

Increased grass cover height in the alleys of apple orchards does not promote Cydia pomonella biocontrol

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Abstract Habitat management such as ground cover implementation in orchards is considered a promising approach to augment natural enemy abundance and diversity. However, the actual effects of grass cover management and specifically mowing frequency, on the abundance of tree natural enemies and thus on the biocontrol of major pests, are still largely unknown. We compared the effect of three heights, tall (no cutting), medium (mean height of 20 cm) and short (mean height of 5 cm), of a spontaneous grass cover in an experimental orchard on natural enemy abundance and predation rates from April to August. Early in the season (from April to May) grass cover height had no effect on codling moth egg predation. However later in

the season (July and August) the predation rate increased more in the short grass cover than in the tall grass (66 vs. 38 %. respectively). The abundance of the earwig Forficula pubescens (Gené) (Dermaptera: Forficulidae) was significantly positively correlated with egg predation in the plots with short grass but negatively correlated in the tall and medium plots suggesting that these predators could find an alternative resource in the taller grass cover.

Keywords Biological control · Natural enemy · Habitat management · Earwig · Forficula pubescens · Cheiracanthium mildei

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Introduction

Habitat management is an ecologically-based approach aimed at favoring natural enemies and enhancing biological control in agricultural systems (Landis et al. [2000](#page-9-0)). Classical examples of habitat manipulation are the management of hedgerows and ground cover in and around the crop to favor natural enemies. The increase in plant diversity is thought to provide natural enemies with alternative resources, i.e., prey, nectar, pollen, honeydew; day shelters to escape disturbances due to environmental changes or pesticides, and overwintering shelters (Landis et al. [2000\)](#page-9-0). Orchards are potentially more adapted to conservation biocontrol than arable crops due to the presence of perennial habitats and of hedgerows and ground cover in a large proportion of the cultivated area in current orchards (Simon et al. [2010](#page-9-0)).

Different types of ground cover have been implemented and tested in orchards: mulch, flower strips, grass crop cover, spontaneous ground cover (Simon et al. [2010](#page-9-0)). Most of these studies compared bare soil with one of this ground cover. This resulted in clear conclusions: the presence of a ground cover favors natural enemies abundance in trees (Markó and Keresztes [2014;](#page-9-0) Rieux et al. [1999;](#page-9-0) Silva et al. [2010](#page-9-0); Smith et al. [1996;](#page-9-0) Wan et al. [2014\)](#page-10-0). Indeed, compared to bare soil, ground cover is a complete change in soil habitat with an important increase in complexity, shelter and resource availability, thereby favoring natural enemies. Different types of non-spontaneous land cover have been also compared between each other but the conclusions were far less clear-cut. For example, Markó and Keresztes [\(2014](#page-9-0)) compared a frequently mowed natural grass cover with a flower cover and observed higher Araneae and Chrysopidae abundances in trees with the flower ground cover, but no difference in Coccinellidae abundance. In contrast, Silva et al. [\(2010](#page-9-0)) compared a spontaneous ground cover with a sown grass cover. The sown grass cover was a cover of plants selected to produce nectar and pollen for an extended flowering period in order to favor natural enemies. They found that the abundance of Araneae, Coccinellidae and Chrysopidae was similar in the two ground covers.

Sowing a ground cover and/or a flower cover in orchard alleys can have some direct economical consequences but also some indirect drawbacks (some pesticides are forbidden when flowers are present to protect pollinators). Manipulating the existing (i.e. "spontaneous") ground cover is a less expensive alternative. Indeed some growers already use this habitat manipulation approach by decreasing mowing frequency. Mowing represents a mechanical disturbance in itself, decreases the height and the complexity of the grass ground cover (Horton et al. [2003](#page-9-0); Kruess and Tscharntke [2002](#page-9-0); Langellotto and Denno [2004\)](#page-9-0). This increase in complexity is thought to influence arthropod numbers in the cover, through increased shelter and also alternative food which decreases intraguild predation. However, an increase in natural enemy abundance in the grass cover does not necessarily translate into a higher abundance in the trees (Horton et al. [2003](#page-9-0)). The movement of natural enemies between the grass cover and the tree canopy could be driven by several factors: the natural enemy's mobility, the physical proximity of the strata, prey availability, anthropic disturbance, the living conditions including microclimate and predation escape (Horton et al. [2009](#page-9-0); Lawton [1983;](#page-9-0) Schellhorn et al. [2014](#page-9-0)). Last, if there is an increase in natural enemies on trees, this is expected to positively impact pest control. However, the relationship between natural enemy diversity and pest control is not straightforward: different studies showed a positive, negative or neutral effect between predator diversity and pest control (Finke and Snyder [2010](#page-9-0); Letourneau et al. [2009](#page-9-0); Straub et al. [2008](#page-10-0)). The relationship between natural enemy diversity and pest control is strongly dependent on the predator species composing the community and their interactions (i.e., intraguild competition, resource competition) (Straub et al. [2008](#page-10-0)).

