Research for Practice
LAIMBURG JOURNAL

Full Paper

The interest of apple orchard vegetation for the pollen supply of honey bees in South Tyrol (Italy).

Die Bedeutung der Vegetation von Apfelanlagen für die Pollenversorgung von Honigbienen in Südtirol (Italien)

L'importanza della vegetazione dei meleti per l'approvvigionamento di polline delle api mellifere in Alto Adige (Italia)

Jacob Geier[1], Edith Bucher[2], Manfred Wolf[1], Benjamin Mair[1], Werner Kofler[3], Jean Nicolas Haas[3]

[1]Laimburg Research Centre, 39040 Auer/Ora, Italy

[2]Autonomous Province of Bolzano/Bozen - Office for Nature, 39100 Bozen/Bolzano, Italy

[3]University of Innsbruck, 6020 Innsbruck, Austria

ABSTRACT

Fruit cultivation is one of the main economic sectors in South Tyrol (Italy) with one of the largest fruit production areas in the EU. In some parts of South Tyrol, more than 75% of the agricultural land is used for the cultivation of fruit trees, such as apples (*Malus domestica*). Almost 3500 beekeepers are managing 38 000 honey bee colonies, mainly in the valleys between 200 and 600 m a.s.l.. In this article, we aim to elucidate to what extent these fruit tree areas are visited and used by honey bees, especially outside the apple tree blossom period. We investigated the importance of apple orchards for the supply of pollen to honey bees during a period of five years (2016-2020), using palynological analyses of honey bee pollen pellets combined with vegetation surveys inside apple orchards. The results clearly indicate that the local apple orchards were the major pollen source for honey bees during the four weeks of apple tree blossom. However, in the weeks before the beginning of the apple blossom, and in the six weeks after, the apple orchards played a minor role in the supply of pollen to honey bees. Throughout the seasons investigated, flowers of a wide range of entomophilous and anemophilous plant species were the main sources of pollen. Broadleaved trees and shrubs contributed major parts of the pollen supply,whereas herbaceous plants, such as *Veronica* sp., *Plantago lanceolata* and *Verbascum* sp. were frequented far less by the honey bees. This suggests, that the honey bees may prefer pollen sources that provide large quantities of pollen, even though their flowers may not always produce nectar.

KEYWORDS

honey bee, pollen pellets, pollination, fruit cultivation

CITE ARTICLE AS

Geier Jacob, Bucher Edith, Wolf Manfred et.al. (2024). The interest of apple orchard vegetation for the pollen supply of honey bees in South Tyrol (Italy). Laimburg Journal 06/2024 DOI:10.23796/LJ/2024.018.

CORRESPONDING AUTHOR

Jacob Geier, Laimburg 6, Pfatten/Vadena, 39040 Auer/Ora, BZ, Italy,
geier.jacob@gmail.com,
+390471969643

Tab. 1: Coordinates and altitude of the locations of the beehives used in this study.

Abbreviation	Location	Coordinates	Altitude
DT	Dorf Tirol	N 46.674671 °, E 11.165658 °	460 m
LN1	Lana (2016-2019)	N 46.619248 °, E 11.144114 °	310 m
LN2	Lana (2020)	N 46.626026 °, E 11.165850 °	280 m
RB	Rabland	N 46.674004 °, E 11.054841 °	560 m

INTRODUCTION

The European honey bee (*Apis mellifera*) is the most commonly exploited insect species for crop pollination worldwide, except in Antarctica [1] [2] [3] [4] [5] [6]. Honey bees are generalists and pollinate a broad range of plant species, from trees and shrubs to herbs [7]. According to Ollerton et al. [8], approximately 87.5% of the angiosperms are pollinated by animals. Even though lots of crop species do not need pollinators to produce fruits or seeds, McGregor [4] estimated that around one-third of the human plant diet needs insect pollination. Apart from the fact that this ecosystem service increases yield quantity, there are several studies showing that the quality of the yield is also improved [9] [10] [11]. According to Gallai et al. [12], the economic value of crop pollination by insects corresponded to 14.2 x € 10^9 within the European Union (EU-25) in 2005. Apple trees (Malus domestica) are one of the main crops to depend heavily on insect pollinators [13].

According to the South Tyrolean Agriculture and Forestry report [14], after Poland, Italy was the second largest apple producing country (2.2 x 10^6 t) in the EU in 2019. Almost half of all apples produced in Italy were cultivated in the province of South Tyrol, representing the biggest contiguous apple tree cultivation area in the EU with 18 333 ha [14] [15]. In South Tyrol there are almost 3500 beekeepers, taking care of 38 000 honey bee colonies, all of which are potential pollinators of apple orchards [14]. In some parts of this region, the apple orchards take up more than 75% of the agricultural land in the valleys [14]. During the main apple tree blossom period in spring, many beekeepers place their hives in or near apple orchards, as the flowers of apple trees are an excellent source of pollen and nectar for honey bees [16]. Although honey bees can fly several kilometres to a fodder resource, they often prefer to forage in the vicinity of their hives [17]. Therefore, it is likely that in the main apple cultivation areas the orchards' function as a floral resource is not just limited to the apple tree blossom but lasts for the vegetation period before and afterwards. Geier et al. [18] investigated the impact of understory management in apple cultivation on potential pollen sources for honey bees in the understory of apple orchards and according to Granata et al. [19], the management of the understory strongly affects the abundance of flower visiting insects in apple orchards. Russo and Danforth [20] studied the pollen preferences of different bee species visiting apple orchards of *Malus pumila* in New York (USA), but their investigations were limited solely to the apple tree blossom period of a few weeks in spring. Another study conducted in South Tyrol and nearby Trentino investigated the botanical origin of pesticide residues in pollen pellets collected by honey bees during and two weeks after the apple tree blossom period, but not for the beginning of the vegetation period [21]. Mair and Wolf investigated the development of honey bee populations in South Tyrol [22] and examined corbicular pollen loads of these honey bee colonies for pesticide residues and their botanical origin [23]. However, they did not provide any information on the abundance of these pollen sources in apple orchards. Therefore, overall, there is only a small amount of data available regarding the importance of apple orchards for honey bees outside the main apple tree blossom period.

This study aims to investigate the botanical origin and amounts of pollen gathered by honey bees in four localities around the town of Meran in South Tyrol between 2016 and 2020. We hypothesised that honey bees also visit apple orchards before and after the blossom period to gather pollen from plant species growing in the understory, due to a lack of pollen resources in the surrounding vegetation. Therefore, we compared the botanical origin of the corbicular pollen loads collected with survey data about the understory vegetation in apple orchards in order to understand the importance of apple orchards as floral resources and pollen fodder for honey bees from the beginning of the vegetation period in March until the end of June.

METHODS

DATA AQUISITION

Palynological data on the botanical origin and amount of pollen gathered by honey bees were compiled for the years 2016-2020, whereas vegetation surveys were only conducted in 2017, 2019 and 2020 [18] [24]. Since several people were involved in gathering the data, such as the palynological analyses and vegetation surveys, there were minor differences in the methods utilised and the level of taxonomic determination for pollen types and plant species. For this reason, the higher plant taxonomic levels were used for all data elaboration if different taxonomic levels were recorded.

