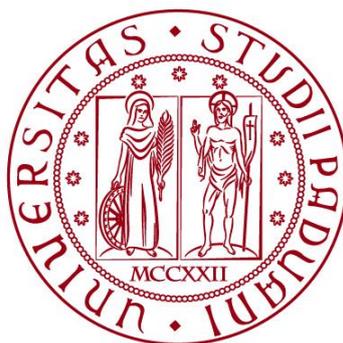


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Corso di Laurea magistrale in Environmental Sustainability and Education



**TESI DI LAUREA**

**Effects of habitat type on pollinators and potential role  
of hoverflies as bioindicators in the Euganean Hills  
Regional Park**

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## Abstract

Pollinator insects are in decline, mostly due to habitat loss and fragmentation. Preserving different habitat types is crucial for pollinator protection, because different habitat types, such as forests or calcareous grasslands, maintain different pollinator communities. The aim of this thesis was to study the diversity and abundance of hoverflies and bees in the Euganean Hills Regional Park in three different types of habitats: abandoned quarries, calcareous grasslands and forest margins. To do so, 24 sites have been selected and sampled in four different rounds between May and July using transects. Overall, we found 819 individuals of bees and 365 of hoverflies and 24 hoverfly species. Our results showed that the three habitat types differed in terms of vegetation with the forest margin being the habitat type with the lowest flower cover and highest vegetation height among the three. Furthermore, the forest margin was the habitat type with the highest abundance of hoverfly species, while abandoned quarries and calcareous grasslands had both comparable values. Also, we found that the percentage of semi-natural habitat had a positive effect on hoverfly abundance and diversity. Bees instead, did not respond to the considered environmental drivers. In addition, we found that two hoverfly species, *Episyrphus balteatus* (De Geer, 1776) and *Melanostoma scalare* (Fabricius, 1794), were strongly associated with the habitat type of the forest margin. In conclusion, our results confirmed that to support pollinator diversity, it is recommended to maintain a high diversity of habitat types, and in particular a high amount of forest margins for hoverflies, and a high percentage of semi-natural areas in the landscape.

## Riassunto

Gli insetti impollinatori sono in declino, principalmente a causa della perdita e frammentazione dei loro habitat. La conservazione di diversi tipi di habitat è cruciale per la protezione degli impollinatori perché habitat diversi, come ad esempio boschi o vegri, sostengono comunità di impollinatori estremamente differenziate. Lo scopo di questa tesi è stato quello di studiare la ricchezza e l'abbondanza di sirfidi e api in tre diversi tipi di habitat del Parco Regionale dei Colli Euganei: cave abbandonate, vegri e margini di bosco. A tal proposito sono stati selezionati 24 siti che sono stati campionati attraverso l'utilizzo di transetti in quattro diversi round tra i mesi di maggio e luglio. Nel complesso abbiamo campionato 819 esemplari di api e 365 di sirfidi, e trovato 24 specie di sirfidi. I nostri risultati hanno dimostrato che i tre tipi di habitat differivano in termini di vegetazione, e in particolare il margine di bosco era il tipo di habitat con la minor copertura florale e la vegetazione più alta tra quelli considerati. Inoltre, il margine di bosco è il tipo di habitat in cui è stata trovata la maggiore abbondanza e diversità di specie di sirfidi, mentre in cave abbandonate e vegri abbiamo ottenuto valori comparabili. È stato anche dimostrato che la percentuale di habitat seminaturale ha un effetto positivo sull'abbondanza di sirfidi. Le api invece, non hanno risposto alle stesse variabili ambientali considerate. In aggiunta, sono state trovate due specie di sirfidi, *Episyrphus balteatus* (De Geer, 1776) e *Melanostoma scalare* (Fabricius, 1794), fortemente associate al margine di bosco. In conclusione, i nostri risultati hanno confermato che per supportare la diversità degli impollinatori, è consigliato conservare un'elevata diversità di tipi di habitat, e in particolare di margini di bosco per i sirfidi, e un'alta percentuale di aree seminaturali nel territorio.

## **Introduction**

### **Pollinator insects**

In plants, sexual reproduction involves pollination, which is the transfer of pollen from the anthers, the male part of the flower, to the stigma, the female part (Gullan and Cranston, 2014). This process can come through many means such as wind (anemophily) or water (hydrophily), but many plants rely on insects (entomophily). Insects visit flowers to feed on nectar and pollen, but in some cases, mostly in Orchidaceae, the flower attracts them through shapes and odors resembling those of the female counterpart of the pollinator insect that pollinates it in the attempt to mate with it (Gullan and Cranston, 2014).

Insect pollination is very efficient and has many advantages such as reducing pollen wastage, being successful when the conditions are not suitable for other forms of pollination and maximizing the number of plant species in a certain area. Among insects, the major anthophilous orders are Hymenoptera (bees, wasps and ants), Diptera (flies), Lepidoptera (butterflies and moths), Coleoptera (beetles) and Thysanoptera (thrips) (Gullan and Cranston, 2014).

### **Bees**

Bees belong to the order of Hymenoptera, suborder Apocrita, and have diversified into a group of 7 families, 28 subfamilies, 67 tribes, 529 genera and over 20,000 species known and described so far (Danforth et al., 2019). “Bees” can be defined as any lineage of aculeate wasps whose larvae feed on pollen rather than arthropod preys (Danforth et al., 2019). Based on their behavior, we can divide the species of bees in:

- i. **Social bees:** sociality in bees is not defined by the mere aggregation of many individuals in the same place, since communal nests are also common among some species of solitary bees (Danforth et al., 2019). Social taxa are characterized by a series of co-operative behaviors which consist in: first, a reproductive division of labor with a caste system where sterile or non-reproductive individuals, known as “workers”, build and defend the nest, gather pollen and nectar for the development of the offspring and assist the reproductive females generally known as “queens”; second, they exhibit

cooperative brood care, meaning that female colony members help raising offsprings that are not their own; finally, there is an overlap of generations where “mothers” and “daughters” occupy the same nest, and they all contribute to the colony functioning (Gullan and Cranston, 2014; Danforth et al., 2019). We can further divide social taxa in eusocial societies (“true social” insects) and cooperative breeding societies: they share all the points of social behavior but in eusociality the reproductive division of labor is permanent, while in cooperative breeding it is only temporary, meaning that in eusociality once a female becomes a sterile worker it retains that caste for the rest of its life. Eusocial bees are honeybees, bumble bees, stingless bees, some halictid bees and two species of allodapine bees, and they all present permanent, caste-based societies (Danforth et al., 2019).

- ii. **Parasitic bees:** bees with parasitic behavior thrive at the expense of a host. They can be divided into two broad categories: brood parasites and social parasites. Brood-parasitic bees, also called cleptoparasites or “cuckoo” bees, do not build their own nest nor do forage for pollen and nectar. Instead, they enter the nest of other bees, lay an egg in an open or closed brood cell, and after the adult female or the first-instar larva kills the host egg, the cleptoparasitic larva will feed on the pollen and nectar provided by the host. This kind of parasitic bees can attack both social bees and solitary bees (Danforth et al., 2019). Social parasites are very different from the latter and attack only social bee species. The female social parasite enters the nest of the host and replaces the queen becoming the egg-laying female. Then, the former queen can either be killed or remain within the nest ceasing to lay eggs, while the female parasite claims the workers making them raise her own offspring, which will become social parasites themselves (Danforth et al., 2019).
- iii. **Solitary bees:** are the vast majority of bees on Earth, comprising more than 75% of the known bee species. In solitary bees all females can produce offspring, they forage for food resources to sustain their prole, primarily pollen and nectar, and they build and maintain their own nest. Usually solitary bees live alone, but in some cases multiple reproductively active females may share the same burrow forming communal nests. Even in this

cases there is no actual cooperation between nest mates, and each female effectively behaves as a solitary bee (Danforth et al., 2019).