In South-Eastern France, the current grass covers found in orchard alleys shelter a community of natural enemies with the potential to control apple pests (Simon et al. [2007\)](#page-9-0). The main apple pest is the codling moth Cydia pomonella (Linnaeus) (Lepidoptera: Tortricidae), which is targeted by 12 insecticide applications on average (Sauphanor et al. [2009\)](#page-9-0) unless alternative methods such as mating disruption are used. Codling moth has two or three yearly larval generations in South-Eastern France and the egg stage is the most crucial for biocontrol to avoid damage to the fruit by the larvae. Both egg predation and, to a lesser extent, parasitism (Monteiro et al. [2013\)](#page-9-0) could contribute to this early-stage biological control. The guild of generalist predators active against Lepidoptera eggs has been already documented: earwigs (Forficula spp., Dermaptera: Forficulidae) (Glen [1977\)](#page-9-0), spiders (Araneae) (Glen [1977](#page-9-0); Pearce et al. [2004](#page-9-0)), ladybirds (Coleoptera: Coccinellidae) (Chang and Snyder [2004;](#page-9-0) Evans [2009](#page-9-0); Glen [1977](#page-9-0)), and true bugs (Hemiptera: Miridae) (Glen [1977](#page-9-0)). This guild is present during the presence of codling moth eggs in the trees. Aphids, including the rosy apple aphid Dysaphis plantaginea (Passerini) (Hemiptera: Aphididae), are alternative prey that can be shared by these predators (Dib et al. [2010\)](#page-9-0).

In this experimental study, we assessed the abundance and diversity of some important natural enemies and the predation of codling moth eggs in the apple trees according to three heights of ground cover. We addressed two different questions: (i) which natural enemies do contribute to the predation along the season? and (ii) when the predation rate is the highest, what is the impact of the ground cover height on the relationship between natural enemies and predation rate? The sentinel eggs technique (Glen [1977](#page-9-0)) was used to assess the natural enemy predation activity. Three heights of grass cover were compared in the orchard: tall (no mowing), medium (20 cm height maintained throughout the whole season) and short (5 cm height maintained throughout the whole season). These three grass heights represent three levels of physical disturbance (due to mowing frequency), complexity and resource availability for natural enemies during the growing season within the orchard alleys.

Materials and methods

Experimental orchard and study grass covers

The study was conducted in a National Institute for Agricultural Research (INRA) experimental apple orchard (0.2 ha) in Avignon (south-eastern France) from 25 April to 22 August 2013 (one sampling date per month). The orchard was planted in 2007. It was under a minimal pesticide management program. Only two pesticides were applied during the study: bupirimate (7 May) to control the powdery mildew fungal disease and C. pomonella granulosis virus (7 July) to reduce codling moth population. The orchard had six rows of 60 trees. The rows were spaced 4 m apart and the apple trees were spaced 1.5 m apart within each row.

