IOBC-WPRS Working Group "Landscape Management for Functional Biodiversity"

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Edited by:

Graham Begg, Felix Bianchi, Nick Birch, Bärbel Gerowitt, John Holland, Daniela Lupi, Camilla Moonen, Mark Ramsden, Paul van Rijn

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The Convenors and Organizing Committee would like to thank the following organizations for their generous support of this meeting:









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#### Welcome note

I would like to welcome everyone to the 7<sup>th</sup> meeting of the Working Group kindly organised by Dr Graham Begg of the James Hutton Institute. As with previous meeting our aim is to bring together the latest advances in research and development on landscape management for functional biodiversity, to provide the opportunity to exchange information and to coordinate common research and identity of the main areas for future research. I hope you enjoy the varied programme of presentations, discussions and field visit but that you also contribute with your ideas and thoughts so we can have some lively debates. This will be my last conference as convenor and I thank all of you that have helped over the years, especially Prof Bärbel Gerowitt my co-convenor and those that have helped as editors of the bulletin or as local organisers. Now to enjoy the Scottish hospitality, not least a dram or two.

John Holland (WG-convenor)

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**Session II** 

# How economic thresholds could improve the uptake of conservation biological control in arable landscape – an industry perspective

## Mark Ramsden<sup>1</sup>, Steve Ellis<sup>2</sup>

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**Abstract:** The management of agricultural landscapes to better support the natural enemies of crops pests is an underused component of integrated pest management (IPM). Natural enemies can slow the rate of pest population growth and help limit economic damage to crops. However, a lack of floral resources, w inter habitat, additional alternative prey, and refuges from harmful crop management at field and landscape scales can reduce their impact. Providing these additional resources will enable natural enemies to make a more reliable and consistent contribution to pest control. To achieve this, farmers need to have confidence in the benefits of supporting natural enemies, and understand how targeted provision of additional resources will help to develop a more robust IPM programmes. This paper summarises evidence of the contribution of natural enemies to the control of key invertebrate arable crop pests, and discusses the additional resources they require. We consider how the impact of natural enemies can be incorporated into arable crop management through greater use of economic thresholds.

Key words: Additional resources, conservation biological control, crop pests, economic thresholds, IPM

## Introduction

Infestations of invertebrate arable pests are difficult to predict and can cause substantial yield losses (Culliney, 2014). Risk averse pest management favouring prophylactic insecticide application is common in arable crop management, as the cost of yield losses associated with severe pest outbreaks are far greater than the cost of applying sprays, and insecticides have a clear impact on pest infestations (Figure 1, Garthwaite *et al.*, 2015). However, the Sustainable Use Directive (SUD), SUD2009/128/EC requires farmers and agronomists within the European Union to follow the principles of IPM. This entails a reduction of insecticide use combined with greater used of non-chemical approaches such as crop rotation, development of pest resistant crop varieties, and improved contribution of predators and parasitoids (natural enemies) (Hillocks, 2012). As non-chemical methods have different modes of action to insecticides, the transition from insecticide dominated programmes to IPM is not straightforward. In Conservation Biological Control (CBC) for example, natural enemies slow pest population growth, reducing the likelihood of it reaching damaging levels in the crop. Despite a well-developed ecological understanding of pests and their natural enemies, CBC has had limited uptake in arable crop management. We summarise the role of economic

thresholds, and the mode of action of CBC, and then discuss how economic thresholds can facilitate reduced insecticide use through greater application of alternatives such as CBC in arable crops.

## **Economic thresholds**

Economic thresholds are a valuable method of assessing whether or not action is necessary to prevent pests from causing financial losses in a crop. They are usually defined in terms of the number of a pest per unit area, per plant, or per part of plant, above which action should be taken, and are available for most UK arable pests - though many have not been peer reviewed (Ramsden *et al.*, 2017). Economic thresholds should account for the tolerance of the crop to pest injury, which varies with factors such as crop variety, growth stage, crop density, and environmental stress. These factors will affect the pest density at which control measures are required (Ellis & Berry, 2012). While economic thresholds are currently used as a tool for deciding whether or not to apply insecticides, they can also function as a tool for demonstrating the impact of non-chemical options, such as CBC; i.e. where natural enemies have a significant impact, thresholds are less likely to be exceeded.

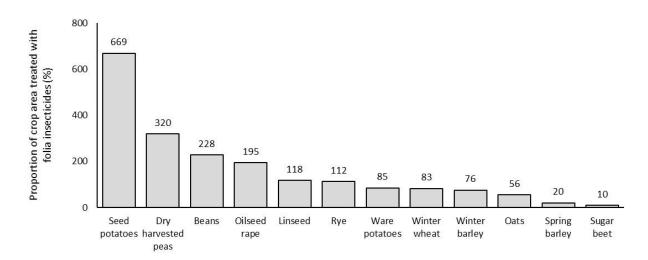


Figure 1. The proportion of crop area treated with foliar insecticides in UK arable crops in 2014, adapted from Garthwaite *et al.*, 2015. Applications greater than 100% indicate that multiple treatments have been applied.

#### The mode of action of conservation biological control

Both ground dwelling and flying natural enemies are capable of reducing pest population growth rates (Holland *et al.*, 2012; Ramsden *et al.*, 2016, Figure 2). This can reduce the pest density during periods when the crop is particularly vulnerable to attack, or delay the timing of peak pest abundance until the crop has reached a tolerant growth stage. The increased impact of natural enemies is therefore dependent on the size of natural enemy populations and the timing of their arrival in the crop; early and/or large natural enemy populations have a greater impact in slowing pest population growth.

Unlike the majority of arable pests, which complete their lifecycle within the host crop or other widely available habitats (e.g. grass field margins), most natural enemies use a diverse range of habitats over the course of their lifecycle, and their populations may be bottlenecked by a lack of one or more key resources (Ramsden et al., 2014). In order to increase the size of the natural enemy population at an early stage of pest infestation, targeted resource provision is required within agro-ecosystems throughout the year. Winter habitat, especially undisturbed grassland and herbaceous margins, is important for providing food and/or shelter for overwintering natural enemies, from which they can migrate into arable crops in the spring (Geiger et al., 2009). Adults of many largely carnivorous larval species feed mostly or entirely on floral resources, using floral and extra-floral nectar for sustenance, and pollen for proteins and additional nutrients (Wäckers and van Rijn, 2012), making this an important resource during the spring and summer months. Pest infestations in crops can be relatively short lived, leaving long periods of time when prey may be scarce, as a result natural enemies must migrate between prey in crop and non-crop habitats as the seasons progress. A lack of alternative prey can therefore limit natural enemy populations in the summer and autumn, affecting how many go on to overwinter (Frank, 2010).

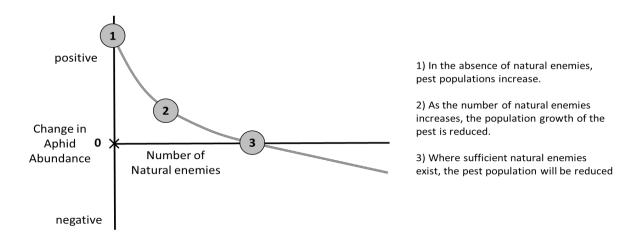


Figure 2. The relationship between pest population growth and abundance of natural enemies in arable crops, adapted from Ramsden *et al.*, 2016.

## **Applying conservation biological control**

Providing targeted additional resources across agro-ecosystems will support a robust and diverse natural enemy population, which in turn will contribute to suppression of pest populations (Olson and Wäckers, 2006). However, there is little guidance on how farmers can incorporate the benefits of landscape diversification into crop management (Pywell *et al.*, 2017; Zhang *et al.*, 2017). The most widespread application of CBC in the UK has been the installation of 'beetle banks' under Environmental Stewardship schemes (Collins *et al.*, 2003). Such schemes are difficult to link to reductions in insecticide applications because their impact is not quantified with respect to the information farmers and agronomists use to decide whether or not to take action. Economic thresholds provide a method for incorporating non-chemical approaches into this decision making process; i.e. the impact of beetle banks can be quantified in terms of their ability to prevent a given pest from exceeding its economic threshold.

## Conclusion

To improve the uptake of IPM in arable crops, it is important to be able to demonstrate the benefits of its adoption to farmers. This could involve showing how the contribution of natural enemies can reduce the number of occasions when the economic threshold is exceeded. However, it will first be important to ensure that thresholds are widely used in arable farming. The widespread occurrence of resistance of a number of pests to inexpensive synthetic pyrethroid insecticides may help to encourage the adoption of economic threshold in making spray decisions. Although alternative chemical control options to pyrethroids are available they are significantly more expensive. This is likely to make farmers question the need to spray because of the potential impact it may have on gross margins. Only when thresholds are adopted on a wide scale will it be possible to start to demonstrate the benefits of IPM and reduce reliance on prophylactic insecticide applications.

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# Ecological Focus Areas and beneficial insect (Coleoptera: Carabidae) conservation for pest control: habitat suitability and farm specific factors

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Abstract: Ecological Focus Areas (EFAs) were introduced as a component of the Common Agricultural Policy (CAP) 'greening' measures. Different options are available depending on those activated by individual Member States, but across Europe as a whole, they range from cover crops and agroforestry, to the protection and enhancement of boundary features. Eligible farms must have a minimum arable area (including fallow land, temporary grassland and crop land) of 15 ha, for which EFAs need to comprise 5% of this area. The overall environmental benefits delivered by a given EFA option will depend on farm specific factors, and will vary between farms and Member States. In order to help account for this variability, the Joint Research Centre (JRC) commissioned the development of a software tool, the EFA calculator, to assist land managers in the selection of EFA elements appropriate to their farm, in order to optimise ecosystem services, biodiversity and farm management. A simple bespoke scoring system underpins the calculator but enables the distillation of complex and data-intensive parameters into a readily interpretable and user friendly format. Although multiple ecosystem services are accounted for by the tool, this paper will focus on pest control, specifically the impact of EFAs on ground beetles (Coleoptera: Carabidae) and their potential role as beneficial insects. Crop cover, frequency of cultivation, field size and the quality and connectedness of field boundaries are key factors taken into account.

