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Impact of Agroforestry on Major Pests of Oilseed Rape and Their Bio-Control

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ABSTRACT

Agroforestry systems are traditional land use systems that were and are still used in Europe. They can be defined as those land use systems which involve two main components – trees/shrubs and an agricultural crop (which could also be pasture). This research took place at an alley cropping agroforestry system that combined winter oilseed rape with poplar tree rows near Dornburg, Germany and at an open cropland oilseed rape system used as reference. Winter oilseed rape crop is one of the main crops in Germany which is cultivated for many different purposes, and improvements of pest control of oilseed rape is important for future cultivations. Four observation locations at different distances from the tree rows were established in agroforestry system (tree row, 1 m, 7 m and 24 m distance) to compare to the reference location (open cropland). After different methods to assign the effect of agroforestry systems on different pests, beneficial organisms, and infestation levels, results show that there is a not beneficial side of agroforestry systems in general, however, it has some positive effects at some levels especially being an additional shelter for the parasitoid species gave them an option to migrate. As conclusion agroforestry systems can be facilitated to combine with oilseed rape fields in future to use it in integrated pest management.

1 INTRODUCTION

1.1 Agroforestry Systems

Agroforestry can be defined as a sustainable way of land management which integrates both agricultural and forestry practices on the same land management base. Agroforestry system practices have been defined by different authors (Nair, 1993) as practices which involve “the deliberate integration of trees with agricultural crops and/or livestock either simultaneously or sequentially on the same unit of land”. The International Centre for Research in Agroforestry (ICRAF) and the World agroforestry Centre (WAC) define the term agroforestry as “a dynamic, ecologically based natural resources management system that, through the integration of trees in farmland and rangeland, diversifies and sustains production for increased social, economic and environmental benefits for land users at all levels” and “A land-use system in which woody perennials (trees, shrubs, palms, bamboos) are deliberately used on the same land management unit as agricultural crops (woody or not), animals or both, either in some form of spatial arrangement or temporal sequence. Finally, the AFTA (Association for Temperate Agroforestry) (1997) in USA also defines Agroforestry systems as “an intensive land management system that optimizes the benefits from the biological interactions created when trees and/or shrubs are deliberately combined with crops and/or livestock”.

Following the definitions given by AFTA (1997) and Alavalapati and Nair (2001) there are currently five basic types of agroforestry practices in temperate areas: windbreaks, alley cropping, silvopasture, riparian buffers and forest farming. This classification is mostly based on the main practices developed in America. Although the situation is different in Europe, this classification is still valid, but it should be slightly changed and increased in scope: silvoarable agroforestry, forest farming, riparian buffer strips, silvopasture, improved fallow and multipurpose trees (Mosquera-Losada et al., 2009). As conclusion, Agroforestry has been proposed as a sustainable agricultural system over conventional agriculture and forestry, conserving biodiversity and enhancing ecosystem service provision while not compromising productivity and agroforestry can enhance biodiversity and ecosystem service provision relative to conventional agriculture and forestry in Europe and could be a strategically beneficial land use in rural planning if its inherent complexity is considered in policy measures (Torralba et al., 2016).

Seminatural habitats, such as forests, hedgerows, field margins, fallows, and meadows support a large number of pest and natural enemy species, as they provide a more stable environment than annual crops (Rusch et al., 2010). Different studies have elucidated the range of supportive effects of semi-natural habitats on both pest and natural enemy species. (Rusch et al., 2010) summarise the effects of seminatural habitats as those are being alternative host environment, providing food sources such as pollen and nectar, providing shelter and alternative overwintering areas.

1.2 Oilseed Rape *(Brassica napus* L.*)*

1.2.1 Oilseed Rape History and Dispersion

Domestication of members of the *Brassica* family to oil producing crops occurred during different periods of history and in many locations throughout the world. In the old civilizations of Asia and the Mediterranean, rape oil was used for lighting purposes. Rape crops were cultivated in India 3000 years ago and were introduced to China and Japan about 500 to 200 BC (Krzymañski, 1998). In Europe, seeds and pods have been recovered from excavations of Bronze Age sites.

Today oilseed rape is cultivated and processed for many different purposes: oil for human nutrition, as a renewable raw material for the chemical industry, as a source of regenerative energy, as a source of high energy and protein content for animal nutrition in the form of rape cake and meal, as a catch crop for green manuring and as a forage crop.

1.2.2 Oilseed Rape Botany

Brassica napus is self-compatible although both wind and insect pollination can occur. Large amounts of pollen are released, and this is likely to contribute to both cross- and self-pollination (Harding & Harris, 1997).

Brassica napus grows to 200 cm in height with hairless, fleshy, pinnatifid and glaucous lower leaves which are stalked whereas the upper leaves have no petioles.

Rapeseed flowers are bright yellow and about 17 mm across. They are radial and consist of four petals in a typical cross-form, alternating with four sepals. They have indeterminate racemose flowering starting at the lowest bud and growing upward in the following days. The flowers have two lateral stamens with short filaments, and four median stamens with longer filaments whose anthers split away from the flower's centre upon flowering.

The rapeseed pods are green and elongated siliquae during development that eventually ripen to brown. They grow on pedicels 1 to 3 cm long and can range from 5 to 10 cm in length. Each pod has two compartments separated by an inner central wall within which a row of seeds develops. The seeds are round and have a diameter of 1.5 to 3 mm. They have a reticulate surface texture and are black and hard at maturity.

1.2.3 The Use of Oilseed Rape

Oilseed rape is a crop with very diverse uses and today rape oil is coveted as never before. The reason for this lies in the widespread utilization of rape oil in a variety of sectors including human nutrition, as an alternative regenerative fuel, as an environmentally friendly lubricating oil used for very different purposes or as raw material in the chemical industry. Furthermore, the residues from oil production are used as a valuable animal food providing a high energy and a high protein content (Orlovius & Kirkby, 2000).

1.3 Rape Stem Weevil *(Ceutorhynychus napi* Gyll.*)*

The Rape Stem weevil *Ceutornychus napi* is 3.2–4 mm long with a long rostrum. It is greyish with three rows of whitish hairs between the longitudinal furrows of the elytra (Alford et al., 2003). Adults of *C. napi* overwinter in cocoons in the soil around plants, where they complete their development (Dechert & Ulber, 2004); Juran et al., 2011). They emerge from the following early spring and migrate to oilseed rape fields. On winter oilseed rape crops, weevils can appear very early, in February and March. Females usually start laying their eggs on growing stems in March/April (Dechert & Ulber, 2004), exceptionally in February too (Central Institute for Supervising and Testing in Agriculture, ÚKZÚZ 2021). The process of egg deposition later results in the twisting and splitting of tissue, followed by distortion and considerable disruption to growth. Eggs hatch one to two weeks after deposition and the larvae then feed within the pith for three to five weeks eventually dropping to the ground and pupating in the soil. The adult stage is reached about one month later, but, unlike those of related species, they remain within their earthen chambers and do not emerge until the following spring (Alford et al., 2003).

1.4 Cabbage Stem Weevil *(Ceutorhynychus pallidactylus* Marsh.*)*

The Cabbage Stem Weevil *Ceutorhynchus pallidactylus* is 2.5–3.5 mm long. It is greyish-brown with brown-red legs and has scattered white scales over its body and fine hairs on the elytra, those concentrated centrally on the base of the elytra forming a white rectangular spot (Williams, 2010a). Adults of *C. pallidactylus* emerge from the soil gradually as the oilseed rape ripens and overwinter in shallow layers of soil or under leaves and other plant remains at places which surround fields and pastures (field banks, margins of field tracks, wood margins). Oviposition begins later than at *C. napi* and most females usually lay their eggs during April and often in May and June (Buchs, 1998); Dechert & Ulber, 2004); Juran et al., 2020). The reason for the delayed start of oviposition of *C. pallidactylus* (in comparison to *C. napi*) is the difference in progress of the male and female's migrations to crops from hibernating sites. The proportion of females was found to increase gradually in yellow water traps during the monitoring of flight activity in winter oilseed rape, so the ratio of males to females present in crops equals out substantially later in *C. pallidactylus* populations than in the case of *C. napi* (Seidenglanz et al., 2022).

1.5 Cabbage Stem Flea Beetle *(Psylliodes chrysocephala* L.*)*

The Cabbage Stem Flea Beetle *Psylliodes chrysocephala* is 4–5 mm long, usually black with a blue-green metallic sheen (Williams, 2010a), a brown variant also occurs (Bonnemaison & Jourdheuil, 1954). It has large hind femurs enabling it to jump. The antennae have 10 segments (Williams, 2010a). Adult Cabbage Stem Flea Beetle are present throughout most of the European continent. They are oval in shape, 3.2–4.6 mm long (Bonnemaison & Jourdheuil, 1954), and have 10 antennal segments and thickened hind femurs to enable them to jump to avoid predators (Furth, 1988; Ruan et al., 2020). They are usually black with a blue-green metallic sheen although a brown variant also occurs (Bonnemaison & Jourdheuil, 1954). The beetles feed on the cotyledons and young leaves of plants and after a period of 2 weeks, they start to mate and oviposition begins (Alford et al., 2003; Bonnemaison & Jourdheuil, 1954; Såringer, 1984). Oviposition usually peaks in autumn, when temperatures are between 2 and 16°C, but continues until early spring in mild conditions (Bonnemaison, 1965; Bonnemaison & Jourdheuil, 1954; Mathiasen, Sørensen, et al., 2015; Meuche, 1940; Såringer, 1984). Eggs are oval, orange, 0.6 mm long and 0.4 mm wide (Bonnemaison & Jourdheuil, 1954) and are laid in batches in the soil near the host plant (Såringer, 1984; Vig, 2003). Eggs hatch from September onwards (Alford, 1979; Johnen et al., 2010) but larvae are sensitive to cold winter frosts which could limit their distribution in the furthest north areas of Europe (Mathiasen, Bligaard, et al., 2015; Mathiasen, Sørensen, et al., 2015). There are three larval instars (Bonnemaison, 1965). From late February to June, third instar larvae tunnel out of the plant, drop to the ground, and create a small cavity a few centimeters under the soil surface to pupate (Williams & Carden, 1961). Pupation lasts 8–12 weeks depending on temperature; new generation adults start to emerge in May within the OSR crop where they stay to feed on the stems and the exterior of pods (Såringer, 1984; Williams & Carden, 1961). In late summer, adults undergo a period of aestivation (prospective diapause; (Såringer, 1984)where they stop feeding and either remain in the crop (Sivcev et al., 2016; Vig, 2003) or migrate to sheltered areas such as hedgerows and woodlands (Bonnemaison & Jourdheuil, 1954). By the end of August, when temperatures have cooled, the beetles become active again and migrate into newly sown crops, reaching the population peak by early September, although this varies with weather conditions (Såringer, 1984; Vig, 2003).