Keeping in mind bees' essential role in sustaining plant communities, it is necessary to know that there are important differences in the ecology of social bees and solitary bees. First, period of activity and plant preferences are different. Social bees can be active for a period of time spanning months since they are multivoltine, having two or more broods of offspring per year, and they are polylectic, meaning that they have very broad host-plant preferences (Danforth et al., 2019). In solitary bees the period of adult activity is much shorter than it is for eusocial bees, lasting from a few weeks to a month, and usually it coincides with the availability of floral resources which varies depending on the species since they can have both narrow and broad host-plant preferences (Danforth et al., 2019). It is believed that solitary bees evolved oligolecty, a narrow host-plant preference, because it restricts resource competition between species by distributing the bees in space (different kinds of flowers) and in time (different periods of the year based on the flowering time of the host-plant). However, in times of pollen shortage due to a lower presence of the host-plant, this can lead to intraspecific competition (Linsley, 1958). Second, the foraging range is much wider in social bees than in solitary bees. The ability to communicate among workers allows colonies of some eusocial bees to benefit from resources that are kilometers from their nest site. By contrast, the foraging range of a solitary bee covers usually a maximum of 500 meters from the nest because the individual solitary bee must rely on its knowledge of the surrounding vegetation and resource availability (Danforth et al., 2019). Finally, solitary bees and social bees differ also in their biogeographical distribution. The species richness of solitary bees is highest in arid, Mediterranean climate regions, characterized by a high diversity of plant species and highly seasonal rainfall patterns, with which bees often time their emergence to coincide with peak flowering of their host-plant and to avoid droughts. Furthermore, since many solitary bees are ground-nesting, arid soils are less susceptible to the proliferation of fungi that can be dangerous for the bees' brood cells. Instead, social bees' diversity is highest in the humid tropics because, having multiple worker generations during the same year, they need floral resources over a much longer period of time (Danforth et al., 2019).

## **Hoverflies**

Hoverflies belong to the order of Diptera and comprise two families: Syrphidae and Microdontidae. Syrphidae, counting over 6000 species from all continents are one of the largest families of Diptera, and 96 genera and around 980 species of hoverflies can be found in Europe (Sommaggio, 1999; Sarthou et al., 2023).

A distinctive characteristic of adult hoverflies is that even though they are harmless they imitate the appearance of bees and wasps to deceive predators. This is an example of Batesian mimicry: an innocuous species evolves to resemble closely an abundant harmful model. Since both hoverflies and bees are pollinators and visit similar habitats, the hoverflies' mimetic form is advantageous because predators will associate the color pattern with some nasty experiences they had with the model and will tend to avoid them (Gilbert, 2005) [Figure 1].

Hoverflies have recently gained much attention, especially in agroecosystems, because the larvae of some species can provide pest control services, while the adults provide pollination services to wild flowers and flowering crops (Meyer et al., 2009). Hoverfly larvae can occupy very different ecological niches and depend on specific macro- and micro habitats, which provide them both food resources, essential for their highly differentiated feeding habits, and overwintering refugia (Meyer et al., 2009). Based on their diet we can divide the larvae into: phytophagous, which feed on plants, including stems, bulbs and roots; microphagous, which feed on fungi or on micro-organisms present in water loaded with organic matter; saprophagous, which feed on decomposing matter (plants or animals) and can be considered part of the microphagous category; zoophagous, which feed on living animals, mainly aphids, and are the ones considered important auxiliaries for pest control (Sarthou et al., 2023). Hoverflies, once they become adults, feed on pollen and nectar [Figure 2]. Pollen is mainly consumed by females because provides nutrients essential for ovarian development and egg production, while nectar is consumed by both males and females to provide energy for flight while searching for a mating partner. Compared to bees, which are restricted to a limited home range and to providing food also for their offspring, hoverflies can carry pollen over longer distances while foraging covering from a few meters to 2 kilometers per day, and over considerably longer distances while migrating (Doyle et al., 2020; Moquet et al. 2018).

a)



b)



*Figure 1. Specimen of Eristalis sp. (a) mimicking a honeybee (b)*



*Figure 2. Specimens of Episyrrhus balteatus (De Geer, 1776) feeding on a flower*

### **Habitat type effect on pollinator insects**

The main local drivers of pollinator diversity and abundance are floral and nesting resources (Grundel et al., 2010). Different habitat types can offer a different amount and quality of floral and nesting resources. For example, hedgerows, i.e. linear seminatural features in agricultural landscapes, provide an important forage and dispersal resource for pollinator insects, especially where intensive agriculture depauperates the landscape of semi-natural habitats (Garrat et al., 2017). Another example are urban gardens, patches of human-managed habitat in private properties or in public green spaces, which can mitigate the negative effect of habitat loss due to urbanization on wild pollinators (Majewska and Altizer, 2020). Urban gardens strongly affect abundance and species composition when they provide flowering plants and areas with low soil disturbance for soil-nesting sites (Speak et al., 2015; Matteson et al., 2008). Flower species differ in the amount of nectar and pollen and their chemical composition. Structurally different flower species will provide a greater array of foraging niches supporting a greater diversity of pollinator functional groups (Cole et al., 2017). In addition, floral resources differ across habitat types depending on the season, meaning that their profitability as foraging habitats also changes over time. Habitat effects are not only accredited to the different floral resources they provide, but also to the proportion between areas exposed to sunlight and shaded shelters for thermoregulation (Ricarte et al. 2011; Cole et al., 2017). Overall, the heterogeneity of habitat types increases the variety of resources to meet the necessities of a greater diversity of species, since each species needs different resources at a certain point in time or at distinct lifecycle stages (Cole et al., 2017).

Habitats for bees may differ depending on the bees' size, their vegetation, their physical structure and their micro-climate. Many bees are highly seasonal, meaning that they have to time their emergence to coincide with the peak of flowering of the host-plant in their specific habitat to find the right food sources (Westrich, 1996). Furthermore, the habitat has also an effect on parasitic bees because since they rely completely on their hosts for the brood care of their own offspring, they can proliferate only if their host has all the resources it needs to survive (Westrich, 1996). The basic conditions that a habitat must fulfill for a nesting-bee (all the non-parasitic bees) to produce their offspring are:

- A sufficient amount of food plants providing for pollen and nectar. Pollen is the essential component of larval food and since many species of solitary bees are oligolectic, depending on a narrow selection of species of host-plants, they must find enough pollen sources in the vicinity of their nest to sustain their brood. Nectar is an energy-rich food that bees need to fuel all their activities. Usually, bees are not confined to specific nectar sources, and even oligolectic bees can look for nectar in plants from which they do not gather pollen (Westrich, 1996).
- Specific nesting sites. The majority of bees dig their nest into the ground, and they generally prefer south-facing banks, cliffs or slopes with few vegetation. Some bees prefer sandy soils, others clay. Other bees build their nests inside irregular cavities found in rocks or dead wood. Carpenter bees can excavate their own tunnels in dead trees. Some species are specialized in nesting inside empty snail shells. Certain bees dig their burrows in the pith of stems of some plants such as *Rubus* or *Verbascum* (Westrich, 1996).
- Specific materials to build the nest. Most of the mining bees cover the interior of their brood cells with secretions coming from the Dufour's gland to make them waterproof and fungus-resistant. Other bees are specialized in gathering materials that they find in the vicinity of their nest, such as clay, leaves, resin or petals, sometimes mixed with their own saliva, and they use them to build their nests on the surface of stones or on the bark of trees, or to line their brood cells (Westrich, 1996).

It is important that the combination of these essential resources must be available within the home range of a nesting female. The distance that a foraging female can cover during its trips is species specific, so it is yet to be known in many species described, but it is thought to be influenced by the species' size and specialization, and by the conditions of the respective habitats (Westrich, 1996).

Hoverfly species have larval stages in flower-poor forests usually depending on specific vegetation types, and adults disperse to open habitats to look for floral resources (Ricarte et al., 2011; Moquet et al., 2018). Hoverfly larvae will be linked to the habitats which satisfy their food necessities the best: saproxylic and phytophagous larvae will need specific trees and plants; zoophagous larvae, feeding

mostly on aphids, will need the presence of the prey's host-plants; saprophagous larvae, which can be also aquatic, will need a particularly humid environment where they can find microorganisms and decaying matter, but also temporary or permanent water bodies (Ricarte et al., 2011; Moquet et al., 2018). Adult hoverflies tend to visit habitats that are often very different from those of their larvae. They are less tied to specific species of plants and can push themselves towards more open areas rich in flowering plants from which they obtain nectar and pollen (Ricarte et al., 2011).