Key words: beneficial insect, Carabidae, Ecological Focus Area, ground beetle

## Introduction

Ecological Focus Areas (EFAs) have been introduced as part of the so called 'greening' measures of the Common Agricultural Policy (CAP). Member States can select (activate) the elements that they wish to apply within the area for which they are the competent authority. Eligible farms, those with an arable area of 15 ha or more, select from the EFAs activated in their Member State. They need to account for 5% by area of the total arable land declared, including fallow land, temporary grassland and crop land. It does not include permanent grassland or permanent crops. The introduction of EFAs on the farm has the potential to enhance ecosystem service delivery (Tzilivakis *et al.*, 2016). Several studies have identified carabid beetles (Coleoptera: Carabidae) as beneficial insects able to consume both pests and weed seeds in agricultural areas (Kromp, 1999; Holland & Luff, 2000). The agricultural landscape is essentially a mosaic of disturbed habitat (the cultivated area) interspersed with areas of relative stability (field margins and hedgerows). The cultivated area of the field is subject to frequent disturbance and application of agro-chemicals, both of which may be

detrimental to the survival of carabid populations (Kromp, 1999). This paper reports on the preliminary development of a simple habitat suitability scoring system as applied to EFAs for carabid beetles and the value of EFA elements to carabid beetles and potential pest control.

## Material and method

The EFAs available for implementation and selected for assessment in this paper represent those available in England with additional options (agroforestry, liquorice as a nitrogen fixing crop) selected for illustrative purposes. Some EFA elements, for example land left fallow and nitrogen fixing crops, have a further choice of management available (e.g. species mixture or land cover). The tillage frequency refers to the potential maximum period that a given species or mixture may typically be grown without re-establishment as a function of its biology rather than EFA specifications. Unless otherwise stated, autumn tillage is presumed to be undertaken between August and October, with spring tillage between January and February. Tillage in catch and cover crops is limited to early autumn since both require establishment by 31<sup>st</sup> August in England (Rural Payments Agency, 2016). A second cultivation may occur in catch crops when removed from October onwards during the same year, or the spring of the following year for cover crops. The influence of EFA elements and options on crop microclimate has been included as a function of leaf size and canopy height.

A review of the literature identified 11 species of carabid relatively ubiquitous in cultivated areas (Holland & Luff, 2000; Kromp, 1999; Warner *et al.*, 2008). These 11 species have been selected for further analysis and the derivation of habitat suitability scores associated with individual EFA elements. The life-cycle and behavioural traits considered to be of greatest importance in determining their abundance in arable land are summarised in Table 1.

For each factor in Table 1, the impact of each EFA element and option where applicable, has been given a semi-quantitative score of between 0 and 100, where 0 is a strong negative impact, 100 a strong positive impact. Crop management practices with a potential negative impact include tillage (Holland & Luff, 2000). Where adult carabid activity coincides with the autumn tillage period (August to October) a lower score is allocated to denote a potential risk to abundance. The magnitude of temporal coincidence between carabid activity and the potential tillage period (0 to 3 months) varies between species and is adjusted accordingly. The larvae of spring breeding species or summer / autumn breeders are present during the summer and winter respectively (Thiele, 1977), the latter within the cropped area (Holland and Luff, 2000). Pterostichus madidus is capable of activity within the deeper soil layers, potentially below the impact of cultivation. This species is allocated a positive impact score for this behaviour characteristic, which increases the score allocated to the impact of cultivation overall. A Principle Components Analysis (PCA) of the scores for each carabid species has been undertaken using the Idiographic Analysis with Repertory Grids (Idiogrid<sup>®</sup>) Version 4 (2008) software (Grice, 2002). The PCA calculates the degrees of similarity between species or EFA elements for the purpose of discussing the underlying data used within the EFA calculator tool.

	Ad	Bl	Во	Hr	Lp	Nbr	Nbg	Pc	Pmd	Pml	Tq
Spr adult activity	Apr- Jul	Apr- Aug	-	-	May- Jun	Apr- May	Jan- May	-	-	-	-
Aut adult activity	Sept- Oct	-	May- Sept	May- Sept	Aug- Sept	<sup>1</sup> Sept- Nov	Sept- Dec	Jun- Sept	May- Oct	May- Oct	Apr- Oct
Breeding period	Spr	Spr	Aut	Aut	Spr	Aut	Spr/ Aut	Spr	Sum/ Aut	Sum/ Aut	Sum/ Aut
Larvae present	Jul- Sept	Jul- Sept	Sept- May	Sept- May	Jun- Sept	Nov- Apr	May- Sept	Jul- Sept	Oct- May	Oct- May	Oct- Apr
Activity at depth	no	no	no	Yes	no	no	no	no	yes	yes	yes
Micro- climate	Xr	Xr	Xr	Xr	Xr	Xr	Xr	Xr	Xr	Xr	Sh
Winter habitat	В	В	В	F	В	В	F	B/F	F	F	F

Table 1. Carabid species found in arable land and key life-cycle and behavioural traits.

Ad: Anchomenus dorsalis (Pontoppidan); Bl: Bembidion lampros (Herbst); Bo: Bembidion obtusum (Serville); Hr: Harpalus rufipes (De Geer); Lp: Loricera pilicornis (Fabricius); Nbr: Nebria brevicollis (Fabricius); Nbg: Notiophilus biguttatus (Fabricius); Pc: Poecilus cupreus (Linnaeus); Pmd: Pterostichus madidus (Fabricius); Pml: Pterostichus melanarius (Linnaeus); Tq: Trechus quadristriatus (Schrank); Xr: xerophilic, Sh: shade, B: boundary, F: field, <sup>1</sup>Nbr aestivation period in the cropped area between May and August.

## **Results and discussion**

The similarity in habitat preference, given the life history, behavioural and morphological traits between the 11 carabid species under assessment (Table 1) are given in Figure 1a. The closer that two species are grouped on the PCA graph, the greater the similarity between them. The proximity to the descriptions on the perimeter of the PCA graph represent a closer affinity of that species toward a given characteristic. In Figure 1b the proximity of an EFA element to the carabid species listed on the perimeter of the PCA graph indicates a greater potential suitability of that option to a given species of carabid.

Figure 1a clusters species that utilise the field boundaries as hibernation sites that are influenced by boundary quality and field size, although *P. cupreus* may also hibernate in the field. *Nebria brevicollis* is present in the crop as larvae during the winter and has a summer aestivation period as an adult (Thiele, 1977). A second cluster contains species active as adults in the cropped area later into the season, and in the deeper soil layers (Holland & Luff, 2000). Activity within the crop from January onwards renders adult *N. biguttatus* at greater risk to spring tillage. A key determinant of the impact of an EFA element on potential carabid abundance (Figure 1b) is the frequency and timing of tillage, coupled with crop microclimate. The approved mixtures for nitrogen fixing crops are variable in their potential re-

establishment requirements, ranging from annual (e.g. chickpea or beans), biennial (e.g. sweet clover) to perennial (e.g. birds foot-trefoil). The selection of species requiring less frequent reestablishment are those most likely to benefit carabid populations in agricultural land. The scores derived for each carabid species are then aggregated for individual EFA elements and options, and utilised within the EFA calculator tool to assess the benefit of a given element to ecosystem service provision, in this case beneficial insects and pest control.

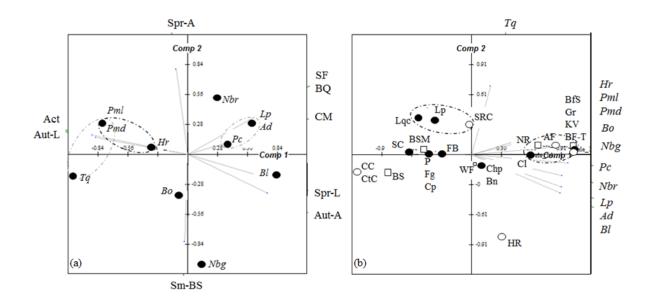


Figure 1. (a) Similarity of habitat preference and vulnerability of adults and larvae to EFA management interventions for 11 carabid species frequent on arable land mapped onto the first two principal components (cumulative variance = 70.63%) of PCA (Act: activity at depth; Aut-A: lower risk autumn tillage – adult; Aut-L: lower risk autumn tillage – larvae; BQ: boundary quality; CM: crop microclimate; SF: small field size; Sm-BS: small body size; Spr-A: lower risk spring tillage – adult; Spr-L: lower risk spring tillage – larvae). (b) Similarity in habitat suitability between selected EFA elements and options for 11 carabid species frequent on arable land (cumulative variance = 90.46%) of PCA ( $\Box$  fallow land; • nitrogen fixing crop;  $\circ$  other; Gr: sown grass; WF: sown wildflower; BSM: sown bird seed mix; NR: natural regeneration; BS: none (bare soil); KV: kidney vetch; ChP: chickpea; Lqc: liquorice; BF-T: birds foot-trefoil; Lp: lupin; SC: sweet clover; Bn: bean; P: pea; Cl: clover; Fg: fenugreek; FB: faba bean; Cp: cowpea; CC: cover crop; CtC: catch crop; BfS: buffer strips; SRC: short rotation coppice; AF: agroforestry; HR: hedgerows).