1.6 Pollen Beetle *(Brassicogethes aeneus* F.*)*

The Pollen Beetle, *Brassicogethes aeneus* (formerly *Meligethes aeneus*) Fabricius (Coleoptera: *Nitidulidae*) is a univoltine species and a pest of oilseed rape (OSR) (*Brassica napus* L., Brassicaceae) in Europe (Skellern & Cook, 2017). The Pollen Beetle *Brassicogethes aeneus* F. is the dominant species of pollen beetle found on rape crops throughout Europe. It is a small black beetle (1.9 – 2.5 mm long and 1.3–1.5 mm wide) with foreshortened elytra. The antennae have 11 segments with a compact 3-segemented club (Williams, 2010a). Larvae and adults of this insect are florivorous and feed on pollen. Adult pollen beetles emerge in the spring and are generalist feeders eating pollen from several plant families, but they are specialized on brassicaceous plants for oviposition and larval development (Kirk-Spriggs 1996; Ouvrard et al., 2016). Two larval instars have been described. The first instar usually develops in closed floral buds while the second instar feeds in open flowers and frequently moves between younger flowers (Free & Williams, 1978).During their development, pollen beetle larvae eat OSR pollen which is rich in proteins, starch, and free amino acids (Evans et al., 1991; Cook et al., 2004; Hervé et al., 2014). Once mature, larvae drop from the plant to pupate in the soil and adult emergence occurs during the summer. After emergence, adults feed on pollen in flowers before migrating to forests where they overwinter under the litter layer (Williams et al., 2010).

1.7 Parasitoid Wasps *(Tersilochus spp.)*

Tersilochines occur in all terrestrial biotopes in Europe from steppes and wet forests to alpine meadows and tundra, but usually are more abundant in humid forests. Most of species are rather small and have a body length 3.0-7.0 mm, but some species may exceed 10 mm. Species of this subfamily may easily be recognized by their characteristic forewing venation lack of areolet, thickened intercubitus, first and second abscissae of radius angled about 90°, and a large pterostigma and with maxillary and labial palpi 4 and 3 segmented, respectively (Çoruh & Khalaim, 2013)

Almost all *Tersilochinae* are larval koinobiont endoparasitoids of various Coleopteran species. Some species are used in biological control of pests. The two parasitoids *T. microgaster* and *T. obscurator* are recognized as key parasitoids of *P. chrysocephala* and *C. pallidactylus*, respectively, in mainland Europe (Ulber, 2003; Ulber & Williams, 2003). Like their hosts, both are univoltine. Adults migrate to the crop in spring and females oviposit into their host larvae within leaf petioles or stems. Although the parasitoid egg hatches within the host larva while it is in the plant, further development of the parasitoid larva is delayed until its host is mature and has left the plant to pupate in the soil. The larva then develops to adulthood over the next few weeks. The adult wasp remains inside the cocoon in diapause and emerges from the soil of previous years' oilseed rape fields the following spring (Ferguson et al., 2004).

In this research *T. fulvipes* and *T. heterocerus'* parasitism has not been evaluated on Rape Stem Weevil larvae and Pollen Beetle larvae because the presence of Rape Stem Weevil was at lower levels and the parasitism level of collected Pollen Beetle larvae was too low to evaluate to get a significant result. However, *T. microgaster'* parasitism on Cabbage Stem Flea Beetle larvae collected from leaf samples and *T. obstractor'* parasitism on Cabbage Stem Weevil larvae collected from both leaf and stem samples have been evaluated.

1.8 Aim of the Research

This study aimed to determine the effect of agroforestry system on major pests of oilseed rape and their bio-control agents in different life stages. Rape Stem Weevil (*Ceutorhynychus napi* Gyll.), Cabbage Stem Weevil (*Ceutorhynychus pallidactylus* Marsh.), Cabbage Stem Flea Beetle *(Psylliodes chrysocephala* L.) and Pollen Beetle (*Brassicogethes aeneus* F.) are focused on this project as major pest species and *Tersiloch* species focused as main specialist parasitoid species.

2 MATERIALS & METHODS

2.1 Study Site and Layout

Various methods and materials were used in this research to obtain a deeper knowledge of the effect of agroforestry systems on pest infestation and parasitoid species activity in a field with oilseed rape between two poplar tree lines in Dornburg, Germany and to compare the effect of agroforestry to a conventional oilseed rape field located in Dornburg near the experimental agroforestry field. The agroforestry systems had north-south orientated 12 m-wide tree rows of poplar clones (clone Max 1 *Populus nigra* × *Populus maximowiczii*). The aboveground biomass of the trees was harvested the last time in spring 2015 after plantation in 2007. Oilseed rape was planted at the end of August 2022 in the field between forest trees and in the conventional field.

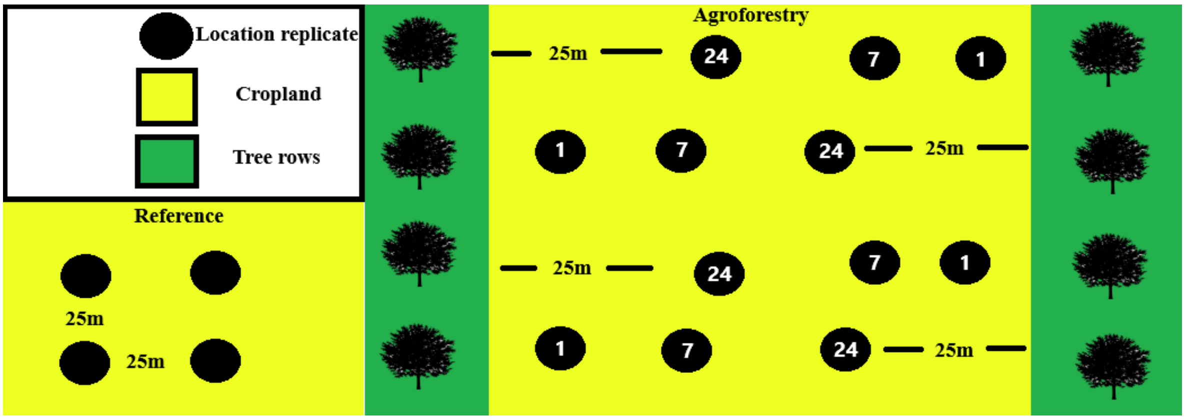
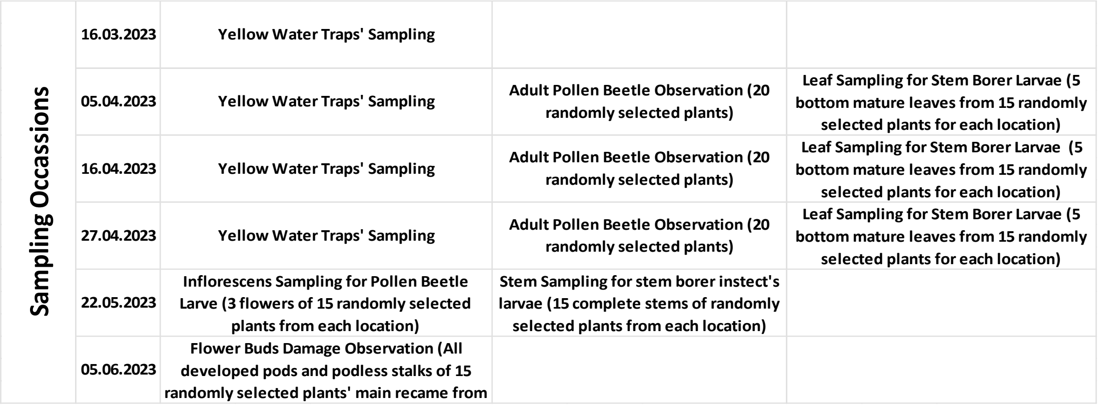


Fig. 1 Demonstration of the locations of sampling points (made by the author)

6 different repetitions of field trips have been done for sampling and observing to evaluate the different phases of this research as shown in Table 1.



Sampling Occassions	16.03.2023	Yellow Water Traps' Sampling		
	05.04.2023	Yellow Water Traps' Sampling	Adult Pollen Beetle Observation (20 randomly selected plants)	Leaf Sampling for Stem Borer Larvae (5 bottom mature leaves from 15 randomly selected plants for each location)
	16.04.2023	Yellow Water Traps' Sampling	Adult Pollen Beetle Observation (20 randomly selected plants)	Leaf Sampling for Stem Borer Larvae (5 bottom mature leaves from 15 randomly selected plants for each location)
	27.04.2023	Yellow Water Traps' Sampling	Adult Pollen Beetle Observation (20 randomly selected plants)	Leaf Sampling for Stem Borer Larvae (5 bottom mature leaves from 15 randomly selected plants for each location)
	22.05.2023	Inflorescens Sampling for Pollen Beetle Larve (3 flowers of 15 randomly selected plants from each location)	Stem Sampling for stem borer instect's larvae (15 complete stems of randomly selected plants from each location)	
	05.06.2023	Flower Buds Damage Observation (All developed pods and podless stalks of 15 randomly selected plants' main recame from		

Table. 1 Sampling Occasions

2.2 Yellow Water Trap Monitoring

To monitor the immigration of adult pest insect species and estimate first flight times yellow water traps were located in both agroforestry system field and conventional field at each distance replicate. Each yellow water trap position (Fig. 1) indicates a different sampling point used for the further assessments additionally.

Fig. 2 Yellow Water Traps

Four repetitions of yellow water trap evaluation have been done on March 16th, April 5th ,16th and 27th 2023for this monitoring phase.

Identification of the caught pests and parasitoids has been done by following their morphological traits as mentioned in section 1.3, 1.4, 1.5, 1.6 and 1.7.

2.3 Adult Pollen Beetle Observation

Following the assessment of immigrating insects by using yellow water traps the numbers of adult Pollen Beetle were counted on the plants.