In this thesis, we considered three habitat types different in terms of resources offered to bees and hoverflies. The first habitat type is the calcareous grassland, i.e. grasslands that occur on steep, calcareous outcrops on hilly domes or mountainous regions, and are characterized by specific environmental conditions (Butaye et al., 2005). This habitat type is known for its high plant species richness, mainly herbaceous xerophilous plants typical of hot and dry environments, which also reflects a high arthropod diversity. Because of the huge biodiversity found in calcareous grasslands, they are integrated into the "Natura 2000" network and protected by the Habitats Directive 92/43/EEC (Butaye et al., 2005; [www.parcocollieuganei.com](http://www.parcocollieuganei.com)) [Figure 4a]. The second habitat type is the forest margin, i.e. marginal areas between a forest and a grassland (Cary, 1996). Forest margins, being interfaces between the forest and the adjacent land cover, can help preserving the biodiversity of the forest interior offering protection from adverse conditions that can dominate outside the forest, but also can provide suitable habitat conditions for specialist and generalist species usually found inside the forest. The forest margin's microclimate differs from the forest's one, and gradients in temperature, humidity, light and wind are all affected by the margin's structure and the forest's tree species composition (Meeussen et. Al., 2020). The forest margins considered for this study were always between a forest and cultivated land (mainly vineyards). Therefore, it is important to take into consideration the fact that, in comparison to the forest interiors, forest margins can be influenced by a higher influx of herbicides and pesticides, but also higher levels of atmospheric nitrogen coming from the adjacent cultivated fields (Meeussen et. Al., 2020) [Figure 4b]. Finally, we took into account the habitat type of abandoned quarries. Abandoned quarries are the result of the abandonment of a once-exploited site by mining

industries, and in many regions can be prominent landscape features often occupying larger areas than natural reserves (Novák and Konvička, 2006). This habitat type is usually characterized by the quarry cliff side, a peculiar and extreme habitat associated with increased soil erosion and unsuitable for plant colonization, and by a more stable quarry floor (Yuan et al., 2006). Spontaneous colonization of places heavily affected by human activity by species of conservation interest has been reported for both plants and animals, and it is particularly important for quarries since restoration via spontaneous succession is often the cheapest and easiest way. However, some studies are trying to assess how vegetation surrounding quarries can influence the course of succession, to see if it can be channeled towards a more specific vegetation reducing the costs of a post-excavation restoration, developing valuable habitats for the conservation of biodiversity (Novák and Konvička, 2006) [Figure 4c].

### **Landscape effect on pollinators**

Besides the local effect of the habitat type, pollinator diversity and abundance are affected by the landscape composition and configuration (Senapathi et al., 2017; Betts et al., 2019; Gillespie et al., 2022). Changes in the landscape, involving the conversion of natural habitats to anthropogenic land-use and agricultural intensification, are one of the main drivers of biodiversity loss and in particular pollinators decline (Aguirre-Gutiérrez et al., 2015; Senapathi et al., 2017).

Considering that different pollinator groups need different feeding and nesting resources to satisfy their needs, it is expected that the response to habitat fragmentation will differ depending on the group, and that landscapes with higher diversity of suitable habitats are more prone to have a higher richness in pollinator species (Aguirre-Gutiérrez et al., 2015).

The proportion and intensity of land dedicated to agriculture in the landscape tends to be negatively related to pollinator abundance and species richness. In intensive agricultural landscapes, many species of insects may end up confined into small non-cultivated areas that usually are unable to provide the floral resources needed to sustain viable populations of pollinators (Patrício-Roberto and Campos, 2014; Senapathi et al., 2017). An agricultural landscape that seems to benefit pollinators are mass flowering crops, but their limited flowering season fails to provide

longevity of resources (Senapathi et al., 2017). Something more useful to pollinators would be creating ecological corridors that connect the various patches of natural habitats fragmented by the agricultural landscape. These ecological corridors, if composed by different species of plants which blossom throughout the year, integrated with hedgerows and tree trunks, may offer additional foraging and nesting habitats that can benefit pollinator communities (Patrício-Roberto and Campos, 2014).

Urban land cover is globally increasing, resulting in more habitat loss and fragmentation. As we have already seen for agricultural intensification, pollinators richness and abundance are negatively related to increasing urbanization. However, not all taxa respond in the same way to urbanization, and some particular functional groups seem to dominate in different urban landscapes. For example, cavity nesting bees and specialist bees are rare in cities, but bumblebees seem to really benefit from domestic gardens. Several studies show that hoverflies seem to be more negatively affected by urbanization than bees, but this topic is still under-researched and further data are needed to improve urban habitat management (Senapathi et al., 2017).

### **Protected areas and pollinators**

To buffer the negative effects of habitat loss and deterioration, protected areas (PAs) are often designed. For many species, PAs have become a last refuge where they can take shelter from threats caused by human activity (Chowdhury et al., 2023; Cooke et al., 2023).

Compared to other taxa, there are relatively few studies that explore insect representation in PAs. Creating areas that humans can no longer exploit for their needs and calling them “protected” just for the sake of it, is not enough. Protected areas need to be accurately designed and must take into consideration all the factors that may have both positive and negative effects on the protected species (Chowdhury et al., 2023). In fact, insects often depend on specific host-plants, meaning that minor changes in the composition of plant communities in the PAs might have huge effects on the habitability of the area, and they may also be endangered by the presence of non-native parasites and predators. Furthermore, PAs sometimes can mitigate against harmful pressures such as habitat loss and intensive

management, but this may result in the retention of some sensitive species that are highly vulnerable to more pervasive threats such as climate change, disease and pollution, leading to a “protection paradox” (Cooke et al., 2023). Another important characteristic of PAs is their size and spatial configuration. In pollinator conservation seems that small-size PAs are more valuable than the large ones (Gutiérrez-Arellano and Mulligan, 2020). Larger-sized PAs normally include a higher number of habitats and hence a higher species richness being able to effectively sustain their populations, preventing or slowing down species extinction. However, smaller-sized PAs provide more efficient pollination services because of their proximity to the crops, and they are comparable to daily movements of most pollinator insects. In addition, a wise spatial distribution of these PAs can create a mosaic of crops and adjacent natural and seminatural habitats, that may provide pollinator communities with enough food resources and suitable nesting sites. (Diks et al., 2016; Gutiérrez-Arellano and Mulligan, 2020).

Having said that, after protected areas are created, to be truly effective they require both appropriate management and regular monitoring, as well as different management interventions depending on the species that they host and their needs during the various stages of their life cycles (Gutiérrez-Arellano and Mulligan, 2020; Chowdhury et al., 2023).

### **Aims of the thesis**

The overall aim of this thesis is to study the effects of three different habitat types on pollinators in the protected area of the Euganean Hills Regional Park. The three habitat types considered are: abandoned quarries, calcareous grasslands and forest margins. First, we considered the habitat type effect on hoverfly and bee abundance and hoverfly diversity. Second, focusing on the hoverflies, we tested if these particular pollinator insects can be used as effective bioindicators of the three habitat types. Hoverflies have three main characteristics that can make them good bioindicators: first, the highly differentiated larvae’s habits and environmental requirements, even among larvae with similar diets; second, hoverflies are fairly common in almost every terrestrial ecosystem; third, their identification, at least at the genus level, it is not difficult (Sommaggio, 1999). However, their use as bioindicators is still poorly developed probably because of some drawbacks such

as the short flight season of some species, which can make more complicated the choice of the right sampling time, and the high mobility of adult hoverflies allows them to rapidly recolonize stressed habitats (Sommaggio, 1999).

## **Materials and Methods**

### **Study area**

The study area was the Euganean Hills Regional Park, located in the Northeastern part of Italy. The peculiarity of this area is its geological nature: there are both sedimentary rocks, from what once was the seafloor, and effusive magmatic rocks, from past volcanic activity. The vegetation of the Euganean Hills is also particular since here live thermophilic plants, which are typical of hot and dry climates, but also microtherm plants, which are typically found in montane and submontane ecosystems. Therefore, given the various ecological conditions among the different hills, we can come across calcareous grasslands, the pseudo-Mediterranean scrub, chestnut woods, oak woods and Robinia bushes. This great variety of habitat types provides shelter for a huge number of vertebrate and invertebrate species, especially when compared to the ones found in the surrounding plains ([www.parcocollieuganei.com](http://www.parcocollieuganei.com)).

The Euganean Hills Regional Park is part of the “Natura 2000” network, which is a network of areas destined to the conservation of biodiversity in the European Union territory, instituted by the article 3 of the Habitats Directive 92/43/EEC on the conservation of natural habitats and of wild fauna and flora. The “Natura 2000” network allows all its members to apply the concept of protection of biodiversity, recognizing the different biotic, abiotic and anthropic elements, and granting the right equilibrium among them. Moreover, since the 5<sup>th</sup> of July 2024, the park has officially been proclaimed Biosphere Reserve “Euganean Hills” into the program MAB UNESCO, during the 36<sup>th</sup> session of the International Coordinating Council of the Man and the Biosphere (MAB). This was possible thanks to the exceptional natural and cultural value of the area, and thanks to the effort shown by the park promoting sustainable development practices which respect and value the local environmental resources ([www.parcocollieuganei.com](http://www.parcocollieuganei.com)).