STUDY SITES

The study presented here was conducted in the main apple cultivation area of the province of South Tyrol, Italy (Fig. 1; Tab. 1) near three villages around the town of Meran: Dorf Tirol (460 m a.s.l.), Lana (280-310 m a.s.l.) and Rabland (560 m a.s.l.). The study sites were very

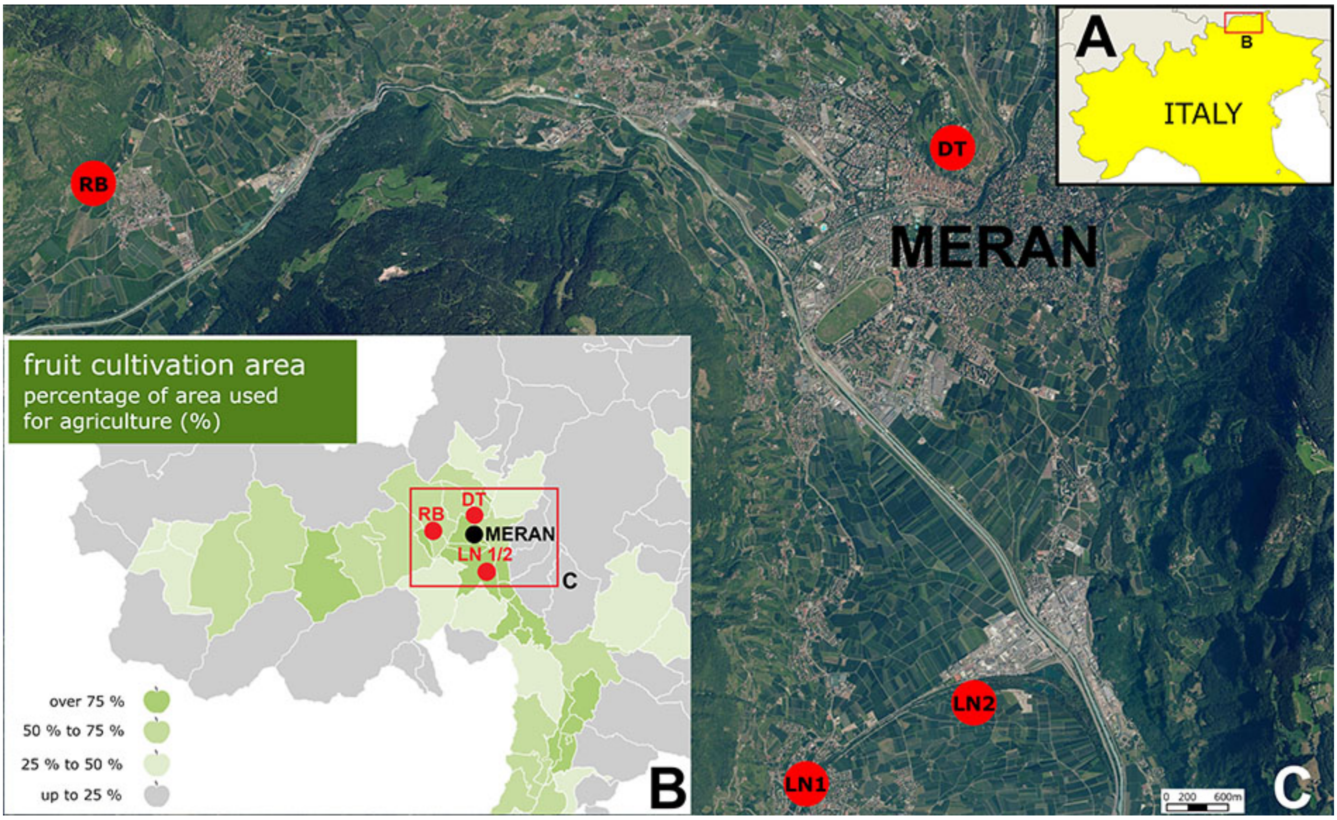

Fig. 1: A. Map showing the research area of South Tyrol in Italy, and B. map of the fruit cultivation area with land percentages used for fruit tree orchards (including the abbreviations of our research locations), and C. landscape context of the three research locations around the town of Meran. Abbreviations: DT = Dorf Tirol; LN = Lana; RB = Rabland (source: QGIS.org, 2021).

close to apple orchards but differ by nearly 300 m in their altitude, therefore also differing in the blossoming time of the apple trees and the understory plant species. Furthermore, there were also differences in the surrounding vegetation outside the apple orchards in the study. At each location, a) two honey bee colonies of the taxon *Apis mellifera* ssp. *carnica* were installed for pollen pellet sampling, and b) ten apple orchards, each close to these beehives, were selected for the detailed vegetation surveys. In 2020, the location of the beehives in Lana had to be changed because of altered management plans, and the beehives were positioned about 1.8 km northeast of the previous site at an apiary with very similar conditions (Fig. 1, LN1 and LN2). However, the apple orchards used for the vegetation surveys were the same as during the previous years (2017, 2019).

VEGETATION SURVEY

The vegetation surveys were conducted in ten apple orchards per study site in the years 2017, 2019 and 2020 [18] [24] and focused on dicotyledonous angiosperms in the understory vegetation. The orchards investigated were the same for all of the years, except for the two sites at Rabland and one at Lana. These three orchards had to be replaced because of changed management plans, i.e. they were cleared and newly planted with apple trees in spring 2019. Therefore, an exact comparison of the vegetation understory of the orchards was only possible for the years 2019 and 2020.

Once a week, a set of different parameters was recorded in the field for each orchard: ground cover, height of the according vegetation, mowing events, generative status of apple trees, as well as the percentage of the vegetative cover and the flowering status of plant species in the understory. To ensure that the area examined was the same every week, a concrete pillar was marked. Then, a 2 m long folding rule was placed next to a pile to define the length of the sampling area; the width was defined by the adjacent rows of apple trees. The plant cover of the understory was quantitatively recorded using the cover-abundance scale defined by Braun-Blanquet [25], and additionally, information was recorded about whether a plant species had open flowers or not.

The method used for the vegetation survey in 2017 was similar, with a few minor differences: the length of the sampling area was 1.5 m, the plant cover was estimated only for flowering species, and the presence of non-flowering plants was just noted.

The values for the number of orchards were calculated as means over the three years 2017, 2019 and 2020, considering the data from all the investigated apple orchards in the three locations. However, the

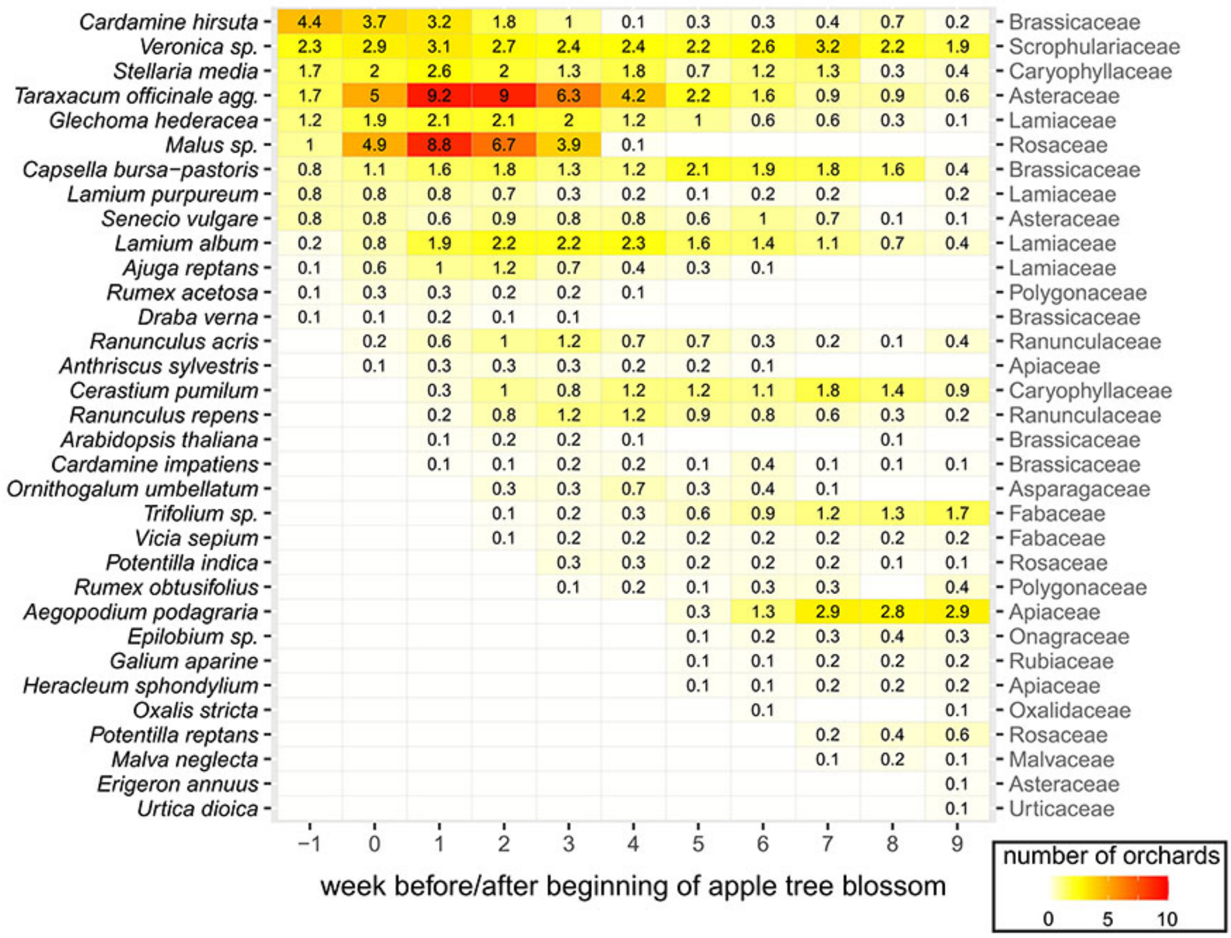

Fig. 2: Average number of apple orchards with the listed plant species flowering at a given week within the three investigated years (2017, 2019 and 2020) in the research area of South Tyrol, Italy. The red colour corresponds to a plant species flowering in almost all orchards, the white colour indicates that the plant species was not flowering in any of the orchards during the investigated seasons from March to June.

data were calibrated using the day of the beginning of the apple tree blossom period each year as a reference point ("day 0"), in order to minimise the impact of the variance in phenology of the different yearly seasons, caused by abiotic factors such as climate.

