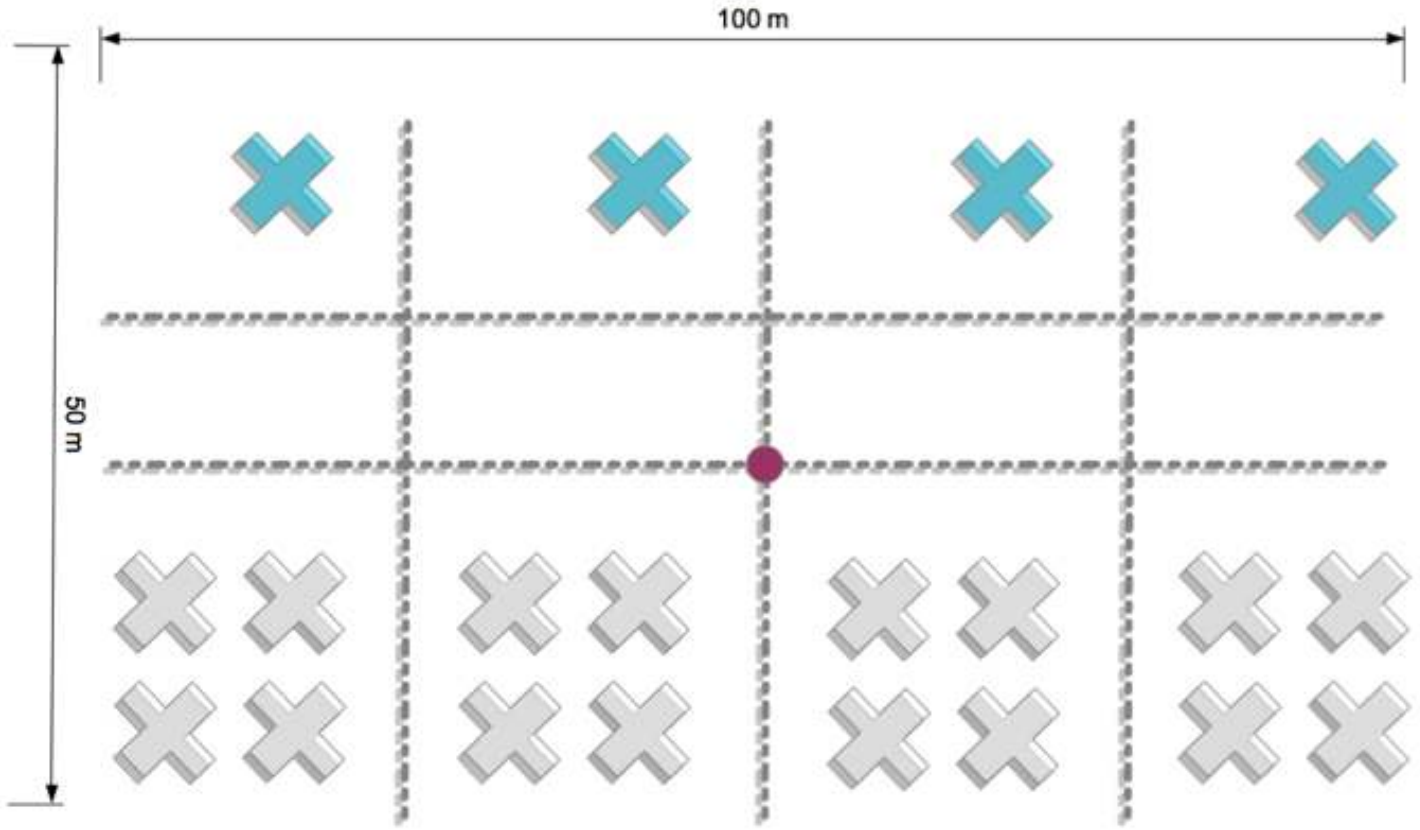

Figure 2: Layout of Sampling Design

A 100 x 50 m sampling site was constructed in each apple orchard with two sampling transects (crosses) and a set of intervening trees (dashed lines). The pollen supplementation transect (turquoise crosses) ran parallel to the bee observation transect (grey crosses). A pair of temperature/humidity and temperature/light data loggers was placed in the centermost tree of the site (red circle).

temperature influence flowering and fruit development (Dennis 1979), so we also monitored these variables at two minute intervals from May 5th until May 19th, 2012 using HOBO pendant temperature/light loggers and HOBO Pro v2 temperature/relative humidity loggers (Onset, Bourne, Massachusetts, USA). We placed the HOBOs within the centermost tree at each sampling site, with the temperature/relative humidity loggers shielded from direct insolation and the temperature/light loggers pointed towards the sun on an exposed branch. Note that the temperature readings taken from the unshielded Pro V2 loggers were not used in characterizing microclimatic conditions at the sampling sites.

Evaluating Pollinator Efficacy and Bee Diversity

The sampling of bee diversity in the orchards took place from May 11-14th, 2012, during the peak bloom of McIntosh apples. Each site was visited only once while McIntosh apples trees were in flower. To augment our observations of bee diversity and behavior, we conducted supplemental surveys in orchards from May 15th-18th on late blooming varieties (primarily Lobo and Courtland). Bees were observed under clear to lightly overcast conditions from 9:00 AM – 6:00 PM Eastern Standard Time when ambient air temperatures were above 15°C, and wind speeds were below 3.3 m/s. The average number of apple flowers per tree for a site was estimated by counting the flowers on a branch containing approximately one tenth of the flowers on the tree, and repeating this process for five trees. Since dandelion (*Taraxacum officinale* Wigg) abundance in the orchard understory can distract bees from visiting apple flowers (Free 1968), we counted all dandelion heads that fell within a hoop (diameter=1.5 m) randomly placed nine times between the rows and then an additional nine times beneath any of the apple trees within the confines of the site.

Adapting the approach of Boyle and Philogene (1983), we visually assigned bees to one of seven morphospecies: 1) honey bees (*Apis mellifera*), 2) bumble bees (*Bombus* spp. Latreille), 3) green metallic halictids (mostly *Augochlora* spp. (Say)),

4) large *Andrena* (*Andrena carlini (*Cockerell), A. *duningii* Cockerell, *A. erythronii* Robertson*, A. milwaukeensis* Graenicher, *A. regularis* Malloch*, A. vicina* Smith, *Colletes* spp. Say), 5) *Osmia spp*. Panzer, 6) small *Andrena* (all other *Andrena* spp. Fabricus), and 7) small black bees (*Ceratina* spp. Say, *Halictus* spp. Latreille, *Lasioglossum* spp. Curtis). *Colletes* were grouped with large *Andrena* because of their morphological similarities. All morphospecies other than honey bees constituted the 'wild bee' group. Feral honey bees have been observed visiting apple orchards (Chang & Hoopingarner 1990); however, it is impossible to visually distinguish feral honeybees from their domesticated conspecifics.

Bee pollinator efficacies were assessed in terms of floral visitation rates, tendencies to sidework, and activity patterns in varying climatic and temporal conditions. Floral visitation rates were prioritized over pollen deposition (Kremen 2004) given that visitation is an easily observed indicator for pollinator efficacy (Thomson and Goodell 2001). We used three complementary and co-occurring sets of observations to characterize aspects of pollinator behavior as well as to assess bee diversity across all the studied orchards. Observations took place within 40 minute time periods when sampling bees on McIntosh apple trees, and 20 minute time periods when surveying late blooming varieties. All observations were made on bees actively visiting apple blossoms, as opposed to herbs in the orchard understory

One observer made timed observations of bee behavior, and thus functional traits effecting pollinator efficacy. A given bee recognized to morphospecies was observed for a maximum of four minutes, and the number of flowers visited during this time period was counted to estimate bee 'foraging rate'. The average amount of time the bee expended while drawing nectar and/or pollen per visit was also recorded as a measure of 'foraging duration'. The start time was marked when the bee landed on its first flower. Notes were made on bee 'foraging type' when visiting the blooms, and whether the bee was gathering pollen or nectar or both. Moreover, the 'approach' of the bee was described as 'sideworking' (drawing nectar without

contacting stigma), 'frontworking' (drawing nectar or pollen while contacting the stigma) or both (start by sideworking, but then move across the stigma).

A second, '*stationary*' observer counted bees visiting apple trees along a transect of four evenly spaced clusters of McIntosh trees extending the length of the site (Figure 2), assigning each observation to morphospecies. There were four trees per cluster and each tree was sampled for 30 seconds, sequentially, until all four trees in that area of the transect at the site were sampled. This was repeated three times per cluster so that each tree was observed for a total of 1.5 minutes. The whole procedure was then replicated for the remaining three clusters of trees. The time of day was noted for each 1.5 minute interval during which bees were observed so that results could be cross-referenced with environmental conditions recorded by data loggers. This also allowed us to track activity patterns of morphospecies as a function of the time of day and weather conditions.

A third '*opportunistic*' observer passed between the orchard rows with a sweep net to estimate bee diversity at the site. Honey bees were identified on the fly and tallied using a counter; wild bees were either caught with a sweep net and placed into a universal tube or visually counted given their morphospecies identity. Netted specimens were later frozen for processing and species-level identification.

Captured bees were identified to species using pertinent literature for bee genera of eastern North America (Stephen 1954; Mitchell 1960, 1962; LaBerge 1971, 1978; LaBerge & Bouseman 1973; Roberts 1973; Bouseman & LaBerge 1978; Laverty & Harder 1988; Packer, Genaro & Sheffield 2007; Gibbs 2010, 2011; Colla, Richardson & Williams 2011) as well as http://www.Discoverlife.org/. Specimens were compared with reference collections at the American Museum of Natural History, the Cornell University Insect Collection, and the Lyman Entomological Museum. Specialists John Ascher, Bryan Danforth, and Jason Gibbs verified species identifications. As an index of bee mass and foraging range (Greenleaf et al. 2007), we measured the distance between the wing bases, which is formally referred to as

the inter-tegular (IT) span (Cane 1987). Following Cane (1987), we measured a subsample of one to five specimens per species to estimate IT spans.

Estimation of Fruit and Seed Set

We conducted a pollen supplementation experiment on McIntosh apple trees to estimate the degree of pollen limitation in the system, and to correlate the incidence of fruit set of un-manipulated flowers with bee FDis. Four evenly spaced McIntosh apple trees were identified per site in a separate row from that used by the stationary observer (Figure 2). There were two experimental treatments (hand pollinated flowers and open pollinated flowers) per study tree, each comprising 20 flowers. Given that the apple inflorescence consists of a cluster of five florets, only the centermost 'king' flower was studied as it is the least likely to be aborted by the apple tree if fruit is set (Dennis 2003). Hand-pollinated flowers were tagged and then fecundated with commercially available Red Delicious pollen (Firman Pollen Inc., Wakima, Washington, USA) using a camel hair paintbrush. Only a single pollen application was made per flower. Tagged, open-pollinated flowers were freely visited by available pollinators. Flowers were chosen that were exposed and located along the outer limbs of the trees, and branch height from the ground was measured as a covariate (Dennis 1979).

Fruit set of McIntosh apples was evaluated from May 23rd -25th, 2012, and was noted as having occurred if the fruit remained fixed to the spur when pedicels were lightly tugged upon. Sites that applied chemical thinners, which artificially abort apples, were asked not to do so with study trees; to minimize potential complications we also measured fruit set prior to the normal period of chemical thinning in the orchards.

We randomly selected an additional sample of eight McIntosh pomes from a constant height throughout each of the study trees at each site to estimate seed set. We collected these samples from June 4th and 6th and from June 18th-20th, when the

apples in a sampled orchard had achieved 1.0 cm in diameter and seed set was readily distinguishable. Seed set was assessed by cutting apples in half and counting the total number of fully formed seeds as per Brault and Oliveira (1995).

Landscape Characterization

To determine an appropriate spatial scale at which to study landscapes surrounding our orchard sites, we estimated the average foraging range of the wild bee community. With two adjustments noted below, the IT span measurements of bee specimens were converted to bee foraging ranges (Greenleaf et al. 2007) and weighted by the abundance of each morphospecies across sites to yield an average foraging range of 680 m. This estimate, however, excluded honey bees as they were artificially introduced into the orchards and removed following the apple bloom. During the sampling period we captured only bumble bee queens, which are much larger than the workers that characterize this morphospecies through the season. Hence in calculating the spatial scale at which the landscape around each orchard would be studied, we estimated bumble bee dispersal at 1.75 km from data for workers of *Bombus terrestris* (Walther-Hellwig et al. 2000) rather than the 6.26 km estimated from captured queens. Land cover classes were interpreted and analyzed from aerial imagery only within this 680 m foraging range around each orchard, a total area of 145 ha.

After preliminary analyses of many land cover types (Table A1), we focused on three metrics to characterize the landscape surrounding each studied orchard: the areas (m^2) of forest, meadow and orchard within a 680 m foraging range. To clarify, the calculation of area covered by orchard in the 145 ha included all orchards, not only the studied orchard. Orchards of dwarf and standard trees were not distinguished when estimating orchard area. Forests had closed canopies and understories of varying heights and densities, including both primary and secondary growth deciduous forests. Meadows were areas greater than 10x10 m dominated by grasses and forbs. We tallied the total area of each land cover type in the 145 ha

foraging area around each site. We also calculated a 'mean distance' to forest and meadow patches by averaging the distances along the radial vectors from the site epicenter to all points of contact within the meadows and along the edges of forest patches in the 145 ha foraging area around each site. This 'mean distance' metric gauges the length of foraging paths taken by bees relative to each orchard and the total area of the three land cover types gauges the availability of foraging and nesting resources surrounding an orchard.

We used ArcGIS 10.1 (Environmental Systems Research Institute, Redlands, CA, USA) for landscape analyses. We interpreted land classes from a composite of satellite and orthophoto imagery taken by the *Ministère des ressources naturelles et de la faune du Québec* (30 cm resolution) for the Montérégie region in 2009, *DMTI Spatial* (60 cm resolution) for Mont-St-Bruno in 2006 and *Imex ltée* (30 cm resolution) for Mont Saint Hilaire in 2007. Interpretations were cross-referenced with data from the Quebec Forest Inventory Service (MRNF 2003) and were verified by field-based validation from June 22nd-25th, 2012.

Data Analysis

All statistical analyses were conducted with R version 2.15.3 (R Project for Statistical Computing release 2013-03-01 http://www.r-project.org).