The studies were conducted in collaboration with Esapolis and the Butterfly arc, two structures dedicated to the dissemination in the naturalistic field, focusing on the world of insects, which are collaborating with the park on various projects on the biodiversity of the Euganean Hills. In particular, some of their aims are to

characterize the hoverfly population in this protected area, classify them at a systematic level, and to verify if they can be used as valuable bioindicators of biodiversity.

### Sites selection

For this study were selected 24 different sites inside the Euganean Hills Regional Park area: 7 abandoned quarries, 9 calcareous grasslands, 8 forest margins. The sites were found using Google Earth Pro 7.3.6.9796 (64-bit) and were chosen keeping a distance of at least 500m between each other [Figure 3]. The distance between each site was particularly important because it took into consideration the foraging area of solitary bees, to avoid sampling individuals that could visit more than one site among the selected ones. Even if abandoned quarries were quite common in the area, they were not always accessible, therefore each site was then scouted in person to see if it was actually good for sampling and if it was easily accessible. Finally, a unique ID code has been assigned to each site, composed of letters identifying the habitat type and numbers from 01 to 09 (CG01 to CG09 for calcareous grasslands, AQ01 to AQ07 for abandoned quarries and FM01 to FM08 for forest margins).

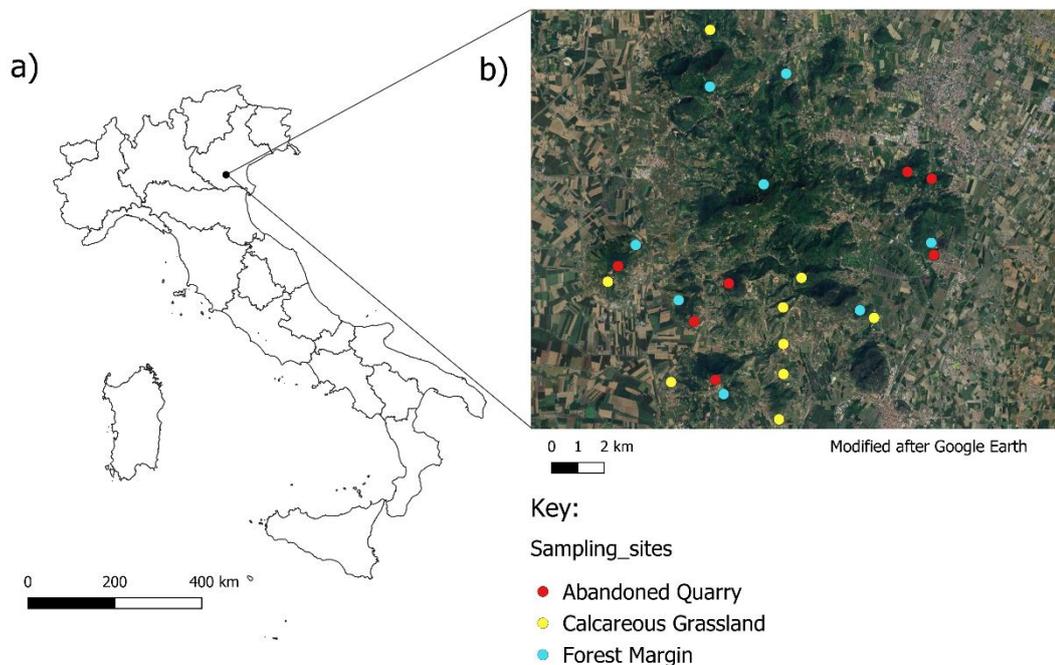


Figure 3. a) Location of the Euganean Hills Regional Park in Italy. b) Satellite view of the Euganean Hills Regional Park showing the location of each sampling site.

a)



b)





*Figure 4. Examples of: Calcareous grassland (a); Forest margin (b); Abandoned quarry (c)*

### **Sampling method and data collection**

Each site was sampled using transects. The transects were standardized both in length (40mX20m) and time (30 minutes per site). If the area was particularly small, as it happened for some abandoned quarries, it was sampled as a whole. Pollinator insects were sampled at sight using nets and preserved in falcon tubes filled with alcohol diluted at 75%. Only wild bees and hoverflies were sampled, while honeybees and bumblebees were counted and taken note of. In each falcon tube was inserted a label with a code indicating the site where the specimens were sampled and the sampling round. The sampling rounds were 4 and were performed in different periods from May to July, waiting at least two weeks between one round and the next one (round 1: 09/05 – 13/05; round 2: 05/06 – 07/06; round 3: 27/06 – 02/07; round 4: 22/07 – 26/07).

Once on the field, some environmental data was collected compiling field data sheets, while temperatures were registered using data loggers. Each field data sheet contained information about:

- Site ID
- Sampling round
- Date
- Starting time and finishing time of the sampling
- Sunshine % time
- Shadow %
- Wind force (on a scale from “no wind” to “high”)
- Flower cover %
- Green vegetation cover %
- Herbaceous vegetation height (estimation of the mean height in cm)
- Management of the site
- The number of honeybees and bumblebees seen during the sampling
- A list of all the flowering plant species and their abundance (as a % of the total flowering plants found) inside the considered transect.

All these data were then rewritten in an Excel spreadsheet to be used later for the statistical analyses.

### **Species identification**

First thing first, the wild bees were separated from the hoverflies, always keeping note of the round and the site in which each specimen was sampled. Then, we counted all individuals collected on each site.

Concerning the hoverflies, first the specimens were identified at the genus level with the help of the “Illustrated key to the hoverfly genera of Europe (Syrphidae and Microdontidae)” (Sarthou et al., 2023) dichotomous key. The abundance of each genus of hoverflies in each site for each round was then registered in a matrix in an Excel spreadsheet. After that, with the help of another dichotomous key, “Hoverflies of Northwest Europe. Identification keys to the Syrphidae” (Van Veen, 2004), the specimens were identified at the species level, and once again it has been taken note of the abundance of the number of different species in each site for each round on an Excel spreadsheet.



Figure 5. Sampling with net along a transect



Figure 6. Setup for species identification

### **Landscape predictors**

The determination of the land cover at the landscape scale in the sampling sites was done using QGIS and Google Earth Pro. Around each site we have drawn a circular area (about 500m in diameter) called “buffer”. Inside each buffer, using new layers and the function “polygon” we have determined the main different land covers that were found in the area which were: “Woodland percentage”; “Tree groups and groves percentage”; “Vineyard percentage”; “Quarry area percentage”; “Rural house percentage”; “Olive trees percentage”; “Grassland percentage”; “Arable land percentage”; “Urban green areas percentage”; “Water courses, canals and waterways percentage”; “Robinia bushes percentage”; “Trees in rows percentage”; “Calcareous grassland percentage”; “Lakes and water basins percentage”. Both the area and the perimeter of each polygon have been calculated. We pooled the data of these categories to summarize the land use into fewer categories and used: “Seminatural percentage”, “Perennial percentage”, “Quarry area percentage”, “Urban percentage”, and “Annual percentage”. Finally, all the data were brought to an Excel spreadsheet to do further analyses and calculations.

### **Statistical analyses**

Statistical analyses were executed using R Software version 4.4.1. and RStudio 2024.09.0+375.

The function `lm()` was used to fit linear regression models to test the relationship between the flower cover and the different habitat types and between the herbaceous vegetation height and the different habitat types. In addition, the package `ggplot2` was used to create graphs showing the mean flower cover and the mean herbaceous vegetation height in each habitat type considered. Moreover, we tested the relationship between the sampling round and the recorded temperature, using a linear regression model with sampling round as explanatory variable and recorded temperature as response variable. We considered correlations among landscape predictors which were “Seminatural percentage”, “Perennial percentage”, “Quarry area percentage”, “Urban percentage”, and “Annual percentage” using the function `cor.test` in R. We decided to use “Seminatural percentage” as landscape predictor for all further analyses.

The function `lm()` was used to fit three different linear regression models, showing the effect of the independent variables “Round”, “Habitat” and “Seminatural area” on the dependent variables “hoverfly abundance”, “hoverfly species richness” and “bee abundance”. The three response variables were transformed to a logarithmic scale. The function `qqplot()` was used to visually assess if the model residuals followed a normal distribution. Then the function `allEffects()` of the package `effects` was used to calculate the marginal effects of all the independent variables of the model.

We identified the indicator hoverfly species, i.e. the species that among the specimens sampled responded closest to the different environmental conditions of the habitat types considered. The indicator values were obtained using the `multipatt()` function of package `Indicspecies`. The `multipatt()` function allows to not only measure the association between a species and a group of sites, but using an extension of the original `IndVal` method it looks for indicator species in both individual site groups and combination of site groups.