PALYNOLOGY

Sampling of pollen pellets

Self-made, front-mounted external pollen traps were installed in front of the beehives to harvest the pollen pellets collected by honey bees from March to June. The traps were activated once a week in the morning, with favourable weather conditions for honey bees (no rain or heavy winds), and deactivated/emptied after 24 hours. Thereafter, the collected pollen pellets were stored at -25 °C. At least one sample was available per week and location and subsequently analysed; however, for few weeks no samples were available at all - especially in 2016 and 2018.

Palynological analyses

The palynological analyses were performed partially in the Biological Laboratory of the Environment Protection Agency in Bolzano, and partially at Laimburg Research Centre (both in South Tyrol). The laboratories used different methods to prepare the slides and quantify the pollen types.

A) Biological Laboratory of the Environment Protection Agency in Bolzano (Bolzano, Italy):

A subsample of 1 g of pollen was taken and diluted with 30 mL of Milli-Q water and then agitated at 100 rps for 30 min. 1 mL of the solution was further diluted in 6 mL of Milli-Q water and again agitated at 100 rps for 30 min. Two drops of 60 mL were placed separately on a microscope slide. After drying, one drop of glycerine jelly with fuchsine (Lanzoni S.r.l) was placed over each pollen sediment and covered with a thin glass sheet. Afterwards 1000 grains (500 + 500) of pollen were counted for each slide. The percentages of grains belonging to the different pollen types were used to indicate the composition of the sample.

B) Laimburg Research Centre (Ora, Italy):

A subsample of 0.5 g was taken from a sample. Each pellet was then placed on a slide and mixed with some drops of soapsuds. After all the water evaporated from the slides, the mounting medium ("gel-

Fig. 3: survey sampling area inside an apple orchard in Lana. Both pictures show the same sampling area, but in different times of the season. a) 10.4.2020 during the apple tree blossom, b) 8.5.2020 after the apple tree blossom.

vatol", consisting of polyvinyl alcohol, A. dest. and glycerol) containing basic fuchsine was added. Every slide was analysed under the microscope to determine the pollen type of the pellet used. The numbers of pollen pellets per pollen type were then used to calculate the percentage of the sample.

C) Determination of pollen grains:

The determination of the pollen grains was performed using a light microscope (Olympus BX50 or Leica DM2500 LED), analysing different morphological traits of the pollen grains (shape, size, apertures, surface etc.). Generally, 400X magnification was used, however, in some cases 1000X magnification with oil immersion and sometimes phase-contrast was necessary to identify the pollen types. In addition to palynological determination literature [26] [27] [28] and online databases [29] [30] [31] [32], the scientists' own pollen reference slide collection (located at Laimburg Research Centre) of plant species characteristic to South Tyrol was used. The level of taxonomic determination varies between different plant species; for some pollen, the determinable level by light microscopy is limited to the family level, for others it is possible to identify the plant genus or species [33]. In a few cases, no final palynological determination was possible. The figure showing the main pollen sources (see below) was created using Tilia software [34]. All other graphics were created with RStudio [35].

The pollen values determined were also calculated as means over all investigated years (2016-2020) for all beehives investigated in all four locations. Here again, the data were calibrated using the day of the beginning of the apple tree blossom period in every year as a reference point ("day 0").

RESULTS

FLOWERING PLANT SPECIES IN THE UNDERSTORY

One of the main goals of this study was to identify the plant species growing in the understory in apple orchards that honey bees could use as a source of fodder. Figure 2 shows the number of apple orchards in which the observed plant species were flowering in a given week for the period from March to June before/after the apple tree blossom period. It is evident that most plant

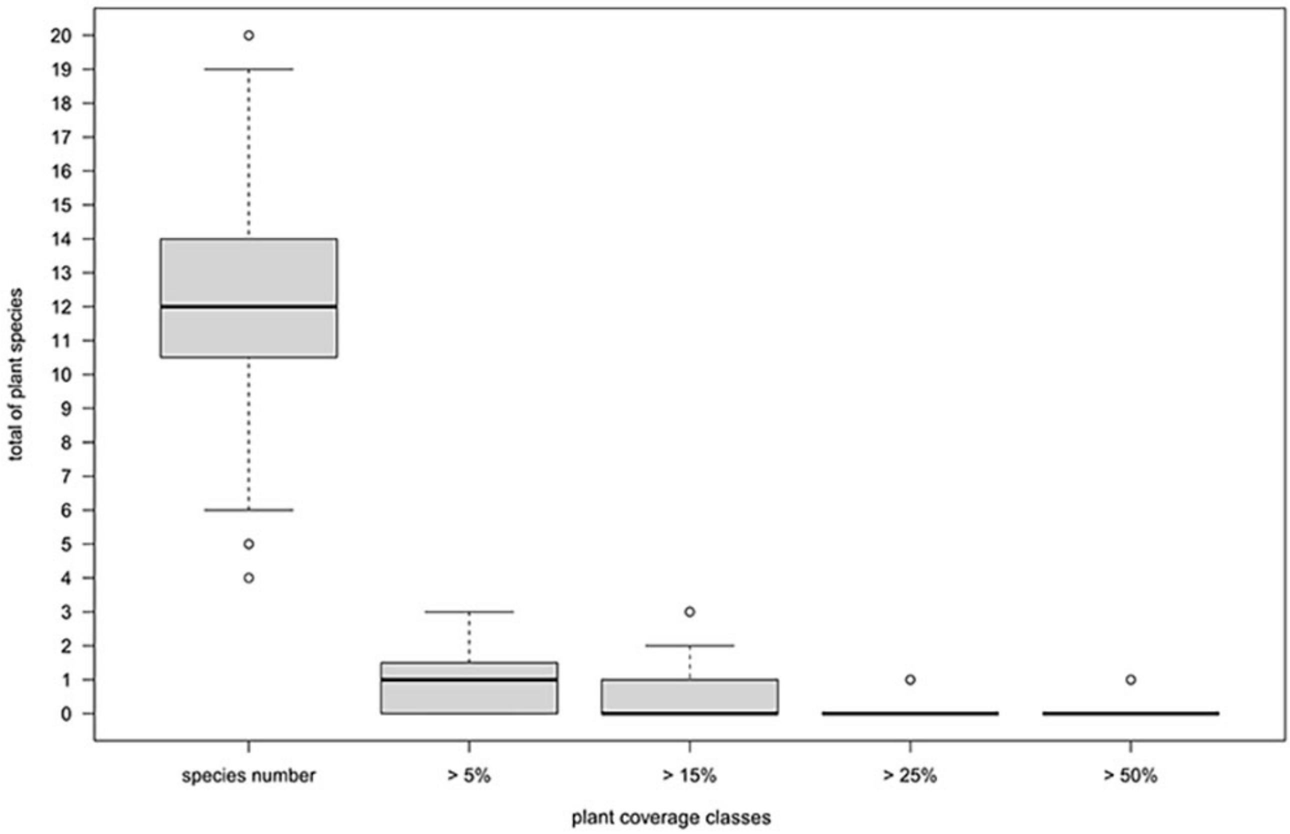

Fig. 4: Boxplot representing the vegetative ground cover of plant species found in the understory of apple orchards within two of the investigated years (2019, 2020) in the research area in South Tyrol, Italy. The total number of plant species per orchard is shown on the left, followed by how many of them reached the different plant coverage classes of >5, 15, 25, and 50% (as defined by Braun-Blanquet 1964 [25]). The bold line shows the according median for each coverage class, the box represents the 75th and 25th percentile, and the whiskers indicate the minima/maxima. Circles define outliers.

species flowered in a specific time range and mostly just in a few orchards. However, eight species/-genera (*Cardamine hirsuta*, *Veronica* sp., *Stellaria media*, *Taraxacum officinale* agg., *Glechoma hederacea*, *Capsella bursa-pastoris*, *Senecio vulgare* and *Lamium album*) flowered consistently in several apple orchards over the years studied (Fig. 2). Only *Taraxacum officinale* agg. flowered in most orchards at the same time during its main blossoming period and this overlapped with the apple tree blossom for several weeks (Fig. 2). Compared to this dandelion, all of the other plants mentioned above flowered only in a limited number of apple orchards, even though they flowered during the whole observation period. However, outside of the apple blossom period, there were always several flowering species present in some of the orchards (Fig. 2, Fig. 3).