Calibrating a Measure of Functional Diversity

In order to measure the functional diversity of the bee community present in each apple orchard, we sought to examine bee functional traits related to: 1) variation among bee morphospecies in their respective pollinator behaviors, 2) patterns in bee activity according to environmental conditions. We then consolidated findings into a functional trait table and assessed which traits were most correlated with fruit set and seed set in orchards. The ultimate objective of this analysis was to parameterize our measure of functional dispersion with those functional traits most associated with apple pollination services. Functional

dispersion could then be used to model fruit set and seed set with generalized linear mixed models (GLMM).

How do bees vary in their behavior when pollinating apples blossoms?

To validate whether there was a relationship between morphospecies identity (*n*) and qualitative bee behaviors (*p*: including sideworking, frontworking, both), and foraging type (*q*: nectar, pollen, both), we conducted a chi-square test on two separate *n* x *p* and *n* x *q* contingency tables. The two analyses were considered separately because data on bumble bee foraging types were not available due to observational limitations. We tested the null hypothesis of independence between descriptors with a test statistic simulated using Monte Carlo methods ('stats' package; R core team 2013). Freeman-Tukey deviates were considered significant if they were higher than $[\nu\chi^2_{[1,\alpha/\text{no.cell}]}/(\text{no.cells})]^{1/2}$, where ν is the degrees of freedom of the contingency table, α the significance level, and no. cells the number of cells being tested (Sokal & Rohlf 1995). We only tested cells with expected values greater than five.

We analyzed differences among morphospecies in foraging rates (flowers visited per minute) as well as foraging duration (average time spent visiting each flower), having first checked whether bees varied in their behavior in relation to the weather (Peat & Goulson 2005), floral densities (Heinrich 1979; Waddington 1980), apple variety, or foraging type (Thomson and Goodell 2001). We began by using two GLMM's (calculated with the 'lme4' package by Bates et al. 2012) for foraging rate and foraging duration, respectively. We assumed a Gaussian error distribution for both response variables and modeled them separately as a function of fixed effects that included: morphospecies, average temperature, average humidity, time of day, average apple tree basal diameter, apple tree variety and orchard tree density. The abiotic parameters were specific to when bees were being observed at each orchard. The two variables describing orchard management (average apple tree basal

diameter and orchard tree density) were included in this and subsequent analyses because they were found to approximate floral densities at each site but were measured with less error. We also included additive random effects for the intercept with respect to sampling sites in these two models. Finally, we tested if there was a significant interaction between foraging type and morphospecies identity (excluding bumble bees) on foraging rate and duration, respectively, using two additional GLMM's ('lme4' package by Bates et al. 2012) with the same random effect structures. Observations across sites, environmental conditions, varieties and behaviors per bee group could then be pooled together if no significant confounding effects were found. A Kruskal-Wallis comparison between treatment rank means with a Bonferonii correction was used for the pairwise testing of both foraging rate and duration as a function of morphospecies.

How does bee activity vary with time of day and environmental conditions in orchards?

Since pollinator activity in orchards is affected by abiotic parameters (Boyle-Makowski & Philogene 1985), we tested for preferences among bee morphospecies in respect to humidity, light, and temperature conditions as well as temporal patterns of activity. We used a partial redundancy analysis (RDA; 'vegan' package; Oksanen et al. 2012) to this end, which is best understood as a multiple-linear regression of a principle component analysis (Legendre & Legendre 2012). Morphospecies level observations were taken only from the stationary observer, and were collapsed to provide a single estimate for each of the four clusters of trees per site. This matrix of morphospecies by tree cluster, **Y**, was subjected to a Hellinger transformation (Legendre & Gallagher 2001) to alleviate the dominant effect of honey bees, which was found to mask variance in non-*Apis* bees. The **Y** matrix was scaled to unit variance and regressed on an environmental matrix, **X,** consisting of the temperature, time of day, humidity and light conditions during which each cluster was observed. The site in which tree clusters were found as well as the apple tree variety sampled were coded as a dummy variable and treated as a

matrix of covariables, **W**, in order to partial out site and apple variety effects. In this way we were able to observe preferences of each bee morphospecies for particular temporal and environmental conditions, while accounting for the covariance between morphospecies. The first three dimensions of the RDA were tested for significance by treating each previously tested axis as a covariable (Legendre, Oksanen & ter Braak 2011).

Which bee functional traits are most correlated with apple pollination services?

Data on bee behavior were consolidated to create a functional trait matrix. Functional traits were described for both species captured and morphospecies observed on McIntosh trees across orchards. If a species-level trait was not available, we would use the value of the morphospecies to which the species belonged. When referring to traits of both bee species and morphospecies taken together, we will use the generic term 'bee type'. Foraging rates and durations were estimated by their average values for each bee type. Since bee size has been linked to pollinator behavior (Stout 2000) and pollen deposition (Hoehn et al. 2008), we also estimated average IT span for each bee type. We estimated 'percent stigma contact' -- the percentage of visits upon which each bee type made contact with the stigma -- from our observations on the frequency of sideworking and frontworking; foraging type could not be used because data were not available for bumble bees. Temporal and environmental preferences were synthesized as an 'environmental index' by taking the projection of each bee morphospecies vector on the first RDA axis, which explains the greatest variance in the ordination. We noted pollen carrying habit (Michner 2000; Thorp 2000) for each bee type, which refers to whether a bee carries pollen packed moist into corbiculae and hence unavailable for pollination or packed dry and hence available for pollination. Two additional traits characterizing bee behavior were taken from the literature: sociality (solitary or eusocial) and flight period (early: active from April-July; late: April-October) after Mitchell (1960, 1962), Packer, Genaro & Sheffield (2007), and Gibbs (2010, 2011).

We used a fourth corner analysis ('ade4' package; Legendre, Galzin, Harmelin-Vivien 1997; Dray, Dufour & Chessel 2007; Dray and Legendre 2008; Ter Braak, Cormont and Dray 2012) to screen functional traits and identify those most correlated with fruit set and seed set. This analysis considers three tables simultaneously: 1) a table **L** (n x p) containing the abundance of p bee species caught and morphospecies observed at n sites; 2) a table **R** (n x m) with the pollination service vectors m (average fruit set and seed set values) for the n sites, and 3) a table **Q** (p x s) describing s functional traits for the p species. Table **L** included species level as well as morphospecies observations of bees visiting McIntosh apple trees. Morphospecies observations were summed across both the stationary and opportunistic observers; the two sets of observations exhibited high concordance in a multiple factor analysis (MFA; see Appendix B). Through the inflation of matrices to a correspondence table (Dray and Legendre 2008) we were able to correlate bee functional traits with seed and fruit set variables, as mediated by the abundance of each bee type across sites. To test the strength of the link between traits and services against a null model, we assayed two permutation techniques for table **L** appropriate for our datasets, which contain random variables in tables **R** and **Q**. The first model permutes the rows and columns of table **L** simultaneously as proposed by Dolédec et al. (1996); the second permutes the rows and columns of table **L** separately and adopts the higher p-value from the two tests as proposed by Ter Braak, Cormont and Dray (2012). The functional traits found to be significantly correlated with seed set and fruit set were then used to establish a metric describing bee functional diversity.

Functional diversity of the bee community present at each site was quantified using functional dispersion (FDis in the 'FD' package; Laliberté & Legendre 2010; Laliberté & Shipley 2011). The analysis measures the mean distance in multidimensional trait space of individual species to the centroid of all species (Figure 3). Functional dispersion accounts for relative abundance by shifting the centroid toward the more abundant species. Functional dispersion was calculated from table **L** in relation to a Gower dissimilarity matrix (Gower & Legendre 1986;

Legendre & Legendre 2012) of trait variables selected through the fourth corner analysis. Orchards with high values of FDis have many different bees groups exhibiting distinct functional traits, while low values of FDis have only a single type of bee or different bee types all exhibiting the same traits.

Modeling Ecosystem Services

We compared fruit set of hand- versus open-pollinated flowers to identify the degree of pollen limitation in the orchard system (Corbet, Williams and Osborne 1991). A logistic GLMM ('lme4' package by Bates et al. 2012) was adopted to predict the probability of fruit set as a function of pollination treatment (hand- versus open-pollinated) as a fixed effect. We included observed trees nested within sites as random effects to account for the hierarchical structure of the sampling design. If hand-pollinated flowers were found to set fruit more often than open-pollinated flowers, this would indicate the orchards were generally pollen limited.

Two separate GLMM analyses ('lme4' package by Bates et al. 2012) were used to study factors influencing the incidence of fruit set and the number of seeds produced per apples, respectively. In both analyses trees nested in sites were treated as random effects; we assumed fruit set followed a binomial error distribution, and seed set a Poisson error distribution. Fixed effects were organized into four variable groups: bee FDis, environmental covariates, landscape metrics, and orchard management covariates (see Table 1 for the list of variables). The environmental parameters chosen were average values taken from dataloggers for temperature and light conditions over the entire monitoring period encompassing flower development, pollination and early fruit development. Basal trunk diameters were specific to the trees being studied for fruit and seed set. Branch heights were only available for the fruit set analysis as apples sampled for seed set were taken at a constant height. Variables with correlation coefficients greater than 0.5 were not included in the same model (Booth, Niccolucci and Schuster 1994); as such, meadow area could not be considered in these analyses because of collinearity with FDis.

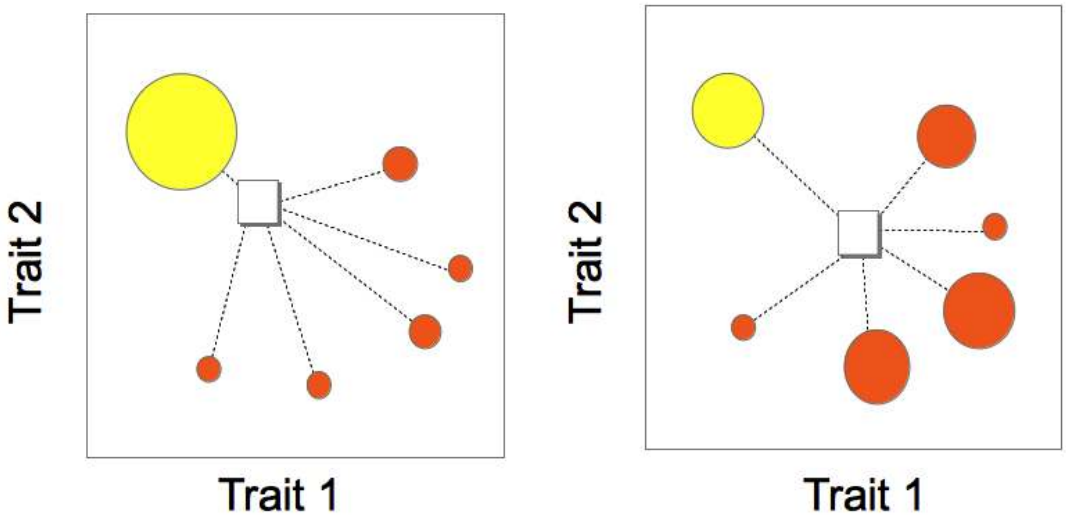

Figure 3: Functional Dispersion

This figure is adapted from Laliberté and Legendre (2008) to illustrate the nature of functional dispersion (FDis). Circles represent bee morphospecies in a simple two-dimensional trait space; in our analyses the trait space has more dimensions, but the nature of the measure does not change with dimensionality. The size of each circle is proportional to the bee's relative abundance in a given sampling site and the color indicates whether the bees are domesticated (yellow) or wild (orange). The grey square is the community centroid, the position of which will shift towards the more abundant species. Functional dispersion measures the mean distance in the multidimensional trait space of the individual species to the centroid of all species, as weighted by relative abundance. Functional dispersion will take a low value in the left figure because the community is dominated by honey bees that all exhibit the same traits. In the right hand figure, FDis will be higher because other bee groups exhibiting a variety of different traits are relatively more abundant.

Explanatory variables were standardized to a mean of zero and a standard deviation of one to allow for the comparison of model parameter estimates.

To study the relationship between bee functional diversity and the surrounding landscape, we used multiple linear regression to model variation in FDis with landscape metrics as well as environmental and orchard management covariates (see Table 1). Functional dispersion was a normal continuous variable. We did not employ a mixed model because FDis was calculated at the site level. In these analyses environmental covariates were averaged only over the period when bees were being sampled in each orchard. Similarly, trunk diameter was taken as the average of study trees per site. We checked for collinearity, normality and standardized explanatory variables by the same means as in the analysis of fruit set and seed set.

A model averaging approach was taken in model selection, which allowed us to study the uncertainty when quantifying the precision of a given coefficient (Burnham & Anderson 2002). We screened all possible level two interactions of interest using an automated model selection program ('glmulti' package; Calcagno & Mazancourt 2010; Calgano 2012). The function was implemented on each variable group (environmental, landscape, orchard management variables) separately for the fruit set, seed set and FDis analyses. The principle of marginality was respected, and a maximum of five parameters were generated per permutation. These measures were taken as a means of restricting model complexity. We ranked models according to their AIC values calculated from maximum likelihood criteria, and estimated associated Akaike weights and relative importance scores ('MuMIn' package; Bartoń 2013). Relative importance is calculated as the sum of the Akaike weights over all of the models in which the parameter of interest appears (Johnson & Omland 2004). None of the interactions had relative importance scores greater than 0.60 were thus not retained for further analysis. Selected variable were consolidated and taken as the 'saturated model' from which all variable combinations were again generated. Interactions between variables were not

Table 1: Model Parameters

Parameter specifications for linear models predicting fruit set, seed set and functional dispersion.

	Fruit Set	Seed Set	Functional Dispersion
Functional Dispersion	✓	✓	-
Orchard Environment			
Bee Sampling Light Intensity (lum/ft)	-	-	✓
Bee Sampling Temperature (°C)	-	-	✓
Monthly Mean Light Intensity (lum/ft)	✓	✓	-
Monthly Mean Temperature (°C)	✓	✓	-
Time of Day	-	-	✓
Wind Rank (1-3)	-	-	✓
Orchard Landscape			
Total Forest Area (m^2)	✓	✓	✓
Mean Forest Distance (m)	✓	✓	✓
Total Meadow Area (m^2)	-	-	✓
Mean Meadow Distance (m)	✓	✓	✓
Total Orchard Area (m^2)	✓	✓	✓
Orchard Management			
Average Trunk Diameter (cm)	-	-	✓
Branch Height (m)	✓	-	-
Dandelion Abundance	-	-	✓
Study Tree Trunk Diameter (cm)	✓	✓	-
Trees Density (trees/m^2)	✓	✓	✓
Distribution	Binomial	Poisson	Normal Continuous
Linear Model	GLMM	GLMM	Multiple Linear
Random Effect	Tree nested in site	Tree nested in site	None

considered this time , as they were judged biologically meaningless among variable groups. Model-averaged partial regression coefficients for each covariate were then estimated as well as their 97.5% confidence intervals. Covariates were considered important if their summed Akaike weights were above 0.60 and significant if their confidence intervals did not include zero. We subjected the most parsimonious 'top models' (i.e. ΔAICc from the best model < 2.0) to standard diagnostic testing (Appendix C).