## Results

### Pollinator descriptive results

Overall, we found 819 specimens of wild bees and 365 specimens of hoverflies. The hoverfly species found were 24. The number of species found in each habitat type and the abundance of each species in each habitat type have been listed in Table 1.

Table 1. List of hoverfly species sampled with their abundance in each habitat type, number of species found in each habitat type and number of specimens found in each habitat type.

Hoverfly Species	CG	AQ	FM	Tot
<i>Chrysotoxum cautum</i> (Harris, 1776)	8	1	6	15
<i>Syrphus ribesii</i> (Linnaeus, 1758)	1	1	3	5
<i>Syrphus vitripennis</i> (Meigen, 1822)	0	2	0	2
<i>Syrphus nitidifrons</i> (Becker, 1921)	0	0	2	2
<i>Sphaerophoria scripta</i> (Linnaeus, 1758)	49	56	41	146
<i>Eristalis similis</i> (Fallén, 1817)	1	0	5	6
<i>Eristalis tenax</i> (Linnaeus, 1758)	16	10	21	47
<i>Eristalis arbustorum</i> (Linnaeus, 1758)	7	6	2	15
<i>Merodon albifrons</i> (Meigen, 1822)	6	2	1	9
<i>Merodon clavipes</i> (Fallén, 1781)	1	0	0	1
<i>Merodon avidus</i> (Rossi, 1790)	10	4	4	18
<i>Xanthogramma pedissequum</i> (Harris, 1780)	1	2	4	7
<i>Xanthogramma</i> sp.	1	0	1	2
<i>Episyrphus balteatus</i> (De Geer, 1776)	1	2	13	16
<i>Myathropa florea</i> (Linnaeus, 1758)	0	2	4	6
<i>Paragus pecchiolii</i> (Rondani, 1857)	1	8	4	13
<i>Paragus albifrons</i> (Fallén, 1817)	1	2	1	4
<i>Paragus bicolor</i> (Fabricius, 1794)	0	1	1	2
<i>Paragus</i> sp.	2	0	0	2
<i>Syritta pipiens</i> (Linnaeus, 1758)	5	9	8	22
<i>Melanostoma mellinum</i> (Linnaeus, 1758)	0	2	9	11
<i>Melanostoma scalare</i> (Fabricius, 1794)	0	1	7	8

<i>Cheilosia</i> sp.	0	1	0	1
<i>Helophilus pendulus</i> (Linnaeus, 1758)	2	2	1	5
Number of species	17	19	20	24
Number of specimens	113	114	138	365

Note: CG: calcareous grassland; AQ: abandoned quarry; FM: forest margin

### **Vegetation height and flower cover in different habitat types**

The mean vegetation height for calcareous grasslands (CG) was 53,47cm  $\pm$  3,73 (SEM), for abandoned quarries (AQ) was 53,75cm  $\pm$  4,62 (SEM) and for forest margins (FM) was 69,06cm  $\pm$  5,87 (SEM). The difference among habitat types resulted statistically significant: forest margin was the habitat type where herbaceous vegetation was the highest, while calcareous grasslands and abandoned quarries had shorter herbaceous vegetation of comparable heights [Figure 7].

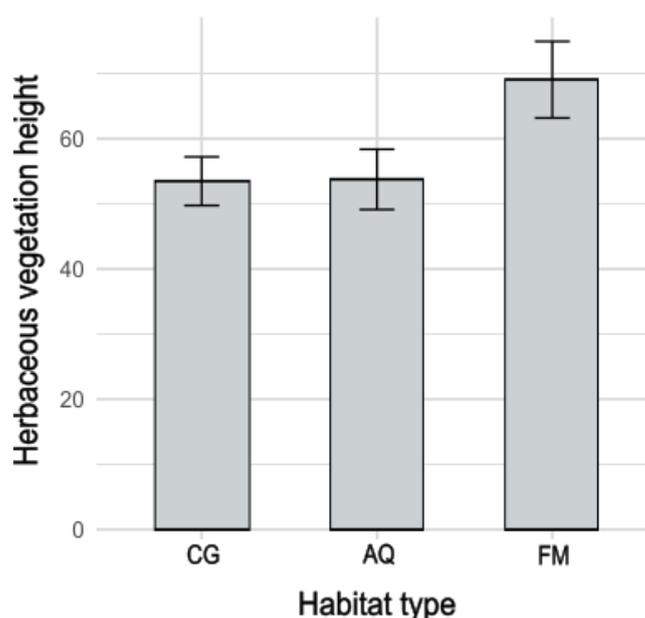


Figure 7. Mean herbaceous vegetation height measured in each habitat type throughout the whole sampling period. CG: calcareous grassland; AQ: abandoned quarry; FM: forest margin.

The mean flower cover in calcareous grasslands (CG) was 39,4% ± 3,95 (SEM), in abandoned quarries (AQ) was 39,1% ± 5,41 (SEM) and in forest margins (FM) was 33,7% ± 4,63 (SEM). The difference was not statistically significant [Figure 8].

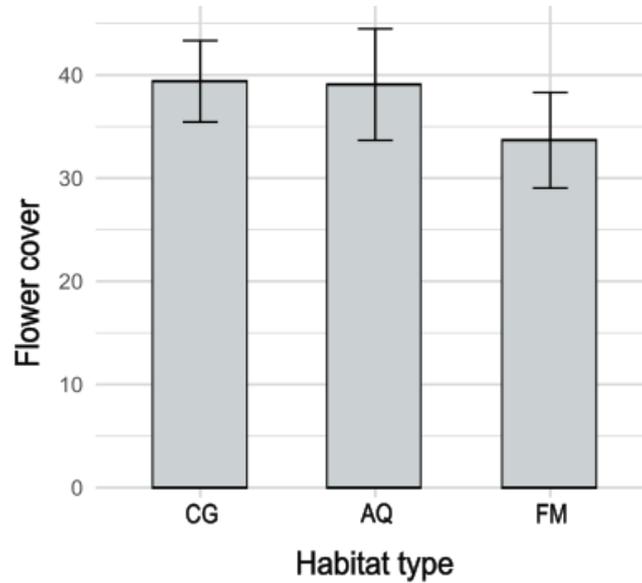


Figure 8. Mean flower cover in each habitat type throughout the whole sampling period. CG: calcareous grassland; AQ: abandoned quarry; FM: forest margin.

### Sampling round and temperature

The results of the linear regression [Table 2] show that there was a significant relation between Temperature and Round, and that there were significant differences between different rounds. The temperature increased during the sampling. Temperature in round 1 was lower than in round 2 (p value = 0,0138), 3 and 4 (p values <0,001).

Table 2. Results of the linear regression (lm) showing the relationship between Temperature and Sampling round.

	Estimate	Std. Error	t value	Pr (> t )
Round 2	23,526	0,937	2,511	<b>0,0138 *</b>
Round 3	59,920	0,937	6,396	<b>&lt;0.001 ***</b>
Round 4	61,044	0,937	6,516	<b>&lt;0.001 ***</b>

Note: Significant results (p value <0,05) in bold character. Round 2, Round 3, Round 4 stand respectively for different sampling rounds performed between the months of June and July.

### Hoverfly abundance

The graphs [Figure 9] show the effect of different variables on the hoverfly abundance. There were significant effects of the sampling round: round 2 had a significant positive effect on the abundance of hoverflies ( $p$  value = 0,002) and round 3 had a significant positive effect on their abundance as well ( $p$  value = 0,015). Among the different habitat types, the forest margin had a significant positive effect on hoverfly abundance ( $p$  value = 0,048) with respect to the calcareous grassland, which had the lowest abundance. Also, the percentage of semi-natural habitat had a significant positive effect on the abundance of hoverflies ( $p$  value = 0,008) [Table 3].