The alley vegetation was a four meter wide spontaneous ground cover. It was composed mainly of Trifolium repens, Festuca pratense and Taraxacum officinale mixed with some Trifolium pratense, Lolium perenne, Convolvus arvensis, Plantago lanceolata, Plantago major, Picris echioides, Crepis sancta, Bellis perenis, Veronica persica and Dactylis glomerata. The ground cover was mowed using a line trimmer. Ground cover in both the row and alley were managed in the same way. Three management regimes and corresponding ground cover heights were defined: tall, no mowing during the whole study period; medium, mowed every two weeks to maintain the grass cover height at 20 cm on average; and short, mowed every week to maintain the grass cover height below 5 cm. In the tall cover height, i.e. 'no mowing' regime, the height of the ground cover regularly increased and it was taller than 40 cm from May to September. Since both the rows and alleys were uncut, during summer the tallest plants in the ground cover ''overlapped'' with the lowest tree branches. The experimental design was a randomized block with four replicates per moving regime. Each plot included 30 trees in total with ten trees in three successive rows. At the first sampling date (i.e., April), the ground cover was not yet tall enough to be considered in the tall plots.

Predation rate of codling moth sentinel eggs in the apple canopy

We used eggs of a susceptible C. *pomonella* laboratory strain reared on an artificial diet (INRA, Avignon, France) to assess egg predation (Glen [1977](#page-9-0)). Egg laying sheets (30 \times 30 cm) were placed in the rearing cages at dusk and females were allowed to lay eggs for 12 h. The following morning, the eggs were sterilized under a UV lamp (20 W) for 20 s (Monteiro et al. [2013\)](#page-9-0). The sheet was cut into cards containing an average of $8.75 \pm SD$ 1.28 eggs. Ten cards were placed in each plot (forty cards in total for each grass cover height) excluding trees at the border of plots. Each card (one per tree) was stapled to the lower side of a leaf at the outside of the canopy at 1.5 m height. After 2.5 days exposure to natural enemies, the cards were removed and the predation rate was assessed by determining the ratio of the number of remaining eggs to the number of exposed eggs per card. Egg predation was accessed in five successive months, on 25 April, 24 May, 27 June, 19 July and 22 August 2013.

Earwigs and spiders abundance and diversity

The abundance of earwigs and spiders in the tree canopy was estimated with corrugated cardboard band traps. In each plot, ten corrugated cardboard band traps, i.e., forty for each grass cover height were wrapped on the same trees where the predation rate was estimated. They were wrapped around a branch at a height of 1.20–1.50 m above ground level. The traps were 10 cm wide, 20 cm long, with 13 corrugations per 10 cm (Pekar [1999](#page-9-0); Simon et al. [2011\)](#page-9-0). The traps were placed one week before the exposition of sentinel eggs and removed one week after the exposure, during the day. To avoid possible interactions between the trap and the sentinel eggs on the same tree, these were never placed on the same branches. Once removed, the traps were individually closed in a plastic bag then opened under laboratory conditions and any arthropods stored in 70 % ethanol for further identification. According to the relative abundance of sampled arthropods, we defined operational taxonomic units (OTU). The two species of earwigs, Forficula auricularia (Linnaeus) and Forficula pubescens (Géné) (Dermaptera: Forficulidae), were two distinct OTU. Araneae families were considered as OTU: Salticidae, Gnaphosidae, Clubionidae, Miturgidae, Thomisidae and Philodromidae.

Statistical analysis

To determine whether the different grass cover heights (tall, medium and short) influenced egg predation and the abundance of predators, we applied a generalized linear model (GLM) at each date with, respectively, a Binomial error distribution with logit link function and a quasi-Poisson error distribution followed by an analysis of deviance. A multiple post-hoc test (Tukey test) was performed to detect any significant difference between the moving regime. The relationships between codling moth egg predation and the abundance of natural enemies were studied using two approaches. In the first approach, we addressed the question: which natural enemies do contribute to the predation along the season? For this purpose, all data were considered. In the second approach, we addressed the question: when the predation rate is the highest, what is the impact of the height of ground cover on the relationship between natural enemies and predation rate? For this purpose, only data collected in July and August were used. We assessed the effects of the grass cover height on the abundance of natural enemies and on the predation rate. For both approaches the same method was applied. A GLM was used with a Binomial error distribution and a logit link function. An information-theoretic approach including all the candidate models (based on the fit of all possible variable combinations) was used. The best model was selected using the Akaike Information Criterion (AIC) which measures goodness of fit and model complexity (Zuur et al. [2009](#page-10-0)). The estimated parameters, SE, Z-values, and p-values of the best models were given. The Z-value is the Wald statistic testing the hypothesis that the corresponding parameter (regression coefficient) is zero. For all GLM procedures, model parameters were considered to be significant at a level of 1 % (Zuur et al. [2009](#page-10-0)). Generalized linear models were performed with the lme4 package (Bates et al. [2014\)](#page-9-0), and the information-theoretic approach with the package MuMin (Barton [2013](#page-9-0)) in the R 2.14.1 software (R Development Core Team [2010](#page-9-0)).