Results

Field sampling results

A total of 4686 bees were observed on McIntosh apple trees, 858 (18%) of which were wild bees and 3828 (82%) of which were honey bees. The total number of wild bees captured for species level identification on McIntosh by the opportunistic sampler was 261, from which 36 species spanning eight genera and five families (Table 2.1, 2.2) were identified as having visited McIntosh trees. The most abundant and diverse genus of wild bee was *Andrena* (627 individuals and 17 species) and the most abundant species was *Andrena duningii* Cockerell (26 individuals). Bumble bees (176 individuals) and small black bees (55 individuals) were less frequent. The green metallic halictids (three individuals) and *Osmia* (four individuals) were so seldom seen that neither of these groups were considered in the morphospecies level analyses. An additional five species were identified from supplemental surveys during which 183 wild bees were captured, for 41 species in total. The pollination behavior of 293 bees across the different morphospecies was estimated. A total of 137 tree cluster observations were made on the co-occurrence of morphospecies during different times of day and weather conditions. For the fruit set experiment, 2508 flowers were treated for either hand- or open-pollination. Finally, a sample of 698 fruits was gathered for seed set estimates.

Table 2.1: Wild Bee Species List (1/2)

Species list for wild bees caught on McIntosh apples, as well as on late blooming varieties from supplementary surveys in 2012.

		2012		
Family	Species	McIntosh	Late	Total
Andrenidae	*Andrena barbilabris* (Kirby)	10	1	11
	*Andrena carlini (*Cockerell)	11	3	14
	Andena crataegi Robertson	21	4	25
	Andrena cressonii Robertson	1	4	5
	Andrena duningii Cockerell	26	35	61
	Andrena erythronii Robertson	3	0	3
	Andrena forbesii Robertson	9	4	13
	Andrena hippotes Robertson	12	13	25
	Andrena imitatrix Cresson	2	0	2
	Andrena milwaukeensis Graenicher	13	11	24
	Andrena miserabilis Cresson	14	16	30
	Andrena nasonii Robertson	3	9	12
	Andrena nivalis Smith	0	1	1
	Andrena regularis Malloch	4	0	4
	Andrena robertsonii Dalla Torre	0	1	1
	Andrena rufosignata Cockerell	6	6	12
	Andrena rugosa Robertson	23	24	47
	Andrena vicina Smith	8	9	17
	Andrena w-scripta Viereck	1	0	1
Apidae	*Bombus bimaculatus* Cresson	16	1	17
	Bombus griseocolis (DeGeer)	8	3	11
	Bombus impatiens Cresson	8	0	8
	Bombus perplexus Cresson	3	0	3
	Bombus rufocinctus Cresson	0	2	2
	Bombus ternarius Say	2	0	2

Table 2.2: Wild Bee Species List (2/2)

Species list for wild bees caught on McIntosh apples, as well as on late blooming varieties from supplementary surveys in 2012.

		2012		
Family	Species	McIntosh	Late	Total
Apidae	*Bombus vagans* Smith	2	1	3
Colletidae	*Colletes inaequalis* Say	22	13	35
Halictidae	*Augochlora pura* (Say)	3	3	6
	Halictus confusus Smith	3	2	5
	Halictus rubicundus (Christ)	4	2	6
	Lasioglossum cinctipes (Provancher)	3	2	5
	Lasioglossum coeruleum (Robertson)	2	2	4
	Lasioglossum foxii (Robertson)	0	1	1
	Lasioglossum heterognathum (Mitchell)	1	0	1
	Lasioglossum laevissimum (Smith)	3	5	8
	Lasioglossum leucozonium (Schrank)	2	0	2
	Lasioglossum lineatulum (Crawford)	3	4	7
	Lasioglossum quebecense (Crawford)	5	0	5
	Lasioglossum versans (Lovell)	0	1	1
Megachilidae	*Osmia proxima* Cresson	1	0	1
	Osmia pumilla Cresson	3	0	3
Grand Total		261	183	444

Bee Functional Traits Mediating Pollination Services

Globally, there was a significant relationship between morphospecies identity and type of approach to apple flowers (χ^2 = 76.08, p-value < 0.001) as well as foraging type (χ^2= 28.31, p-value < 0.001). Significant Freeman-Tukey deviates (p-value<0.05) indicated that small *Andrena* forage for pollen more than expected and for nectar less than expected, while the opposite was true of large *Andrena*. For foraging approach, only frontworking had expected values greater than five, but none of these deviates were significant. Only honey bees exhibited sideworking more than expected, not making contact with the stigma for 37.8% of all visits. Apart from honey bees, only the small black bees (5.3%) and small *Andrena* (2.6%) sideworked.

Environmental conditions did not significantly influence either bee foraging rates or durations (p-value>0.05 in each case) and neither did the interaction between morphospecies and foraging type (p-value>0.10 in each case). The Kruskal-Wallis rank sum test was globally significant for foraging rate (χ^2 = 111.42, df = 4, p-value< 0.001) and foraging duration (χ^2= 129.52, df = 4, p-value< 0.001), indicating that morphospecies were generally distinct in these behavioral traits. Results from the pairwise comparison of group mean foraging rates, durations and remaining functional traits studied are summarized in Table 3. Bumble bees visited the most flowers per minute and spent the least amount of time on each flower. Both *Andrena* groups and small black bees generally visited the least amount of flowers per minute and spent the most amount of time on each flower. Honey bees were intermediate between bumble bees and other morphospecies.

The RDA analysis revealed significant environmental effects on the number of bee observations in the orchards (F-value=9.05, p-value<0.01). We dropped relative light levels as an environmental parameter because it was not significant (F-value=0.55, p-value>0.10) in a preliminary analysis, but other environmental variables were significant when tested by permutation (n-perm=9999), including

Table 3: Functional Trait Table

Average foraging duration (time feeding from each flower, sec) and foraging rate (number of flowers visited per minute in a foraging bout) per morphospecies, as well as associated standard errors and sample sizes (n) from the pairwise comparison of group means. Significantly different means (p-value<0.05) are indicated with superscript. Additional functional traits listed include: flight period (early: active from April-July; late: April-October); average inter-tegular (IT) span; percentage of visits where contact is made with stigma; if pollen in corbiculae is moistened or left dry; their position along a relative humidity-time of day to temperature gradient (Environmental Index); and if the bee was solitary or social.

	n	Foraging Duration (sec)	Std. Error	Foraging Rate (flr./min)	Std. Error	Flight Period	IT span (mm)	Percent Stigma Contact	Pollen Carrying Habit	Env. Index	Sociality
Bumble bee	43	2.86[a]	0.003	14.28[a]	0.805	Late	5.53	1.00	Moist	-0.38	Social
Honey bee	56	8.73[b]	0.016	6.42[b]	0.429	Late	3.25	0.62	Moist	0.65	Social
Large Andrena	50	17.05[c]	0.037	4.61[c]	0.439	Early	2.69	1.00	Dry	1.12	Solitary
Small Andrena	47	20.94[cd]	0.045	3.63[c]	0.305	Early	2.17	0.97	Dry	0.87	Solitary
Small Black Bee	41	24.90[d]	0.051	3.22[c]	0.314	Late	1.62	0.95	Dry	0.09	Social*

*Albeit that the small black bee group comprised both solitary and social bee species, the morphospecies was approximated as being social given that all species captured were social.

relative humidity (F-value= 3.67, p-value<0.01), time of day (F-value= 4.62, p-value<0.001) and temperature (F-value= 12.04, p-value<0.001). The first RDA axis explained 49.4% of the variance (F-value=20.15, p-value<0.001, n-perm=9999), RDA2 9.7% (F-value= 3.97, p-value<0.01, n-perm=9999), and RDA3 7.4% (F-value=3.02, p-value<0.05, n-perm=9999). Examination of the triplot (Figure 4) shows that bumble bees were most associated with cooler, more humid conditions later in the day, while remaining bee groups variously preferred warmer less humid environments earlier in the day. The first RDA axis and thus the environmental index was interpreted as ordering bee observations along a gradient polarizing time of day and relative humidity against temperature. Higher index values indicate a greater preference for warmer, less humid conditions earlier in the day while the converse is true of lower values.

The Dolédec et al. (1996) permutation model in the fourth corner analysis showed that early flight period, long foraging duration, high percent stigma contact, dry pollen carrying habit, and solitary sociality were positively and significantly (p-value<0.05) related to seed set and fruit set (Table 4 and 5). All these traits characterize the two *Andrena* morphospecies. When the Ter Braak, Cormont and Dray (2012) permutation model was used, however, only percent stigma contact was found to be significant for fruit set (r=0.21, p-value<0.05) and seed set (r=0.22, p-value<0.05). The Ter Braak, Cormont and Dray (2012) method provides the more conservative estimate and points to percent stigma contact, reflective of honey bee sideworking, as the telltale functional trait limiting pollination services in this system. Nonetheless, results from the Dolédec et al. (1996) method were implemented in the calculation of FDis, given that this generates a more comprehensive and nuanced weighting of bee functional diversity relevant to assessing the contributions of wild bees.

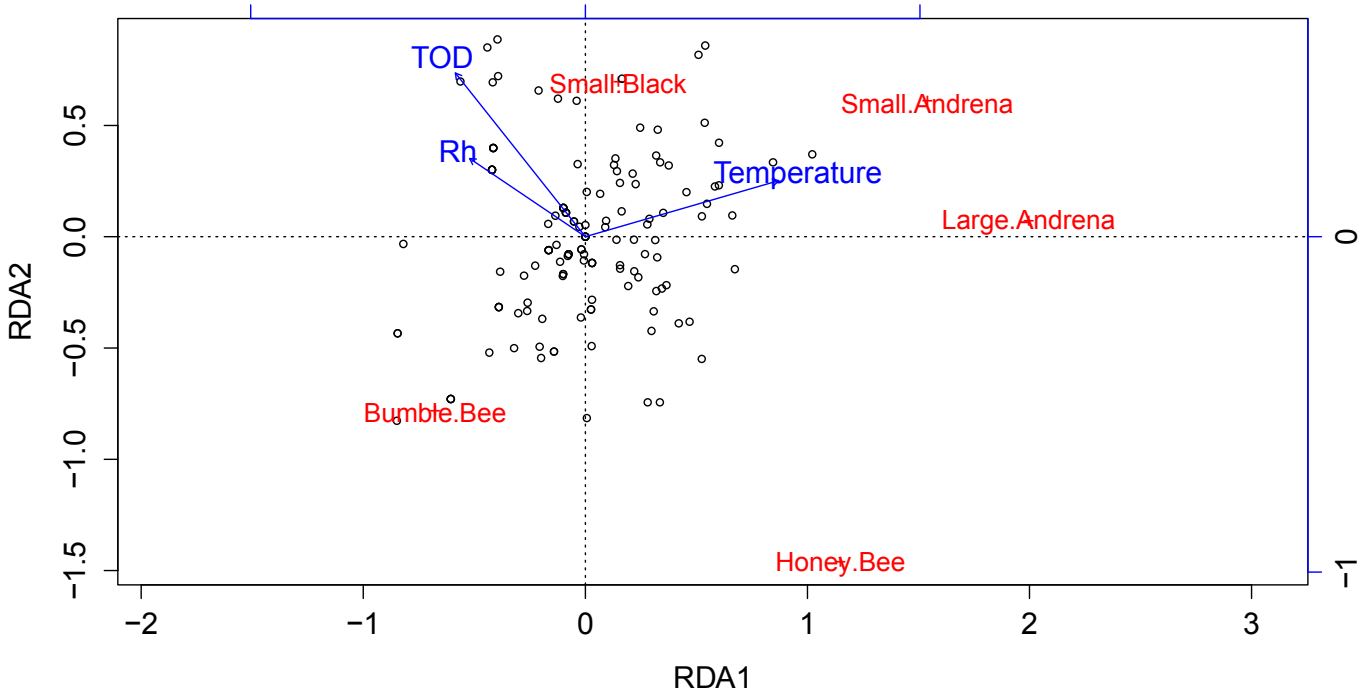

Figure 4: Redundancy Analysis Triplot

Triplot for the redundancy analysis of the bee morphospecies matrix across timed observations periods (black circles) as a function of environmental variables, including time of day (TOD), relative humidity (Rh; %) and temperature (°C). Morphospecies at opposite ends of an RDA axis are negatively correlated in their relative abundances across sites, and those at the same end of an RDA axis are positively correlated. The more distant the morphospecies in the space, the less similar they are in their environmental affinities. The vectors for environmental variables are positively associated with the influence of that variable on the placement of morphospecies in the space. For example, small *Andrena* are negatively associated with bumble bees, inasmuch as small *Andrena* are relatively more abundant in warmer conditions and bumble bees are relatively more abundant in cooler conditions

Table 4: Results from the Fourth Corner Analysis

The table provides the relationships between bee functional traits and the average values of fruit set and seed set across orchards as mediated by the relative abundance of each bee species or morphospecies to which the functional traits are attributed. Correlations (r) are given for continuous variables and F-values for factors (* p-value<0.05; ** p-value<0.01). All factors were assessed as a set of binaries (early/late, dry/moist, solitary/social); those positively related with fruit set and seed set are listed while their converse was negatively related.

Functional Trait	**Fruit Set**	**Seed Set**
Factors	**F-value**	
Pollen Carrying Habit (Dry)	**206.46****	**208.72****
Sociality (Solitary)	**185.89***	**193.53***
Flight Period (Early)	**165.75***	**168.59***
Continuous variables	**r**	
Percent Stigma Contact	**0.21****	**0.22****
Foraging Duration	**0.20****	**0.20****
IT Span	-0.12	-0.10
Foraging Rate	-0.10	-0.078
Environment Index	0.07	0.039

Model Averaging for Pollination Services and Bee Diversity

Open-pollinated flowers set fruit half as often (0.54 fewer times) as hand-pollinated flowers (p-value<0.001), indicating pollen limitation is a significant factor determining fruit set and seed set of open-pollinated flowers across sites (Table 5). Considering patterns of bee diversity (Table 6), it is clear that in this pollen limited system functional dispersion is an important predictor of fruit set (w=0.72, β=0.39±0.37). Since an additional test regressing fruit set in hand-pollinated flowers on FDis was not significant (β=1.67, p-value>0.10), we can conclude that pollen limitation was not confounded by factors such as nutrient limitation or plant growth. Seed set was also positively determined by FDis (w=0.80, β=0.081±0.07), and negatively predicted by distance from meadow (w=0.98, β=-0.12±0.06) and by total orchard area (w=0.76, β=-0.07±0.06). Model averaging indicated that FDis is most positively related to total meadow area (w=1.00, β=0.11±0.05) and total forest area (w=0.86, β=0.05±0.04). All remaining covariates were considered non-significant as they had relative importance scores below 0.60 and confidence intervals associated with the partial coefficients that overlapped zero.