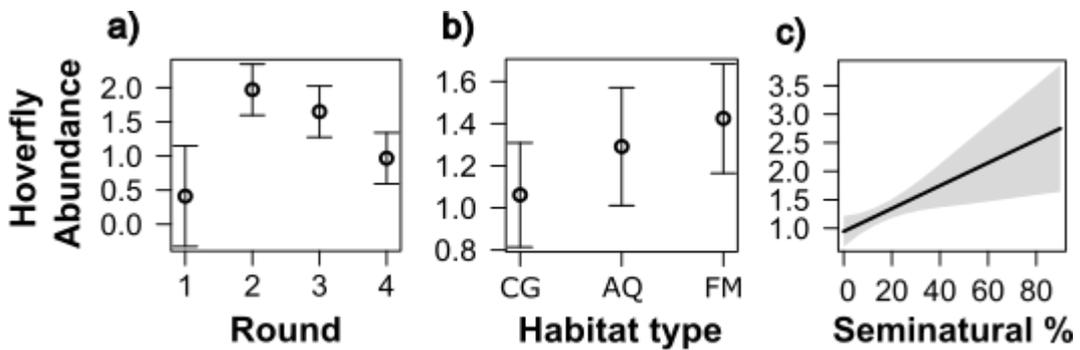


Figure 9. Effect of Sampling round (a), Habitat type (b) and Seminatural % (c) on the logarithmic transformed abundance of hoverflies. Regarding the habitat types in graph (b) CG: calcareous grassland; AQ: abandoned quarry; FM: forest margin.

Table 3. Results of the linear regression (lm) showing the relationships between the abundance of hoverflies and the Sampling round, the Habitat type and the Seminatural %

	Estimate	Std. Error	t value	Pr (> t )
Round 2	1,561	0,499	3,126	<b>0,002 **</b>
Round 3	1,239	0,499	2,482	<b>0,015 *</b>
Round 4	0,556	0,499	1,113	0,269
Abandoned quarry	0,230	0,190	1,208	0,230
Forest margin	0,363	0,181	2,003	<b>0,048 *</b>
Seminatural %	0,020	0,007	2,703	<b>0,008 **</b>

Note: Significant results ( $p$  value <0,05) in bold characters

### Hoverfly species richness

The graphs [Figure 10] show the effect of different variables on the hoverfly species richness. Regarding the rounds, only round 2 showed a significant positive effect on species richness ( $p$  value = 0,022), while round 3 seemed to have a marginally significant effect ( $p$  value = 0,064). As for the habitat types, the forest margin had a significant positive effect on species richness ( $p$  value = 0,036) with respect to the calcareous grassland, which had the lowest richness in species. The seminatural percentage had also a significant positive effect on species richness ( $p$  value = 0,015) [Table 4].

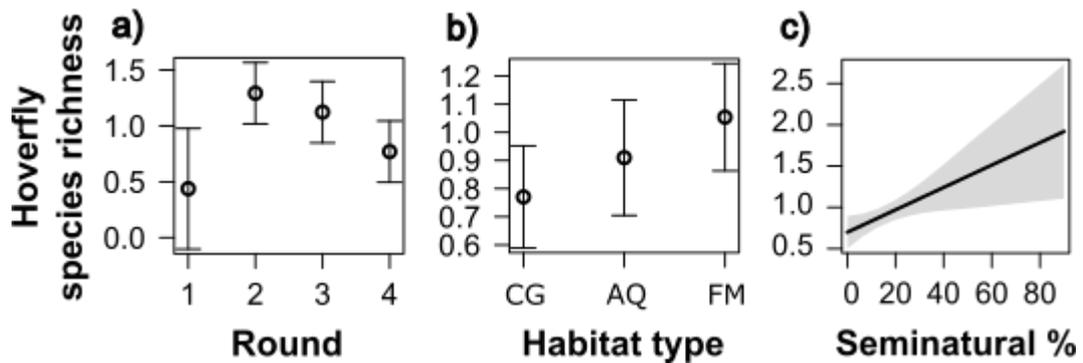


Figure 10. Effect of Sampling round (a), Habitat type (b) and Seminatural % (c) on the logarithmic transformed richness of hoverfly species. Regarding the habitat types in graph (b) CG: calcareous grassland; AQ: abandoned quarry; FM: forest margin.

Table 4. Results of the linear regression (lm) showing the relationships between the abundance of hoverflies and the Sampling round, the Habitat type and the Seminatural %

	Estimate	Std. Error	t value	Pr (> t )
Round 2	0,855	0,366	2,336	<b>0,022 *</b>
Round 3	0,685	0,366	1,873	0,064 .
Round 4	0,332	0,366	0,908	0,367
Abandoned quarry	0,139	0,139	1,001	0,319
Forest margin	0,283	0,133	2,132	<b>0,036 *</b>
Seminatural %	0,014	0,005	2,493	<b>0,015 *</b>

Note: Significant results ( $p$  value <0,05) in bold characters

### Bee abundance

The results of the linear regression model [Table 5] show that there are no variables that had a significant statistical effect on bees' abundance. The abandoned quarry was the habitat type that had a marginally significant positive effect, failing in exceeding the significance threshold ( $p$  value =  $0,075 > 0,05$ ).

Table 5. Results of the linear regression (lm) showing the relationships between the abundance of wild bees and the Sampling round, the Habitat type and the Seminalural %

	Estimate	Std. Error	t value	Pr (> t )
Abandoned quarry	2,700	1,500	1,799	0,075 .
Forest margin	0,617	1,433	0,431	0,668
Round 2	-1,562	3,944	-0,396	0,693
Round 3	-2,562	3,944	-0,650	0,518
Round 4	-4,270	3,944	-1,083	0,282
Seminalural %	-0,039	0,059	-0,672	0,503

### Bioindicator analysis

The results [Table 6] show that of the 24 hoverfly species considered, two species were significantly associated with the habit type forest margin. The two species are: *Episyrphus balteatus* ( $p$  value =  $0,014$ ) and *Melanostoma scalare* ( $p$  value =  $0,009$ ).

Table 6. Results of the "multipatt" function showing which hoverfly species could be possible bioindicators for a determined habitat type

Group forest margin #sps. 2		
	stat	p.value
<i>Episyrphus balteatus</i>	0,419	<b>0,014 *</b>
<i>Melanostoma scalare</i>	0,401	<b>0,009 **</b>

Note: Significant results ( $p$  value  $< 0,05$ ) in bold characters

Considering the other habitat types, there were no species associated with calcareous grasslands and abandoned quarries that showed statistically significant associations of habitat types [Table 7].

Table 7. Results of the “multipatt” function showing which hoverfly species could be possible bioindicators for a determined habitat type or combinations of different habitat types.

Group calcareous grassland		
	stat	p.value
<i>Paragus</i> sp.	0,236	0,346
<i>Merodon clavipes</i>	0,167	1,000
Group abandoned quarry		
	stat	p.value
<i>Syrphus vitripennis</i>	0,189	0,338
<i>Cheilosia</i> sp.	0,189	0,291
Group forest margin		
	stat	p.value
<i>Episyrphus balteatus</i>	0,419	<b>0,014 *</b>
<i>Melanostoma scalare</i>	0,401	<b>0,009 **</b>
<i>Syrphus ribesii</i>	0,236	0,502
<i>Eristalis similis</i>	0,230	0,317
<i>Syrphus nitidifrons</i>	0,177	0,641
Group calcareous grassland + abandoned quarry		
	stat	p.value
<i>Eristalis arbustorum</i>	0,349	0,328
<i>Merodon albifrons</i>	0,332	0,240
Group calcareous grassland + forest margin		
	stat	p.value
<i>Chrysotoxum cautum</i>	0,308	0,39
<i>Xanthogramma</i> sp.	0,171	1,00
Group abandoned quarry + forest margin		
	stat	p.value
<i>Paragus pecchiolii</i>	0,353	0,113
<i>Melanostoma mellinum</i>	0,316	0,134
<i>Myathropa florea</i>	0,289	0,190
<i>Xanthogramma pedissequum</i>	0,270	0,455
<i>Paragus bicolor</i>	0,183	0,526

Note: Significant results ( $p$  value  $<0,05$ ) in bold characters.

## **Discussion**

The overall aim of this thesis was to determine the effect of three different habitat types on pollinators in the protected area of the Euganean Hills Regional Park. We found that the three considered habitat types differed in terms of vegetation. Pollinator insects need floral resources to satisfy the needs of both adults and larvae, and many solitary bees need also specific materials to build their nests (Danforth et al., 2019; Moquet et al., 2018; Westrich, 1996). Calcareous grasslands and abandoned quarries seemed to have very similar flower covers, while the forest margin seemed to have a slightly lower flower cover with respect to the other two habitat types. However, the main difference between the considered habitat types was their vegetation height. Calcareous grasslands and abandoned quarries had comparable vegetation heights, while the forest margin showed the presence of taller vegetation than the one in the two other habitat types. These two results suggested that the plant species and the management in these three habitat types were different, and this could have influenced the populations of pollinators found, since different plants can reward the pollinators with different pollen and different amounts of nectar, that are resources particularly important for oligolectic solitary bees and for adult hoverflies (Westrich, 1996; Moquet et al. 2018). In addition, vegetation height can influence microclimatic conditions. Pollinator insects are able to actively look for microclimates in order to regulate their body temperature. Behavioral thermoregulation can happen at every stage of the insect's life cycle: the larvae can move to a preferred microclimate on the plant where they are feeding and can also look for the most appropriate microclimate for their pupation; adults may look for the best microclimate for egg deposition and will look for the best location and time for their daily activities (Johnson et al., 2023). Habitat types with these characteristics prove to be really important for pollinator insects, allowing them to maintain the right body temperature and avoid heat stress. If temperatures are elevated, and there are no places nearby or inside the habitat type considered with more favorable microclimates, this may result in shifts of the pollinators' activity period toward cooler times, shortening the available time for foraging and mating (Johnson et al., 2023).