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# Simultaneous use of winter cress (Brassicaceae) in trap cropping, conservation biological control, and pollinator conservation

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Abstract: Winter cress, Barbarea vulgaris R. Br. (Brassicaceae), has been proposed as trap crops for the diamondback moth, Plutella xylostella L. (Lepidoptera: Plutellidae). In bloom, B. vulgaris could also be used to attract beneficial insects. In this study we tested the attractiveness of flowering B. vulgaris to P. xylostella and to two of its parasitoids, Diadegma insulare Cresson (Hymenoptera: Ichneumonidae) and Diadromus collaris Gravenhorst (Hymenoptera: Ichneumonidae). We also tested whether the presence of flowering B. vulgaris planted in the field border could affect densities of insect pests and beneficial coccinellids in adjacent cauliflower plants. Flowering B. vulgaris did not change the densities of insect pests found in adjacent cauliflower, except for Eurydema ornata L. (Hemiptera: Pentatomidae). Coccinellids were more abundant on flowering B. vulgaris than on cauliflower plants. Chrysomelids were also more abundant on flowering B. vulgaris than on cauliflower plants, while hemipteran and lepidopteran pests were more abundant on cauliflower than on B. vulgaris plants. In plots with flowering B. vulgaris, P. xylostella pupae suffered 1.7 and 4.0 times more parasitism by D. insulare and D. collaris, respectively, than in plots without flowering B. vulgaris. Flowering reduced the attractiveness of B. vulgaris to P. xylostella, making it lose its effectiveness as a trap crop. Flowering B. vulgaris plants were visited by hoverflies, such as Sphaerophoria scripta L. (Diptera: Syrphidae), by pollen-feeding beettles, such as *Psilothrix viridicoerulea* Geoffroy (Coleoptera: Melyridae), and by mining bees of the genus Andrena (Hymenoptera: Andrenidae). Being biennial, B. vulgaris could be used as a trap crop for P. xylostella the first year, and to lower the populations of E. ornata, increase parasitism of *P. xylostella*, and attract pollinators when it flowers the second year.

Key words: Barbarea vulgaris, trap cropping, conservation biological control, pollinators

## Introduction

Trap crops are plant stands that are, per se or via manipulation, deployed to attract, intercept, retain, and/or reduce targeted insects or the pathogens they vector in order to reduce damage to the main crop (Shelton & Badenes-Pérez, 2006). One of the plant species proposed as a trap crop for the diamondback moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae), is *Barbarea vulgaris* R. Br. (Brassicaceae), commonly known as winter cress and yellow rocket (Badenes-Pérez *et al.*, 2004; Badenes-Pérez *et al.*; 2005, Idris & Grafius, 1994; Lu *et al.*, 2004; Shelton & Nault, 2004). Given the choice between *B. vulgaris* and different cruciferous crops, *P. xylostella* prefers to oviposit on *B. vulgaris*, even though its larvae do not survive on it (Badenes-Pérez *et al.*, 2014b; Badenes-Pérez *et al.*, 2004; Idris & Grafius, 1994; Lu *et al.*, 2004; Shelton & Nault, 2004). *Barbarea vulgaris* is also very attractive to the flea beetles *Phyllotreta cruciferae* Goeze and *P. striolata* F. (Coleoptera: Chrysomelidae), suggesting that

this plant could also be used as a trap crop for their management (Root & Tahvanainen, 1969). Although *Phyllotreta* spp. show distinct preferences for host plants (Metspalu *et al.*, 2014; Root & Tahvanainen, 1969), the use of trap cropping to manage *Phyllotreta* spp. has yielded mixed results (Altieri & Gliessman, 1983; Parker *et al.*, 2016). For other economic pests, such as *Eurydema ornata* L. (Hemiptera: Pentatomidae) (Trdan *et al.*, 2006), nothing is known about the effect that a flowering *Barbarea* companion plant would have on their population dynamics on an adjacent cauliflower crop.

Laboratory experiments with *B. vulgaris* have shown that flowering and non-flowering *B. vulgaris* are equally attractive to ovipositing *P. xylostella* (Lu *et al.*, 2004). Flowering *B. vulgaris* has also been shown to be a good nectar source for *Diadegma insulare* Cresson (Hymenoptera: Ichneumonidae), a parasitoid of *P. xylostella*, as well as for bees and hoverflies (Dailey & Scott, 2006; Idris & Grafius, 1997; Idris & Grafius, 1995). Thus, our hypothesis is that flowering *B. vulgaris* could be used as a trap crop for different insect pests found in a cauliflower crop, in conservation biological control, and to attract pollinators.

## Material and methods

#### Trap cropping capability of flowering Barbarea vulgaris

Experiments were conducted in the field in Arganda del Rey (Spain). Cauliflower, cultivar 'Snowball' (Intersemillas, Quart de Poblet, Spain) and G-type *B. vulgaris* var. *arcuata* (collected in Denmark and donated to us by Dr. Niels Agerbirk) were used. A control treatment with a solid planting of cauliflower was compared to a treatment with cauliflower that had flowering *B. vulgaris* planted in two opposite borders. The control plots had 8 rows, while the plots with flowering *B. vulgaris* had two additional rows with flowering *B. vulgaris*. Each row had 20 plants. Rows were separated by 1.0 m with 0.5 m spacing between adjacent plants within rows. Sampling of insects was conducted weekly from the middle of April till the end of June 2011 (Badenes-Pérez *et al.*, 2017). The insects sampled included the coleopterans *Adalia bipunctata* L. (Hemiptera: Pentatomidae), and *Phyllotreta* spp. and *Psylliodes* spp. (Coleoptera: Chrysomelidae); the hemipterans *Aleyrodes proletella* L. (Hemiptera: Aleyrodidae) and *Brevycoryne brassicae* L. (Hemiptera: Aphidiae); and the lepidopterans *Pieris brassicae* L. and *P. rapae* L. (Lepidoptera: Pieridae), and *P. xylostella*.

#### Parasitism of P. xylostella plots with and without flowering B. vulgaris

Pupae of *P. xylostella* were collected in June 2011 to compare parasitism rates in plots with and without flowering *B. vulgaris*. At least 10 pupae were collected from each of the 8 plots where the trap cropping experiment was conducted (Badenes-Pérez *et al.*, 2017).

#### Floral visitors

In May 2011, at the peak of bloom in *B. vulgaris*, floral visitors were recorded by observing sets of 5 plants for 5 minutes on three different sampling dates. Insect visits were recorded between 9:00 and 12:00 h.

## **Results and discussion**

#### Trap cropping capability of flowering Barbarea vulgaris

Except for *E. ornata*, we found no significant differences in insect densities between cauliflower plots with flowering *B. vulgaris* and control cauliflower plots (P > 0.05). *Eurydema ornata* was found in lower densities on the cauliflower plants in the plots with flowering *B. vulgaris* than on the control plots (Wald  $\chi 2 = 6.28$ ; df = 1; P = 0.012). When comparing insect densities on flowering *B. vulgaris* and adjacent cauliflower, *A. proletella*, *B. brassicae*, *E. ornata*, *P. brassicae*, *P. rapae*, and *P. xylostella* were more abundant on cauliflower than on flowering *B. vulgaris* plants. *Adalia bipunctata* and *C. septempunctata*, as well as *Psylliodes* spp. and *Phyllotreta* spp. were significantly more abundant on flowering *B. vulgaris* than on cauliflower (Badenes-Pérez *et al.*, 2017).

#### Parasitism of P. xylostella plots with and without flowering B. vulgaris

We found two species of parasitoids attacking *P. xylostella*: *D. insulare* and *Diadromus collaris* Gravenhorst (Hymenoptera: Ichneumonidae). Parasitism by *D. insulare* was 22.8 and 38.4% for pupae collected in control plots and in plots with flowering *B. vulgaris*, respectively (z = 2.23; P = 0.013). Parasitism by *D. collaris* was 1.8 and 7.2% for *P. xylostella* pupae collected in control plots and in plots with flowering *B. vulgaris*, respectively (z = 1.43; P = 0.076) (Badenes-Pérez *et al.*, 2017).

#### Floral visitors

The two most common floral visitors of *B. vulgaris* flowers were the hoverfly *Sphaerophoria scripta* L. (Diptera: Syrphidae) (0.4 insect visits/plant/min) and the pollen-feeding beettle *Psilothrix viridicoerulea* Geoffroy (Coleoptera: Melyridae) (0.3 insect visits/plant/min). Other floral visitors included the pollen-feeding beetle *Heliotaurus ruficollis* F. (Coleoptera: Tenebrionidae), mining bees of the genus *Andrena* (Hymenoptera: Andrenidae), the hoverfly *Episyrphus balteatus* De Geer (Diptera: Syrphidae), and the wheat stem sawfly *Trachelus tabidus* F. (Hymenoptera: Cephidae). Besides being pollinators as adults, larvae of the hoverflies *E. balteatus* and *S. scripta* are aphidophagous (Bargen *et al.*, 1998; Jauker & Wolters, 2008; Khan *et al.*, 2016).