Figure 4 summarises the number of species per apple orchard reaching the different cover classes according to the cover-abundance scale by Braun-Blanquet [25]. On average, only 0.88 species per orchard reached a cover of more than 5% at least once. This number decreased drastically for higher cover classes, with only 0.47 taxa, which corresponds to about one species in two orchards, reaching a cover of at least 15%; the number of plant species per orchard reaching a cover of 25% was 0.12, and only 0.05 species, corresponding to one species in 20 orchards, covered more than 50% of the investigated area.

However, the overall understory was generally higher, as grasses, which were not included in the vegetation surveys of this study, covered most of the ground between the rows of apple trees. In the research sites in Dorf Tirol and Lana, the ground cover was over 70% on half of the observed research fields, with Lana showing a slightly higher variation. The apple orchards in Rabland had more than 80% ground cover during 50% of the observations, and at this location too, the variation was high. The total of our locations had a median of 70% ground cover, being 60% or more on three quarters of the observation fields.

PALYNOLOGY

Overall, 158 different pollen types were identified by analysing 177 different honey bee pollen pellet samples collected between 2016 and 2020. The 32 main pollen taxa and plant resources are illustrated in Figure 5. Only pollen types that exceeded the threshold of 10% in a pollen pellet at least once were considered as main pollen sources. Again, the data was calibrated, using the day of the beginning of the apple tree blossom period each year as a reference point. Figure 5 represents the majority of the pollen collected, since the graph "other species" does not exceed 20% in most cases. It is clearly evident that the main sources of pollen are the flowers of trees and shrubs. Only four of the main pollen sources are herbaceous plants: *Veronica* sp., *Plantago lanceolata*, *Verbascum* sp. and *Knautia* sp.. In some cases it was not possible to determine the pollen type. Figure 5 also shows the seasonal progress of the pollen types collected by honey bees. Some pollen were almost always only collected at the beginning of the season (*Populus* sp., *Parrotia* sp., *Acer negundo*, *Veronica* sp.), whereas others were not collected earlier than 40 days after the apple trees began to blossom (*Castanea sativa*, *Verbascum* sp.). The pollen type reaching the highest percentage was that of *Acer negundo*, with a maximum of almost 77%, closely followed by Malus-type, with a maximum of 76%. Interestingly, the honey bees even collected pollen from flowers of anemophilous trees (*Populus* sp., *Parrotia* sp., *Acer negundo*, *Pinus* sp., *Quercus ilex*, Quercus-type). Finally, most pollen types were collected over several weeks, with some exceptions (*Parrotia* sp., *Verbascum* sp.).

In addition to the graphs of the main 32 pollen types, Figure 5 shows the proportion between pollen collected outside apple orchards, and pollen potentially collected inside apple orchards. It is evident that at the beginning of the plant flowering season in March, most of the pollen was collected outside the apple plantations. With the start of the apple tree blossom period (April), the percentage of pollen possibly collected inside the apple orchards increased from 10% to 81% within ten days. After the apple tree blossom period (second half of May) the percentage of pollen collected inside the orchards started to decrease again and stayed under 50% for the rest of the season (June), with one single exception, possibly related to climatic parameters for the years of the study: 56 days after the beginning of the apple tree blossom period, the amount of pollen collected inside apple orchards reached 60%.

DISCUSSION

Flowering plants growing between rows of apple trees inside apple orchards have both beneficial and unfavourable effects according to Merwin [36]. On the one hand, they can stunt the growth of young trees or decrease the apple yield, as they compete with the apple trees for essential resources [37] [38] [39]. On the other hand, serving as understory or ground cover, flowering plants can improve water infiltration [40], nutrient retention [41], and crop/fruit quality [42]. Such flowering plants can even help to protect the soil from erosion [43] and shearing or compressing forces [36]. Flowering understory vegetation can also have a positive effect on the biological controls of pests, by providing nectar, pollen or habitats to beneficial arthropods [44]. Accordingly, understory vegetation can be considered to be common weeds, having negative effects on crops in some cases but can also be very useful under other circumstances. In this study, the understory vegetation cover of apple orchards examined was between 60% and 85% in half of all vegetation surveys (Fig. 6). However, only a very few of the species found in the understory exceeded a maxi-

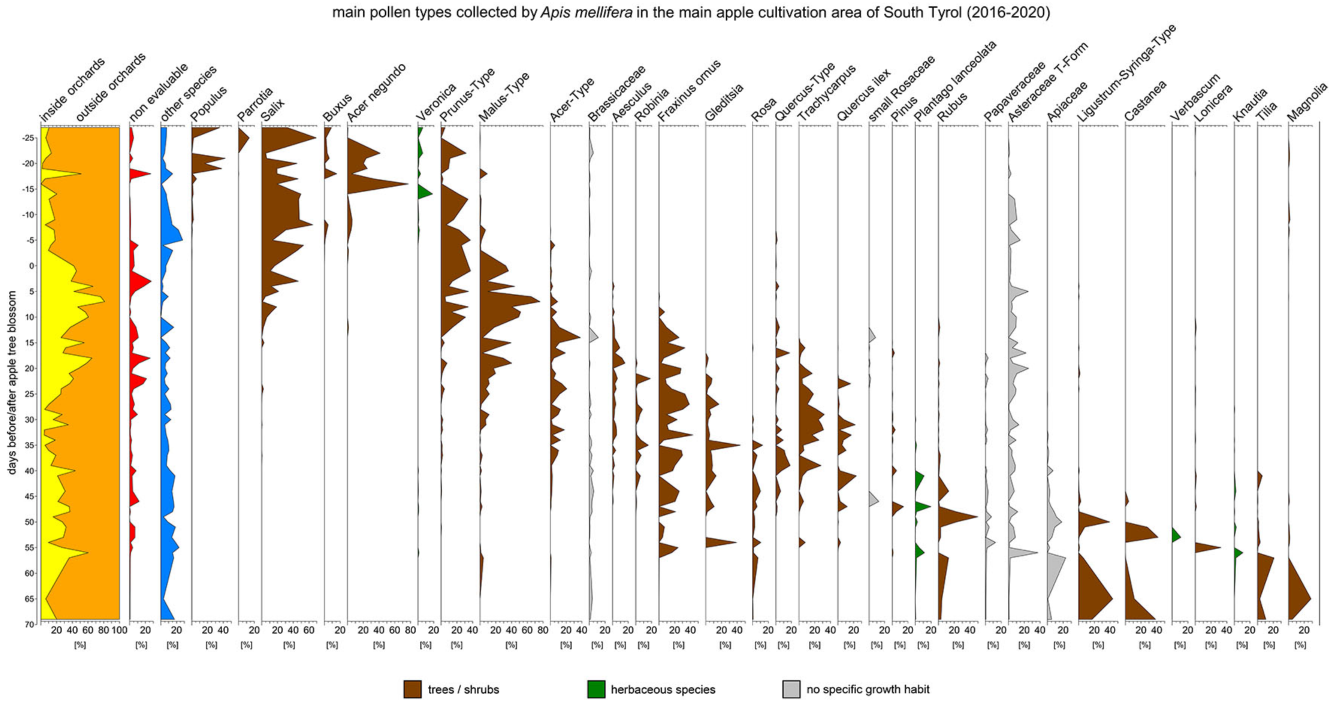

Fig. 5: Pollen collected by honey bees (*Apis mellifera* ssp. *carnica*) in the apple tree (*Malus domestica*) cultivation area of South Tyrol (Italy) within the investigated years (2016-2020) as determined to plant species, genera or family level. The Y-axis shows the days before or after the apple tree blossom, and the X-axis indicates (from left to right) i) the proportion of pollen collected inside (yellow) and outside apple orchards (orange), ii) the frequency of pollen types that were not determinable (red), iii) other pollen taxa (blue), and iv) the pollen frequency (in percentages) for 32 plant taxa (trees/shrubs in brown, herbaceous plants in green, and other plant families in grey) in the honey bee pellets aligned on the time axis.

mum vegetation cover of more than 5% (Fig. 4) and the flower abundances, which are the most interesting for insect pollinators, may be even smaller.