For this phase of the research, the inflorescences with flower buds on the upper parts of the plants were shaken into wide tray. Dropped adults have been counted immediately before they escape by flying. 20 randomly plants were selected for each location for 3 repetitions on April 5th ,16th and 27th 2023. For one of these sampling repetitions (16th April) unfortunately, the temperature was close to 0°C and too cold for counting adult Pollen Beetle numbers on the plants.

2.4 Leaf Sampling and Extraction

Stem borer insects are one of the main focuses of this research. However, Cabbage Stem Flea Beetle larvae start and complete their feeding, pupating phase before than Cabbage Stem Weevil larvae. To assess the Cabbage Stem Flea Beetle larvae infestation and the beginning time of

Cabbage Stem Weevil larvae feeding activity mature leaves of oilseed rape were sampled with sharp knifes and stored in perforated plastic bags. As they start feeding and boring to the stem from bottom leaves it was important to start sampling them before sampling plant stems. Sampled bottom leaves were placed into extraction boxes (Fig. 3) and the larvae which dropped into the bottom part of the boxes were collected regularly into glass tubes with 70% ethanol for the further larvae dissection phase of the research.

Fig. 3 Extraction Boxes

For this part of the research 3 sampling dates have been done on April 5th ,16th and 27th 2023, with collecting 5 bottom mature leaves of 15 randomly selected plants from each sampling point. For each date, the larvae dropping to the bottom of the boxes were collected every other day periodically and sampled leaves stored in the boxes 8-12 days total.

2.5 Stem Sampling

To assess the stem borer insects the next phase of the research was sampling the complete stems of the plants in order to dissect the stems and collect the stem borer larvae inside of them. Collected stems were dissected with a sharp knife and pest larvae collected into glass tubes with 70% ethanol for the further larval dissection phase of the research.

For this phase of the research 15 complete stems of randomly selected plants were sampled for each sampling location on May 22nd 2023.

Fig. 4 Dissection of Stem

2.6 Inflorescences Sampling

Adult Pollen Beetles lay their eggs inside of the petals of flower buds. Larvae start feeding on pollen within buds after the eggs hatch. To assess the number of pollen beetle larvae in buds and flowers, 3 inflorescences of the main branch of 15 randomly selected plants per sampling location were collected on May 22nd 2023 and stored in perforated plastic bags. In the lab, the pollen beetle larvae were collected from these flowers and stored in 70% ethanol for later dissection.

To determine the level of feeding damage caused by adult pollen beetles to flower buds, all developed pods and all podless stalks were counted on the main raceme of 15 randomly selected main stems per sampling location on June 5th 2023. The percentage of podless stalks was calculated based on the total number of pods plus podless stalks.

2.7 Larval Dissection

At both leaf sampling and stem sampling phase of this research collected larvae of Cabbage Stem Weevil and Cabbage Stem Flea Beetle were classified and stored separately following the descriptions from published literature. The larvae of Cabbage Stem Flea Beetle are up to 8 mm long, creamy-white with three pairs of legs, a black head and a black dorsal plate on the final abdomen segment (Williams, 2010a). On contrast Cabbage stem weevil larvae are white with a creamish head, legless and up to 6 mm long (Williams, 2010a). The key trait to differentiate between the larvae of these two different species was the presence of legs.

Collected and stored larvae of Cabbage Stem Weevil, Cabbage Stem Flea Beetle have been dissected with thin dissection needles and a proper microscope to assess the level of larval parasitism (%) of pest larvae at various sampling positions. Parasitized pest larvae have an egg on larva from *Tersilochus spp*. inside of their body. However, super-parasitized pest larvae have two or more eggs from same *Tersilochus spp*.'s females inside of their body. From the bottom leaf samples 1159 Cabbage Stem Weevil larvae and 257 Cabbage Stem Flea Beetle larvae were caught in extraction boxes, and 672 Cabbage Stem Weevil larvae caught in stems from stem samples to assess the parasitism and super-parasitism level by dissection of these larvae. All the analysis has been done by dissecting the caught larvae. In the analysis, the number of parasitized and super-parasitized larvae has been evaluated, and the percentage of parasitism and the percentage of super-parasitism represent the ratio of parasitized and super-parasitized larvae to all dissected larvae, respectively.

Fig. 5 Dissection Petri

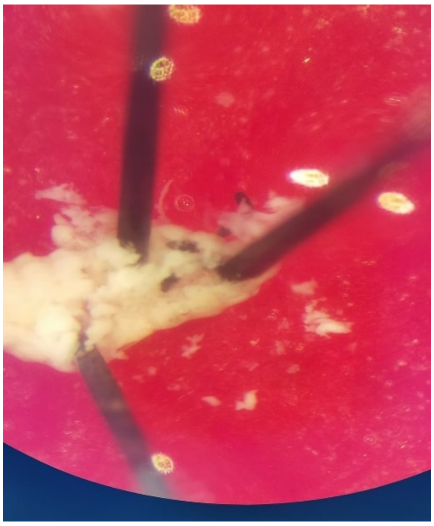


Fig. 6 Dissection of a Parasitized Pest Larva

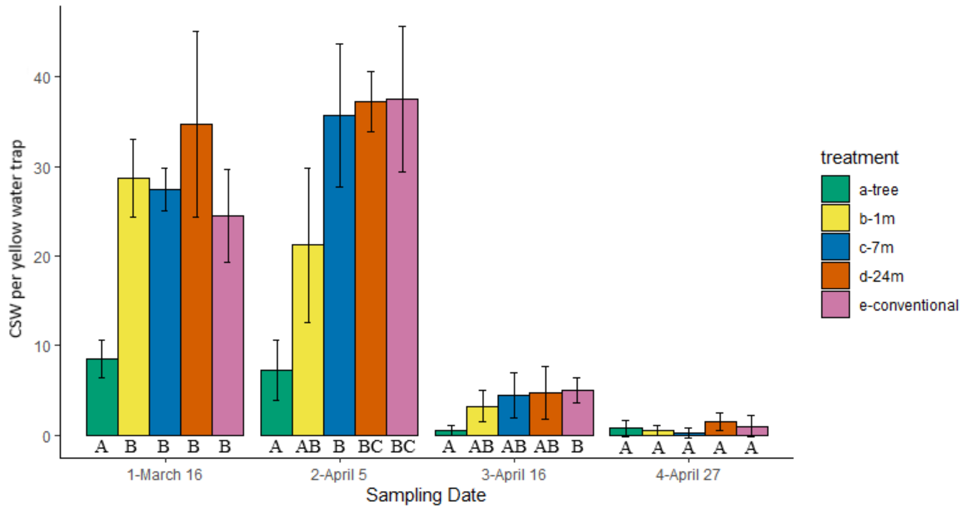
2.8 Statistical Analysis

The primary data from all the phases of the research has been processed in R. For all sets of data, confidence interval tests by p value=0,05 were performed. A 95% confidence interval (CI) of the mean is a range with an upper and lower number calculated from a sample. Because the true population mean is unknown, this range describes possible values that the mean could be. To assess the effect of agroforestry between different distances and between the conventional field one-way ANOVA test applied to data. When significant F value were encountered the Tukey test was applied the data which has significant F value in ANOVA results to assess the significant differences between different treatments.

3 RESULTS

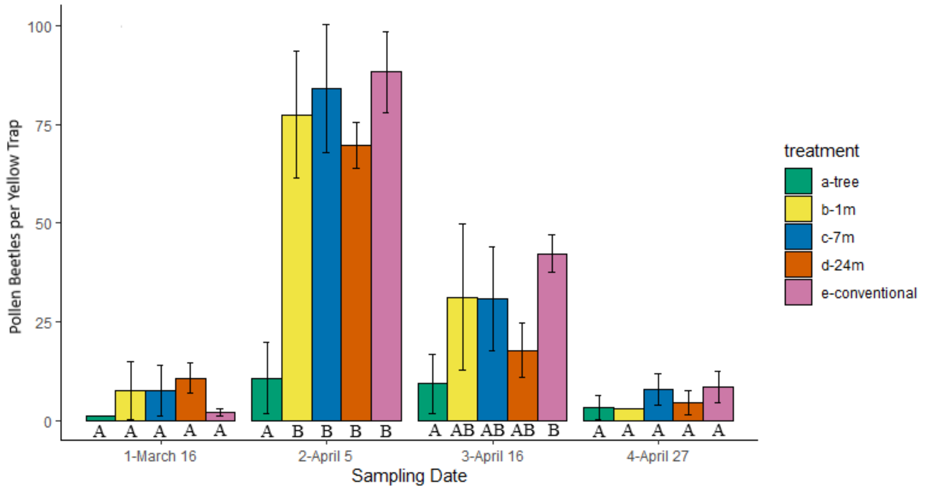
3.1 Yellow Water Trap Samples' Analysis

The study observed that the peak population densities and flight activities of the studied pest insect species increased towards the end of March and the beginning of April (Fig. 8, 9, and 10). Unlike other pest insect species, the Cabbage Stem Weevil was observed to increase its population density before the middle of March and commence its flights earlier than the first observation date of March 16 (Fig. 7). Among the main focused pest species Rape Stem Weevil and Cabbage Stem Flea Beetle are the least common species (Fig. 9 and 10). However, parasitoid species started flying around in Dornburg, Germany later than the pest species and first occured between 5th and 16th April. Analyses of yellow water trap samplings show that there are significant differences between tree rows and other distances/conventional field. Results show that Cabbage Stem Weevils and Pollen Beetles fly around in the agroforestry field (crop rows) and conventional field rather than flying in tree rows when their population density was higher. Therefore, significant differences were found from one-way ANOVA at 16th March, 5th April and 16th April between tree lines and other distances in agroforestry field and also in conventional field for Cabbage Stem Weevil (Fig. 7). Moreover, results also show that significant differences for Pollen Beetles at 5th and 16th April between tree lines and other distances in agroforestry field and also conventional field (Fig. 8). However, there were no significant differences for Rape Stem Weevil and Cabbage Stem Flea Beetle (Fig. 9 and 10). There are also no significant differences for parasitoid species, however, it can clearly be seen from Figure 11 they first started to fly in and around tree lines between 5th and 16th April and they migrate to conventional field 2 weeks later which can be seen from the result of 27th April's counting.