Results

The natural enemy community

A total of 20,537 natural enemies were caught on the five sampling dates. Earwigs and spiders were the most abundant groups, representing 96.75 and 3 % of the total arthropods, respectively. The other natural enemy groups (Coccinellidae, Miridae) were found in percentages below 0.01 %. Two species of earwig were found: F. auricularia (26.5 % of the earwigs) and F. pubescens (73.5 %). The spider community consisted of Salticidae (74.4 % of the Araneae community), Gnaphosidae (9.70 %), Miturgidae (only one species: Cheiracanthium mildeı¨ (L. Koch) (Araneae: Miturgidae), 3.63 %), Philodromidae (3.64 %), Thomisidae (1.82 %) and Clubionidae (0.30 %). Salticidae were the most common Araneae family in the tree canopy, they were present during the whole season with a peak of abundance in April. Gnaphosidae and Miturgidae were present late in the season (i.e., August). Very few Thomisidae and Philodromidae were present in the tree canopy (Table [1\)](#page-4-0). There was no difference in the abundance of any Araneae family (i.e., Miturgidae, Clubionidae, Gnaphosidae, Thomisidae, Philodromidae and Salticidae) among the three grass cover heights at any sampling date (Table [1\)](#page-4-0).

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Fig. 1 Temporal dynamics of a the mean abundance per trap of F. auricularia ($+SE$), **b** the mean abundance per trap of F. pubescens $(+SE)$ and c predation rate of codling moth eggs $(+SE)$ in the tall, medium and short grass cover plots in apple orchards. Bars bearing different letters are significantly different $(p < 0.05$; each date was tested separately)

The mean abundance of *F. auricularia* (\pm SE) per tree steadily increased from April to July (peak of presence) and decreased in August. However, the height of the grass cover did not have a significant effect on their abundance at any time from April to August (Fig. 1a). The mean abundance $(\pm SE)$ of F.

pubescens per tree increased slowly until June $(6.81 \pm 0.60, 5.90 \pm 0.7 \text{ and } 6.30 \pm 0.7 \text{ in the tall},$ medium and short plots, respectively) with no difference among plot types $(F = 0.46, df = 2, 113,$ $p = 0.63$). The peak of abundance was observed in July with a significantly higher abundance in the tall (117.72 ± 11.27) plots than in the short plots (64.45 ± 7.11) (F = 8.30, df = 2, 113, p < 0.001). F. pubescens abundance decreased in August with a higher abundance in the tall (28.13 ± 3.11) and medium (26.72 ± 4.03) than in the short plots (10.1 ± 1.24) (F = 14.65, df = 2, 113, p < 0.001) (Fig. 1b).

Predation rates

The predation rate of codling moth eggs was low (below 0.2) from April to June, then increased significantly in July and August (between 0.35 and 0.70) (Fig. 1c). The height of the grass cover had a significant impact on the predation rate. In June, the predation rate (mean \pm SE) in the tall plots (0.16 ± 0.05) was significantly higher than in the short plots (0.05 ± 0.02) $(\chi^2 = 19.6, \text{ df} = 2,$ $p < 0.001$). In July, the predation rates in the short and medium plots $(0.66 \pm 0.07; 0.58 \pm 0.07, \text{respec}$ tively) were significantly higher than in the tall plots (0.38 ± 0.07) $(\chi^2 = 55.39, df = 2, p < 0.001)$. In August, the predation rates decreased and were significantly higher in the short (0.49 ± 0.07) than in the medium plots (0.37 ± 0.07) $(\chi^2 = 9.90,$ $df = 2$, $p = 0.007$).