The most parsimonious models in the' top model' set (i.e. ΔAIC from the best model < 2.0) were used to generate land use recommendations enabling increased pollination services (Figure 5). Fruit set was modeled by FDis (β=0.35, p-value<0.05). The FDis linear model (R^2_{adj}=0.70, p-value<0.001) had total meadow area (β=0.13, p-value<0.001) and total forest area (β =0.05, p-value<0.05) as explanatory variables. Seed set was a function of FDis (β=0.09, p-value<0.01) and mean distance from meadow (β=-0.11, p-value<0.001). A level of seed set indicative of high fruit quality (Brault & Oliveira 1995) is associated with a mean distance to surrounding meadow

Table 5: Model Averaging for Pollination Services Analyses

Estimated coefficients (β), their 95% confidence intervals and importance values (*w*) are given per model parameter for fruit set and seed set analyses. Significant terms with confidence intervals not overlapping with zero are in bold.

	Fruit Set				Seed Set			
	w	***β***	**Lower CI**	**Upper CI**	***w***	***β***	**Lower CI**	**Upper CI**
Functional Dispersion	**0.72**	**0.39**	**0.02**	**0.77**	**0.80**	**0.08**	**0.01**	**0.15**
Landscape Metrics								
Mean Meadow Distance (m)	0.33	-0.18	-0.56	0.20	**0.98**	**-0.12**	**-0.18**	**-0.06**
Total Orchard Area (m^2)	0.24	-0.03	-0.42	0.35	**0.76**	**-0.07**	**-0.13**	**-0.01**
Mean Forest Distance (m)	0.33	-0.17	-0.53	0.19	0.21	0.01	-0.05	0.07
Total Forest Area (m^2)	0.29	0.13	-0.24	0.50	0.27	0.03	-0.03	0.09
Orchard Management Covariates								
Trunk Diameter (cm)	0.24	0.03	-0.21	0.27	0.36	-0.03	-0.08	0.02
Branch Height (m)	0.35	0.13	-0.11	0.38	-	-	-	-
Trees Density (trees/m^2)	0.33	0.18	-0.23	0.59	0.30	0.03	-0.04	0.11
Environmental Covariates								
Light Intensity (lum/ft)	0.28	0.07	-0.37	0.52	0.25	-0.00	-0.09	0.08
Average Temperature (°C)	0.28	-0.12	-0.47	0.22	0.20	-0.00	-0.06	0.05

Table 6: Model Averaging for Bee Functional Dispersion Analysis

Estimated coefficients (β), their 95% confidence intervals and importance values (*w*) are given per model parameter for the bee functional dispersion analysis. Significant terms with confidence intervals not overlapping with zero are in bold.

	Bee Functional Dispersion			
	w	***β***	**Lower CI**	**Upper CI**
Landscape metrics				
Total Meadow Area (m^2)	**1.00**	**0.11**	**0.06**	**0.16**
Total Forest Area (m^2)	**0.86**	**0.05**	**0.01**	**0.09**
Mean Forest Distance (m)	0.21	-0.02	-0.07	0.02
Total Orchard Area (m^2)	0.11	0.01	-0.04	0.05
Mean Meadow Distance (m)	0.10	0.00	-0.04	0.05
Orchard management covariates				
Trunk Diameter	0.45	0.04	-0.01	0.08
Trees Density (trees/m^2)	0.16	-0.01	-0.06	0.02
Dandelions Abundance	0.11	0.01	-0.04	0.05
Environmental covariates				
Temperature (°C)	0.19	0.02	-0.02	0.07
Light Intensity (lum/ft)	0.12	-0.01	-0.06	0.03
Wind Rank (1-3 scale)	0.11	-0.00	-0.05	0.05

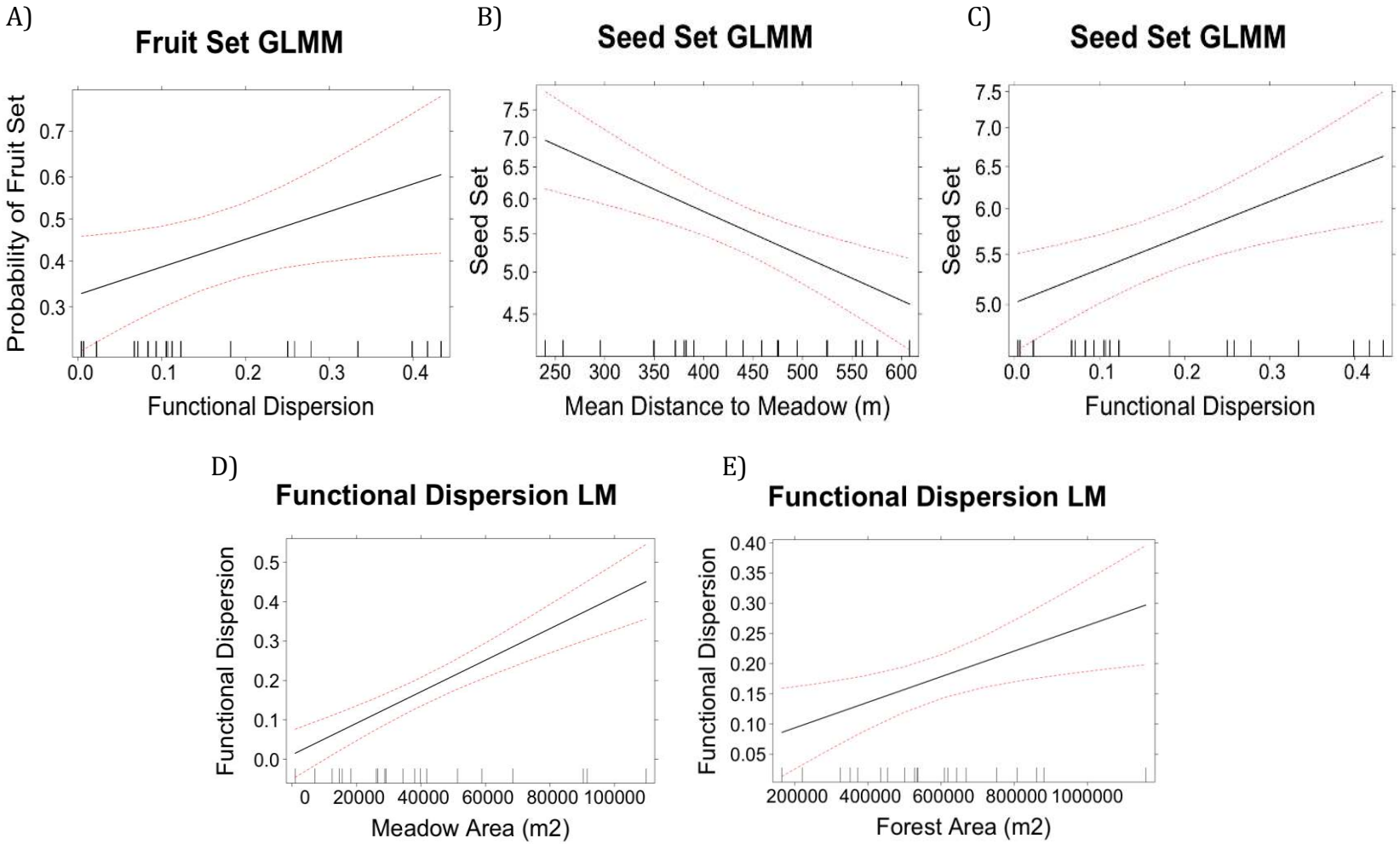

Figure 5: Added Variable Plots

Subfigure A: probability of apple fruit set versus functional dispersion (FDis) GLMM ('lme4' package by Bates et al. 2012); subfigure B and C: apple seed set versus FDis and mean distance to meadow (m) GLMM; subfigures D and E: FDis versus meadow area (m^2) and forest area (m^2) linear model (LM). Dashed lines indicate 95% confidence intervals. An increase in FDis indicates an increase in the relative abundance of different bee morphospecies exhibiting different functional traits associated with apple fruit set and seed set. Graphs were generated with the 'effects' package (Fox 2003) in R 2.15.3.

of 400 m, FDis of 0.25, and in turn a total meadow area of 6 ha, and total forest area of 100 ha in the landscape within a radial distance of 680 m of an orchard. Land use recommendations through fruit set analyses are less transparent given the complications in determining a threshold value for the probability of fruit set as it relates to final orchard yield.

Discussion

The fundamental challenge to apple pollination is the transfer of compatible pollen to flowers during the brief period of floral receptivity in spring. Honey bees have been a mainstay in orchard pollination in that they are easily transportable, loyal to the crops they visit, and can be maintained at high densities at the onset of the season when other pollinators are less abundant (Gould & Gould 1988; Free 1993). Renting honey bee hives seemingly bypasses the need for wild bees as pollinators. . However, our orchards were generally pollen limited despite honey bees having been maintained at recommended densities. To an extent, the quality of supplemental hand-pollination in our experiment comprised entirely of outcrossed pollen would be higher than anything that could be expected from real-world pollination performed by natural pollinators (Aizen and Harder 2007). Establishing that unmanipulated control flowers are setting fewer fruits or seeds per fruit than supplementally pollinated flowers does not mean that service by better pollinators could eliminate the observed pollination deficit. The benefit of wild bee pollination is that it produces greater fruit set and seed set apart from an unnaturally high standard established by the pollen supplementation experiment. It therefore raises the question why honey bees are providing insufficient pollination services, and what aspects of wild bee pollination potentially can compensate for honey bee deficiencies.

The inadequacy of apple pollination performed by honey bees was associated with their well-studied sideworking behavior (Dennis 1979; Robinson 1981; Kuhn & Ambrose 1982; DeGrandi-Hoffman Hoopingarner & Baker 1985; Free 1993; Westwood 1993; Schneider *et al.* 2002, 2004). We observed that honey bees failed to contact the stigma directly for 38% of all floral visits. Robinson and Fell (1981) estimated sideworking to decrease the proportion of fruit set by over 40% in 'Delicious' apples. Techniques have been developed to diminish honey bee sideworking in apple pollination (Schneider 2002), but the utility of such options is questionable in light of recent honey bee declines (Johnson et al. 2009; Currie, Pernal & Guzmán-Novoa 2010; Neumann & Carreck 2010).

Andrena species were the most abundant of the wild bee genera in our orchards and, in contrast to honey bees, they exhibited many traits positively associated with fruit set and seed set. *Andrena* species typically gather pollen to provide for their progeny when visiting apple flowers (Brittain 1933) and in the process make contact with the stigma. Notably, we found that small *Andrena* gathered pollen more often than other bees while large *Andrena* favored nectar visits, however neither group tended to sidework. Both andrenid groups had long foraging durations, which has been positively associated with greater pollen deposition in other systems (Thomson 1986, Thøstesen & Olesen 1996). Pollen adhering to the dense and specialized pubescence of all the *Andrena* species we observed (Michner 2000; Gardner & Ascher 2006) is readily transferred to the stigma (Thorp 2000), much in contrast to pollen packed moist into the hind leg scopae of honey bees (Westerkamp 1991). Discounting the poorly transferable corbicular pollen pellets in honey bees, andrenid bees carry more apple pollen on their bodies (Kendall and Solomon 1973). Although *Andrena* species generally are considered to provide high quality pollination to apple flowers compared with other bees (Brittain 1933; Johnson 1984; Boyle–Makowski 1985; Jacob-Remacie 1989; Cane 1996), they have a slow foraging

rate that may limit their capacity to provide sufficient pollination services during the brief period of apple stigma receptivity.

Bumble bees have functional traits that potentially can offset deficiencies in both honey bees and andrenids. Consistent with observations by Jacob-Remacie (1989), bumble bees in our orchards never sideworked and had the highest floral visitation rates of the bees we observed. Thomson and Goodell (2001) reported that bumble bees deposited more pollen grains on apple stigmas than sideworking honey bees. Since bumble bees can be active at times of day and weather conditions unfavorable for both honey bees and andrenids they can increase the potential for pollination of apples despite the vagaries of spring weather (Paarmann 1977; Pouvreau 1984; Free 1993). This potential, however, is offset by their relatively low abundance; all the bumble bees when apples are in bloom are queens emerging from winter dormancy and have yet to establish hives with abundant workers (Gardner & Ascher 2006).

To summarize, the groups of bees most active in our orchards when apples were blooming had important and complementary differences in foraging behavior and activity patterns that enhanced overall pollination service. Honey bees were abundant but less effective pollinators, *Andrena* efficacious and abundant but slow foragers, and bumble bees fast and able to forage in unfavorable weather but in low numbers. We showed that a measure of this functional dispersion in the bee community positively predicted fruit set and seed set in our orchards. Pollination services were enhanced in orchards with a greater mix of bees exhibiting different, complimentary traits as opposed to orchards that were dominated by honey bees alone.

A key question then is what can be done to increase the abundance of the andrenids and bumble bees that enhance pollination services in apple

orchards. This is basically a question not simply of orchard practice but rather of managing the landscape surrounding an orchard. While honey bee hives can readily be brought to an orchard during the apple bloom, increasing the abundance of wild bees to increase functional dispersion in the bee community requires that suitable foraging resources and nesting sites be provided in the surrounding landscapes prior to and following the apple bloom.

During springtime the understory herbs and shrubs in forests of eastern North America are important pollen and nectar resources for both *Andrena* species and bumble bee queens (Schemske et al. 1978; Bierzychudek 1982; Giles & Ascher 2006; Taki, Kevan & Ascher 2007). The closing of the forest canopy by late May coincides with the end of apple blossoming in our region, at which point the shaded forest understory also becomes less favorable for flowering and bee foraging activity (Romey et al. 2007; Winfree, Griswold & Kremen 2007; Mandelik et al. 2012). Meadows, which have many flowering species later in summer, provide a natural complement to forests (Payette and Oliveira 1989), providing critical floral resources for the *Andrena* species we studied before they enter dormancy in July (Mitchell 1960). Similarly, bumble bees forage in meadow habitats throughout the growing season in addition to using them as overwintering, and nesting sites (Plath 1934; Goulson 2003; Heinrich 2004). Because current bee populations derive from resources available to the prior generation (Danforth et al. 1999, Minckley et al. 2000; Roulston and Goodell 2011), floral resources in forests and meadows one year influence bee abundances in orchards the following year. Therefore, it is necessary to consider the temporal and spatial variability in bee foraging resources within and across generations to promote pollination services by wild bees in orchards.