Hoverfly abundance responded to the considered environmental drivers, while bees' abundance did not. The habitat type preferred by the hoverflies was the forest

margin, followed by the abandoned quarries and calcareous grasslands. One possible reason may be the fact that being a sort of transitional habitat between the forest and a grassland it can offer a variety of resources that can satisfy the foraging needs of both larvae and adults: the flower-poor forest can offer more microhabitats that may be optimal for the different ecological niches of the hoverfly larvae, while the flower-rich grassland offers the resources needed by the adults (Ricarte et al., 2011; Moquet et al., 2018). Another reason why the forest margin was the preferred habitat type by hoverflies could be the fact that it was the habitat type where were registered slightly lower temperatures with respect to the others. This characteristic is important because adult hoverflies do not thrive well in hot and dry environments (Miličić et al., 2018) and higher temperatures could disturb the hoverfly larvae development (Noel et al., 2022), meaning that having the possibility of taking shelter from the heat in the forest's shadow can be beneficial to both hoverfly adults and larvae. Another result showing a statistically significant effect on hoverfly abundance was the percentage of semi-natural habitat. This could be explained by the very well-known positive effect of semi-natural areas on pollinator insects in general (Gillespie et al., 2022). While herbaceous semi-natural habitats may offer higher amounts of flowers for adult hoverflies, woody semi-natural habitats provide hoverflies with more suitable resting sites, granting protection from harsh weather conditions and from predators, and with overwintering sites (Schirmel et al., 2018). Many hoverfly larvae are phytophagous or feed on aphids, meaning that the higher is the vegetation cover the higher is the probability for the larvae to find the host-plants they need or the host-plants of their prey (Ricarte et al., 2011; Moquet et al., 2018). Non-aphidophagous species further benefit from semi-natural habitats and their complexity because they are the only places where hoverflies can find the right resources for larval development and oviposition sites in agricultural landscapes. Aphidophagous species, which at first sight may benefit from the presence of crops where their larvae can find a higher food density, once adults cannot be sustained by the very short flowering time of crops, but they need semi-natural habitats where they can find abundant floral resources that offer continuous supplies of nectar and pollen (Schirmel et al., 2018; Meyer et al., 2009). Regarding the effects of the different sampling rounds, the results showed that after the second round they started to sharply decrease, confirming once more that higher temperatures have a

negative effect on hoverflies. An explanation why the first round had such a low hoverflies abundance, even if the temperatures were the most suitable for those pollinators, could be the heavy rainfalls that happened during the month of May 2024 that could have disturbed the hoverflies' lifecycle and daily activity. It is known that during periods of rain pollinators' activity decreases. Rainfall can increase the chances of pollen degradation and nectar dilution, which are the main floral rewards for the majority of pollinators. Moreover, other than posing mechanical difficulties for flight, rain can alter the behavior of pollinators altering their perception and sensory intake, and through the removal of flowers' scent and volatiles from the atmosphere it can obfuscate olfactory signals (Lawson and Rands, 2019).

The analysis on the hoverfly species richness showed results that were very similar to the ones on the hoverfly abundance. Once again, the preferred habitat type was the forest margin. Also, the percentage of semi-natural habitat had a positive effect on the species richness. Finally, the species richness followed the same trends as the hoverfly abundance during the four sampling rounds. These results suggest that since the forest margin is the habitat type with the highest abundance of hoverflies, it is also the habitat type where it is easier to find different hoverfly species, probably because the nearby woodlands offer different microhabitats for many hoverfly species increasing species richness (Meyer et al., 2009). And the same goes for the other two habitat types, abandoned quarries and calcareous grasslands, that having lower abundance of hoverflies have also less species. However, since it has been proven that hoverfly species richness is positively affected by resource heterogeneity and hoverfly density depends on resource quantity, calcareous grasslands may prove themselves as important habitat types for hoverflies. If embedded in a highly diverse landscape matrix, calcareous grasslands, thanks to their richness in plants species and flower abundance, may help sustain diverse hoverfly communities (Meyer et al., 2009). The same thing could be possible for abandoned quarries, but further studies on the matter are needed.

From the results of the bioindicator analysis, two hoverfly species emerged as statistically significant indicators for the forest margin habitat type: *Episyrphus balteatus* (De Geer, 1776) and *Melanostoma scalare* (Fabricius, 1794).

*E. balteatus* is the only species of the genus *Episyrphus* in Europe and it is an almost ubiquitous and highly anthropophilic hoverfly. Its flight period is from February to November, but overwintering adults can be occasionally seen flying during colder months on exceptionally mild and sunny days. Adults usually visit a wide range of white and yellow flowers found in open spaces, but their preferred habitat is the forest where they can find resting and hibernating refugia in agricultural landscapes. The larvae are mainly aphidophagous and find their prey on low vegetation and crops, but they can also feed on a wide range of other arthropods (Speight, 2017). *M. scalare* is one of the six species of the genus *Melanostoma* found in Europe. It is mainly found in humid/mesophilous forests, but being highly anthropophilic can be also found along edges of farmlands and gardens or parks. Its flight period is from March to September, and it flies low over ground vegetation and bushes, but also along streams and poorly drained locations. The larvae are mainly aphidophagous, looking for their prey in the woodland litter, but they can also prey on other arthropods such as other co-occurring hoverfly larvae (Speight, 2017). In an agricultural landscape, species like *E. balteatus* and *M. scalare* are very important allies because they are effective antagonists of crop infesting aphids as larvae, and pollinators in flowering crops as adults (Meyer et al., 2009). Furthermore, they can be used as bioindicators to assess if the ecosystem is stressed (Sommaggio, 1999). *E. balteatus* is particularly sensitive to the use of pesticides in crop fields, which can lead to a 100% mortality of the larvae and to a decrease in fecundity of the females that have developed in treated crops (Speight, 2017). The larvae of *E. balteatus* and *M. scalare* do not solely depend on aphids, but they can integrate their diet with alternative prey usually found in nearby forest margins and woodland litter (Speight, 2017; Bortolotto et al., 2016). Moreover, since adults depend on the amount of flower resources, the lack of semi-natural habitats for more crop fields may result in a limiting factor for their feeding habits (Meyer et al., 2009). Therefore, the absence of these species could mean that one or more of these conditions are not satisfied and can be used as bioindicators to determine the state of the habitat type they are associated with.

## Conclusions

Hoverfly abundance and hoverfly species richness both responded to the same environmental drivers. They were the highest in the forest margin habitat type, they decreased over the sampling rounds with the increase in temperature, and they were positively affected by the percentage of semi-natural habitat present in the landscape. Regarding wild bees' abundance, it did not respond to the considered environmental drivers in a statistically significant way, but it should be noted that we could not take into account the species richness of bees. From the bioindicator analysis, two species: *Episyrphus balteatus* and *Melanostoma scalare* resulted to be strongly associated with the forest margin; probably because forest margin is the habitat type that best satisfies their ecological needs.

Our results confirmed the positive effect of the amount of semi-natural habitat on pollinators abundance, suggesting that land managers could increase the amount of semi-natural habitats and their diversity between crop fields to benefit pollinators. Doing so, pollinators such as wild bees and hoverflies could use these patches of semi-natural habitats to find the needed foraging and nesting resources and to move more easily and more freely between cultivated fields, increasing the efficiency of their ecosystem services.

For further studies, it could be interesting to carry out more sampling rounds in early spring and autumn which are the two seasons in which fly the highest amounts of hoverfly species (Speight, 2017). Finally, using the database from Syrph the Net, it could be interesting to see the characterization of different habitat types based on their richness in hoverfly species, and to see if there are even more hoverfly species that could be used as bioindicators for the biodiversity of the considered habitat types.