An increase in vegetational diversity can often reduce insect pest incidence in crops, but there are also reports of no changes and increases in insect pest densities with increased vegetational diversification (Ratnadass et al., 2012; Veres et al., 2013). In our study, presence of flowering B. vulgaris did not significantly alter the abundance of insects on adjacent cauliflower, except in the case of E. ornata. As no E. ornata adults and egg masses were found on B. vulgaris plants, the decrease in E. ornata densities could not be caused by a preference for *B. vulgaris*, but rather by an interference with finding its cauliflower host. We had hypothesized that flowering B. vulgaris could be effective as a trap crop for Phyllotreta spp. and *P. xylostella*, but this was not the case. Reduced attraction to *Phyllotreta* spp. and P. xylostella adults to flowering B. vulgaris could be due to a reduction in glucosinolate and saponin content in the plant with the onset of flowering. In B. vulgaris, glucosinolate and saponin concentrations have been shown to decrease with increasing leaf age (Badenes-Pérez et al., 2014a). We found greater parasitism of P. xylostella in plots with B. vulgaris, yet this did not result in a significant reduction of P. xylostella larvae in those plots. Flowering B. vulgaris plants were visited by hoverflies, pollen-feeding beetles, and mining bees. As B. vulgaris can be used as a trap crop for P. xylostella in a pre-flowering stage on its first year (Badenes-Pérez et al., 2005), being biennial, B. vulgaris could be used as a trap crop for *P. xylostella* the first year, and to reduce infestations of *E. ornata* and attract beneficial insects when it flowers the spring of the following year.

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# Banker plants promote functional biodiversity in cabbage

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Abstract: Natural enemies of the cabbage whitefly *Aleyrodes proletella* do currently not regulate whitefly populations sufficiently. Biological control methods in field crops have often been neglected, although offering promising tools to increase the abundance and diversity of natural enemies. For instance, the release and promotion of natural enemies with banker plants is one way to increase functional biodiversity and biological control services in field crops. This field study investigated the following two banker plant systems against A. proletella: (1) the greenhouse whitefly Trialeurodes vaporariorum on pumpkin and (2) the honeysuckle whitefly A. lonicerae on European columbine. Both systems were inoculated with the parasitoid Encarsia tricolor. We evaluated the effect of the banker plant systems on A. proletella parasitism rates as well as the abundances of adult E. tricolor and predators on neighbouring Brussels sprouts plants. Both, average parasitism rates and adult E. tricolor increased by at least 50% by either banker plant systems. Furthermore, the abundance of hoverfly larvae was 63% higher in the treatment with pumpkin as banker plant compared to the control, whereas 11-12% fewer spiders were found in both treatments with banker plants. In conclusion, especially the banker plant system with T. vaporariorum and E. tricolor on pumpkin promoted functional biodiversity on cabbage plants and showed promising potential as banker plant system in cabbage crops.

**Key words:** *Aleyrodes proletella*, Aleyrodidae, biological control, cabbage whitefly, *Encarsia tricolor*, natural enemies, predators

## Introduction

The cabbage whitefly *Aleyrodes proletella* (Hemiptera: Aleyrodidae) has developed to a major pest on several *Brassica* crops in Europe (Trdan & Papler, 2002; Nebreda *et al.*, 2005). Especially organic vegetable producers lack successful management strategies against this pest, mainly because the few efficient chemical agents on the market are restricted to conventional production (Richter & Hirthe, 2014). Furthermore, the impact of the dominating whitefly parasitoid *Encarsia tricolor* (Hymenoptera: Aphelinidae) and the abundance of important generalist predators like hoverfly larvae or whitefly specialists like *Clitostethus arcuatus* (Coleoptera: Coccinellidae) is currently not sufficient to downregulate *A. proletella* populations significantly (Bathon & Pietrzik, 1986; Pütz *et al.*, 2000; Laurenz *et al.*, 2016). Alternative or supplementary tools are desired to improve the efficiency of biological control of *A. proletella*. Banker plants for instance have proven to increase natural enemy abundances and biological control services in the greenhouse, but also in the field (Pickett *et al.*, 2004; Huang *et al.*, 2011). This study evaluates the potential of two newly composed banker plant systems with *E. tricolor* as natural enemy to promote the functional biodiversity of *A. proletella* natural enemies.

## Material and methods

#### **Banker plant production**

European columbine (*Aquilegia vulgaris*, 324 plants) and pumpkin plants (*Cucurbita maxima* 'Uchiki Kuri', 36 plants) were grown in two separate gauze tents inside a greenhouse. As soon as 1-3 true leaves were fully developed, European columbine and pumpkin plants were evenly infested with the alternative hosts, i.e. 3,000 adult females of the honeysuckle whitefly *A. lonicerae* or the greenhouse whitefly *Trialeurodes vaporariorum*, respectively (Goolsby & Ciomperlik, 1999; Pickett *et al.*, 2004). The natural enemies, i.e. 375 adult *E. tricolor* females per plant species, were introduced two weeks after infestation with the alternative hosts and allowed to deposit eggs for 11 days. Before transplanting them to the experimental plots, all banker plants were kept for five days under outdoor conditions for hardening.

#### Experimental design

Each experimental plot consisted of two Brussels sprouts fields (each 4 m x 2.4 m). The area between these two fields (2.4 m x 2 m) carried the three treatments. It was either covered with black mulch film for the entire experimental period (control treatment) or planted with one of the two banker plant systems, *A. lonicerae* and *E. tricolor* on European columbine (AEC) (54 plants per plot) and *T. vaporariorum* and *E. tricolor* on pumpkin (TEP) (six plants per plot). Plots were 14 m to 17 m apart and allocated in a randomized block design with six replicates. Grass was sown between and around the plots and was kept short by regular mowing.

Eight cabbage plants per plot were evaluated bi-weekly from  $6^{th}$  July to  $12^{th}$  October 2015. The numbers of parasitized and unparasitized whitefly puparia (last nymphal stage) were counted per plant to calculate parasitism rates. In addition, adult *E. tricolor* as well as the type and number of predators were determined for each plant.

#### **Statistics**

Data were processed in IBM SPSS Statistics 24. All determined parameters were analysed in a multivariate general linear model (GLM) with repeated measures over time followed by a post hoc test after Tukey for multiple comparison between treatments, if applicable.

#### **Results and discussion**

#### **Parasitoids**

Average parasitism rates of *A. proletella* nymphs were 51% and 53% higher in the AEC and TEP treatment, respectively, than in the control (both p < 0.001) (Figure 1). Additionally, the average numbers of adult *E. tricolor* per cabbage plant increased by 53% and 50% in the AEC and TEP treatment compared to the control (both p < 0.001). There was no difference in parasitism rates or adult *E. tricolor* between the two banker plant treatments (p = 0.113 and p = 0.989). An increase of whitefly parasitism (*Bemisia argentifolii*) on an outdoor crop (cantaloupe) by banker plants inoculated with parasitoids (*Eretmocerus* spp.) was also reported by Pickett *et al.*, 2004. They even observed three to five times higher parasitism rates by banker plants compared to a control.

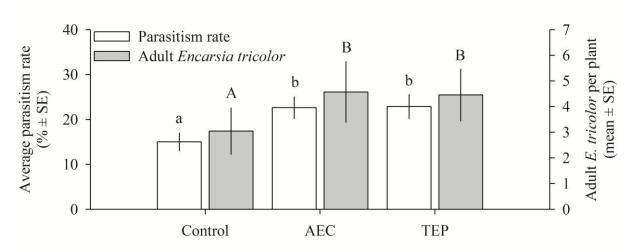


Figure 1. Parasitism rates and numbers of adult *E. tricolor* per cabbage plant and day as average values over the entire experimental period (AEC = banker plant system with *A. lonicerae/ E. tricolor* on European columbine, TEP = banker plant system with *T. vaporariorum/ E. tricolor* on pumpkin, different letters indicate significant differences between treatments, GLM with repeated measures over time and Tukey test,  $\alpha = 0.05$ ).

#### **Predators**

On the one hand, 63% more hoverfly larvae were found on cabbage in the TEP treatment compared to the control (p = 0.033) (Table 1). The AEC treatment did not differ from the control or the TEP treatments in respect to the number of hoverfly larvae (p = 0.751 and p = 0.162, respectively). On the other hand, spiders were 11% and 12% less abundant on cabbage in the AEC and TEP treatment than in the control (p = 0.018 and p = 0.042). Other predatory groups were not affected by the banker plants and only found in relatively low numbers.

Table 1. Average predator numbers per cabbage plant and day over the entire experimental period (mean  $\pm$  SE) (AEC = *A. lonicerae*/*E. tricolor* on European columbine, TEP = *T. vaporariorum*/*E. tricolor* on pumpkin, different letters indicate significant differences between treatments, GLM with repeated measures over time and Tukey test,  $\alpha = 0.05$ ).