The main blossom period of the understory plants overlaps strongly with that of the apple tree blossom (Fig. 2), if we compare it in weeks 0-3 from the start. The increase in pollen collected inside the apple orchards (Fig. 5, left graph, yellow area, days 0-21 after the standardised “day 0”, where day 0 correlates to the start of the apple tree blossom) occurred synchronously. During the apple tree blossom, apple farmers usually do not mow the understory, and any application of pesticides, dangerous to honey bees is - in principle - prohibited by law [45]. In addition, the flowering plants in the understory of orchards, and the apple trees do not only compete for soil nutrients and water resources, but also for insect pollinators and pollination. As the peak blossom times of both dandelions (*Taraxacum officinale* agg.) and apple trees (*Malus domestica*) occur almost simultaneously (Fig. 2), it is likely that these two plant species compete strongly for insect pollinators. This could therefore also lead to the apple trees being insufficiently pollinated, with negative effects on the fruit quality [10]. Our palynological results demonstrate that Malus-type is clearly the main pollen being collected by honey bees during the apple tree blossom period (Fig. 5). Nevertheless, Free [46] argued that this type of competition also existed when considering other nectar collecting foragers and suggested that farmers should remove dandelions (*Taraxacum officinale* agg.) using selective herbicides to improve insect pollination. This clearly indicates that nectar collection by honey bees, wild bees, or other insect foragers should be considered more carefully when quantifying and analysing the pollination behaviour of honey bees.

Generally, in March, just before the apple tree blossom period, only a few flowering plants were recorded in the apple orchards. This situation changed dramatically as the season and apple tree blossom period progressed, with the number of flowering plant species increasing enormously. Together with the high variance in the numbers of plant species found in the apple orchards (Fig. 4), the vegetation understory must be considered to be very heterogeneous. However, outside the apple tree blossom period in April and early May, there were always flowering plant species present in some of the orchards, but mainly with a plant cover of less than 5%.

As insect generalists, honey bees are known to collect pollen from many different plant species [7]. However, during a foraging flight, honey bees usually tend to visit only one plant species [47]. Indeed, this was also observed in our study: pa-

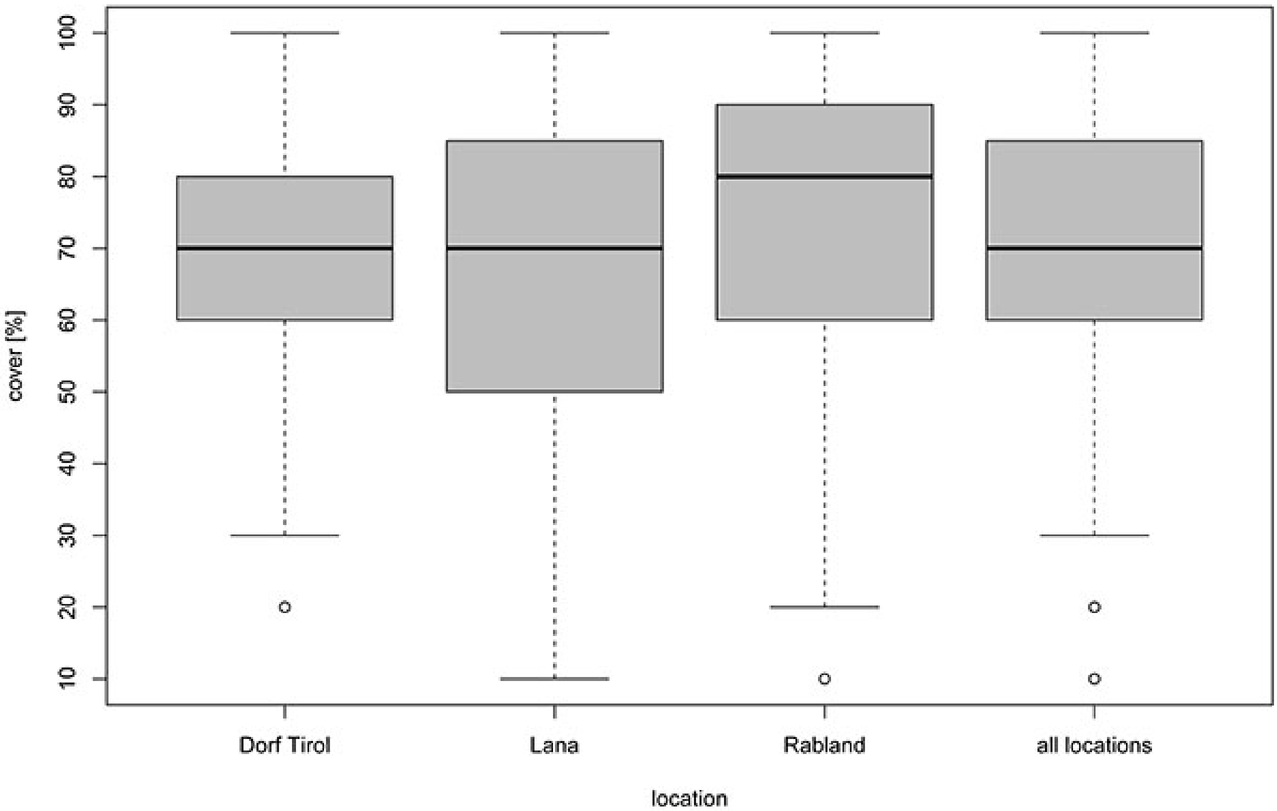

Fig. 6: Boxplot representing the vegetative ground cover inside apple orchards within the three investigated yeas (2017, 2019, and 2020) in the research area of South Tyrol (Italy). The bold lines show the according median for each coverage, the box represents the 75th and 25th percentile; the whiskers indicate the minimum/maximum and the circles are outliers.

lynological analyses of honey bee pollen pellets often revealed only one pollen-type. Interestingly, it is well known that the diversity and quality of pollen collected by honey bees strongly influence the physiology of the hives of these social insects [48], although it is still not clear what controls the honey bees' choice concerning their fodder resources. Some studies suggest that it may be the quality of the pollen [49] [50] [51], but other research implied that visual (colour), tactile and chemical signals emitted by flowers and their anthers may be crucial [52] [53]. Either way, it is clear that honey bees show preferences for certain flowers in order to collect particular pollen types [54], a fact also revealed in our present study.

As pollen is the main natural protein source for honey bees, this has a strong impact on the fitness and strength of a honey bee colony. An adequate pollen supply is crucial for the development of the honey bee larvae, but adult honey bees also benefit from a diet that includes pollen [55]. Here, the results of this study show that trees and shrubs were the main pollen sources for honey bees throughout the period under observation (Fig. 5). This coincides with the results of Donkersley [54], suggesting that this phenomenon could be determined by the higher amount of flowers in wind and insect-pollinated tree and shrub species in particular. In addition, Donkersley [56] even showed that in increasingly grassland-dominated landscapes, where trees are less abundant, honey bees do still consistently forage on the latter. In our study, the flowers of different wind-pollinated species were also visited very frequently, i.e. their pollen was collected in large quantities (Fig. 5), which compares well with other studies [57] [58] [59]. Anemophilous plants produce high amounts of pollen grains, which leads to high pollination and high yields [60]. Hence, anemophilous plants could be a very efficient way to secure a pollen supply for a honey bee colony, and therefore attract foraging honey and wild bees, even though no nectar is provided, and the flower morphology is not designed to attract insect pollinators. However, and on average, zoophilous or insect-pollinated plants do not have a higher pollen protein content than anemophilous plants [61]. This may indicate that honey bees do not actively select for protein-rich pollen. If that were the case, a difference in the pollen protein content would have to be expected [62]. Although pollen is the main and often only protein source for honey bees, it also contains other ingredients that may be of nutritional value [55]. According to van der Moezel et al. [63], clearly the abundance of flowering plant species is more important for pollen foraging, than the nutrient composition of the pollen. However, abundance of flowering plants alone does not seem to be the driving factor, as in some cases, some very abundant plant species were completely ignored by honey bees [64] [65], possibly due to their toxicity. It is a fact that the spectrum of flowering plant species in the honey bee pollen pellets collected does not fully represent the composition of vegetation surrounding the beehives [62]. So, over the course of the season, the abundance and temporal availability of the pollen resources are determining factors for the types of pollen found in the pollen pellets collected. Importantly, the flowering time of plants differs significantly between plant species [66] and mainly depends on different genetic, environmental, seasonal and climatic factors [6] [67]. Pollen from a specific flowering plant is therefore only available during a specific period or season, which is reflected in the spectrum of pollen collected (Fig. 5). In most of the analysed palynological samples in this study, there were just a few flowering plant species representing the major part of the collected pollen, which seems to be a common phenomenon [62]. Honey bees probably tend to visit the most available plants in terms of quantity daily, and the most attractive pollen sources, instead of less prominent ones.