## Bibliography

- Aguirre-Gutiérrez, J., Biesmeijer, J. C., van Loon, E. E., Reemer, M., WallisDeVries, M. F., & Carvalheiro, L. G. (2015). Susceptibility of pollinators to ongoing landscape changes depends on landscape history. *Diversity and Distributions*, *21*(10), 1129-1140.
- Betts, M. G., Hadley, A. S., & Kormann, U. (2019). The landscape ecology of pollination. *Landscape Ecology*, *34*, 961-966.
- Bortolotto, O. C., Menezes Júnior, A. D. O., Hoshino, A. T., & Campos, T. A. (2016). Distance from the edge of forest fragments influence the abundance of aphidophagous hoverflies (Diptera: Syrphidae) in wheat fields. *Acta Scientiarum. Agronomy*, *38*, 157-164.
- Butaye, J., Adriaens, D., & Honnay, O. (2005). Conservation and restoration of calcareous grasslands: a concise review of the effects of fragmentation and management on plant species. *BASE*, *9* (2), 111-118.
- Cary, L. E. (1966). *A study of forest margins*. University of Wyoming.
- Chowdhury, S., Jennions, M. D., Zalucki, M. P., Maron, M., Watson, J. E., & Fuller, R. A. (2023). Protected areas and the future of insect conservation. *Trends in Ecology & Evolution*, *38*(1), 85-95.
- Cole, L. J., Brocklehurst, S., Robertson, D., Harrison, W., & McCracken, D. I. (2017). Exploring the interactions between resource availability and the utilisation of semi-natural habitats by insect pollinators in an intensive agricultural landscape. *Agriculture, Ecosystems & Environment*, *246*, 157-167.
- Cooke, R., Mancini, F., Boyd, R. J., Evans, K. L., Shaw, A., Webb, T. J., & Isaac, N. J. (2023). Protected areas support more species than unprotected areas in Great Britain, but lose them equally rapidly. *Biological Conservation*, *278*, 109884.
- Danforth, B. N., Minckley, R. L., & Neff, J. L. (2019). *The solitary bees: biology, evolution, conservation*. Princeton University Press.

Dicks, L. V., Viana, B., Bommarco, R., Brosi, B., Arizmendi, M. D. C., Cunningham, S. A., ... & Potts, S. G. (2016). Ten policies for pollinators. *Science*, 354(6315), 975-976.

Doyle, T., Hawkes, W. L., Massy, R., Powney, G. D., Menz, M. H., & Wotton, K. R. (2020). Pollination by hoverflies in the Anthropocene. *Proceedings of the Royal Society B*, 287(1927), 20200508.

Garratt, M. P., Senapathi, D., Coston, D. J., Mortimer, S. R., & Potts, S. G. (2017). The benefits of hedgerows for pollinators and natural enemies depends on hedge quality and landscape context. *Agriculture, Ecosystems & Environment*, 247, 363-370.

Gilbert, F. (2005, October). The evolution of imperfect mimicry. In *Symposium-Royal Entomological Society of London* (Vol. 22, p. 231).

Gillespie, M. A., Baude, M., Biesmeijer, J., Boatman, N., Budge, G. E., Crowe, A., ... & Kunin, W. E. (2022). Landscape-scale drivers of pollinator communities may depend on land-use configuration. *Philosophical Transactions of the Royal Society B*, 377(1853), 20210172.

Grundel, R., Jean, R. P., Frohnapple, K. J., Glowacki, G. A., Scott, P. E., & Pavlovic, N. B. (2010). Floral and nesting resources, habitat structure, and fire influence bee distribution across an open-forest gradient. *Ecological applications*, 20(6), 1678-1692.

Gutiérrez-Arellano, C., & Mulligan, M. (2020). Small-sized protected areas contribute more per unit area to tropical crop pollination than large protected areas. *Ecosystem Services*, 44, 101137.

Gullan, P. J., & Cranston, P. S. (2014). *The insects: an outline of entomology*. John Wiley & Sons.

Jean-Pierre Sarthou, Véronique Sarthou, Martin C.D. Speight (2023) *Illustrated key to the hoverfly genera of Europe (Syrphidae and Microdontidae)*.

- Johnson, M. G., Glass, J. R., Dillon, M. E., & Harrison, J. F. (2023). How will climatic warming affect insect pollinators?. In *Advances in insect physiology* (Vol. 64, pp. 1-115). Academic Press.
- Lawson, D. A., & Rands, S. A. (2019). The effects of rainfall on plant–pollinator interactions. *Arthropod-Plant Interactions*, *13*(4), 561-569.
- Linsley, E. (1958). The ecology of solitary bees. *Hilgardia*, *27*(19), 543-599.
- Majewska, A. A., & Altizer, S. (2020). Planting gardens to support insect pollinators. *Conservation Biology*, *34*(1), 15-25.
- Matteson, K. C., Ascher, J. S., & Langellotto, G. A. (2008). Bee richness and abundance in New York City urban gardens. *Annals of the Entomological Society of America*, *101*(1), 140-150.
- Meeussen, C., Govaert, S., Vanneste, T., Calders, K., Bollmann, K., Brunet, J., ... & De Frenne, P. (2020). Structural variation of forest edges across Europe. *Forest Ecology and Management*, *462*, 117929.
- Meyer, B., Jauker, F., & Steffan-Dewenter, I. (2009). Contrasting resource-dependent responses of hoverfly richness and density to landscape structure. *Basic and Applied Ecology*, *10*(2), 178-186.
- Miličić, M., Vujić, A., & Cardoso, P. (2018). Effects of climate change on the distribution of hoverfly species (Diptera: Syrphidae) in Southeast Europe. *Biodiversity and Conservation*, *27*, 1173-1187.
- Moquet, L., Laurent, E., Bacchetta, R., & Jacquemart, A. L. (2018). Conservation of hoverflies (Diptera, Syrphidae) requires complementary resources at the landscape and local scales. *Insect Conservation and Diversity*, *11*(1), 72-87.
- Noël, G., Caetano, J., Blanchard, S., Boullis, A., & Francis, F. (2022). High temperatures adversely affect the hoverfly *Episyrphus balteatus* (Diptera: Syrphidae) fitness and aphid prey consumption. *Turkish Journal of Zoology*, *46*(2), 186-193.

- Novák, J., & Konvička, M. (2006). Proximity of valuable habitats affects succession patterns in abandoned quarries. *Ecological Engineering*, 26(2), 113-122.
- Patrício-Roberto, G. B., & Campos, M. J. (2014). Aspects of landscape and pollinators—What is important to bee conservation?. *Diversity*, 6(1), 158-175.
- Ricarte, A., Ángeles Marcos-García, M., & Moreno, C. E. (2011). Assessing the effects of vegetation type on hoverfly (Diptera: Syrphidae) diversity in a Mediterranean landscape: implications for conservation. *Journal of Insect Conservation*, 15, 865-877.
- Scaven, V. L., & Rafferty, N. E. (2013). Physiological effects of climate warming on flowering plants and insect pollinators and potential consequences for their interactions. *Current zoology*, 59(3), 418-426.
- Schirmel, J., Albrecht, M., Bauer, P. M., Sutter, L., Pfister, S. C., & Entling, M. H. (2018). Landscape complexity promotes hoverflies across different types of semi-natural habitats in farmland. *Journal of Applied Ecology*, 55(4), 1747-1758.
- Senapathi, D., Goddard, M. A., Kunin, W. E., & Baldock, K. C. (2017). Landscape impacts on pollinator communities in temperate systems: evidence and knowledge gaps. *Functional ecology*, 31(1), 26-37.
- Sommaggio, D. (1999). Syrphidae: can they be used as environmental bioindicators?. *Agriculture, ecosystems & environment*, 74(1-3), 343-356.
- Song, Y., Zhou, D., Zhang, H., Li, G., Jin, Y., & Li, Q. (2013). Effects of vegetation height and density on soil temperature variations. *Chinese Science Bulletin*, 58, 907-912.
- Speak, A. F., Mizgajski, A., & Borysiak, J. (2015). Allotment gardens and parks: Provision of ecosystem services with an emphasis on biodiversity. *Urban Forestry & Urban Greening*, 14(4), 772-781.
- Speight, M. C. D. (2017). Species accounts of European syrphidae, 2017. *Syrph the net, the database of European Syrphidae (Diptera)*, 97, 294.

Westrich, P. (1996, July). Habitat requirements of central European bees and the problems of partial habitats. In *Linnean Society symposium series* (Vol. 18, pp. 1-16). Academic Press Limited.

Yuan, J. G., Fang, W., Fan, L., Chen, Y., Wang, D. Q., & Yang, Z. Y. (2006). Soil formation and vegetation establishment on the cliff face of abandoned quarries in the early stages of natural colonization. *Restoration Ecology*, 14(3), 349-356.

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