		Treatment			
Natural enemies		Control	Banker plant system		
		Control	AEC	TEP	
Spiders		$4.7\pm0.7^{a}$	$4.2\pm0.5^{b}$	$4.1 \pm 0.6^{b}$	
Hoverfly larvae		$0.27\pm0.05^{a}$	$0.32\pm0.05^{ab}$	$0.44\pm0.08^{\text{b}}$	
Predatory bugs		$0.13\pm0.04^{a}$	$0.21\pm0.05^{a}$	$0.13\pm0.03^{a}$	
Gall midge larvae		$0.11\pm0.09^{a}$	$0.06\pm0.05^{a}$	$0.11\pm0.07^{a}$	
Predatory flies		$0.05\pm0.02^{a}$	$0.03\pm0.01^{a}$	$0.01\pm0.01^{a}$	
Lacewing larv	vae	$0.04\pm0.02^{a}$	$0.04\pm0.01^{a}$	$0.04\pm0.02^{a}$	
Ladybeetle	adults	$0.02\pm0.01^{a}$	$0.02\pm0.01^{a}$	$0.06\pm0.01^{a}$	
	larvae	$0.01\pm0.01^{a}$	$0.02\pm0.01^{a}$	$0.01\pm0.00^{a}$	

## Conclusion

The TEP system promoted the functional biodiversity of *A. proletella* natural enemies by specifically increasing the abundance of *E. tricolor* and hoverfly larvae populations on cabbage. However, data on the effects on *A. proletella* populations or other pests, economic damage and cabbage yield as well as information on the effective distance of banker plants are still required for a final evaluation of the tested banker plant systems. Further improvements like the optimization and standardization of banker plants, combinations with other plant protection measures and the conduction of large scale farm trials and feasibility studies are needed before bringing banker plants to the market. Nevertheless, presented results are a promising first step to make sustainable management of field pests more effective.

#### Acknowledgements

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# Impact of agro-ecological infrastructures on the control of insect pests in apple orchards

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Abstract: The rosy apple aphid, Dysaphis plantaginea, and the codling moth, Cydia pomonella, are major insect pests in apple orchards causing high economic losses. A conservation biological control approach using plant species that provide resources for natural enemies may improve natural regulation and reduce insecticide use. In this study, we compared the influence of wildflower strips, grass strips and spontaneous vegetation on the biological control of aphids and moths by their natural enemies (parasitoids, hoverflies, and ladybirds) in three untreated apple orchards in 2014 and 2015. The sown wildflower strip mixture was successful in increasing floral resource provisioning in both years. Among the major natural enemy groups observed in the margin strips only hoverflies responded positively to the increase of floral resources. Within the orchards, the number of aphid colonies per tree was higher close to the margins, but the number of codling moth larvae smaller. The number of natural enemies observed in aphid colonies was largely driven by aphid number. None of the natural enemy groups observed in D. plantaginae colonies responded to the adjacent strip treatments. This dynamic response may explain the reduction in aphid numbers close to wildflower strips without finding higher overall natural enemy numbers. Similarly, codling moth parasitism was higher close to the margins. These results suggest a positive influence of floral resource provisioning by wildflower strips on aphid predation and codling moth parasitism but they also demonstrate that effect size may be small if species-rich spontaneous vegetation does already occur in margins or inside orchards.

**Key words:** conservation biological control, predation, parasitism, codling moth, rosy apple aphid, floral resources

## Introduction

Flowering strips significantly increase the local abundance and biodiversity of flying predators and parasitoids in the agroecosystem (Marshall & Moonen, 2002). However, only few studies have demonstrated their role in pest control and their impact on the dynamic of arthropod populations and on trophic interaction within the crop (Lavandero *et al.*, 2006; Géneau *et al.*, 2012).

The objective of this work was to test the effect of biodiversity in flowering strips on the biological control of two major insect pests in apple orchard, the codling moth (CM) and the rosy apple aphid (RAA).

## Material and methods

#### Orchard experimental design

The experiment was carried out in 2014 and 2015 in three untreated apple orchards (480 to  $1200 \text{ m}^2$ ) at INRA Montfavet, France (43°54'51.57 "N, 4°52'56.15" E). Three strip treatments (30 to 70 m<sup>2</sup>) were established along the longer edge of each orchard (Figure 1): (a) a wildflower strip mixture comprising 30 vascular plant species (WS) optimized for high and long lasting productions of floral resources, (b) a grass strip mixture (GS) including *Lolium perenne* (28%) and *Festuca arundinacea* (72%), (c) an unsown strip allowing the natural development of spontaneous vegetation (SV). Grass and wildflower strips were sown in orchard edge in March 2014.

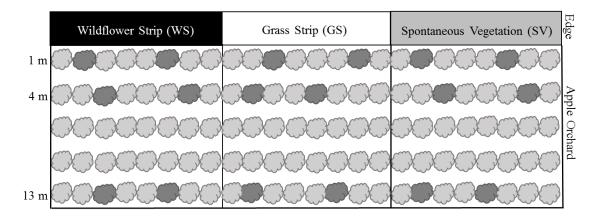


Figure 1. Schema of the experimental design. Shaded symbols indicate the selected trees to monitor apple pests and their enemies in each apple orchard.

#### Entomological observations

The arthropod communities involved in RAA and CM control were monitored at 18 or 27 apple trees per orchard (Figure 1). These observation trees were selected at three distances from the margin strips (1 m, 4 m and 13 m). The same trees were selected in 2014 and 2015. The number of RAA colonies per apple tree was counted at the end of the RAA growing period (19/05/14 and 03/06/15). The occurrence of predators and aphid mummies were recorded in up to six RAA colonies per tree (Table 1; Dib *et al.*, 2016).

	2014	2015
RAA colonies	41.9 [31.8-56.0]	22.3 [11.3-27.4]
Hoverflies	0.1 [0.0-0.4]	0.7 [0.4-0.9]
Ladybirds	0.6 [0.5-0.9]	0.5 [0.2-0.7]
Earwigs	0.6 [0.5-0.9]	0.2 [0.0-0.8]
Parasitoids	0.7 [0.5-1.0]	0.6 [0.3-0.7]

Table 1. Mean number of RAA colonies per apple tree and mean proportions of RAA with an observed predator (hoverflies, ladybirds and earwigs) or a mummy (parasitoids) in 2014 and in 2015. Minimum and maximum values per orchard are indicated in brackets.

CM larvae were collected weekly from May to October in cardboard band traps wrapped around the tree trunks. The number of CM larvae and adult emergences were recorded to infer the parasitism of diapausing and non diapausing larvae (Table 2; Maalouly *et al.*, 2015).

Table 2. Mean number of CM larvae per apple tree, parasitism rates and proportion of parasitoids (*Ascogaster quadridentata, Pristomerus vulnerator* and *Perilampus tristis*) in 2014. A total of 2673 CM larvae were collected (including 927 diapausing). The diapausing samples correspond to the emergences that occurred in 2015. Minimum and maximum values per orchard are indicated in brackets.

	Non-diapausing	Diapausing
CM larvae	27.7 [18.4-33.4]	14.7 [6.9-19.8]
Parasitism rate	0.27 [0.15-0.36]	0.32 [0.15-0.38]
A. quadridentata	0.59 [0.33-0.63]	0.43 [0.21-0.69]
P. vulnerator	0.13 [0.10-0.26]	0.17 [0.00-0.23]
P. tristis	0.27 [0.26-0.40]	0.38 [0.31-0.51]

Furthermore, the percentages of flowering entomophilous plants and the number of predators and parasitoids in each strip treatment were recorded in May of each year to shed light on their respective impact on the control of the apple pests.

#### Statistical analysis

The occurrence of predators in RAA colonies and CM parasitism were analyzed using generalized linear models assuming a binomial distribution (*glmer* function, lme4 package, R version 3.2.4). The strip treatment, the distance to the orchard margin and their interaction were factors tested. All models also included the observation date as an additional fixed factor to account for temporal variation and a random 'orchard' effect to account for a block factor.

## **Results and discussion**

The percentages of flowering entomophilous plants in the margin strips were significantly higher in WS than in SV and in GS both year (Figure 2). We mainly observed hoverflies and ladybirds in the margin strips. The number the hoverflies was higher in WS than SV and GS in 2015 (Figure 2). However, we did not detect significant differences in the number of ladybirds and hymenopteran parasitoids among the margin strips.

The abundances of RAA colonies and CM larvae per apple tree were not homogenously distributed within the orchards. RAA infestations were significantly higher at the closest distances from the margin, but infestations were lower close to WS than close to GS and SV strips. Inversely, the number of CM larvae were significantly lower at the lowest distances from margin strips.

The occurrence of predators and parasitoids in RAA colonies and codling moth parasitism within the orchards were not significantly different among the classes of distances to the margin and among the margin strip treatments. However, the arthropod communities involved in RAA and CM control significantly changed within and between years (Tables 1 & 2).

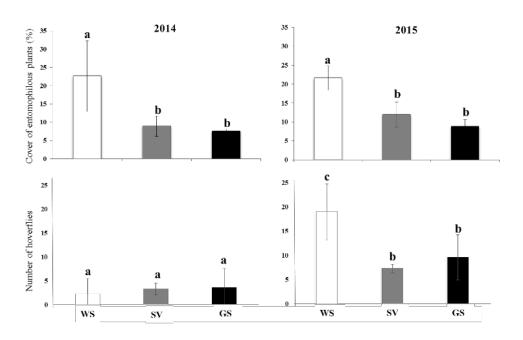


Figure 2. Percentage of entomophilous plants in flower (upper panels) and number of overflies (lower panels) in WS (white), SV (grey) and GS (black) margin strips in May 2014 (left panels) and 2015 (right panels).

These results suggest that provisioning of additional flower resources in orchard margins had a limited impact on the abundance of pest natural enemies both within the orchards and their edges if floral resources are already present in the environment. Selection of entomophilous plants according to temporal natural enemy's requirement could additionally enhance RAA and CM controls in the apple orchards.