The percentage of pollen collected outside the apple orchards by honey bees is probably even higher than the value shown in Figure 5. Also, some pollen types could only be identified at plant family level, whereas unidentified pollen types were considered to be “collected inside orchards”, to make sure that the calculation of the percentages of pollen collected outside the apple orchards was not increased artificially. Therefore, it is quite difficult to determine or differentiate the exact pollen percentages in a pollen pellet from a honey bee collected inside or outside apple orchards. The reason is that some flowering plant species are found both in the understory and outside apple orchards, and therefore it is impossible to know if pollen foraged from these plants was collected inside or outside the apple orchards. Finally, it should be considered that our self-made front-mounted external pollen traps installed in front of the beehives to harvest the pollen pellets collected by honey bees once a week during favourable weather conditions (no rain or heavy winds) from March to June, may have varied in their efficiency, and may even have influenced the foraging behaviour of the honey bee colonies [68].

CONCLUSIONS

The main goal of this study was to investigate the importance of apple orchards for the supply of pollen for honey bee colonies (*Apis mellifera* ssp. *carnica*) positioned in the apple-growing area of South Tyrol, Italy during several foraging seasons (2016-2020). During the apple tree blossom period in April and May, the orchards provided the honey bees with large quantities of pollen as fodder. Before and after that period, the apple orchards played a minor role in the honey bees’ pollen supply. Even though outside the apple blossom period (March and May/June) the amount of pollen collected in the understory of apple tree plantations was low, these flowering plants and their pollen loads remain a very important supply of fodder. In South Tyrol, apple orchards are very abundant, and lots of flowering plant species can be found in the understory throughout the investigated time span (March-June). Astonishingly, flowers from trees and shrubs were the main pollen resources for honey bees over each investigated season, including several anemophilous species. To get a better overview of the foraging behaviours and main pollen and fodder resources of honey bees throughout the whole season, the sampling periods should be expanded until autumn in future. Moreover, analysing the pollen content of honey bee stomachs by morphological or DNA analyses, as well as the differences in pollen spectra in the honey produced [69], could provide further insights into the nectar foraging behaviour of honey bees, not only in Central Europe but worldwide. Furthermore, this could help to understand essential aspects of pollination systems and biodiversity sustainment by honey bees.

ACKNOWLEDGMENTS

We would like to thank Veronika Ungerer who helped with collecting initial understory vegetation data in apple orchards in South Tyrol, and Stefan Tasser for maintaining the bee colonies used for this study. This work uses data collected during the "Palyn I" (Pf-en-19-3) and "Palyn II" (Pf-en-21-3) projects, which were supported and funded by i) the European Union for 2018-2020 in accordance with EU regulation no. 1308/2013, ii) the Autonomous Province of Bozen (Italy; Bureau for Husbandry), and iii) the South Tyrol Apple Consortium (Italy).

ZUSAMMENFASSUNG

Der Obstanbau ist einer der wichtigsten Wirtschaftszweige in Südtirol (Italien), einer Region mit einem der größten zusammenhängenden Obstanbaugebiete in der EU. In einigen Teilen Südtirols werden mehr als 75% der landwirtschaftlich genutzten Fläche für den Anbau von Obstbäumen, vor allem Äpfeln (Malus domestica), genutzt. Außerdem gibt es in Südtirol rund 3500 Imker, welche ungefähr 38 000 Honigbienenvölker bewirtschaften, vor allem in den Tallagen zwischen 200 und 600 m ü.NN. Diese Arbeit beschäftigt sich damit, inwieweit die Obstanbauflächen von Honigbienen aufgesucht und genutzt werden, vor allem außerhalb der Blütezeit der Apfelbäume. Hierfür wurden über einen Zeitraum von fünf Jahren (2016-2020) palynologische Analysen von Pollenhöschen durchgeführt und mit Vegetationsaufnahmen von Apfelanlagen verglichen, um die Bedeutung der Apfelplantagen für die Pollenversorgung von Honigbienen zu verstehen. Die Ergebnisse zeigen, dass die lokalen Apfelplantagen während der vier Wochen der Apfelblüte die wichtigste Pollenquelle für Honigbienen waren. In den Wochen vor Beginn der Apfelblüte und in den sechs Wochen nach der Apfelblüte spielen die Apfelplantagen jedoch eine untergeordnete Rolle für die Pollenversorgung der Honigbienen. Während der gesamten untersuchten Saison dienten die Blüten einer breiten Palette insektenbestäubter und windbestäubter Pflanzenarten als Hauptpollenquellen, vor allem die Blüten von Laubbäumen und Sträuchern. Krautige Pflanzen wie z.B. Veronica sp., Plantago lanceolata und Verbascum sp. wurden von den Honigbienen als Pollenquelle weit weniger besucht. Dies deutet darauf hin, dass Honigbienen für ihre Pollenversorgung Pflanzen bevorzugen, welche eine große Menge an Pollen produzieren, auch wenn deren Blüten kaum oder keinen Nektar enthalten.

RIASSUNTO

La frutticoltura è uno dei settori economici più importanti dell'Alto Adige (Italia), la regione con una delle più grandi aree frutticole contigue dell'UE. In alcune zone dell'Alto Adige, oltre il 75% dei terreni agricoli è utilizzato per la coltivazione di alberi da frutto, soprattutto mele (Malus domestica). In Alto Adige sono presenti anche circa 3500 apicoltori, che gestiscono circa 38 000 colonie di api mellifere, principalmente nelle valli tra i 200 e i 600 m s.l.m.. Questo studio si concentra sulla misura in cui le api mellifere visitano e utilizzano i frutteti, soprattutto al di fuori del periodo di fioritura dei meli. A tal fine, sono state effettuate analisi palinologiche del polline raccolto dalle api per un periodo di cinque anni (2016-2020) e sono state confrontate con analisi della vegetazione dei meleti, al fine di comprendere l'importanza dei meleti per l'approvvigionamento pollinico delle api mellifere. I risultati mostrano che i meleti locali sono stati la fonte di polline più importante per le api durante le quattro settimane di fioritura del melo. Tuttavia, nelle settimane precedenti e nelle sei settimane successive alla fioritura, i meleti svolgono un ruolo secondario nell'approvvigionamento di polline delle api. Durante l'intera stagione studiata, i fiori di un'ampia gamma di specie vegetali impollinate da insetti e dal vento sono stati le principali fonti di polline, soprattutto i fiori di alberi decidui e arbusti. Piante erbacee come Veronica sp., Plantago lanceolata e Verbascum sp. sono state usati molto meno frequentemente dalle api come fonte di polline. Ciò indica che le api mellifere preferiscono piante che producono una grande quantità di polline, anche se i loro fiori contengono poco o niente nettare.

REFERENCES

[1] Garibaldi L.A., Steffan-Dewenter I., Kremen C. et al. (2011). Stability of pollination services decreases with isolation from natural areas despite honey bee visits. Ecology Letters 14 (10), 1062-1072, DOI:10.1111/j.1461-0248.2011.01669.x.

[2] Garibaldi L.A., Steffan-Dewenter I., Winfree R. et al. (2013). Wild pollinators enhance fruit set of crops regardless of honey bee abundance. Science 339 (6127), 1608-1611, DOI:10.1126/science.1230200.