Link between predation and abundance of natural enemies

Table [2](#page-6-0) shows the best models linking the predation rate and natural enemy abundance across the season (first approach) for each grass cover height. In the tall plots, F. pubescens and Miturgidae had a significant positive effect while Philodromidae had a significant negative effect. In the medium plots, F. pubescens and Miturgidae had a significant positive effect. In the short plots, F. pubescens and Gnaphosidae had significant positive effects while F. auricularia had a significant negative effect on the total predation rate.

Table 2 Best generalized linear models selected using the Akaike Information Criterion linking the predation rate and natural enemy abundance in whole season for (a) tall, (b) medium, (c) short height grass cover

Natural enemies with significant effects on predation rate are in bold * p < 0.01 ; ** p < 0.001

Table 3 Best generalized linear models selected using the Akaike Information Criterion linking the predation rate and natural enemy abundance in July and August for (a) tall, (b) medium, (c) short height grass cover

Natural enemies with significant effects on predation rate are in bold * $p < 0.01$; ** $p < 0.001$

Table [3](#page-6-0) shows the best models linking the predation rate and the natural enemy abundance, in July and August only (second approach), for each ground cover height. In the tall plots, Miturgidae had a significant positive effect while Philodromidae had a significant negative effect on the total predation rate. In the medium plots, Miturgidae, F. auricularia and Philodromidae had a significant positive effect while F. pubescens had a significant negative effect on the total predation rate. In the short plots, F. pubescens had a significant positive effect on total predation rate.

Discussion

Earwig and spider communities

Earwigs dominated the community. Spider abundance was much lower and other natural enemies were rare. Our sampling method (corrugated cardboard band traps) affected this result at least in part. First, some predatory arthropods such as coccinellids or mirids are hardly ever sampled by this type of trap. Second, earwigs produce an aggregation pheromone (Sauphanor and Sureau [1993](#page-9-0)) that increases their abundance in traps (Burnip et al. [2002](#page-9-0)).

Forficula auricularia abundance increased steadily from April to August whereas a marked peak in F. pubescens numbers was observed in July. Simon et al. [\(2011](#page-9-0)) observed a similar increase in the total earwig abundance in July in apple orchards in South-Eastern France whereas Romeu-Dalmau et al. ([2012\)](#page-9-0) observed the same pattern for F. auricularia in May in citrus orchards in Spain (i.e., two months earlier than in our study). The difference in temperature between Southern France and the region in Spain could explain the difference in the development of this insect (Helsen et al. [1998\)](#page-9-0). In our study, only F. pubescens was found in higher abundance in plots with tall ground cover whereas the same numbers of F. auricularia were found in the three plot types. Very few literature is available on F. pubescens ecology but some authors think this specie is more polyphagous than F. auric-ularia (Debras [2007\)](#page-9-0). This may explain why the two species did not react the same way in our study. We assumed that F. pubescens found more suitable resource in the tall ground cover than F. auricularia and fed preferentially in this cover to the detriment of the apple tree canopy.

The three grass cover management regimes used in our study also had no effect on spider abundance. In contrast, Horton et al. ([2003](#page-9-0)) found that mowing frequency did affect spider abundance in a pear orchard, with more spiders found in the trees where the alleys were mowed less frequently. In the present study, the abundance of Salticidae, the major family found in the tree canopy, did not increase when grass height increased. This is not surprising because the Araneae community in the studied apple trees is essentially arboreal and very few species (some Gnaphosidae) were found in both the tree canopy and in the grass cover during the season (Bogya et al. [2000\)](#page-9-0).

Which natural enemies do contribute to the predation along the season?