Although the availability and quality of surrounding habitats mediate the supply of wild bee pollinators, the total area of orchard on the landscape dictates the demand for pollinators. Large expanses of blooming apple trees can simply exceed the capacity of wild bees to provide adequate pollination services (Veddeler Klein & Tscharntke 2006; Holzschuh et al. 2011). We found that as the total area of surrounding orchards increased, seed set in our study orchards decreased. There clearly is a balance to be struck between increasing apple production by expanding existing orchards and enhancing wild bee habitat and thus pollination services per unit area.

Synthesis and Application: Our results can be used to generate management objectives that target the promotion of wild bee diversity and pollination services. Given that greater forest area is difficult to achieve in the short term and that farmers may be reluctant to adopt forest restoration projects requiring extensive cooperation among property owners (McKenzie et al. 2013), we recommend the greater integration of open field habitats in regional agroecosystems. Meadow and grassland restoration has been adopted for bee conservation in Europe (Westrich 1996; Carvelll et al. 2004, 2007; Pywell et al. 2005; Albrecht et al. 2007) and floral strips have been found to promote wild bee visitation to almond orchards in California (Klein et al. 2012). These sorts of land stewardship programs for pollinator refugia, which do not currently exist in Quebec (Ouellett 2013), require coordinated efforts among farmers to avoid negative externalities and common pool resource problems (Zhang et al. 2007). Such landscape scale programs require time to plan and develop, but nothing prevents individual farmers from implementing practices more immediately that might increase wild bee pollinators within their own orchard. For example, since *Andrena* species sometimes nest in orchards for multiple years (Osgood 1989; Miliczky and Osgood 1995; Gardner and Ascher 2006; personal observation 2012), it may be possible to encourage nesting at suitably prepared sites within orchards

(Cane 1996). There is encouraging evidence that farmers will be open to local orchard management that promotes bee diversity (Ahnström et al. 2013).

In conclusion, we are not recommending that orchardists abandon the use of honey bees in apple pollination in favor of wild bees. Rather, we found that a mix of managed and unmanaged bees exhibiting complementary functional traits ensured high apple fruit set and seed set. Orchardists and land use managers can adapt the results of our linear models to quantitatively evaluate whether the spatio-temporal complementarity of wild bee habitat surrounding orchards is sufficient to enhance pollination services.

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General Conclusion

We studied pollination services and bee functional diversity in 20 apple orchards in southern Quebec, Canada. Wild bee functional diversity was positively correlated with the level of fruit set and seed set in these orchards. At the landscape level bee functional diversity was positively correlated with meadow and forest area adjacent to the orchards, and pollination service was negatively correlated with distance from meadows and with the area of surrounding orchards.

My thesis accomplished four main objectives as part of the broader scientific challenge in understanding and responding to the causes and consequences of wild pollinator declines:

Objective 1: To document the biodiversity of bee fauna visiting apple orchards

A total of 213 bumble bee queens comprising nine species were observed in apple orchards in 2011. In 2012, we observed 4686 bees visiting McIntosh apples, 858 (18%) of which were wild bees while 3828 (82%) were honey bees. Thirty-six bee species from eight genera and five families were identified as having visited McIntosh (Chapter 1; Table 1.1, 1.2). The most abundant and diverse genus of wild bee was *Andrena* (627 individuals and 17 species) and the most prominent species was *Andrena duningii* (26 individuals). Indicator species for orchards sites rich in native bees were: *A. duningii* Cockerell, *A. milwaukeensis* Graenicher, *A. miserabilis* Cresson, and *Colletes inaequalis* Say (Appendix B). Supplementary surveys in 2012 found a total of 29 species visiting Cortland and Lobo apples. In total, we identified 41 bee species visiting Quebec apple trees, 16 of which had not previously been reported in the province's orchards (Oliveira, Pion & Paradis 1980).

Objective 2: To quantify the relationship between pollination services and bee functional diversity

Honey bees are by far the dominant pollinator in the orchards, yet we found the system to be generally pollen limited. Orchards that had higher functional diversity, measured by "functional dispersion", had 0.39 times higher fruit set and 0.08 higher seed set. No other environmental or orchard management co-variables were significant in explaining this relationship. Moreover, increased total orchard area and average distance from meadow were both negatively correlated with seed set. We observed a similar "dilution effect" driven by increased orchard area affecting the number of bumble bees we observed in 2011.

Objective 3: To establish the bee functional traits implicated in the positive relationship between bee diversity and pollination service

Complementarity in resource use between unmanaged and managed bees was found to drive the positive relationship between bee functional dispersion and pollination services. Honey bees were abundant but less effective pollinators, given that they did not contact the stigma on 38% of their visits to flowers on McIntosh trees. Bumble bees visited the most number of flowers per minute, never sideworked, tolerated unfavorable weather and were active at distinct times of day from other bees but were rare. *Andrena* were abundant and exhibited many traits positively correlated with fruit set and seed set, but their slow foraging rate and temperature dependency may limit their capacity to cover the entire orchard during the brief period that apples are in bloom. We found that a mix of managed and unmanaged bees exhibiting complementary functional traits in terms of flower visitation rates, activity patterns and pollination quality ensured high apple fruit set and seed set in orchards.

Objective 4: To produce statistical models predicting bee diversity in apple orchards as a function of the surrounding landscape

In 2011, bumble bee abundances in orchards were positively predicted by surrounding meadow area but were negatively related with increasing area of surrounding orchard. Meadows were identified as source patches for bumble bee queens, while orchard area adjacent to the focal orchard diluted their abundance. Hedgerows were identified as a management tool that could be used to promote the abundance of the most dominant bumble bee species, *Bombus impatiens* Cresson, in light of this dilution effect.

In 2012, we found that meadow area and forest area were positively correlated with functional dispersion in the bee community as a whole. We conclude that both forests and meadows are important for supporting the greatest diversity of bees given that these two land cover types complement each other in their seasonal flower availability and together provide a more continuous supply of foraging resources. Using our linear models we estimated that to improve McIntosh apple fruit quality, surrounding meadows should on average be within 400 m of the focal orchard. Moreover, the surrounding landscape within 680 m of the focal orchard should ideally contain 6 ha of meadow area and 100 ha of forest area. Given that the requirement for forest area is ambitious and that meadow area and mean distance were the predominant effects in the relationships with bee diversity in both years and for seed set in 2012, we believe a promising management strategy is to integrate open field habitats into the agroecosystems of our region.

We are now in a much better position to respond to inquiries on the value of wild bee pollination in apple orchards, as well as the habitat requirements of wild bees. The predominance of wild bees in orchards depends on the amount of meadowland and forest area in the surrounding

landscape. We are currently projecting our linear models onto the Montérégie as a whole with the aim of creating a "pollination service map". Orchardists will be able to use this map to evaluate whether the composition and configuration of wild bee habitat surrounding their orchard can maintain wild bee populations to complement honey bee pollination.

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Appendix A: Scale Dependency of Bumble Bee Diversity in Apple Orchards

Introduction

We conducted a pilot study on bumble bees in 2011 to begin to understand the landscape requirements of wild bee pollinators in apple orchards. We examined the abundance and diversity of bumble bees in orchards, and related abundance to metrics describing the surrounding countryside. Bumble bees were chosen as a study organism because they have the largest foraging ranges of the bees in our area (Greenleaf et al. 2007) and are thus the most implicated in landscape-level dynamics. The investigation is in itself exploratory and was used as a stepping-stone to the preparation for the 2012 sampling season. The analyses comprised part of the term project for BIOL 6077: Numerical Ecology at University of Montreal taught by Dr. Pierre Legendre. The project was essentially an inquiry into which landscape metrics can be used to explain the variance in both total and interspecific bumble bee abundances in orchards throughout a spatially mixed habitat matrix composed of suburban, rural and forest elements.

Method

Bumble bee Sampling

Bumble bees were sampled in 12 apple orchards across the Montérégie, Quebec, Canada, from May 21st to 24th, 2011 when the orchards were in full bloom. Sampling occurred between 8:55 AM to 6:35 PM Eastern Standard Time, temperatures were above 15°C, and environmental conditions were clear to lightly overcast. We recorded cloudiness class, time of day and wind speed class during sampling (Table A1).

Bumble bees were surveyed by a pair of collectors with sweep nets traversing a one-hectare sampling plots for one hour. Bumble bees were only counted if captured on either apple flowers or understory herbs. Species level identifications were made in the field when possible using Laverty and Harder (1988) and samples thereafter released; unidentifiable individuals were frozen and stored for later identification.

Vegetation surveys were conducted because higher apple floral densities may reduce the likelihood of encountering bees, and because dandelions can act as a distraction from apple flowers for bees (Free 1968). Apple flower abundances per site were estimated by counting the flowers on a branch containing approximately one tenth of the flowers on the apple tree, and repeating this process for five trees. The number of trees in the hectare plot was also counted to yield tree density estimates. Dandelion abundance in the orchard was scored on a 1-3 scale.

Landscape Characterization

Landscape analyses were made with ArcGIS 10.0 (Environmental Systems Research Institute, Redlands, CA, USA). We interpreted land classes from a composite of satellite and orthophoto imagery taken from the *Ministère des ressources naturelles et de la faune du Québec* (30 cm resolution) for the Montérégie region in 2009, and *Imex ltée* (30 cm resolution) for Mont Saint Hilaire in 2007. Interpretations were cross-referenced with data from the Quebec Forest Inventory Service (MRNF 2003) and were verified by field-based validation in June 2012. We assessed patterns of land cover in concentric circles with radii of 250, 500, 750 and 1000 m centered around each of our one hectare sampling plots.

Table A1: List of Variables

Variable	Description
Landscape Metrics (m^2)	
Agriculture	Fields of buckwheat, corn, hay, mixed vegetables, soy, as well as flower nurseries and vineyards
Forest	Coniferous, deciduous and mixed forested areas larger than 1 Ha in size
Forest edge	Area ±5 m from the forest edge
Hedge	Strips of mid-succession habitat lining roads and/or fronted on either side by agricultural fields, no more than 10 m in width
Meadow	Agricultural land having been abandoned between one to five years, with no more than 50% grass cover and largely dominated by forbs and brambles
Suburb	Total area of domestic homesteads, their gardens and lawns, excluding intervening roads
Orchard	Agricultural areas dedicated to apple production
Environmental Covariates	
Cloudiness	Rank of the degree of cloudiness from 1-3
Wind speed class	Rank of the degree of windiness from 1-3
Time of day	Time of day when bee sampling began
Orchard Management	
Dandelion rank	Increasing rank of dandelion cover in orchard understory from 1-3
Apple Flower Abundance	Estimated number of apple flowers in our sampling plot
Tree density	Number of trees per m^2

Results and Data Analysis

Outline of Data Matrices

A total of 213 individual bumble bee queens were captured across the 12 sites, comprising nine species (Table A2). The resultant 12 x 9 (*n* x *p*) data matrix, **Y**, consisted of 64.8% zero values with a very asymmetric abundance class distribution. This was due to the strong dominance of *Bombus impatiens* Cresson, comprising 171 (80.2%) of all bumble bees captured. The row sums of matrix **Y** produced vector **y** describing the total abundance of bumble bees found in each orchard. Matrix **Y** was analyzed by multivariate means to assess the covariance of bumble bee species; a univariate approach was taken with regards to **y**.

Three different sets of explanatory variables were used in data analyses, including: landscape variables, environmental variables, and biotic variables (Table A1). Landscape variables were consolidated in four separate explicative matrices ($\mathbf{X}_{(1000,\ 750\ldots)}$) and included the total area (m^2) of landscape classes across the 12 sites for each spatial scale (r=1000m, 750m…) considered. Environmental variables consisted of the time of day, temperature (°C), cloudiness class (ranked from 1 – 3), and wind speed class (ranked from 1 – 3). Finally, biotic variables comprised dandelion abundances (ranked from 1 – 3) in the orchard understory, the number of apple flowers estimated within the sampling hectare, and the apple tree density (trees/m^2). Both environmental and biotic variables were treated as covariable sets in partial methods of linear analysis to emphasize the role of the landscape in structuring the bee community.

Table A2

Species list for bumble bees caught in apple orchards in 2011.

Family	*Species*	Total
Apidae	*Bombus bimaculatus* Cresson	17
	Bombus borealis (Kirby)	1
	Bombus citrinus (Smith)	1
	Bombus griseocolis (DeGeer)	4
	Bombus impatiens Cresson	172
	Bombus rufocinctus Cresson	1
	Bombus ternarius Say	10
	Bombus terricola (Kirby)	1
	Bombus vagans Smith	3
Grand Total		210

Bumble bee abundance as a function of landscape metrics

Statistical analyses were conducted with R version 2.15.3 (R Project for Statistical Computing release 2013-03-01 http://www.r-project.org).

Linear regression was used to understand the functional relationship between landscape metrics and the vector describing bumble bees abundances across sites. To this end, we implemented ranged major axis (RMA) regression (package 'lmodel2'; Legendre 2013) because: both dependent and independent variables were randomly sampled and measured with error, the data distributions exhibited bivariate normality, the response and explanatory variables were not in the same physical units, and it was reasonable to assume the error variance on each axis was proportional to the variance of the corresponding variable (Legendre & Legendre 2012). Matrices $\mathbf{X}_{(1000,\ 750\ldots)}$ were each subjected to forward selection (package 'packfor; Dray, Legendre & Blanchet 2012) separately in relation to bumble bee abundance **(y)** with a minimum alpha-to-enter of 0.05 for each of the four concentrically circular areas.

For a buffer radius of 1000 m, only the total area of surrounding orchard predicted bumble bee abundance. The relationship was best fitted by a negative power function; both the abscissa and ordinate were log transformed to linearize the relationship. The RMA regression on log-transformed data had an R^2_{adj} of 0.74, and a p-value (999 permutations) of 0.001. The area of meadowland was strongly positively correlated with the number of bumble bees found across sites for radii of both 750 m (R^2_{adj} =0.60, p-value<0.01) and 500 m (R^2_{adj} =0.61, p-perm<0.01). Finally, at a radius of 250 m, there was a strong negative relationship between total orchard area and bumble bee abundances (R^2_{adj} =0.60, p-perm<0.01). These relationships are illustrated in Figure A1.