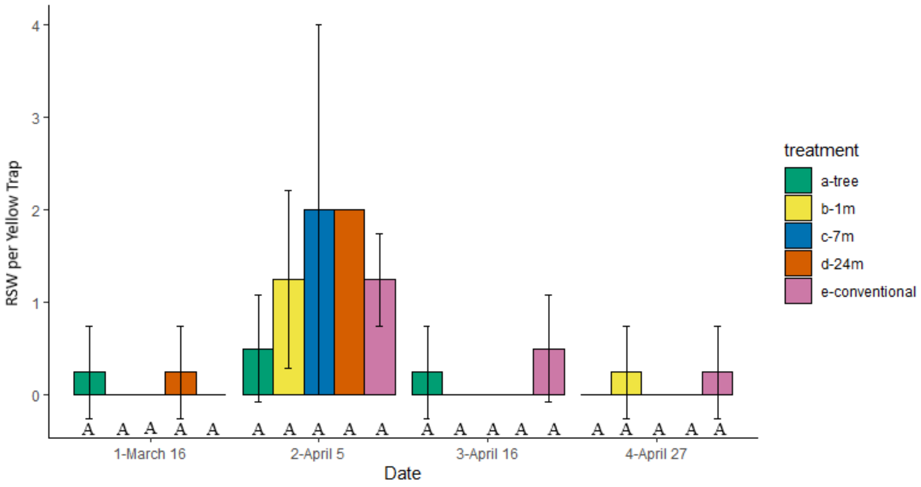
Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05).

Fig. 7 Number of Cabbage Stem Weevils caught in yellow water traps in the agroforestry system (in the tree rows and at 1m, 7m, and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation



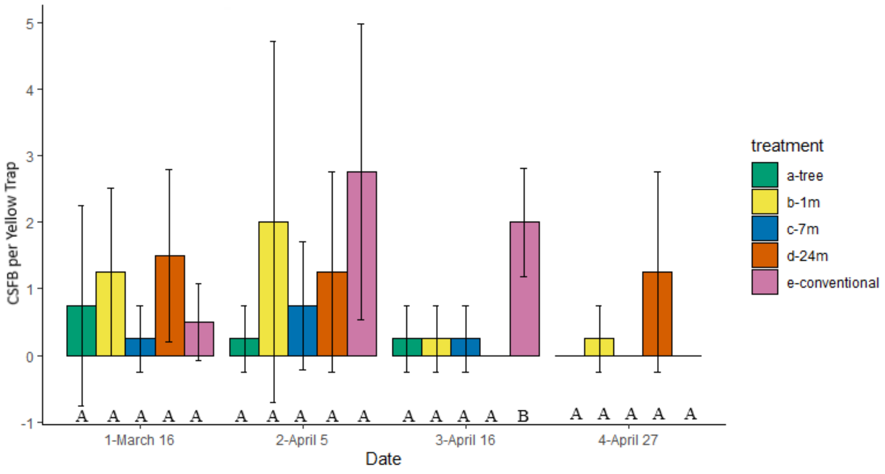
Different uppercase letters of the same font indicate statistically significant differences (p<0.05)

Fig. 8 Number of Pollen Beetles caught in yellow water traps in the agroforestry system (in the tree rows and at 1m, 7m, and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation



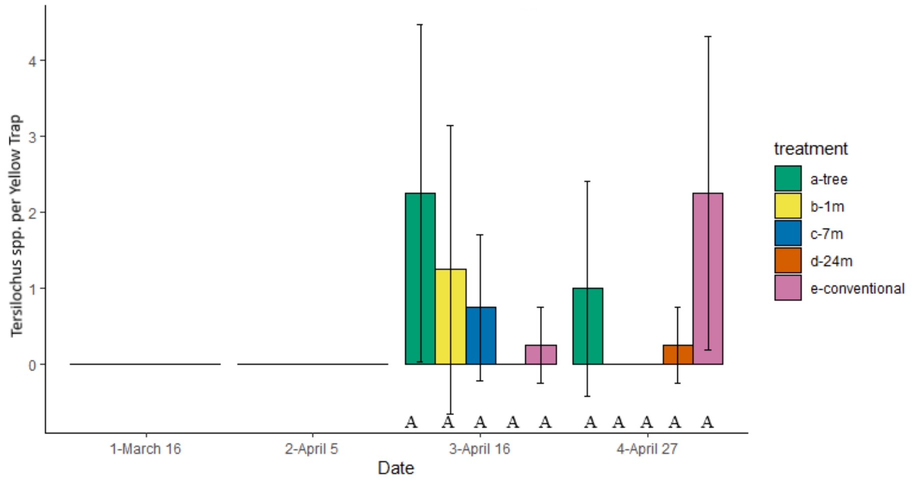
Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 9 Number of Rape Stem Weevils caught in yellow water traps in the agroforestry system (in the tree rows and at 1m, 7m, and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 10 Number of Cabbage Stem Flea Beetle caught in yellow water traps in the agroforestry system (in the tree rows and at 1m, 7m, and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation

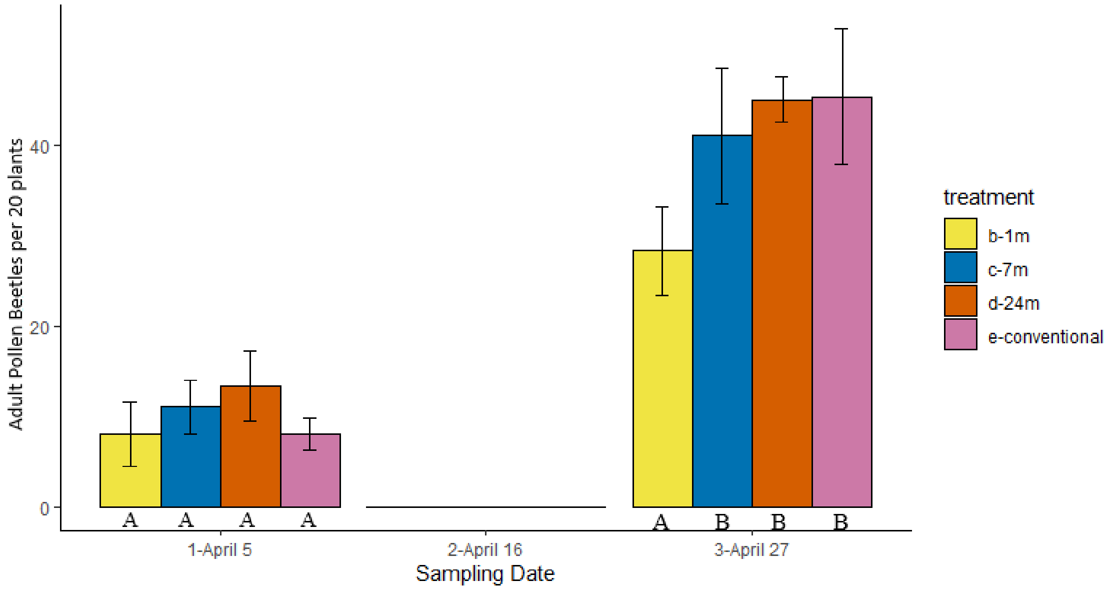


Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

Fig. 11 Number of *Tersilochus spp.* caught in yellow water traps in the agroforestry system (in the tree rows and at 1m, 7m, and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation

3.2 Pollen Beetle Infestation and Plant Damage

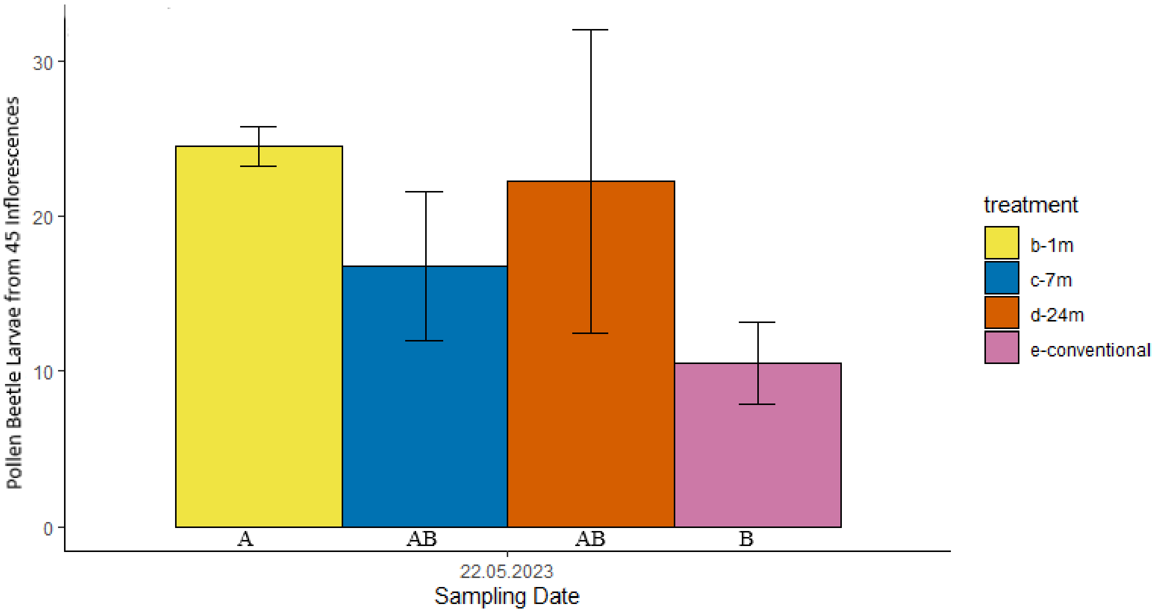
The results of adult Pollen Beetle infestation on the plants in the experimental agroforestry field and conventional oilseed rape field shows that the distribution between different distances to forest tree lines and between conventional locations have not a significant difference at 5th of April, however when population increased between 5th and 27th April the number of Pollen Beetles was significantly lower at 1 meter distance than other treatments (Fig. 12).



Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 12 Number of Pollen Beetles caught in agroforestry system (at 1m, 7m and 24m distance in the oilseed rape plot) and in the conventional system by plant shaking, presented as mean values with 95% confidence intervals calculated based on the standard deviation

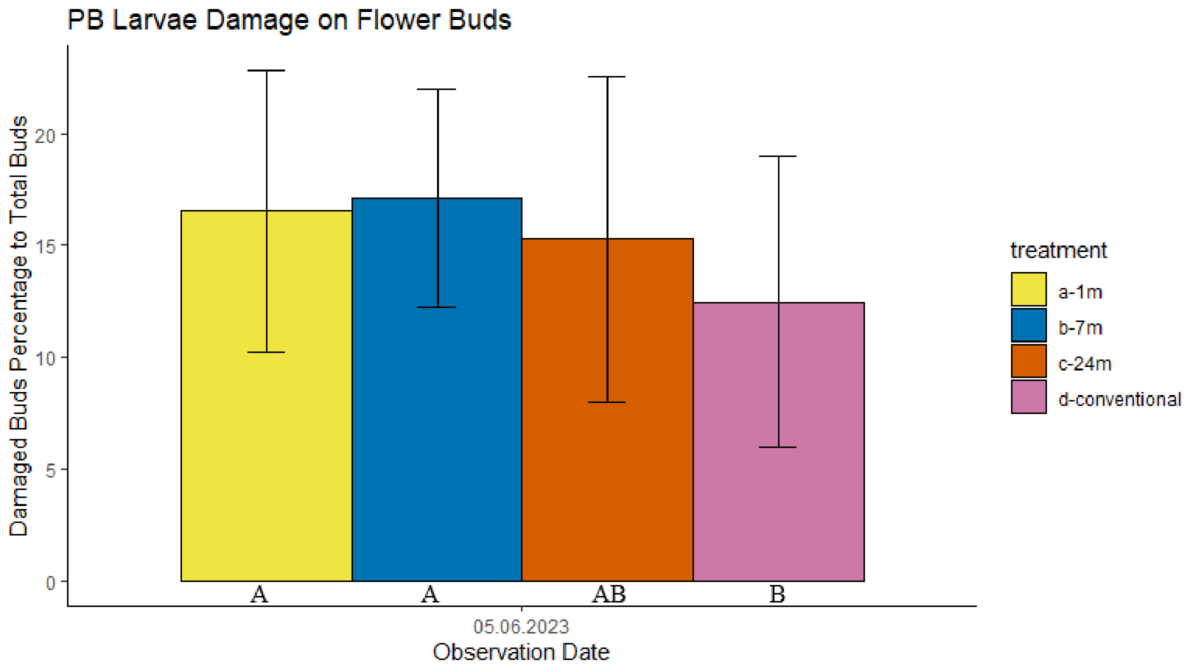
Females of Pollen Beetles lay their eggs into flower buds. When they hatch the larvae remain inside the buds and flowers to feed. For each observation point, flowers from 3 main branches of 15 randomly selected plants were sampled, and the larvae of Pollen Beetles within these flowers were counted. Number of larvae differed between different distances to tree lines and conventional field. As a result of one-way ANOVA test there is a significant difference between the location of 1 meter distance from tree lines and conventional field (Fig. 13).



Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

Fig. 13 Number of Pollen Beetle larvae caught in flower samples in the agroforestry system (at 1m, 7m and 24m distance in the oilseed rape plot) and in the conventional system, presented as mean values with 95% confidence intervals calculated based on the standard deviation

As a result of adult Pollen beetle feeding the oilseed rape plants lose some of their buds. To evaluate the feeding damage of adult Pollen Beetles 15 randomly selected plants' developed flower buds and podless stalks of main branches counted. Analyses show that there a significant difference between different 1m and 7m distances in agroforestry system and between conventional field even though there was no significant difference between 7m and conventional field (Fig. 14).

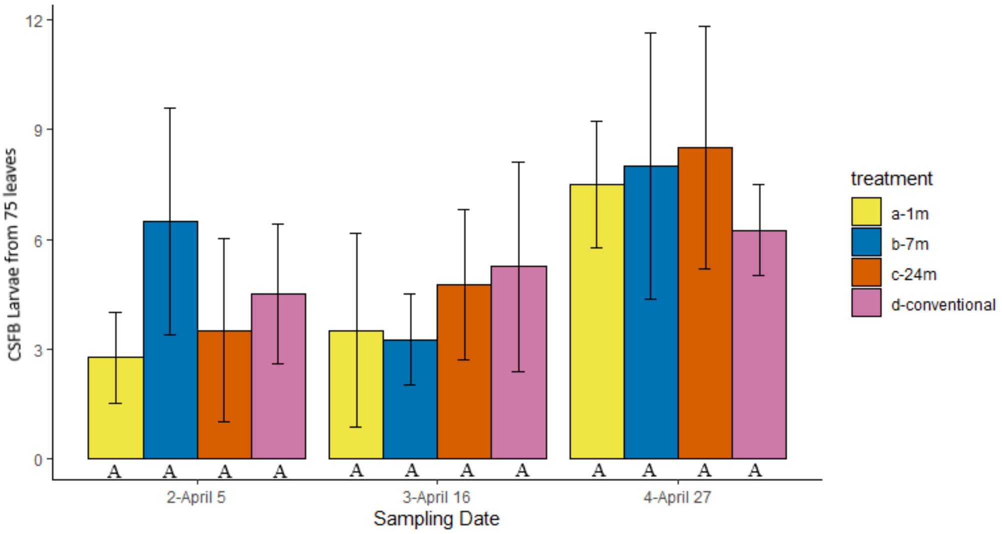


Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 14 Feeding damage of adult Pollen Beetles from 15 randomly selected plants of each location; number of podless stalks related to total number of developed pods + podless stalks, presented as mean values with 95% confidence intervals calculated based on the standard deviation

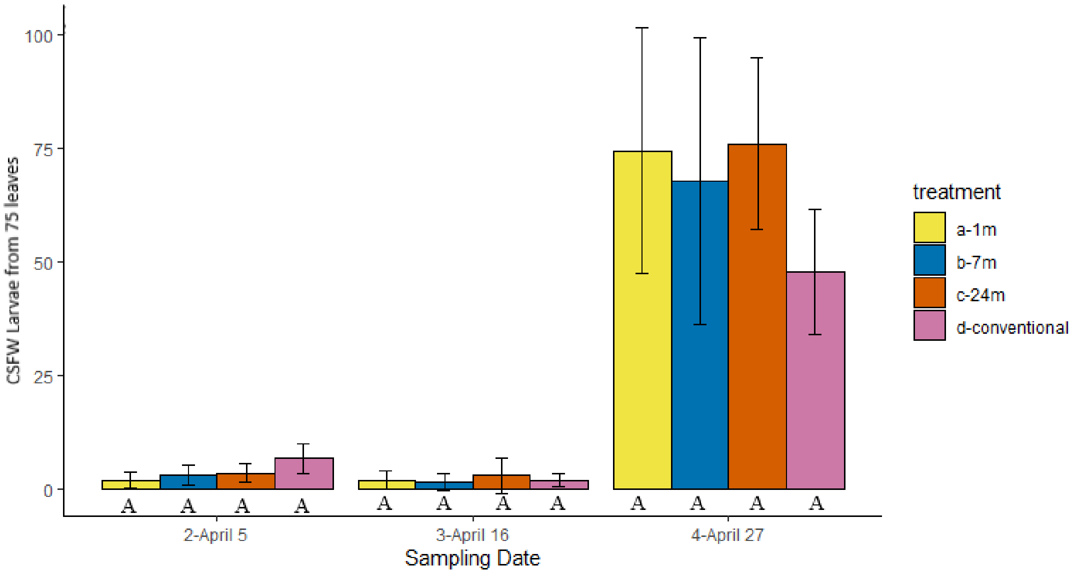
3.3 Plant Infestation by Plant Boring Insects

At this phase of the research Cabbage Stem Weevil larvae and Cabbage Stem Flea Beetle larvae have been sampled from leaves. Cabbage Stem Weevil was the main pest species in both fields and its number is quite bigger than Cabbage Stem Flea Beetle larvae (Fig. 15, 16). However, for both pest species' larvae from the analyses there were no significant effects between the number of larvae at different distances in agroforestry field and conventional field for different sampling dates.



Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

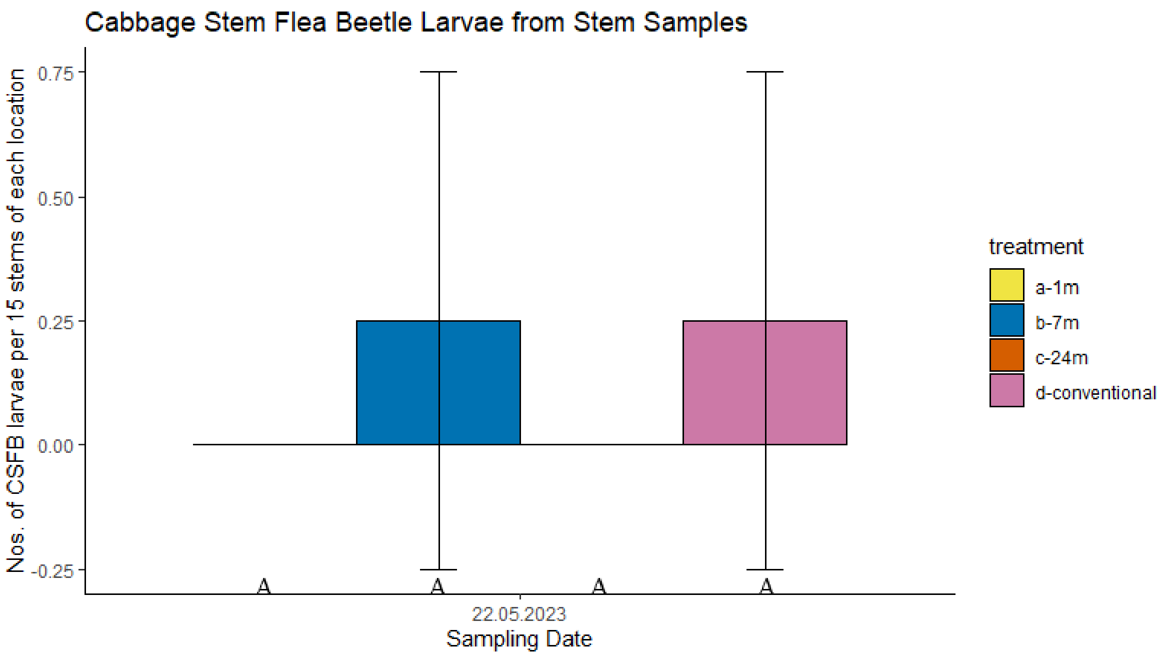
Fig. 15 Numbers of Cabbage Stem Flea Beetle larvae extracted from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