## Acknowledgements

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# Functional agrobiodiversity – a novel approach to optimize pest control in fruit production

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**Abstract:** Fruit growers suffer great economic losses each year due to pest damage. The demand for organic produce is increasing along with the interest from growers to develop sustainable and more resilient production systems and over 20% of the apple production in Denmark is now organic. Available pest management options are limited and prevention is important for resilience. In the project PROTECFRUIT we test the use of functional agrobiodiversity, by promoting the abundance and diversity of natural enemies in ecological infrastructures using perennial, wild flower strips in organic orchards. Rosy apple aphid, *Dysaphis plantaginea*, infestation and damage, and predator abundance and diversity were assessed in organic orchards with flower strips these parameters were also assessed as a function of distance to flower strip. The methodology includes visual observations, beating samples and sentinel prey to estimate predation activity. Field trials were conducted in 2016 and will be repeated again in 2017. Preliminary results show that aphid infestation and fruit damage w ere less in orchards with flower strips than in control orchards.

Key words: apple, Dysaphis plantaginea, flower strip, natural enemies, organic agriculture

## Introduction

Organic apple production has now exceeded 20 percent of the total area of apple production in Denmark. The increasing demand for organic produce and the interest from growers to develop more sustainable and resilient production systems has lead to this increase in organic production. The lack of pest control options has great economic consequences for apple growers each year and less than a fifth of the apples are sold as class A. It is hypothesised that functional biodiversity can promote natural control of pests and contribute to reduce crop losses in organic orchards.

## Material and methods

#### Sampling sites and f lower strips

Field trials were conducted in five organic apple orchards with a flower strip and in three organic apple orchards with no flower strip, serving as control orchards. Three orchards have the flower strip replacing a row of apples, and two orchards have the flower strip in the edge of the field. All assessment methods described below were conducted in the first and third row from the flower strip (2 m and 10 m from the flower strip). The flower strips were established

in 2015, replacing existing strips. The seed mixture was selected based on plant characteristics such as value for natural enemies, flowering time, plant size and tolerance to mulching. The mixture consists of 40 species of perennial native plants: 8 grass species and 32 flowering herbaceous plant species.

### Infestation level and damage of D. plantaginea

Infestation of *D. plantaginea* was assessed by visual observation, four times during the growing season, either as aphid presence in flower clusters or as visual symptoms on long shoots after aphid migration. Time of observation was based on the growth stages of apple trees (BBCH scale) and conducted at pre-flowering (59), at end of flowering (69), at June drop (73), and immediately before first harvest (89). Damage assessments were conducted twice during the season; immediately after June drop and before harvest. Fruit damage by *D. plantaginea* was assessed in ten trees per row, in ten randomly selected apples per tree.

#### Predation activity, predator abundance and diversity

Predators were visually observed four times during the season, at the same dates and on the same flower clusters or shoots as the observations of *D. plantaginea*. Beating samples were conducted three times during the season in order to assess predator abundance and diversity, at end of flowering, at June drop, and before harvest. Three beating samples were conducted per row. Each sample consisted of three beatings per branch on 33 trees. Only one branch per tree was beaten, so in case of shorter rows, fewer samples were possible. Predation activity in the orchards was assessed by sentinel prey. Eggs of *Ephestia kuehniella* were glued onto cards of 2 x 3 cm, using egg white. The predation cards were attached to the underside of apple leaves in the orchard, collected after 24 h and the numbers of eggs eaten were assessed under a stereomicroscope.

### **Results and discussion**

The overall aphid infestation was low in 2016. Preliminary data show that the infestation was highest in the control orchard with about 6% infested clusters/shoots by June drop, whereas in orchards with flower strips the infestation was less than half of that. Likewise, the proportion of apples with damage from aphids was highest in the control orchards (6%), while the proportion of damaged fruits in orchards with flower strips was less than half of that. Figure 1 shows that the proportion of flower clusters/shoots with aphid natural enemies present increased from around 5% during pre- and post-flowering to about 20% later in the season, and was comparable between the control orchards and orchards with flower strips. In the orchards with flower strips the highest proportion was found in the tree row adjacent to the flower strip. Predation activity assessed on sentinel prey peaked by June drop and was highest in the first tree row (2 m from flower strip, almost 100% eggs predated) and less in the third row (10 m from flower strip, 90% eggs predated) and in the control orchard (ca. 80% eggs predated).

Though infestation was low, losses (smaller, misshapen apples) were above what is accepted in conventional production, especially in the control orchards, i.e. the results from the first year points to the contribution of flower strips to reduce the losses by increasing the functional biodiversity. This is corroborated by the higher density and activity of natural enemies near the flower strip. Results are also in agreement with previous findings on the effect of flower strips on codling moths and their natural enemies in apple (Sigsgaard, 2014), indicating that functional biodiversity can increase orchard robustness towards more pest

species. The observed decrease in natural enemy activity with distance from flower strip suggest that a greater effectiveness of functional agrobiodiversity may be achieved with higher plant coverage and proximity of flower strips, as are currently being tested in the project EcoOrchard (Sigsgaard, 2016).

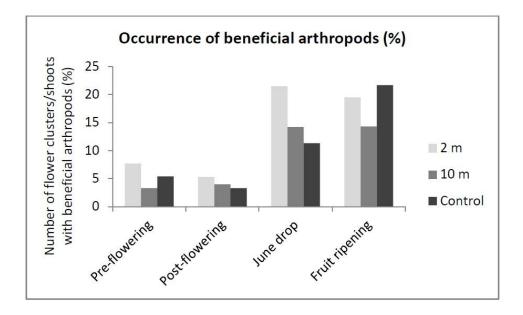


Figure 1. Number of flower clusters/shoots with beneficial arthropods in five organic orchards with flower strips assessed in tree row 2 m and 10 m from flower strip and in three organic orchards without flower strips in 2016.

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# Landscape scale and local crop protection intensity affect the abundance of the codling moth and its predation and parasitism in apple orchards

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**Abstract:** One important feature that may explain variation in pest abundance and pest biocontrol in crops is the land-use intensity in the landscapes. We report results from three studies in which we tried to address this question by assessing whether amount of seminatural habitat and/or pesticide use at both local and landscape scales affected the abundance of codling moth, its parasitism and predation of sentinel eggs in commercial apple orchards of south-eastern France. Our results indicate that in this landscape there is indeed a significant effect of the pesticide use intensity at both the local and the landscape scales on these variables.

Key words: conservation biological control, apple orchard, pesticides, semi-natural habitats

# Introduction

There is increasing recognition that landscape management could contribute to sustainable pest control. However, while on average studies indicate a positive relation between the proportion of semi-natural habitat over the landscape and the abundance and diversity of natural enemies in fields, results about pest abundance are more equivocal (Chaplin-Kramer *et al.*, 2011; Veres *et al.*, 2013). Further, results are variable among studies. One landscape feature that may explain the diversity of results is the land-use intensity in the landscapes.

In the present studies we addressed this question by assessing whether amounts of seminatural habitat and/or pesticide use at both local and landscape scales affected the abundance of codling moth (*Cydia pomonella*, Lepidoptera), its parasitism and predation of sentinel eggs in commercial apple orchards of south-eastern France.

# Material and methods

### Study orchards

The study area is an approximately 80 km<sup>2</sup> pomefruit production area situated in the low Durance valley in South-Eastern France. Apples are mainly grown in conventional orchards (~ 90-85% of orchards), the remaining coming from organic orchards (~ 2-5% of the orchards increasing with time). Because of low fruit prices, some orchards are abandoned (~ 10% of the orchards). Studied orchards had an average area of ~ 0.8 ha. Apple orchards are managed very intensively. In the studied conventional orchards there was an average of approximately 32 treatments, among which approximately 9 insecticides targeted the codling moth. In

organic orchards, the granulosis virus was the main treatment targeting the codling moth. Treatments were frequently complemented by mating disruption against the codling moth (approximately 2/3 orchards).

The landscape is further characterized by the presence of a dense network of windbreak hedgerows that protect the orchards against the prevailing northern winds (Ricci *et al.*, 2009).

#### Sampling

To estimate codling moth abundance and parasitism, we trapped overwintering larvae in autumn from 2006 to 2010 (Table 1) in 10 cm wide cardboard bands wrapped around tree trunks ( $36 \pm 13$  traps/orchard). Larvae were kept in individual vials in an outdoor shelter until emergence.

Predation of codling moth eggs was assessed in 11 commercial orchards (5 organic orchards, 6 conventional). We exposed fresh eggs on thirty cards regularly spaced in each orchard, ten on a border row and the remaining 20 on two rows within the orchard. There were approximately 10 eggs per card. Each card was fixed on the lower side of a leaf at the outside of the canopy at a height of 1.7 m. After three days of exposure, the cards were removed and the predation rate was assessed. Cards were installed twice, in June and August.

Year	# orchards with codling moth	# orchards with parasitoids	# diapausing codling moth larvae	# adult individuals (# parasitoids)
2006	46	10	4853	2815 (112)
2007	45	7	3133	2753 (89)
2008	40	20	3239	2687 (80)
2009	33	13	4786	3990 (181)
2010	38	12	7595	5124 (85)

Table 1: Numbers of orchards, of orchards with parasitism and numbers of codling moths and parasitoids sampled per year.

### Landscape and local characteristics of orchards

Pesticide use in sampled orchards was assessed by surveys to farmers. We also surveyed farmers and technical advisers to map all conventional and organic orchards in the area. Pesticide use intensities in the studied orchards were summarized by frequency treatment indices (TFI). TFIs are calculated as the mean number of treatments, weighted for each product by the ratio of (1) the treated area over the orchard area and (2) the dose used over the recommended dose. We also noted the presence of a ditch or of a spontaneous hedgerow.