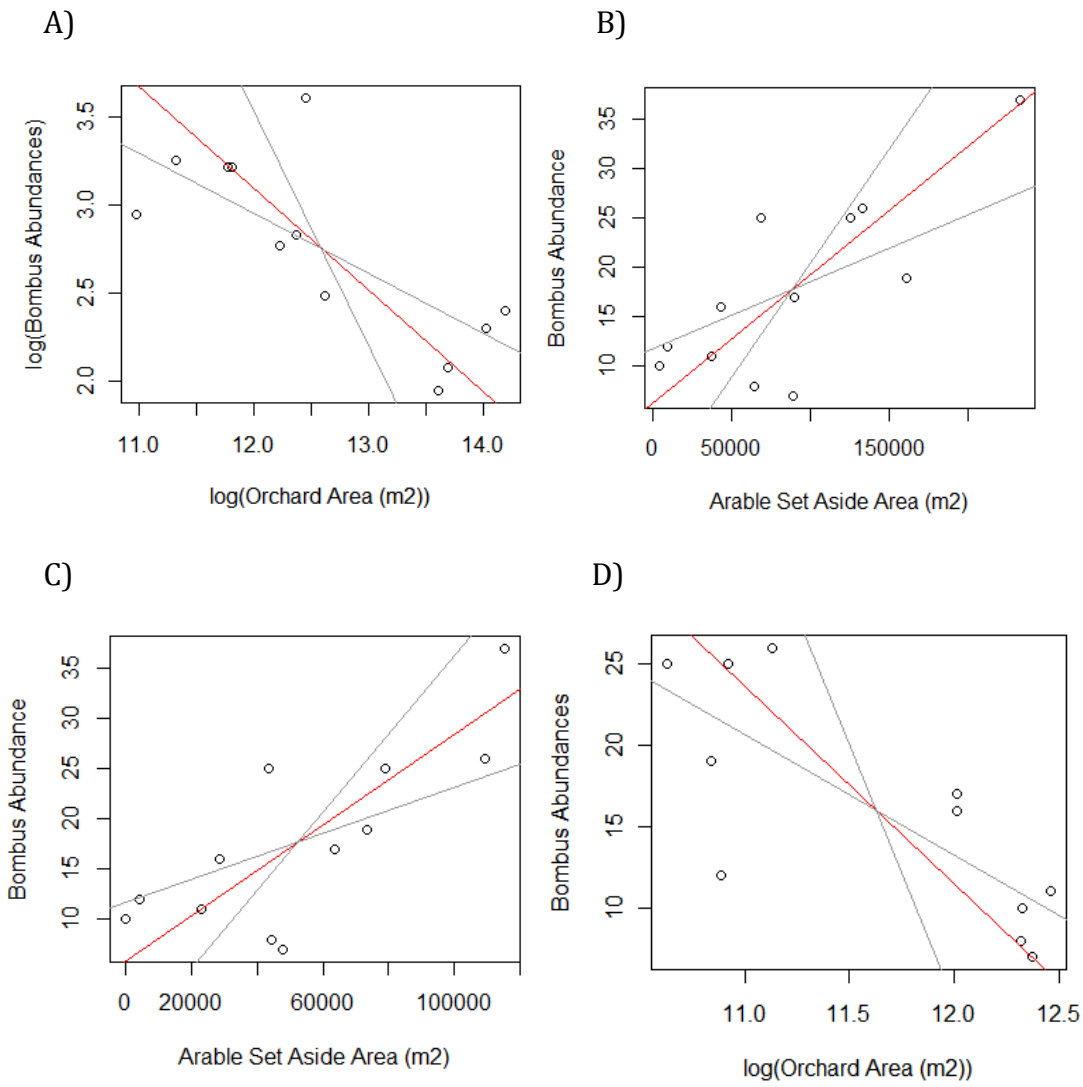


Figure A1: Ranged Major Axis Regression

Ranged major axis linear regression was used to explain the functional relationship between bumble bee abundances and landscape metrics. *Sub-figures a, b, c*, and *d*, were calculated considering landscapes within radial distances of 1000 , 750, 500 and 250 m from sampling sites, respectively.

Interspecific bumble bee landscape preferences

For the multivariate analysis of matrix **Y**, we used transform-based principal components analysis (PCA) to investigate the covariance of bee species vectors across sites (package 'vegan'; Oksanen et al. 2012). We employed transform-based methods because the original matrix **Y** would be prone to the 'double zero problem' when assessing the symmetrical Euclidean distance in a traditional biplot. Type two scaling was used so that the correlation between species would be approximated by the angle between descriptor vectors. We considered four distance measures in the analyses: chi-square, chord, Hellinger, and profiles of relative abundance (Legendre & Gallagher 2001). All these transformations preserve their respective distance metrics when projected with linear methods of analysis in Euclidean space (Legendre & Legendre 2012), including PCA. In interpreting the effects of these alternative distance metrics in the PCA biplots we considered their unique statistical properties, such as the chi-square distance's emphasis on rare species (Legendre & Legendre 2012). Both principal axes in the resultant PCA's were tested for their importance by comparing the amount of variance explained to that of a broken-stick distribution (package 'BiodiversityR'; Kindt & Coe 2005).

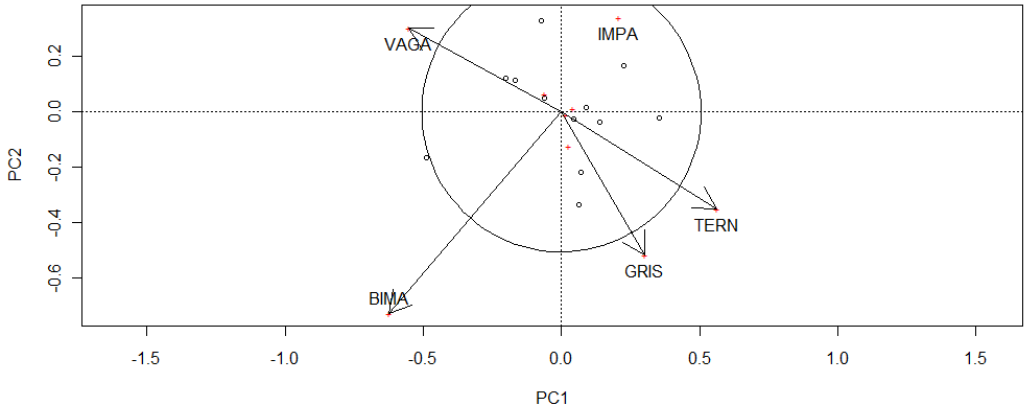
The largest amount of variance was explained by principal components projected with the Hellinger transformation, where both axes were significant and explained 46.3% of the variance on axis 1 and 30.9% on axis 2. We assessed correlation biplots with an equilibrium circle to tease out the meaningful species vectors structuring the ordination (Legendre & Legendre 2012). The biplot of the Hellinger transformation (Figure 2.a) revealed a strong negative correlation between *B. impatiens* and *B. bimaculatus* Cresson as well as between *B. vagans* Smith versus *B. griseocolis (*DeGeer) and *B. ternarius* Say. This ordination provided the most interpretable and significant results, so we chose this Hellinger transformed

data for further analysis with asymmetric linear methods.

We assessed divergent patterns of distribution among bumble bee species in terms of the set of landscape variables at the different spatial scales. To this end, we adopted partial transform-based redundancy analysis (RDA; package 'vegan'; Oksanen et al. 2012). This method can best be explained as a regression of the Hellinger transformed PCA with matrices $\mathbf{X_{(1000,\,750...)}}$ while controlling for a set of covariables. The original site scores as opposed to fitted site scores were projected onto the triplot to preserve the correlations represented in the initial PCA as much as possible. We used a variation inflation factor cutoff of 10 to minimize collinearity between explanatory variables. Although it would have been appropriate to project the variables in $\mathbf{X_{(1000,\,750...)}}$ directly onto the Hellinger-transformed PCA, we favoured RDA because it allows for controlling inter-site variation through partial methods. This was especially important as weather conditions, time of day, and floral densities of both dandelions and apple blossoms could not be held constant and are known to effect bumble bee activity (Boyle & Philogene 1983, 1985; Free 1968).

Matrices $\mathbf{X_{1000}}$, $\mathbf{X_{750}}$, $\mathbf{X_{500}}$ and $\mathbf{X_{250}}$ were each used as explanatory data sets in separate RDA's to model the Hellinger transformed PCA. Time of day and weather conditions, which yielded significant results across scales, were consolidated as matrix **W** and used as covariates for each model. No significant results were found for either $\mathbf{X_{250}}$ or $\mathbf{X_{500}}$; the two larger spatial scales yielded qualitatively similar relationships, which are illustrated by the relationship at a 1000 m radius Figure 2.b. The polarization between *B. ternarius* and *B. griseocolis* versus *B. vagans* is preserved along RDA1, while *B. impatiens* is set against *B. bimaculatus* along RDA2. The overall ordination has a semipartial R^2 of 0.69 and a p-value<0.01 (999 permutations). Both RDA1 and RDA2 were significant by permutation (999 permutations) when

A)



B)

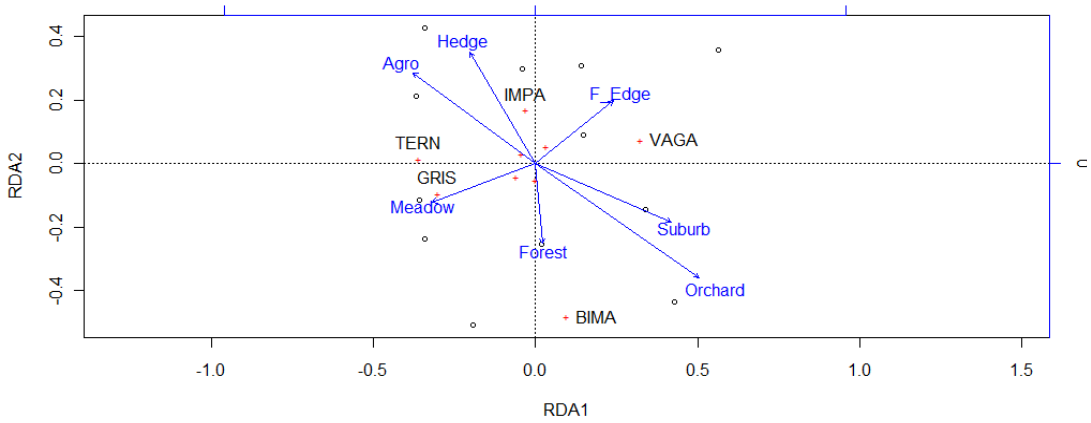


Figure A2: Multivariate Analysis Results

Sub-figure 2.a represents the biplot of a Hellinger transformed principle component analysis with species vectors represented by red crosses and sampling sites by black circles. An equilibrium circle has been included to select the most important species vectors influencing the distribution of sites. Black arrows represent chosen species vectors; *Bombus impatiens* was an exception because it only nearly reaches the perimeter of the circle but was still highlighted given its abundance across the region. *Sub-figure 2.b* is the triplot of a Hellinger transformed RDA between matrices for bumble bee abundances and landscape metrics. Note that fewer species are illustrated to maintain clarity in the figure. The blue vectors each represent explanatory variables describing the total area occupied by each landscape class within 1000 m of sampling sites. BIMA: *B. bimaculatus*; IMPA: *B. impatiens*; *B. griseocolis*; *B. ternarius*; VAGA: *B. vagans*.

treating the previously tested axis as a covariable (RDA1: R^2_{adj} of 0.23, p-value<0.01; RDA2: semipartial R^2 of 0.25, p-value<0.01). Those landscape vectors making the smallest calculated angle from their associated species vector were seen as modeling the polarization between the two bumble bee groups. The forest edge is most correlated with *B. vagans* and arable set aside (denoted as "Meadow") is most in line with *B. ternarius* and *B. griseocolis*; *B. impatiens* is most associated with hedge habitat and *B. bimaculatus* the forest proper.

Discussion

The focus of this pilot study was the scale dependency of landscape preferences by bumble bee queens foraging in apple orchards during the bloom period in spring. Bumble bee abundances were negatively associated with the total area of orchard within a radius of 1000 m around a sampling site, which suggests a 'dilution effect' as the ratio of orchard to favorable habitat increases. Veddeler Klein and Tscharntke (2006) have observed similar bee dilution effects in coffee plantations, as have Holzschuh et al. (2011) in canola.

For a given amount of orchard this dilution effect can be dampened by increasing the area of arable set aside, which is associated with greater bumble bee abundance at distances of 500 m and 750 m. This makes sense in that bumble bees use meadows as nesting grounds (Plath 1934; Goulson 2003; Heinrich 2004) and as foraging sites throughout their lifecycle from spring to late summer (Mandelik et al. 2012). Our analyses indicated bumble bee abundance was favored by not only meadows but also by semi-natural areas (hedgerows, roadside verges), forest and forest edge habitats. Hedgerows were associated with the most abundant bumble bee in the orchards, *B. impatiens*, and are recognized for their potential habitat value for wild bees in general (Hanon & Sisk 2009). Meadows and hedgerows are more

likely to be adopted in farm management than extensive forest restoration projects demanding a high degree of cooperation among property owners (McKenzie et al. 2013).

Like many others (Kennedy et al. 2013), our study has centered on the movement of bees from surrounding habitats to crop fields, but it is noteworthy that considerably less attention has been paid to the countervailing effect of mass flowering crops on the pollination of flora in landscape elements from which bees are derived (Rand, Tylianakis & Tscharntke 2006). For example, Holzschuh et al. (2011) found that bumble bee abundances in grasslands were diluted by adjacent oilseed rape fields, which reduced the seed set of *Primula veris* Linnaeus in the grasslands. Given that apple trees in our region bloom adjacent to and concurrently with forest herbs like *Trillium grandiflorum* (Michaeux) Salisbary, which is bumble bee pollinated (Irwin 2000) and listed as vulnerable in Quebec (Gilber 2005), the interplay between orchards, meadows, and the reproductive success of spring flowers in remnant forests would be an interesting venue for further research.