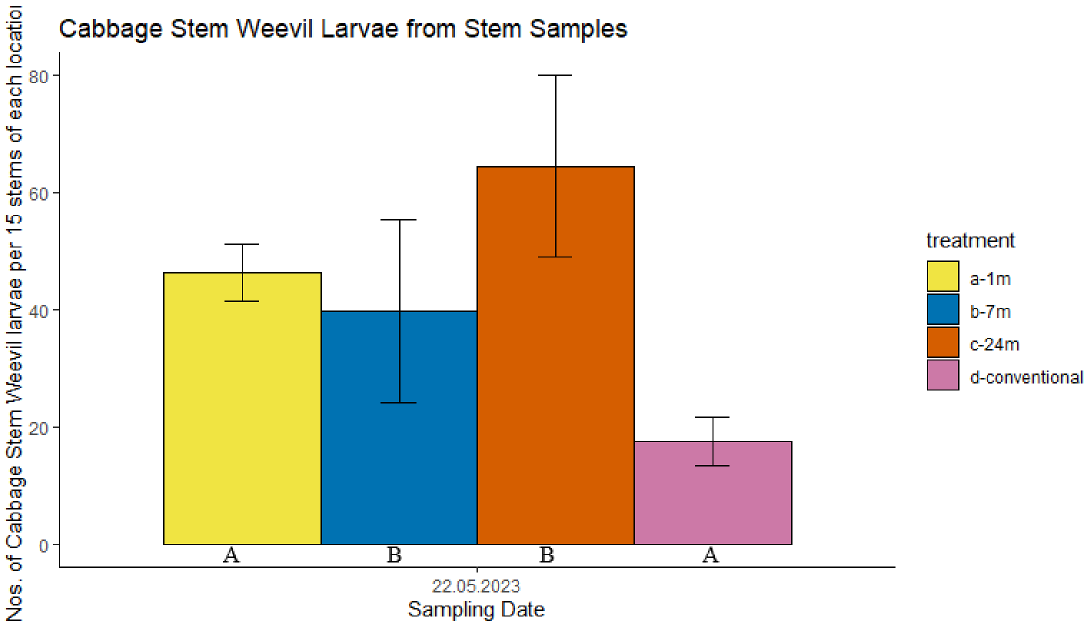
Fig. 16 Numbers of Cabbage Stem Weevil larvae extracted from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation

To assess the level of infestation by stem borer insects of oilseed rape 15 randomly selected main stems of each location were dissected and larvae mining within stems were collected. Results show that Cabbage Stem Flea Beetle larvae were too few to analyse the difference between agroforestry and conventional field (Fig. 17). However, number of Cabbage Stem Weevil larvae differed between distances in agroforestry and conventional field (Fig. 18). One-way ANOVA test shows that there are significant differences between different distances and conventional field shown in figure 18. However, while there was a significantly higher larvae at 1m distance and 24m distance than in the conventional field there were no significant difference between those distances for infestation level (Fig. 19). In addition to comparison of number of larvae caught in stem samples and infestation of stems, for both experiments' results show that there is a significant difference between agroforestry and conventional field. In conclusion, agroforestry has influenced the feeding activity of cabbage stem weevil larvae positively compared to conventional farming system.



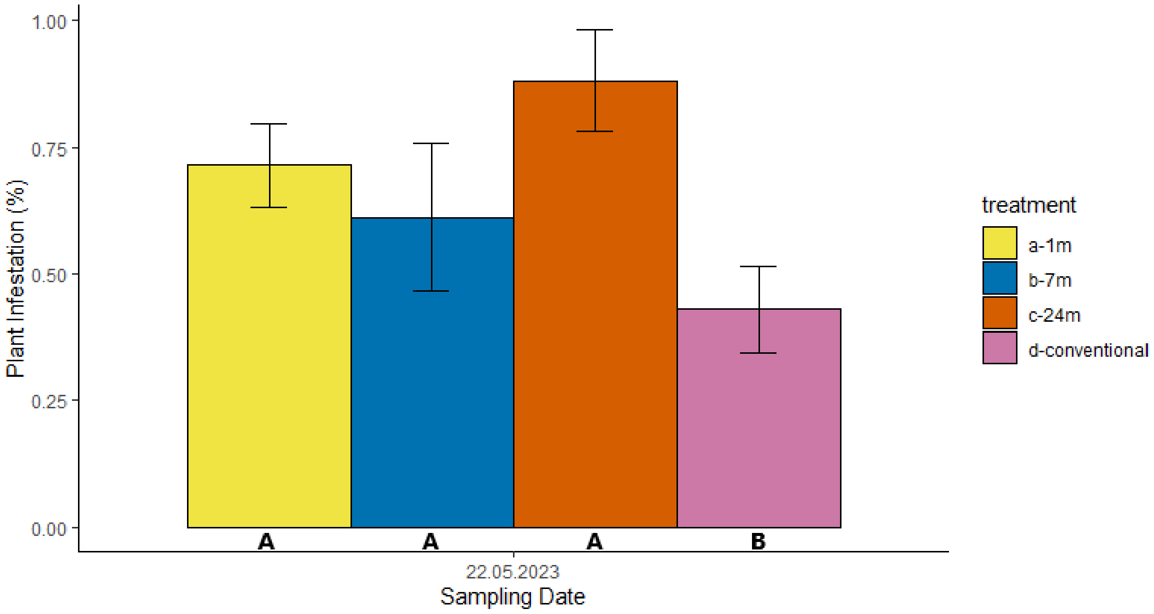
Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

Fig. 17 Numbers of Cabbage Stem Flea Beetle larvae caught from stem samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

Fig. 18 Numbers of Cabbage Stem Weevil larvae caught from stem samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation

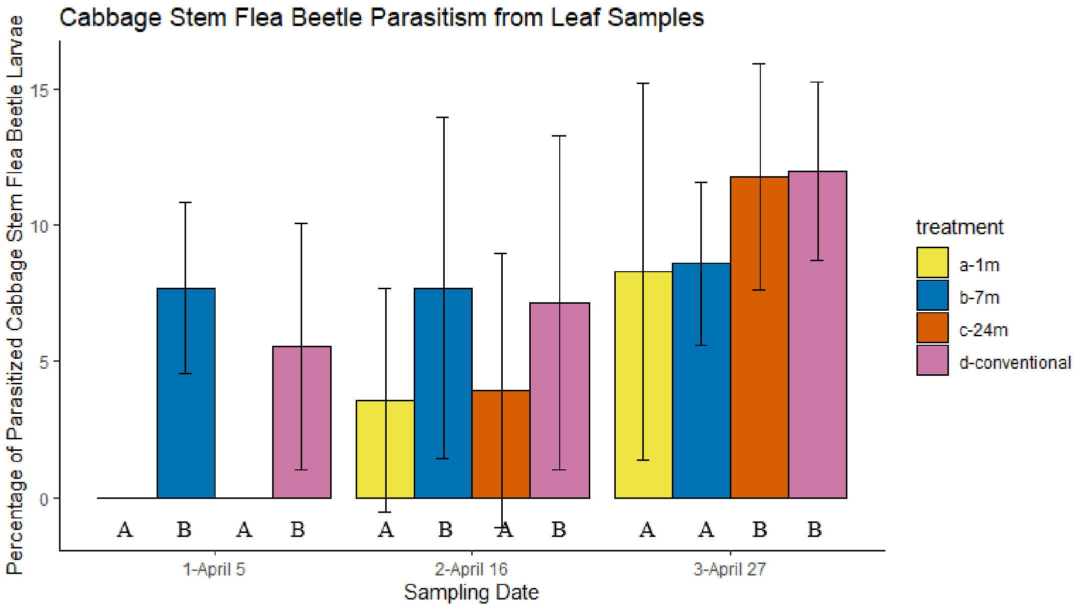


Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 19 Plant infestation percentage of sampled stems, presented as mean values with 95% confidence intervals calculated based on the standard deviation

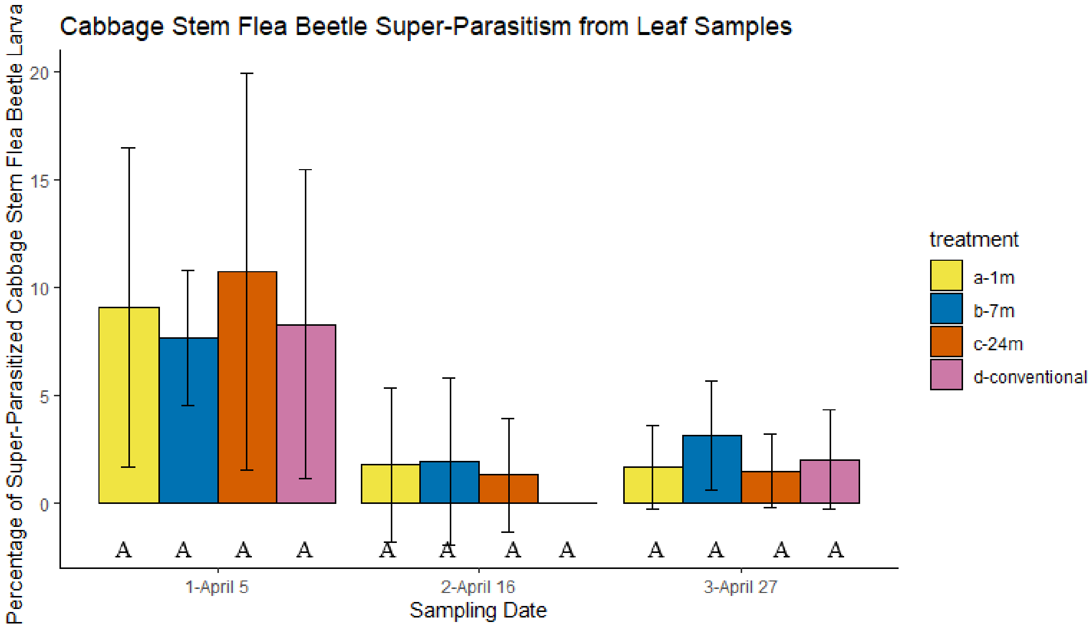
3.4 Larval Parasitism of Stem-Boring Insects