The landscape was described in 50 to 250 m wide buffers around studied orchards. All pomefruit orchards and hedgerows were digitalized manually with ArcView (V 9.1, ESRI) from aerial photographs (BD ORTHO, IGN, 2004 – pixel size: 0.5 m) and yearly field surveys. We calculated for each buffer the length of hedgerows and the proportions of area covered by wood and abandoned or conventional pomefruit orchards.

#### Statistical analyses

Statistical analyses were performed using R 3.2.3. We performed generalized linear models assuming a Poisson distribution for count data and a binomial distribution for proportions. All models included a random 'orchard' effect to account for the nested structure of the data.

Model selection based on AIC values or multi-model inferences were used to assess significant variables. Model residuals were inspected visually (package *arms* for binomial data).

Independent variables included in models are reported in Table 2.

# Results

### Sampling

We collected between 3133 and 7595 codling moth larvae per year. Of these on average 73.4% emerged as adult codling moths, 2.5% as parasitoids and the others died (Table 1).

Predation of sentinel eggs occurred in all orchards and was higher in August  $(48.4 \pm 5.1\%)$  than in June  $(12.4 \pm 3.8\%)$ .

#### Factors affecting codling moth predation and parasitism

There was a strong effect of the pesticide use intensity at both the local and the landscape scales on the abundance of the codling moth (Ricci *et al.*, 2009), its parasitism (Maalouly *et al.*, 2013) and sentinel eggs predation (Monteiro *et al.*, 2013) (Table 2). The proportion of conventional orchards in the landscape surrounding the sampled orchard had a significant effect on all the dependent variables. Codling moth larvae abundance, egg predation and larvae parasitism decreased when the proportion of conventional orchards in the landscape increased (Figure 1).

Codling moth larvae abundance in sampled conventional orchards also marginally decreased with the amount of windbreak hedgerows in the landscape (Table 2).

Table 2: Factors affecting *C. pomonella* larvae abundance and parasitism, and sentinel egg predation. \*:  $p \le 0.05$ ; \*\*\*: p < 0.001; ns: p > 0.05; blank cells: not tested.

			local		landscape				
	Date	Organic or conv.	TFI insecticide	Spontan. hedge	Ditches	Conv.	Aband. orchards	Woods	Hedges
C. pomonella	ns 2006-2010	***				*** 250m			
C. pomonella (conv. orchards)	* 2006-2010					*** 250m			*250m
Predation	*** June/August		ns			*** 50 m			
Parasitism		*	ns	ns	ns	* 250 m	ns	ns	ns

### Discussion

Our results, compiled from different studies carried out in the "Low Durance valley", highlight that it was important in this landscape to take into account the management of orchards over the landscape to understand codling moth abundance, predation and parasitism. One methodological issue in these studies is that, overall, organic orchards are grouped over

the landscape so that local pesticide intensity and landscape intensity are partially correlated. However, landscape level pesticide intensity was significant even when only conventional orchards were considered, and even when the local pesticide intensity was included in the statistical model.

Impact of landscape intensity on biodiversity was already reported for biodiversity (e.g. Holzschuh *et al.*, 2008). It might be particularly strong for orchards because they are very intensively managed.

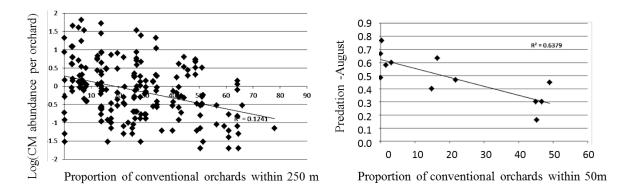


Figure 1. Decrease of codling moth larvae abundance and sentinel eggs predation in August as a function of the proportion of conventional orchards in the landscape.

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# Is higher landscape heterogeneity associated with lower variation of abundances of pests and natural enemies?

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Abstract: While the influence of landscape heterogeneity on responses of both crop pest and natural enemy populations have been extensively studied, impacts on the variation of these responses are not. In this study, we aimed to determine how landscape heterogeneity affects the variation of densities of the soybean aphid and of the guild of natural enemies, and whether temporal variations are observed. We hypothesized that 1) landscape heterogeneity is associated with lower variation of densities of pests and natural enemies; and 2) the effect is stronger during the period of highest aphid population level. Respectively 29 and 31 soybean fields were weekly sampled in 2011 and 2012 in Quebec (Canada). For each field, a coefficient of variation was calculated regarding the density of the soybean aphid (Aphis glycines), of the entire guild of natural enemies, and of the different sub-guilds (predators, parasitoids, entomopathogens) during 1) the population peak of the soybean aphid (August 15-20), and 2) two weeks before (August 1-6). Landscape heterogeneity indices were calculated at a scale of 1.5 km radius around the fields: Crop richness, Crop diversity, Margin density, Landscape patchiness, and Proportion of woodlands. Overall, when considering aphids, predators, pathogens or the entire guild, results are in accordance with our 1<sup>st</sup> hypothesis: less variation in densities observed in heterogeneous landscapes. Finally, in accordance with our 2<sup>nd</sup> hypothesis, landscape heterogeneity has a stronger effect on the variation of natural enemy densities (but not on aphid density) during the population peak of the soybean aphid.

Key words: Spatial context, woodlands, field margins, coefficient of variation, spatial variability

# Introduction

In agroecosystems, landscape heterogeneity influences both pest and natural enemy populations. Fahrig *et al.* (2011) discriminates between functional landscape heterogeneity (based on resource dependencies of species), structural landscape heterogeneity (based on physical characteristics), compositional heterogeneity (e.g., diversity of crop and/or non-crop habitats) and configurational heterogeneity (e.g., landscape patchiness, habitat fragmentation). However, most studies evaluated landscape complexity using the proportions of non-crop or semi-natural habitats in the landscape, where complex landscapes include high proportion of non-crop or semi-natural habitats. Many studies revealed negative effects of landscape heterogeneity on pest density and positive ones on natural enemy density and biocontrol

(Rusch *et al.*, 2016). Especially, landscape complexity showed positive effect on natural enemies in most studies, but less pronounced effects on pest control (Bianchi *et al.*, 2006).

Most of these population studies focused on abundance data instead of variation of abundance data (e.g., coefficient of variation). However, analysing the variance of data can provide relevant information. First, the coefficient of variation can reflect the variability of a population across habitats (spatial), with specialized species showing a higher coefficient of variation of density between habitat types than generalist species (Grez *et al.*, 2013). Second, the coefficient of variation can reflect the variability/stability of a population over time (temporal), with temporal variation of the coefficient of variation observed within a year (Gagic *et al.*, 2014) or throughout the years (Spitzer and Jaroš, 2009). Only few studies have evaluated the effect of the spatial context on the variability of populations. For instance, higher temporal variation of aphid abundance was found in low-intensity managed fields within complex landscapes than in high-intensity managed fields within simple landscapes (Gagic *et al.*, 2014). However, high variation of pest density can also be found between patches of habitat within a same landscape (Bianchi *et al.*, 2010).

In this study, we aimed to evaluate the effect of landscape heterogeneity on the spatial variability of insect populations. We formulated the hypothesis that 1) heterogeneous landscapes are associated with lower variation of densities of pests and natural enemies; and 2) the effect is stronger during the period of highest aphid population density.

### Material and methods

### Field samplings

In 2011 and 2012, respectively 29 and 31 soybean fields were sampled in the Montérégie area of Quebec (Canada). Samplings of soybean aphids (*Aphis glycines*) and natural enemies were performed according to the provincial phytosanitary surveillance network protocol, which includes the weekly observation of 30 soybean plants per field (6 stations, 5 plants each), from the beginning of July until the end of August (see Maisonhaute *et al.*, 2017 for more details). Once a week, the density of aphids and natural enemies (predators, parasitoids, entomopathogenic fungi) were recorded on each plant. The population peak of soybean aphid mainly occurred in mid-August in 2011 and in late August in 2012.

For each field, the within-field coefficient of variation (CV = standard deviation/mean) was calculated for 1) the soybean aphid density, 2) predator density, 3) parasitoid density, 4) entomopathogen density, and 5) the total density of natural enemies (predators, parasitoids, and entomopathogens). The CV was calculated for two periods: the aphid population peak (Peak populations, 2011: week of August 15<sup>th</sup>, 2012: week of August 20<sup>th</sup>), and two weeks before (Early populations, 2011: week of August 1<sup>st</sup>, 2012: week of August 6<sup>th</sup>). Aphid data were log-transformed in 2012 [log<sub>10</sub>(CV)].

#### Landscape and statistical analyses

Landscape heterogeneity was evaluated by analysing the effect of five variables at scale 1.5 km radius around fields: Crop richness (number of different crops), Crop diversity (Simpson index), Landscape patchiness (number of patches of crop and non-crop habitat), Field margin density (mean perimeter-to-area ratio of all fields), and Proportion of woodlands (%).

Statistical analyses were performed using the software R. For both aphid and natural enemy models, a forward selection procedure was used ("forward.sel" function of R package "packfor", 999 permutations, threshold alpha = 0.05) to select variables showing significant effect on the coefficient of variation. Regressions were then carried out with the selected variables, with calculated of adjusted  $R^2 (R^2_a)$  and p-value.

### **Results and discussion**

### Variation of aphid density

In Peak populations, the variation of aphid density was negatively influenced by the field margin density in 2011, while no effect of landscape heterogeneity was found in 2012 (Table 1). In Early populations, the variation of aphid density was not influenced by landscape heterogeneity in 2011, while it was negatively influenced by the field margin density in 2012 (Table 1). These results are in accordance with our  $1^{st}$  hypothesis that a higher landscape heterogeneity is associated with lower variation of densities of pests. Similarly, previous studies showed that more field margins in the landscape increased pest biological control (Östman *et al.*, 2001; Holland *et al.*, 2016). According to contrasting results, it is impossible to conclude for our  $2^{nd}$  hypothesis that the effect of landscape heterogeneity was stronger during the period of highest aphid density.