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Appendix B: Multiple Factor and Indicator Value Analyses

Introduction

It is common in surveys of bee biodiversity to use a combination of bee catching methods to control for potential sampling biases (Roulston, Smith & Brewster 2007). For instance, sweep netting has been complimented by either pan sampling (Fetridge, Ascher & Langellotto 2008; Hopwood 2008; Hendrix, Grundel & Heard 2010; Kwaiser & Heard 2010; Tonietto et al. 2011) or stationary timed counts of bee frequencies (MacKenzie & Eickwort 1996; Winfree et al. 2008). Species richness or abundance values across sampling techniques have typically been collapsed for statistical treatment without considering how biases in sampling methods may vary across sites and taxa.

We adopt multiple factor analysis (MFA; Abdi et al. 2013) as a novel means of qualitatively assessing discrepancies in count estimations. Two observers sampled bee diversity present in 20 apple orchards: one who surveyed the orchard rows opportunistically while the other was stationary and examined the frequency of bee visitation over a predefined set of trees. Multiple factor analysis enabled the cross-referencing of opportunistic and stationary counts into a multivariate space of compromise, affording an unbiased estimate of tree and site level diversity. It was then possible to correlate response and environmental variables of interest with a composite bee diversity gradient. Indicator value analysis (Dufrêne & Legendre 1997) was used to identify bee species significantly associated with orchards that had high wild bee abundance.

Method

Multiple factor analysis (package 'FactoMineR'; Husson et al. 2013) considers various data tables that measure variables on the same set of

observations, using an extension of principle component analysis (PCA; Abdi et al. 2013; Legendre & Legendre 2012). The analysis takes two primary response matrices and standardizes them according to the eigenvalues of their respective first principle components. The two matrices are then concatenated and subjected to a global PCA. The primary matrices used in this MFA were the morphospecies observations across sites for both the stationary and opportunistic observers. Counts for the stationary observer were collapsed across trees and across tree clusters examined per site. These data frames were subjected to a Hellinger transformation to alleviate the dominating effect of honey bees (Legendre & Gallagher 2001), which were found to mask variance in the wild bee community. To understand the synthetic axes upon which site objects (orchards) were being ordered, we calculated the correlation, level of significance and contribution of each morphospecies variable to the dimensions of the joint PCA.

Supplemental tables were projected onto the global PCA, and contained variables pertaining to a) landscape metrics, b) environmental covariates, c) orchard management covariates, and d) pollination services (see Chapter 1, Table 6). The objective was to map explanatory datasets onto the space of compromise delimited by the two modes of observation. The supplemental tables do not contribute to the ordering of site objects along ordinal axes; the method is akin to a correlation as opposed to a regression analysis. Pollination services were estimated as average fruit set and seed set values per site. We calculated the correlation and level of significance of each continuous supplemental variable with the principle components created by the primary matrices. There were two nominal variables added to the analysis, one considering orchard type (dwarf versus standard rootstocks) and the other the degree of suburbanization (predominantly suburban versus rural setting). They were subjected to v-tests (Lebart et al. 2005), which are similar to a one-way analysis of variance of object coordinates for each axis as a function of the categorical variables.

The MFA divided orchards into a two subsets: 12 sites clearly dominated by honey bees and eight sites with a greater proportion of native bees. Indicator value (IndVal) analysis (Dufrêne & Legendre 1997) with the 'labdsv' package (Roberts 2012) was applied to a 20 site by 31 wild bee species matrix, which was classified according to a factor coding for species rich sites. It was then possible to test by permutation which of the species was characteristic of orchards more rich in wild bee species.

Results

The first dimension (PC1) of the resultant joint PCA explained 64.3% of the variance, and dimension two 14.1%. The bee vectors with the highest contribution to PC1 were honey bees (stationary: 5.94; opportunistic: 11.25), large *Andrena* (stationary: 28.18; opportunistic: 12.84) and small *Andrena* (stationary: 10.15; opportunistic: 21.14). Examination of interspecific bee correlations (Table 1) and a correlation circle (Figure 1) show that: 1) honey bees are polarized against native bees in the analysis and 2) orchards are ordered along this gradient. The 12 sites with negative object scores on PC1 are dominated by honey bees, whereas the eight sites with positive values have a greater proportion of native bees. Inspection of the partial points of the individual factor map (Figure 2) reveal a high concordance between the opportunistic and stationary observers for sites dominated by honey bees, but less so as bee diversity increased. The first principle component of the ordination was extracted, which was highly correlated with the functional dispersion (see Chapter 1) of the bee guild ($r=0.967$). Of the vectors from the supplemental tables, those with significant correlations with PC1 are seed set ($r=0.480$, p-value<0.05), fruit set ($r=0.502$, p-value<0.05), temperature ($r=0.565$, p-value<0.01), trunk diameter (0.575, p-value<0.01) and meadow area ($r=0.722$, p-value<0.0001). The R^2 scores for the nominal vectors were 0.46 (p-value<0.0001) for suburbanization and 0.25 (p-value<0.05) for orchard type. Both "suburban" and "standard tree rootstock orchard" factors

Table B1: Results from Multiple-Factor Analysis

The correlations and contributions of each bee morphospecies to PC1 of the multiple factor analysis are given, as well as the level of significance for each correlation (***p-value<0.001, ** p-value<0.01, * p-value<0.05)

	Stationary		Opportunistic	
	Correlation	Contribution	Correlation	Contribution
Bumble bee	0.60**	2.30	0.39	0.99
Honey bees	-0.94***	5.94	-0.94***	11.25
Large *Andrena*	0.87***	28.18	0.81***	12.84
Small *Andrena*	0.74***	10.15	0.94***	21.14
Small black bee	0.46*	3.40	0.63**	3.80

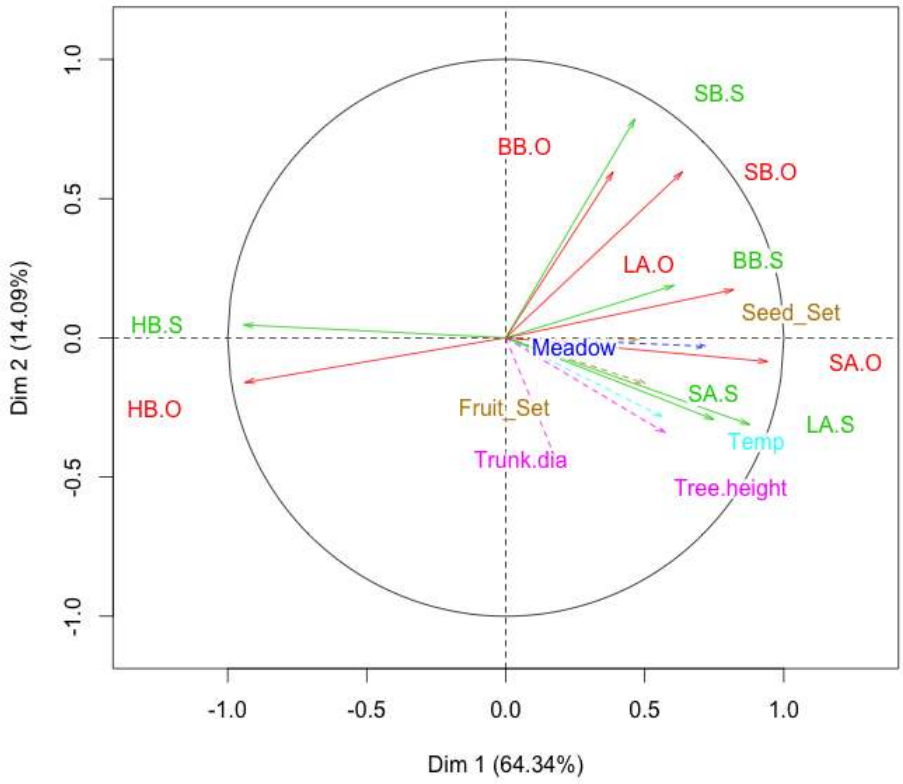

Figure B1: Correlation Circle from Multi-Factor Analysis

Only vectors with correlations greater than 0.50 with PC1 or PC2 are presented in the correlation circle. *Stationary observer*: green; *opportunistic observer*: red; *landscape metrics*: blue; *ecosystem services*: copper; and *landscape management*: pink. Coding for the opportunistic (O) and stationary (S) observers follow: BB: bumble bee, HB: honey bee; LA: large *Andrena*; SA: small *Andrena*; SB: small black bee.

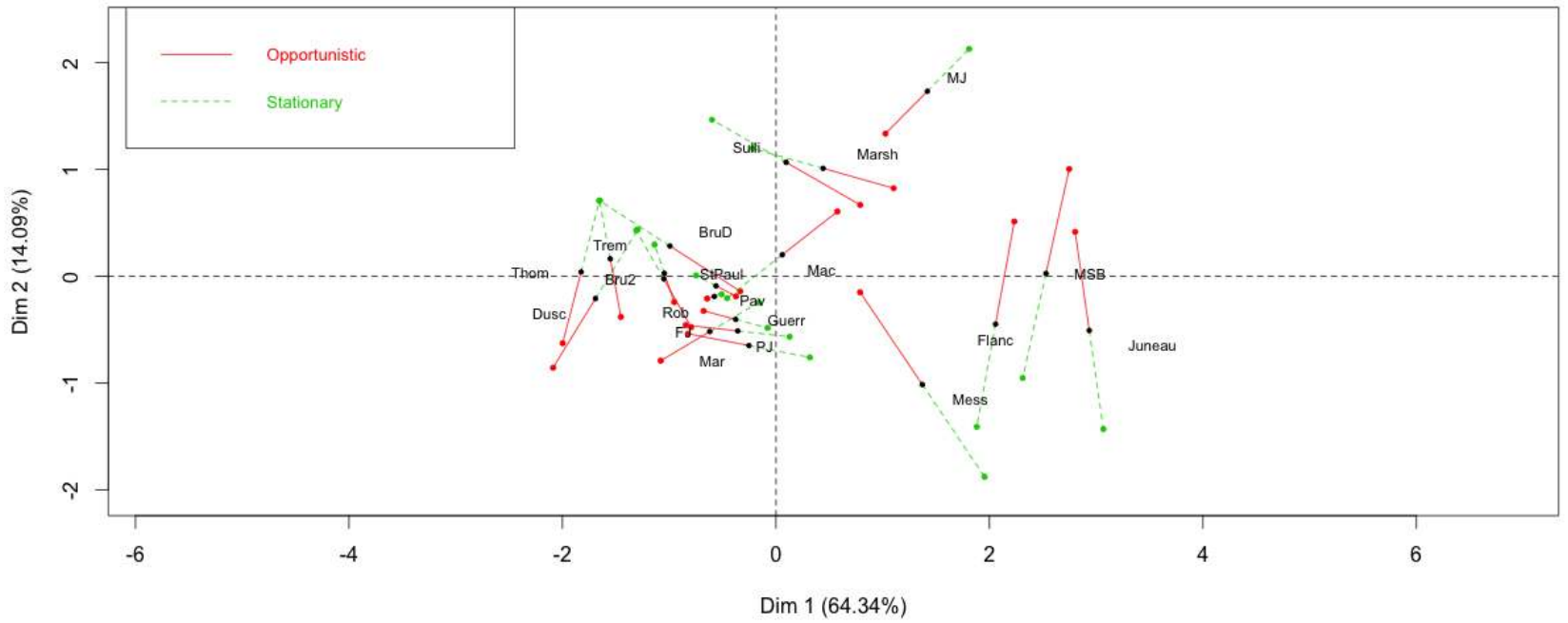

Figure B2: Individual Factor Map

The individual factor map presents orchards (black circles) ordered along the first two dimensions of the global principle components analysis. They are placed at the center of the partial points for the opportunistic and stationary observers, i.e. where either observer alone would have placed the orchards in the ordinal space. The distance between the partial points indicates the degree of concordance between the two modes of observation.

had positive PC1 values while “rural” and “dwarf tree rootstock orchard” had negative PC1 values.

The IndVal analysis identified *Andrena duningii* Cockerell (IndVal: 0.61; p-value<0.05), *A. milwaukeensis* Graenicher (IndVal: 0.56; p-value<0.05), *A. miserabils* Cresson (IndVal: 0.55; p-value<0.05), and *Colletes inaequalis* Say (IndVal: 0.60; p-value<0.01), as all being significantly associated with species rich sites (i.e. those orchards with positive PC1 scores).

Conclusion

The results indicate a high degree of concordance between the stationary and opportunistic observers for sites dominated by honey bees, with partial points held in close proximity for negative values of PC1 (Figure 2). This is expected given that a single, easily recognized species was being identified. The distance between partial points increases for positive values of PC1, for orchards with higher bee diversity. By examining the correlation circle (Figure 1), the discrepancy seems most apparent in the observation of bumble bees and small black bees. Given that the opportunistic observer was mobile, the individual would have been more apt at intercepting fast moving bumble bees and perceiving their larger bodies from afar. The stationary observer then serves as a ready complement, being more capable of examining the diminutive small black bees when standing still. However, the partial points for the opportunistic observer are more closely aligned across species rich sites, suggesting more consistent estimations from this individual.

Results from the supplemental tables indicate that seed set and fruit set in apple orchards are positively correlated with increasing native bee diversity, and specifically the abundance of *Andrena* species. Native bees are highly associated with increasing meadowlands in the landscapes surrounding sites, as well as warm environmental conditions. They are more likely to be found in orchards in suburban settings that have standard tree rootstocks with large basal trunk diameters. On the other hand, orchards with high density dwarf rootstocks dominated by honey bees in rural settings are associated with lower fruit set and seed set.

The IndVal analysis identified species in the large *Andrena* (*A. duningii* Cockerell, *A. milwaukeensis* Graenicher, *C. inaequalis* Say) and small *Andrena* (*A. miserabilis* Cresson) groups as being significantly associated with sites rich in wild bee species, and thus by association with pollination services. These *Andrena* species were all recognized by Gardner and Ascher (2006) for having high pollinator potential for orchards in eastern North America. This was because of their seasonal phenology, which coincides well with the apple bloom, their preference for apple pollen, their large size, and their abundance. *Colletes* are likewise abundant in the early spring and nest in aggregations of thousands of individuals but are primarily associated with *Acer* Linnaeus (maple), and require more specific nesting substrates (Batra 1980).

In conclusion, results from the MFA are very consistent with the more computationally intensive regression analyses found in the main body of the thesis. Multiple Factor Analysis can strengthen ecological surveys through a nuanced inspection of observation biases, while generating synthetic and 'clean' ordinal axes with which environmental and response variables can be correlated.

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Appendix C: Model Diagnostics

Standard diagnostic plots and parameters were examined for each of the best models generated through model averaging for the fruit set generalized linear model (GLMM), seed set GLMM, and functional dispersion linear model (LM).