The percentage of parasitized Cabbage Stem Flea Beetle larvae caught from leaf samples does show some fragmented significant differences between different treatments, however conventional field's parasitism was always significantly different than 1m distances in agricultural field (Fig. 20). Percentage of super-parasitized Cabbage Stem Flea Beetle larvae caught from leaf samples has not shown any significant differences between treatments (Fig. 21). On the other hand, for Cabbage Stem Weevil larvae caught from leaf samples parasitism and super-parasitism have shown some fragmented significant differences when Cabbage Stem Weevil larvae population was lower at 5th and 16th April, however when Cabbage Stem Weevil larvae population was much higher at 27th April the percentage of parasitism and super-parasitism have not shown any significant differences between treatments (Fig. 22 and 23)



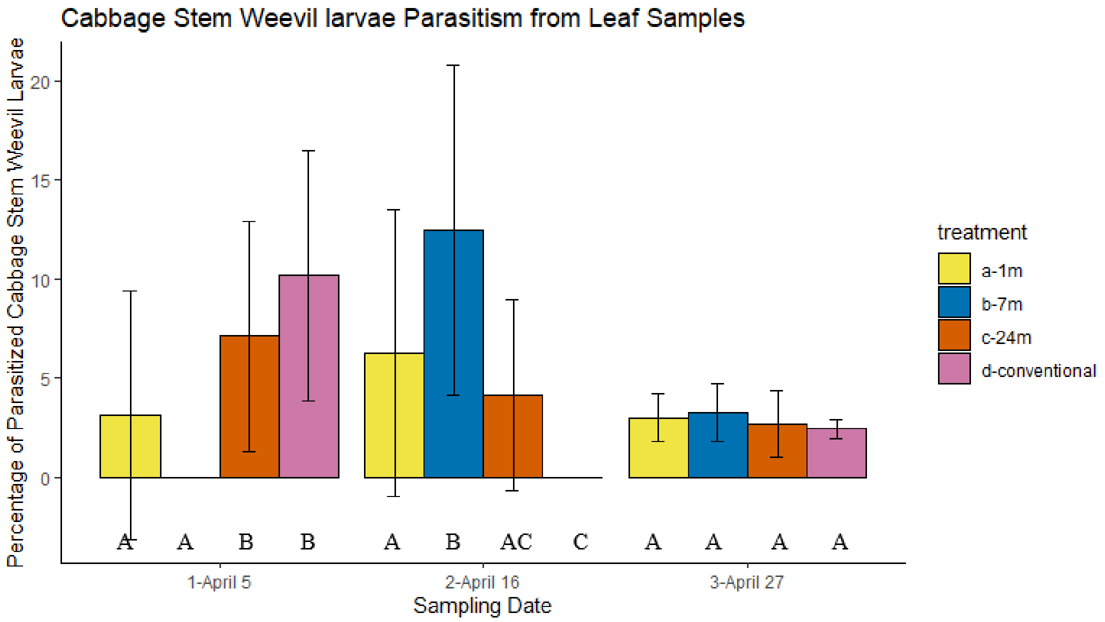
Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 20 Percentage of parasitized Cabbage Stem Flea Beetle larvae caught from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



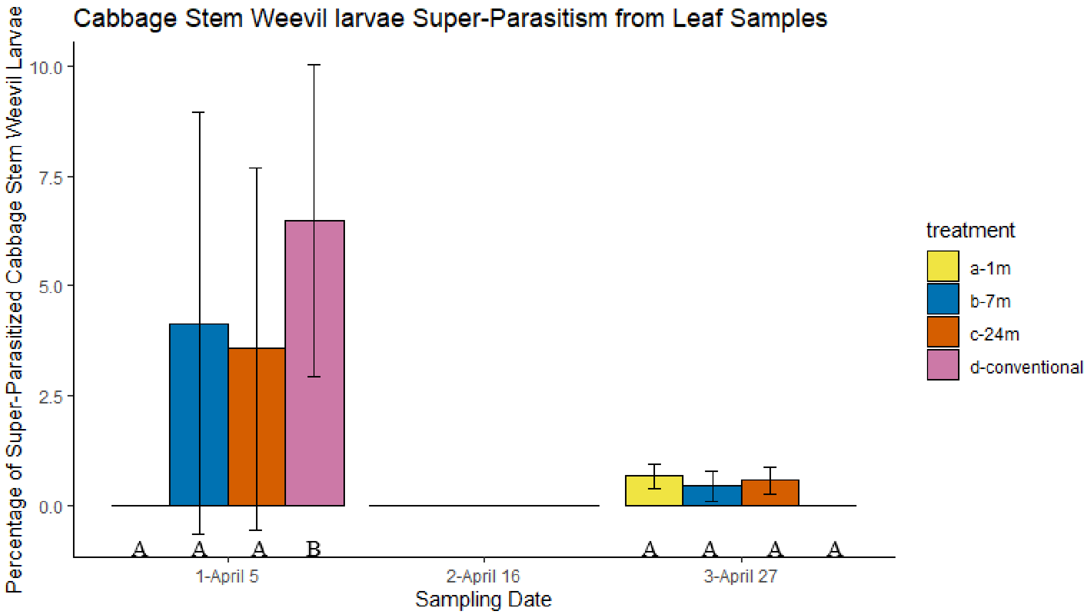
Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 21 Percentage of super-parasitized Cabbage Stem Flea Beetle larvae caught from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences between treatments (p<0.05)

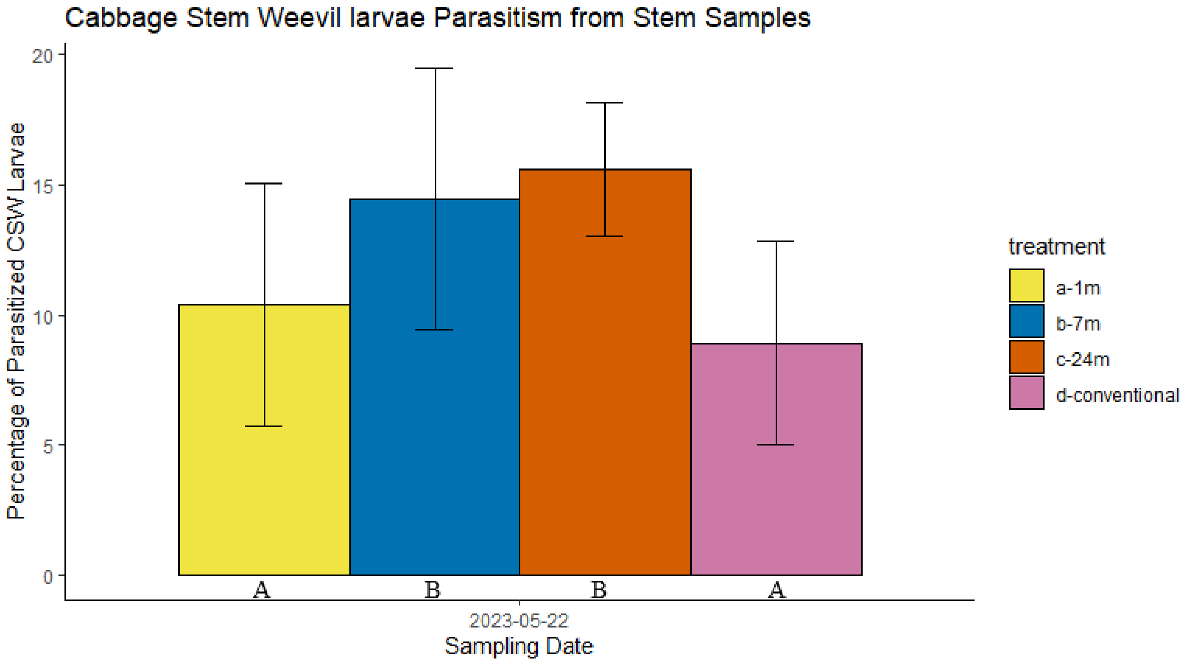
Fig. 22 Percentage of parasitized Cabbage Stem Weevil larvae caught from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences (p<0.05)

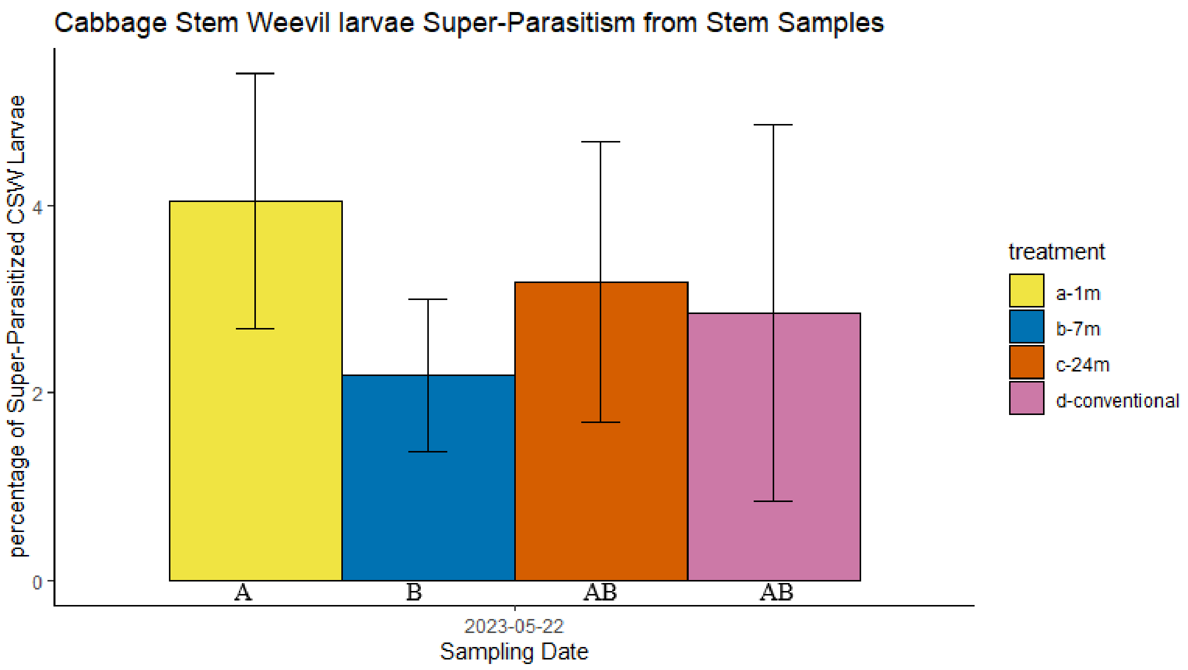
Fig. 23 Numbers of super-parasitized Cabbage Stem Weevil larvae caught from leaf samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation

The stem-borer insect larvae from stem samples in figure 17 shows that Cabbage Stem Flea Beetles have already started pupating into soil at 22th of May. Therefore, assessing the larval parasitism was not possible to have a robust result on Cabbage Stem Flea Beetle larvae caught in stem samples. However, Cabbage Stem Weevils were still present at that sampling date as it shown in figure 18. After the dissection of Cabbage Stem Weevil larvae caught in stem samples to evaluate the larval parasitism and super-parasitism level the results show that percentage of parasitism was significantly higher at 7m and 24m than at 1m and in the conventional field (Fig. 24). The percentage of super-parasitism of Cabbage Stem Weevil larvae caught in stem samples has shown a significant difference between 1m and 7m distance from tree rows (Fig. 25).