[3] Garibaldi L.A., Requier F., Rollin O. et al. (2017). Towards an integrated species and habitat management of crop pollination. Current Opinion in Insect Science 21, 105-114, DOI:10.1016/j.cois.2017.05.016.

[4] McGregor S.E. (1976). Insect pollination of cultivated crop plants. Agricultural Research Service, United States Department of Agriculture, Tucson, Arizona, USA. Retrieved November 10, 2021, from https://books.google.it/books?hl=de&lr=&id=mQoUAAAAYAAJ&oi=fnd&pg=PA1&dq=McGregor,+S.E.+(1976)+Insect+pollination+of+cultivated+crop+plants.+Agricultural+Research+Service,+US+Department+of+Agriculture&ots=AKDbef4dxi&sig=2v7T_VAj1avU5drCYaWge9XV3F0&redir_esc=y#v=onepage&q=McGregor%2C%20S.E.%20(1976)%20Insect%20pollination%20of%20cultivated%20crop%20plants.%20Agricultural%20Research%20Service%2C%20US%20Department%20of%20Agriculture&f=false.

[5] Mortensen A.N., Schmehl D.R., Ellis J. (2013). European honey bee Apis mellifera Linnaeus and subspecies (Insecta: Hymenoptera: Apidae). IFAS Extension, University of Florida, Gainesville, Florida, USA. Retrieved October 20, 2021, from https://edis.ifas.ufl.edu/publication/IN1005?downloadOpen=true.

[6] Oskay G.S, Uygur G.S., Oskay D. et al. (2023). Impact of stress factors internal and external to the hive on honey bees and their reflection on honey bee products. A review. Journal of Apicultural Research, DOI:10.1080/00218839.2023.2247840.

[7] Delaplane K.S., Mayer D.R., Mayer D.F. (2000). Crop pollination by bees. CABI Publishing, Wallingford, UK, here pp. 70-74.

[8] Ollerton J., Winfree R., Tarrant S. (2011). How many flowering plants are pollinated by animals? Oikos 120 (3), 321-326, DOI:10.1111/j.1600-0706.2010.18644.x.

[9] Chautá-Mellizo A., Campbell S.A., Bonilla M.A. et al. (2012). Effects of natural and artificial pollination on fruit and offspring quality. Basic and Applied Ecology 13 (6), 524-532, DOI:10.1016/j.baae.2012.08.013.

[10] Garratt M.P.D., Breeze T.D., Jenner N. et al. (2014). Avoiding a bad apple. Insect pollination enhances fruit quality and economic value. Agriculture, Ecosystems & Environment 184 (100), 34-40, DOI:10.1016/j.agee.2013.10.032.

[11] Wietzke A., Westphal C., Gras P. et al. (2018). Insect pollination as a key factor for strawberry physiology and marketable fruit quality. Agriculture, Ecosystems & Environment 258, 197-204, DOI:10.1016/j.agee.2018.01.036.

[12] Gallai N., Salles J.-M., Settele J. et al. (2009). Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. Ecological Economics 68 (3), 810-821, DOI:10.1016/j.ecolecon.2008.06.014.

[13] Morse R.A., Calderone N.W. (2001). The value of honey bees as pollinators of US Crops in 2000. Bee Culture 128 (3), 1-15. Retrieved November 17, 2021, from https://bp-dc.org/assets/media/documents/pollinators/documents/ValueofHoneyBeesasPollinators-2000Report.pdf.

[14] Autonome Provinz Bozen - Südtirol (2020). Agrar- und Forstbericht 2019. Retrieved November 06, 2021 from https://issuu.com/landsuedtirol-provinciabolzano/docs/535096_agrar-forstbericht_2019_web?fr=sZjkzMzczMDk3Nw.

[15] de Meyer J. (2014). Apple Producing Family Farms in South Tyrol. An Agriculture Innovation Case Study. FAO, Rome, Italy, here p. 1.

[16] Bundesministerium für Ernährung und Landwirtschaft (ed.) (2020). Bienenfreundliche Pflanzen. Das Pflanzenlexikon für Balkon und Garten. BMEL, Berlin, Germany, here p. 11.

[17] Gary N.E., Witherell P.C. (1977). Distribution of Foraging Bees of Three Honey Bee Stocks Located Near Onion and Safflower Fields. Environmental Entomology 6 (6), 785-788, DOI:10.1093/ee/6.6.785.

[18] Geier J. (2021). Pollen provenience in honey bee (Apis mellifera subsp. carnica) pollen pellets from the Merano Region - South Tyrol. (Master thesis). Leopold-Franzens-Universität Innsbruck, Innsbruck, Austria.

[19] Granata E., Mogilnaia E., Alessandrini C. et al. (2025). Management factors strongly affect flower-visiting insects in intensive apple orchards. Agriculture, Ecosystems & Environment 380:109382, DOI:10.1016/j.agee.2024.109382.

[20] Russo L., Danforth B. (2017). Pollen preferences among the bee species visiting apple (Malus pumila) in New York. Apidologie 48 (6), 806-820, DOI:10.1007/s13592-017-0525-3.

[21] Favaro R., Bauer L.M., Rossi M. et al. (2019). Botanical Origin of Pesticide Residues in Pollen Loads Collected by Honeybees During and After Apple Bloom. Frontiers in Physiology 10, 1069, DOI:10.3389/fphys.2019.01069.

[22] Mair B., Wolf M. (2022). Monitoring of the development of honeybee colonies placed near apple orchards in South Tyrol during spring. Journal of Cultivated Plants 74 (07-08), 166-175, DOI:10.5073/JfK.2022.07-08.02.

[23] Mair B., Wolf M. (2023). Studies on the botanical origin and the residues of pesticides in corbicular pollen loads and bee bread of bee colonies in the proximity of apple orchards in South Tyrol. Journal of

Cultivated Plants 75 (09-10), 225-234, DOI:10.5073/JfK.2023.09-10.01.

[24] Ungerer V. (2017). Vegetationskundliche Erhebungen des blühenden Unterwuchses in Südtirols Apfelanlagen und dessen Bedeutung für die Honigbiene. (Bachelor Thesis). Hochschule Weihenstephan-Triesdorf, Freising, Germany.

[25] Braun-Blanquet J. (1964). Pflanzensoziologie. Grundzüge der Vegetationskunde. Springer, Vienna, Austria, DOI:10.1007/978-3-7091-8110-2.

[26] Faegri K., Iversen J., Kaland P.E. et al. (19894). Textbook of pollen analysis. Wiley & Sons, Chichester, UK.

[27] Beug H.-J. (2004). Leitfaden der Pollenbestimmung für Mitteleuropa und angrenzende Gebiete. Pfeil, Munich, Germany.

[28] Bucher E., Kofler V., Vorwohl G. et al. (2004). Das Pollenbild der Südtiroler Honige. (Gredleriana, Supplement). Naturmuseum Südtirol, Bolzano/Bozen, Italy.

[29] Martin A.C., Harvey W.J. (2017). The Global Pollen Project. A new tool for pollen identification and the dissemination of physical reference collections. Methods in Ecology and Evolution 8 (7), 892-897, DOI:10.1111/2041-210X.12752.

[30] AutPal - Society for the Promotion of Palynological Research in Austria (ed.) (2021). PalDat. A palynological database. Retrieved November 22, 2021, from https://www.paldat.org/.

[31] Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria - CREA (ed.) (2021). PollenAtlas. Retrieved November 17, 2021, from https://pollenatlas.net.

[32] Stebler T. (2021). Pollen-Wiki. Der digitale Pollenatlas. Pollen- und Sporensammlung. Retrieved November 5, 2021, from https://pollen.tstebler.ch/MediaWiki/index.php?title=Pollenatlas.

[33] Weber R.W. (1998). Pollen identification. Annals of Allergy, Asthma & Immunology 80 (2): 141-148.

[34] Grimm E.C. (2019). Tilia, Version 2.6.1.. Retrieved November 3, 2021, from https://www.neotomadb.org/apps/tilia.

[35] R Studio Team (2020). R Studio, 1.3.1093. Retrieved Oktober 8, 2021 from https://www.rstudio.com/.

[36] Merwin I.A. (2003). Orchard-floor management systems. In: Ferree D.C. and Warrington I.J. (eds.). Apples. Botany, production and uses, CABI Publishing, Wallingford, UK, pp. 303-318.