Fruit Set Generalized Linear Mixed Model

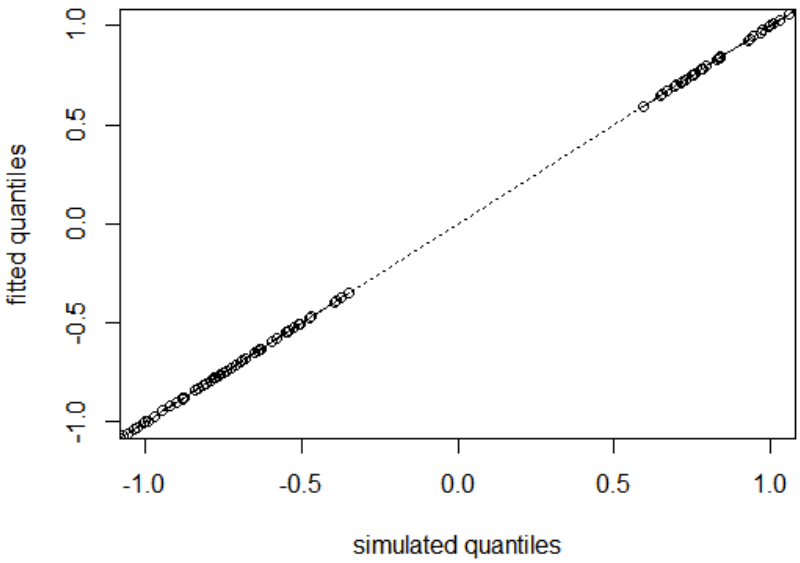


Figure C1: Simulated Quantile-Quantile Plot- Fruit Set Analysis

We used a logistic regression quantile-quantile plot to assess whether the error distribution of the data is modeled correctly and to detect more general departures from model assumptions (Zuur 2009). The graph compares quantiles of residuals assuming the fitted model is the true model, against the actual quantiles of the residuals from the fitted model. The method of Landwehr et al. (1984) was used with code taken from Zuur (2009). Deviations from the 1:1 line would indicate lack of goodness-of-fit. Model assumptions are thus satisfied.

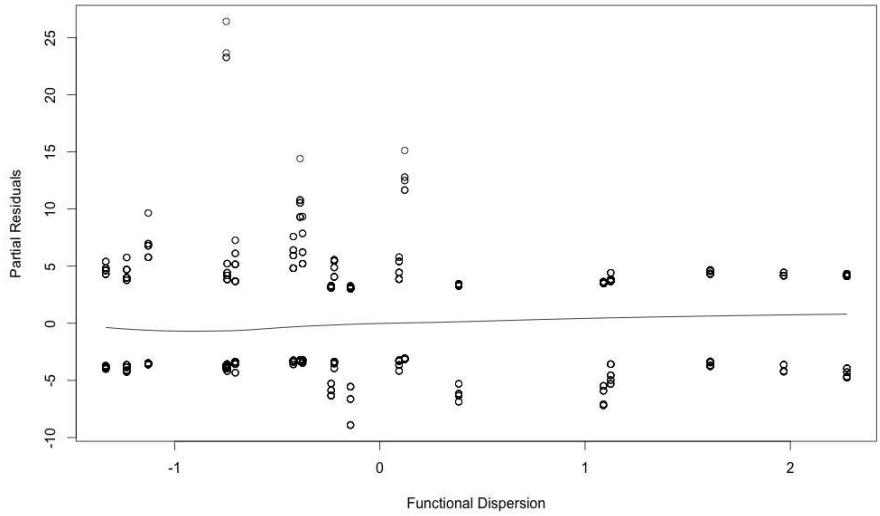


Figure C2: Partial Residuals Plot - Fruit Set Analysis

Partial residual plots consist of a graph of a value of the covariates plotted against their residuals (Zuur 2009). This graph is used to verify the linear assumption of the covariate in logistic regression. Model assumptions are thus satisfied for functional dispersion in the fruit set GLMM.

Seed Set Generalized Linear Mixed Model

The GLMM for fruit set with the lowest AIC score was subjected to model testing following Bolker et al. (2009). The Pearson residuals did not exhibit overdispersion (Chisquare: 558.44, p-value=1.00), and the deviance of the model was 665.55.

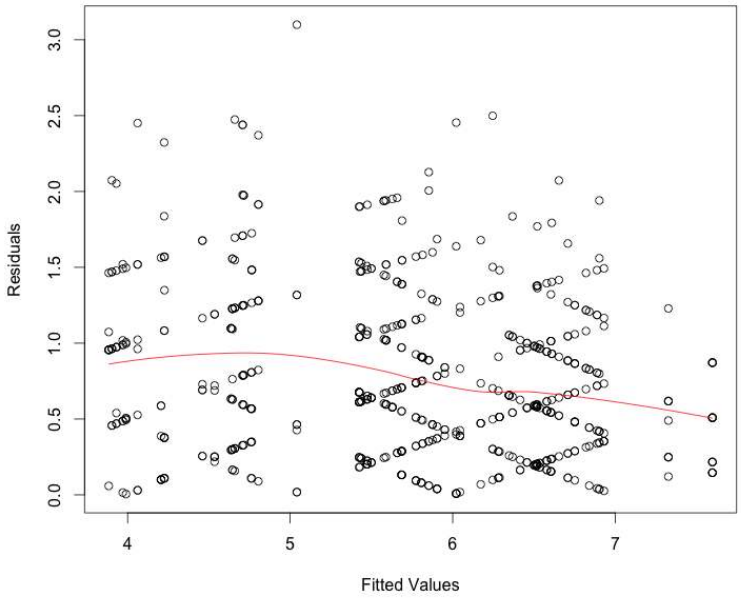


Figure C3: Residuals versus Fitted Values-Seed Set Analysis

When the residuals were plotted against the fitted values, there were no clear indications of heterogeneity in the data.

Functional Dispersion Linear Model

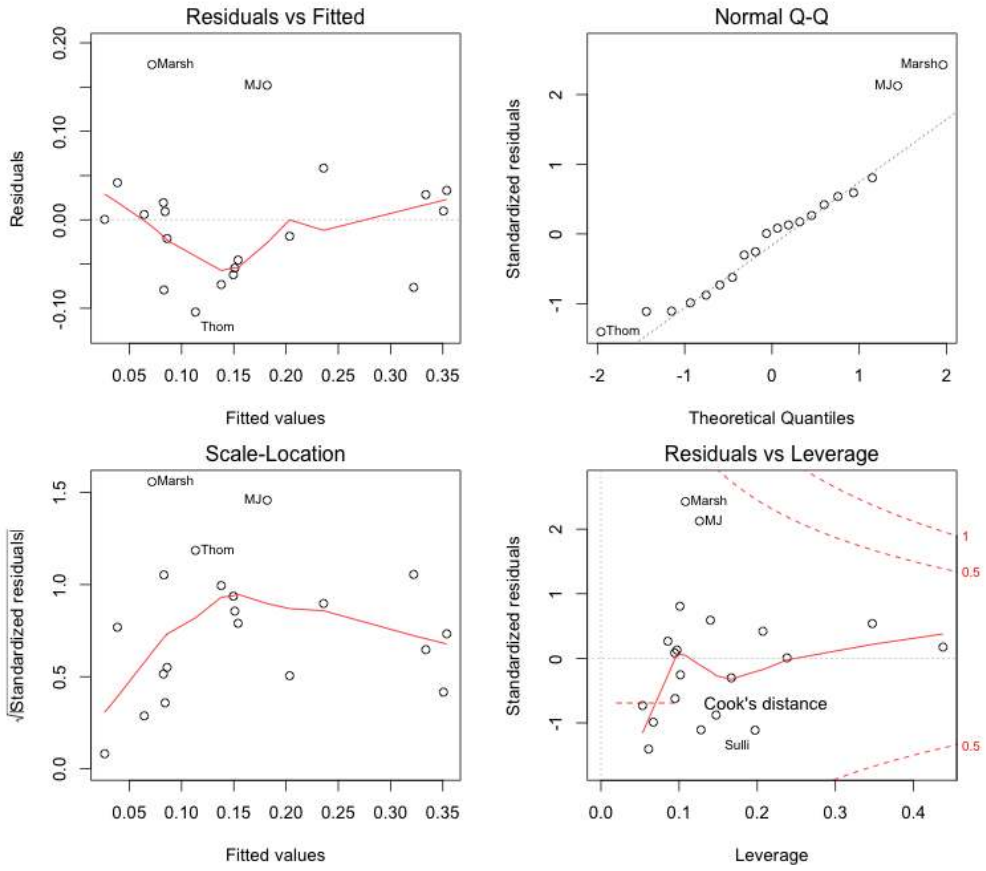
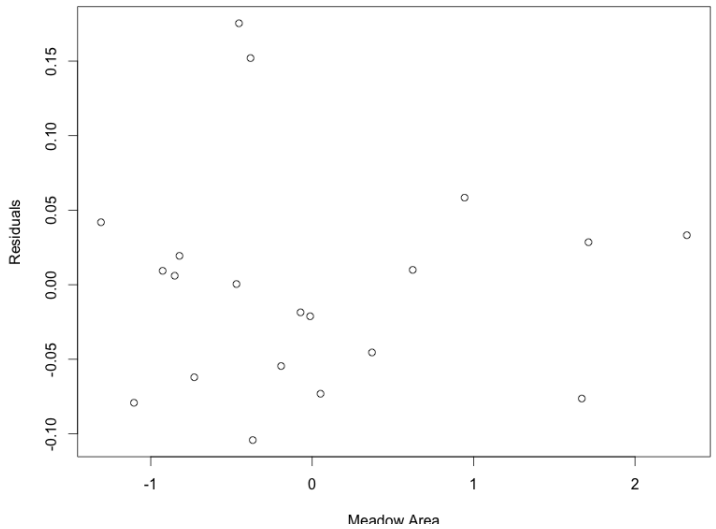


Figure 4: Model Diagnostics –Functional Dispersion Analysis

There are no clear indications of heterogeneity in the residuals, the quantile-quantile plot does not indicate strong deviations from the 1:1 line, and Cook's distances of the individual data points are all below 0.5.

A)



B)

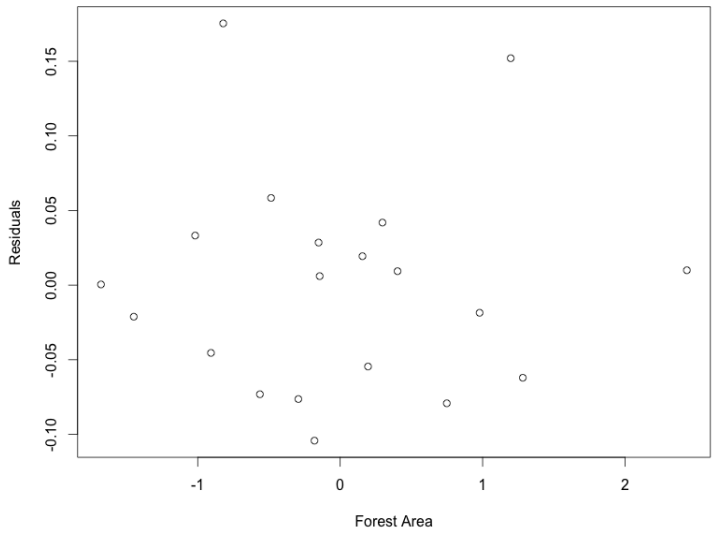


Figure C5: Partial Residuals Plots- Functional Dispersion Analysis

The model residuals of meadow (a) and forest (b) coefficients were each plotted against their original values; there are no clear patterns indicating lack of independence in the residuals.

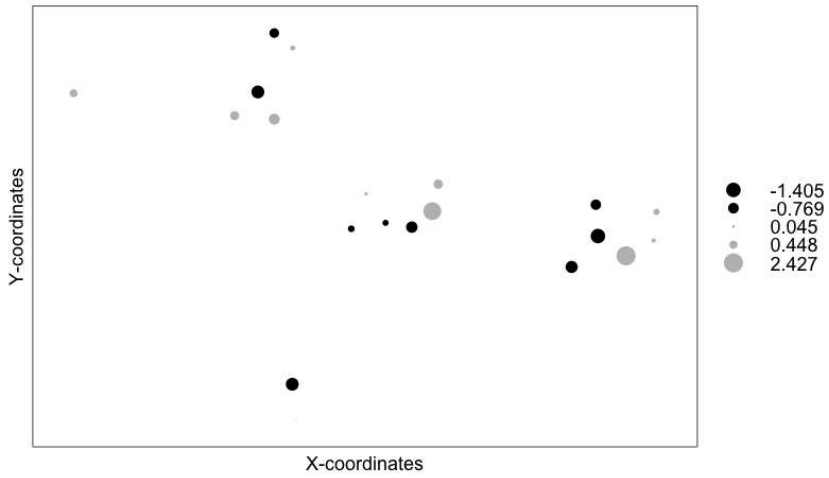


Figure C6: Model Residuals versus Site Coordinates- Functional Dispersion Analysis

The residuals of the functional dispersion LM were projected across the geographic coordinates of the 20 sites. No clear patterns are distinguishable.

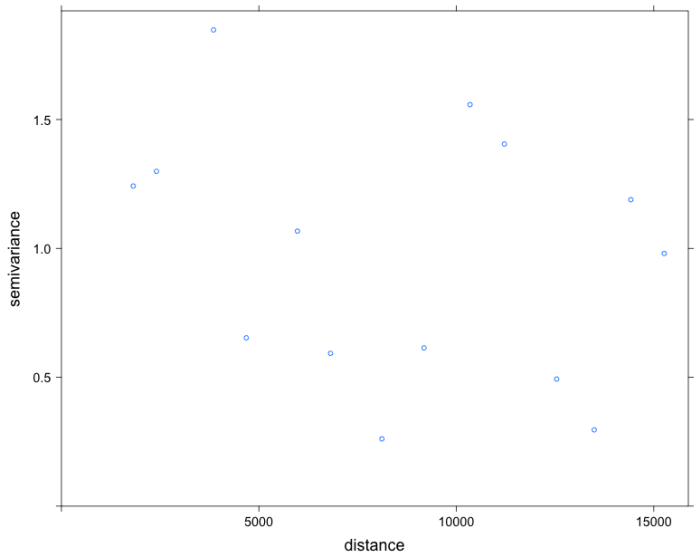


Figure C7: Semi-Variogram- Functional Dispersion Analysis

A semi-variogram was generated with the gstat package (Pebesma 2004) for the residuals of the functional dispersion LM; there is no indication of spatial autocorrelation present.

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