In the present study, we recorded data on the dynamics of egg predation rates in French apple orchards for the first time. The predation rate of codling moth eggs in the studied apple orchard increased from April to July and reached a peak in July and August. These results are consistent with those of Monteiro et al. [\(2013](#page-9-0)) in apple orchards and Atanassov et al. ([2003\)](#page-8-0) in peach orchards. The sequential arrival of natural enemies could explain the temporal dynamics of the predation rate. At the beginning of the season, few natural enemies were present in the tree canopy. Salticidae prevailed among natural enemies but due to their ''sit and wait'' hunting mode, they are not expected to eat static prey such as eggs. Earwigs are known to eat Lepidopteran eggs (Glen [1977](#page-9-0)) and the large increase in F. pubescens abundance in apple trees in July correlated well with the increase in predation rate (Fig. [1](#page-5-0)a, b; Table [2:](#page-6-0) F. pubescens positively correlated with the predation rate regardless of grass height). In contrast, the increase in *F. auricularia* abundance from May onwards did not lead to an increase in the predation rate of codling moth eggs (Fig. [1](#page-5-0)a, c; Table [2](#page-6-0)): F. auricularia was negatively correlated with the predation rate in the areas with short grass cover. Although present in the apple canopy and previously identified as a predator of codling moth eggs (Glen [1977](#page-9-0)), F. auricularia might not have fed exclusively on codling moth eggs at the beginning of the season (from April to June). In spring, F. auricularia is known to feed on rosy apple aphids (Dib et al. [2010\)](#page-9-0) before switching to codling moth eggs (Symondson et al. [2002\)](#page-10-0). This could explain the negative relationship between the predation rate and abundance of F. auricularia. F. auricularia could also disrupt the ability of another natural enemy to consume codling moth eggs (Straub et al. [2008\)](#page-10-0). Even if Miturgidae abundance was very low, the results of both modeling approaches suggested that they were significantly and positively correlated with the predation rate in the tall and medium plots (Table [2\)](#page-6-0). This indicates that they could be predators of codling moth eggs and confirms that their phenology (i.e., presence of active stage) correlated well with the presence of codling moth eggs in apple orchards (Glen [1977](#page-9-0)), making them an efficient predator (Hogg and Daane [2011\)](#page-9-0). In the tall grass cover Philodromidae negatively affected the predation rate suggesting that this grass cover favored these hunting species which can prey on other natural enemies.

Management of grass cover height did not improve the predation rate at the beginning of the season (from April to June). For apple growers, high predation early in the season would be of interest to limit both fruit damage and codling moth populations in the next generation. The abundant aphid community commonly sheltered by the Poaceae grass cover early in the season (Simon et al. [2007\)](#page-9-0) may offer a more easily available resource than prey scattered in the tree canopy. Natural enemies (in particularly omnivorous predators such as F. pubescens) may therefore be diverted from the target prey in the apple tree (i.e. codling moth egg) by prey resources in the grass cover. Several factors and processes related to species behavior and the ecology of the natural enemy complex (sequential arrival, prey preference, habitat preference, insect mobility, intra-guild predation) could be involved in the complex interactions between predation and resource availability.

What is the impact of the height of ground cover on the relationship between natural enemies and predation rate?

The height of the grass cover, however, did appear to influence predation later in the season. Indeed, in July and August, predation rates of the codling moth eggs were lower in the tall plots whereas F. pubescens abundance was higher in these plots than in either of the others. The decrease in mowing frequency and the resulting increase in the height of the grass cover was not favorable to the within-tree predation rate and even modified the link between the predation rate and F. pubescens (Table [3\)](#page-6-0). Miturgidae correlated positively with the predation rate in the tall and medium plots. As an omnivorous predator, F. pubescens can feed on other resources which may thus reduce its predatory activity or redirect it to other species (Robinson et al. [2008\)](#page-9-0). This could be the case in the tall plots where resources provided by the grass cover, notably Senecio spp, were abundant and close to the tree canopy. Thus in this scenario they would eat less target prey. An alternative hypothesis is that the foraging area for localizing prey is larger in the tall modality which would lead to a decrease in the predation efficacy in such a complex multi-strata habitat (Langellotto and Denno [2004](#page-9-0)).

In this study we showed that grass cover height in the alleys of an apple orchard had a significant impact on the abundance and predatory behavior of a generalist predatory arthropod: in July and August tall grass cover was correlated with increased F. pubescens abundance in the apple canopy, but lower egg predation. This result could be due to the presence of alternative resources in the grass and/or trees. Growers must therefore manage the tradeoff between promoting natural enemies (i.e., providing them with resources) and encouraging predators to mostly prey in the tree canopy where pest control is needed. The timing and spatial distribution of mowing (e.g. mowing one alley out of two in the orchard) appears to be crucial and should be studied further. For example growers could use a no mowing regime early in the season to first promote the abundance of natural enemies and then favor arboreal predation by reducing grass resources during key-periods for pest control.

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