[37] Glenn D.M., Welker W.V. (1989). Orchard soil management systems influence rainfall infiltration. Journal of the American Society for Horticultural Science 114 (1), 10-14, DOI:10.21273/JASHS.114.1.10.

[38] Merwin I.A., Stiles W.C. (1994). Orchard Groundcover Management Impacts on Apple Tree Growth and Yield, and Nutrient Availability and Uptake. Journal of the American Society for Horticultural Science 119 (2), 209-215, DOI:10.21273/JASHS.119.2.209.

[39] Welker W.V., Glenn D.M. (1989). Sod proximity influences the growth and yield of young peach trees. Journal of the American Society for Horticultural Science 114 (6), 856-859.

[40] Welker W.V., Glenn D.M. (1988). Growth responses of young peach trees and changes in soil characteristics with sod and conventional planting systems. Journal of the American Society for Horticultural Science 113 (5), 652-656.

[41] Stevenson D.S., Neilsen G.H. (1990). Nitrogen additions and losses to drainage in orchard-type irrigated lysimeters. Canadian Journal of Soil Science 70 (1), 11-19, DOI:10.4141/cjss90-002.

[42] Johnson D.S., Samuelson T.J. (1990). Short-term effects of changes in soil management and nitrogen fertilizer application on 'Bramley's Seedling' apple trees. Journal of Horticultural Science 65 (5), 495-502, DOI:10.1080/00221589.1990.11516085.

[43] Merwin I.A., Stiles W.C., van Es H.M. (1994). Orchard Groundcover Management Impacts on Soil Physical Properties. Journal of the American Society for Horticultural Science 119 (2), 216-222, DOI:10.21273/JASHS.119.2.216.

[44] Bugg R.L., Waddington C. (1994). Using cover crops to manage arthropod pests of orchards. A review. Agriculture, Ecosystems & Environment 50 (1), 11-28, DOI:10.1016/0167-8809(94)90121-X.

[45] Autonome Provinz Bozen - Südtirol (ed.) (2016). Bestimmungen auf dem Gebiet des Pflanzenschutzes - Landesgesetz vom 15. April 2016, Nr .8. Retrieved October 28, 2021, from https://lexbrowser.provinz.bz.it/doc/de/204538/landesgesetz_vom_15_april_2016_nr_8.aspx?view=1.

[46] Free J.B. (1968). Dandelion as a Competitor to Fruit Trees for Bee Visits. Journal of Applied Ecology 5 (1), 169-178, DOI:10.2307/2401281.

[47] Betts A.D. (1920). The Constancy of the Pollen-collecting Bee. Bee World 2 (1-4), 10-11, DOI:10.1080/0005772X.1920.11094543.

[48] Di Pasquale G., Salignon M., Le Conte Y. et al. (2013). Influence of pollen nutrition on honey bee health: do pollen quality and diversity matter? PLoS ONE 8 (8), e72016, DOI:10.1371/journal.pone.0072016.

[49] Cook S.M., Awmack C.S., Murray D.A. et al. (2003). Are honey bees' foraging preferences affected by pollen amino acid composition? Ecological Entomology 28 (5), 622-627, DOI:10.1046/j.1365-2311.2003.00548.x.

[50] Hendriksma H.P., Shafir S. (2016). Honey bee foragers balance colony nutritional deficiencies. Behavioral Ecology and Sociobiology 70 (4), 509-517, DOI:10.1007/s00265-016-2067-5.

[51] Vaudo A.D., Tooker J.F., Patch H.M. et al. (2020). Pollen Protein. Lipid Macronutrient Ratios May Guide Broad Patterns of Bee Species Floral Preferences. Insects 11 (2), 132, DOI:10.3390/insects11020132.

[52] Lunau K. (2000). The ecology and evolution of visual pollen signals. Plant Systematics and Evolution 222 (1), 89-111, DOI:10.1007/BF00984097.

[53] Pernal S., Currie R. (2001). The influence of pollen quality on foraging behavior in honeybees (Apis mellifera L.). Behavioral Ecology and Sociobiology 51 (1), 53-68, DOI:10.1007/s002650100412.

[54] Levin M.D., Bohart G.E. (1955). Selection of pollens by honey bees. American Bee Journal 95 (10), 392-393. Retrieved November 22, 2021, from https://digitalcommons.usu.edu/cgi/viewcontent.cgi?article=1054&context=piru_pubs.

[55] Brodschneider R., Crailsheim K. (2010). Nutrition and health in honey bees. Apidologie 41 (3), 278-294, DOI:10.1051/apido/2010012.

[56] Donkersley P. (2019). Trees for bees. Agriculture, Ecosystems & Environment 270-271, 79-83, DOI:10.1016/j.agee.2018.10.024.

[57] Severson D.W., Parry J.E. (1981). A Chronology of Pollen Collection by Honeybees. Journal of Apicultural Research 20 (2), 97-103, DOI:10.1080/00218839.1981.11100480.

[58] O'Neal R.J., Waller G.D. (1984). On the pollen harvest by the honey bee (Apis mellifera L.) near Tucson, Arizona (1976-1981). Desert Plants 6 (2), 81-109. Retrieved November 10, 2021, from https://repository.arizona.edu/bitstream/handle/10150/552253/dp_06_02-081-094.pdf?sequence=1.

[59] Pearson W.D., Braiden V. (1990). Seasonal Pollen Collection by Honeybees from Grass/Shrub Highlands in Canterbury, New Zealand. Journal of Apicultural Research 29 (4), 206-213, DOI:10.1080/00218839.1990.11101221.

[60] Proctor M., Yeo P., Lack A. (1996). The Natural History of Pollination. (Collins New Naturalist Library : 83). HarperCollins Publishers, London, UK, here pp. 265-266.

[61] Roulston T.H., Cane J.H., Buchmann S.L. (2000). What Governs Protein Content of Pollen. Pollinator Preferences, Pollen-Pistil Interactions, or Phylogeny? Ecological Monographs 70 (4), 617-643, DOI:10.1890/0012-9615(2000)070[0617:WGPCOP]2.0.CO;2.

[62] Keller I., Fluri P., Imdorf A. (2005a). Pollen nutrition and colony development in honey bees. Part 1. Bee World 86 (1), 3-10, DOI:10.1080/0005772X.2005.11099641.

[63] van der Moezel P.G., Delfs J.C., Pate J.S. et al. (1987). Pollen Selection by Honeybees in Shrublands of the Northern Sandplains of Western Australia. Journal of Apicultural Research 26 (4), 224-232, DOI:10.1080/00218839.1987.11100764.

[64] Olsen L.G., Hoopingarner R., Martin E.C. (1979). Pollen Preferences of Honeybees Sited on four Cultivated Crops. Journal of Apicultural Research 18 (3), 196-200, DOI:10.1080/00218839.1979.11099967.

[65] Shawer M.B. (1987). Major Pollen Sources in Kafr El-Sheikh, Egypt, and the Effect of Pollen Supply on Brood Area and Honey Yield. Journal of Apicultural Research 26 (1), 43-46, DOI:10.1080/00218839.1987.11100733.

[66] Fischer M.A., Oswald K., Adler W. (20083). Exkursionsflora für Österreich, Liechtenstein und Südtirol. Bestimmungsbuch für alle in der Republik Österreich, im Fürstentum Liechtenstein und in der Autonomen Provinz Bozen/Südtirol (Italien) wildwachsenden sowie die wichtigsten kultivierten Gefäßpflanzen (Farnpflanzen und Samenpflanzen) mit Angaben über ihre Ökologie und Verbreitung. Biologiezentrum der Oberösterreichischen Landesmuseen, Linz, Austria.

[67] Cho L.-H., Yoon J., An G. (2017). The control of flowering time by environmental factors. The Plant Journal 90 (4), 708-719, DOI:10.1111/tpj.13461.

[68] Keller I., Fluri P., Imdorf A. (2005b). Pollen nutrition and colony development in honey bees. Part 2. Bee World 86 (2), 27-34, DOI:10.1080/0005772X.2005.11099650.

[69] Lucek K., Galli A., Gurten S. et al. (2019). Metabarcoding of honey to assess differences in plant pollinator interactions between urban and non-urban sites. Apidologie 50, 317-329. DOI:10.1007/s13592-019-00646-3.