Different uppercase letters of the same font indicate statistically significant differences between treatments ($p<0.05$)

Fig. 24 Percentage of parasitized Cabbage Stem Weevil larvae caught from stem samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation



Different uppercase letters of the same font indicate statistically significant differences ($p<0.05$)

Fig. 25 Percentage of super-parasitized Cabbage Stem Weevil larvae caught from stem samples, presented as mean values with 95% confidence intervals calculated based on the standard deviation

4 DISCUSSION

4.1 Beneficial Aspects of Agroforestry Systems

Researched benefits of seminatural habitats explains it provides alternative host and prey, alternative food sources, and shelter and overwintering places. Noncrop habitats maintain populations of alternative hosts and prey for the parasitoids and predators of crop pests. This enhances natural pest control by providing the natural enemies of pests with alternative hosts and prey during periods in which host and prey density is low in fields, or by increasing the fitness of natural enemies (Rusch et al., 2010). In our study, it was observed that these seminatural habitats provided shelter to beneficial insects, thereby influenced their migration strategy (Fig. 11). However, habitats providing alternative hosts or prey may also accommodate pest species, thereby increasing pest populations. Pest abundance increased most in the scenarios that freed more pest species from predation (Karp et al., 2013). The higher abundance of Pollen Beetle larvae and bud damage near tree rows (Fig. 13 and 14) may indicate a supporting context for this statement. Initially parasitoid species were more abundant around the tree rows, the Pollen Beetles escaping from parasitoids have undergone reproductive stress to sustain their generations, consequently resulting in increased larvae and bud damage near observation points close to tree rows. The high infestation of Cabbage Stem Weevil larvae near observation points close to tree rows may also have a similar underlying cause. Some plant species increase the fitness of herbivores and parasitoids, whereas other species selectively enhance the fitness of parasitoid. (Rusch et al., 2010). Seminatural habitats also act as sources of pollen and nectar, which are essential for many species (Koptur, 2005; Pickett & Bugg, 1998). In this research the focused parasitoid species as most abundant and specialist were *Hymenopteran* parasitoid wasps. It has also been shown that many *Hymenopteran* parasitoid species feed on floral nectar (Jervis et al., 1993; Wäckers, 2001) and this may lead to higher rates of parasitism (Berndt et al., 2006). It has also been an idea that forest trees in this research may provide shelter for both pest and parasitoid species at extreme conditions such as cold. Woody habitats often provide a more moderate microclimate than the center of fields, protecting natural enemies against extreme temperature variations (Landis et al., 2000; Rahim et al., 1991). The moderate microclimate in combination with presence of nectar sources in wooded edges result in higher parasitoid longevity, early season abundance (Dyer & Landis 1996, 1997) and higher levels of parasitism as compared to field centres (Landis & Haas 1992). However, in our research this could not be observed as our results of larvae infestation by the parasitoids was not significant in general.

4.2 Limitations of Seminatural Habitats to be Beneficial Factors

While there are different benefits provided by forest trees, we should also consider there may be failures to get these benefits to integrate with the pest management. While natural habitat has been shown to increase pest control in many systems, we here identify five hypotheses for when and why natural habitat can fail to support biological pest control, and illustrate each with case studies from the literature: (1) pest populations have no effective natural enemies in the region, (2) natural habitat is a greater source of pests than natural enemies, (3) crops provide more resources for natural enemies than does natural habitat, (4) natural habitat is insufficient in amount, proximity, composition, or configuration to provide large enough enemy populations needed for pest control, and (5) agricultural practices counteract enemy establishment and biocontrol provided by natural habitat (Tscharntke et al., 2016). While agricultural practices held by local farmers in our experimental field hypothesis 5 could be a most possible limitation than the others if local farmers apply some practices which can change the real effect of agroforestry.

4.3 Evaluation of Results from Different Phases of the Research

Assessment of pest density usually requires obtaining actual counts of the pests, and therefore, sampling is important (Binns & Nyrop, 2003). By monitoring the pest and parasitoid species in this research we had information for future predictions of most abundant pests and their parasitism. In this study, the varying emergence time intervals obtained from yellow water trap observations for different pest and parasitoid species could provide insights for future research. Additionally, the fragmented results of larval parasitism could serve as an inspiration for investigating the reasons behind these differences in future studies.

One of the main focuses of this research was evaluate the effect of trees on beneficial arthropods. To assess the pest larval mortality caused by parasitoids the larvae caught from leaf and stem samples dissected to see if they have parasitoid eggs in their body or not. The level of parasitism of target pests was determined from samples of numerous field experiments and commercial crops of oilseed rape by dissection of larvae (Ulber et al., 2010). Percent parasitism of target pests varied between different distances in agroforestry field and between conventional field.

At monitoring phase of this research results obtained from yellow water traps show that pest density is usually higher at further distances (7 and 24 meters higher than tree rows and 1 meter) in agroforestry field and also in conventional field than tree rows (Fig. 7,8,9 and 10). It might show that single cultivated oilseed rape plants provide more benefits than forest trees. On the other hand, parasitoid wasps might have started flying in the fields later than pest species may show that parasitoid wasps are more sensitive to lower temperatures than pest species (Fig. 11). Additionally, even though the number of parasitoid wasps has not shown a significant difference between the distances in agroforestry field and between conventional field (Fig. 10) it can be clearly seen that they first started flying around trees with more numbers of individuals then after 2 weeks they migrated to conventional field. The outcome obtained in this study aligns with the parasitoid migration behaviors proposed in the works of Tscharntke and Brandl (2004) and Tscharntke et al. (2005), suggesting that in extremely diverse landscapes, the impact of on-farm habitat is diminished due to high levels of natural enemy migration into the crop from the surrounding landscape. It can be a sign of parasitoid wasps feed and developed with the food that forest trees provided. As conclusion, parasitoid wasps might prefer the pollens and nectars that from trees more than the provided food from oilseed rape plants.

Agroforestry has also influenced the distribution of adult Pollen Beetles when their number of individuals are high. The reason of this result might be a reproductive stress caused from parasitoid wasps as mentioned before. Although, temperature is the main factor for their population and it can be clearly seen that at 16th of April they were not in the fields because it was below 0°C (Fig. 12).

From the larvae density observation, it can be seen Cabbage Stem Flea Beetle larvae was exist in leaf samples on April (Fig. 15), however, they might have started pupating after and their existence was almost completely decreased in May from on stem samples (Fig. 17). On contrast there was not the same result observed for Cabbage stem weevil larvae. Cabbage stem weevil larvae existed both in April and May (Fig. 16, 18). Therefore, it can be concluded with Cabbage Stem Flea Beetle starts pupating earlier than Cabbage Stem Weevil.

About the larval parasitism Cabbage Stem Flea Beetle larvae caught from leaf samples does show fragmented significant differences for both parasitism level (Fig. 20), but the significant difference of conventional field between different distances of agroforestry field remains still. On the other hand, the Cabbage Stem Weevil larvae caught from leaf samples show also fragmented significant differences at 5th and 16th of April for both parasitism and super-parasitism levels, however when their population reach peek the percentage of both parasitism and super-parasitism have not shown any significant differences between any treatment (Fig. 22,23). It might show even though there might be some influences of agroforestry on parasitoid wasps when host insects' population is lower, the influence might not remain enough for higher population levels of host insects. Additionally, because the percentage of parasitism and super-parasitism of the Cabbage Stem Weevil larvae caught from stem samples has shown some fragmented significant differences between different treatments (Fig. 24, 25), it is not enough to make inferences of the influence of agroforestry system on parasitoid wasps.

4.4 Improvements and Future Studies

Agroforestry has been proposed as a sustainable agricultural system over conventional agriculture and forestry, conserving biodiversity, and enhancing ecosystem service provision while not compromising productivity. However, the available evidence for the benefits of agroforestry is fragmented and does often not integrate diverse ecosystem services into the assessment (Torralba et al., 2016). Comparing the results of Torralba et al. (2016) this research's larval parasitism results were also fragmented, however it showed that nature provides its own benefits to ecological life via being additional shelter to monocultural fields, provides additional food and creating micro-climate to the field. This research that includes monocultural oilseed rape field and agroforestry system has showed that there are small benefits of agroforestry comparing with conventional oilseed rape field. Additionally, in the agroforestry field some of the benefits differ depending on the distance to forest trees in some results. Therefore, agroforestry systems have also different effects in their own fields. Planting trees near crops can diversify the landscape, provide pollinators for crops, and allow predators to control insect pests. However, trees can also compete with crops for light, water, and nutrients (Yang et al., 2019). In our study, the impact of trees on plants may have resulted in varying levels of growth among oilseed rape plants at different distances in our experimental field. It is possible that oilseed rape plants located closer to tree rows were less attractive to pest insects.

This research has been done to evaluate the effect of agroforestry systems on the major pests of oilseed rape and their biological control agents. Various results have been obtained to evaluate the effect of agroforestry systems on major oilseed rape pests and their bio-control agents in different life stages from the aspect of intensity, behaviour of pests and parasitism capacity of their parasitoid species. In conclusion this research contributed to assign the effect of agroforestry systems on pest control in oilseed rape agriculture. For further studies it's effect can be evaluated by species specific with deeper aspects.

5 Conclusion

In conclusion the agroforestry systems has shown significant benefits at some levels compared to conventional farming, however, the most significant results in this research parasitoid wasps started their flights first around tree rows and crops in the agroforestry field and later on they migrated to the conventional field. However, agroforestry systems are not the only variable on pest and bio-control agents' population while temperature, humidity and wind are the main factors as well. Hence agroforestry systems should be studied more in future and can be applied as a supportive method in addition to integrated pest management.

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