Table 1. Effect of landscape heterogeneity on the within-field variation of soybean aphid and natural enemy (NE) densities during the soybean aphid population peak (Peak) and two weeks before (Early). CV: coefficient of variation. NS: non-significant. NA: not applicable.

Year	Period	Group	CV (mean ± SE)	Variables (effect)	$R^2_{a}$	р
2011	Early	Soybean aphid	$0.924 \pm 0.113$	NS	-	-
		All NE indexes	-	NS	-	-
	Peak	Soybean aphid	$0.566 \pm 0.036$	Field margin (-)	0.134	0.029
		Predators	$1.535 \pm 0.110$	NS	-	-
		Parasitoids	$2.302 \pm 0.432$	Patchiness (+) and Diversity (-)	0.224	0.014
		Entomopathogens	$1.610 \pm 0.249$	Field margin (-)	0.173	0.014
		Total NE	$1.428 \pm 0.095$	NS	-	-
2012	Early	Soybean aphid	$0.748 \pm 0.058$	Field margin (-)	0.178	0.010
		All NE indexes	-	NS	-	-
	Peak	Soybean aphid	$0.597 \pm 0.030$	NS	-	-
		Predators	$1.887 \pm 0.203$	Woodland (-)	0.149	0.018
		Parasitoids	$2.117 \pm 0.447$	NS	-	-
		Entomopathogens	-	NA	-	-
		Total NE	$1.845 \pm 0.205$	Woodland (-)	0.102	0.045

### Variation of natural enemy (NE) densities

During aphid Peak populations in 2011, the variation of entomopathogen density was negatively influenced by the margin density, while no significant effect was found for predator density and for the total NE density (Table 1). Higher crop diversity provided lower variation of parasitoid densities, while higher landscape patchiness was associated with higher variability in parasitoid densities. In 2012, the variations of predator density and of total NE density were negatively influenced by the proportion of woodlands, while no effect was found on the variation of parasitoid density. In Early populations in both years, no variable significantly influenced the variation of total NE, predator, parasitoid and entomopathogen densities (Table 1). Data on predators, entomopathogens, and total NE support our 1<sup>st</sup> hypothesis that a higher landscape heterogeneity is associated with lower variation of NE densities, while it is not possible to conclude for parasitoids. In addition, the second hypothesis (stronger effect during the period of highest aphid density) is supported for all groups of NE. According to previous results, landscape heterogeneity, especially woody elements, have beneficial effects on NE (Holland *et al.*, 2016) and provided higher functional diversity of NE, which was associated with higher natural control (Maisonhaute *et al.*, 2017).

Overall, our results show that a heterogeneous landscape provides lower within-field population variability for both pest (aphid) and natural enemies, although the associated landscape variables are different. Finally, aphid natural control is clearly linked to landscape heterogeneity, not only according to the impact on aphid densities, but also according to the variability of their densities in the field. Further studies should be conducted to specifically evaluate the complex links between landscape, entomological variation and biocontrol.

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# Effect of landscape diversity on biocontrol of the millet head miner, *Heliocheilus albipunctella* (Lepidoptera: Noctuidae), in Senegal

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Abstract: The objective of this study was to investigate the effect of landscape complexity on biological control of the millet head miner Heliocheilus albipunctella de Joannis (Lepidoptera: Noctuidae) a key millet pest in West Africa. Field observations were carried out in 2013 and 2014, in a 20  $\times$  20 km area around Dangalma village (14°43'42''N, 16°33'98''E), in the Diourbel region located in the main millet-producing basin in Senegal. Biocontrol was evaluated by excluding natural enemies to infested millet heads. Biocontrol Services Index (BSI) was calculated on a set of 45 millet fields separated by a distance of 2 km from each other. Landscape complexity around each field was assessed yearly using five landscape metrics, calculated from two land cover maps derived from Pleiades satellite images. To study the effect of landscape attributes on BSI values, a generalized linear model was performed at 9 different spatial scales (from 0.250 to 2.250 km around fields). The best statistical model was used to identify the environmental key variables enhancing biological control of the millet head miner (MHM). We found that BSI values increased with landscape diversity, measured as Shannon's Diversity Index (SHDI). Moreover, landscapes dominated by millet fields provided less regulation of MHM than landscapes dominated by semi-natural vegetation. Landscape diversity and composition at a 1750 m around sampling sites explained the greatest proportion of the variation of biological control of the millet head miner. This study indicates that semi-natural vegetation, here mainly trees, have to be conserved to enhance biocontrol of *H. albipunctella*.

Key words: Biocontrol Services Index, millet, landscape metrics, West Africa, Pleiades images

# Introduction

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is one of the main cereal crops in Senegal representing about 60% of the total cereal production, with approximately 600.000 tons per year. Adapted to arid and semi-arid climates, millet production is mostly located in the peanut Basin. The millet head miner (MHM), *Heliocheilus albipunctella* de Joannis (Lepidoptera: Noctuidae) is the major millet pest in West Africa, causing important yield losses up to 85% (Youm & Owusu, 1998). Despite years of research, control strategies developed through agricultural practices as deep ploughing or late planting have shown little success (Youm & Gilstrap, 1993). Recent studies on insect ecology have pointed out the importance of landscape-pest interactions as a crucial determinant of biocontrol success (Hunter, 2002). In

Senegal, main natural enemies have been identified (Gahukar, 1968; Nwanze & Harris, 1992), but their natural habitats are still not well known. To better understand the environmental determinants of biocontrol of the MHM populations, we proposed a landscape approach focusing on the role of natural vegetation. We first used a very high spatial resolution of remote sensing data to map and to quantify the key landscape elements around a set of millet fields. A statistical analysis was then performed to identify environmental factors enhancing natural regulation of the MHM.

# Material and methods

### Study area

The survey was conducted within an area of approximately 20 km x 20 km around the village of Dangalma (14°43'42''N, 16°33'98''E), in the Diourbel region north of the Senegalese Peanut Basin. This area is characterized by a semi-arid climate with annual rainfall varying from approximately 200 mm to 600 mm. The rainy season begins in July and ends in October. The landscape is composed of tree-crop agroforestry systems including *Faidherbia albida* (Del.) A. Chev., *Balanites aegyptiaca* (L.) Delile, *Adansonia digitata* L., *Tamarindus indica* L. and *Acacia seyal* Del. The main cereal crop is millet [*Pennisetum glaucum* (L.) R. Br.], while peanuts (*Arachis hypogaea* L.) are the major leguminous crop.

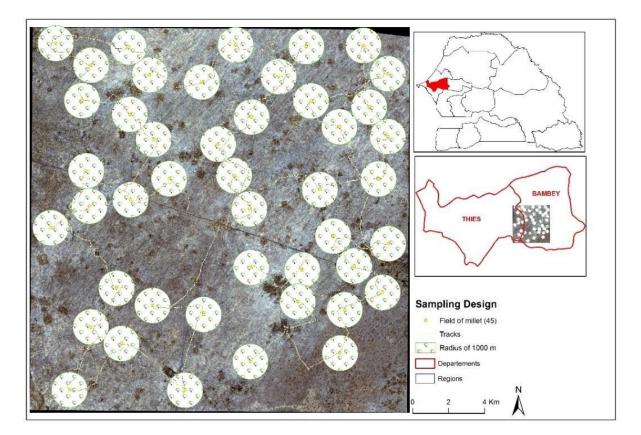


Figure 1. Bambey study area and the set of 45 millet field sites surveyed

### **Biocontrol service**

Biocontrol was evaluated during 2013 and 2014 growing seasons by experimentally excluding access of natural enemies to infested millet heads using mesh bags. Biocontrol Services Index (BSI) proposed by Woltz *et al.* (2012) was calculated on a set of 45 millet fields separated by a distance of 2 km from each other (Figure 1).

### Landscape metrics

Landscape complexity was quantified from two land cover maps derived from Pleiades satellite acquired at the end of the 2013 and 2014 growing season. Based on landscape field data (540 cultures; 1315 trees), the two land cover map were obtained using an object based analysis classification and their accuracy was measured using the kappa index. Based on the bioecology of millet head miner, five landscape metrics (see below) likely to enhance the MHM biocontrol were derived from the land cover maps. Using *Fragstat* software, these metrics were calculated at 9 different spatial scales, from 0.250 to 2.250 km, around the 45 set of millet fields:

# • The millet patch abundance index (MPAI)

We assume that MHM population will be more abundant in areas dominated by millet crop and that biocontrol will be less efficient. Therefore, a Millet Patch Abundance Index (MPAI) which quantifies the relative abundance of millet crops was calculated for each buffer around the 45 millet fields.

### • The tree density index (TDI)

The natural vegetation can provide food and shelter for natural enemies (Otieno *et al.*, 2011). We hypothesize that natural vegetation and more specifically trees will enhance MHM biocontrol. We calculated a TDI by counting the total number of tree patches in each buffer size.

### • Shannon's Diversity Index (SHDI)

Assuming that natural regulation will be more effective in areas characterized by a high vegetation biodiversity, we calculated the Shannon's diversity index. The SHDI is a mathematical measure of species diversity which combines their composition and abundance (Shannon and Weaver, 1949).

### • The Patch Proximity Index (PPI)

Considering that connectivity of insect's habitats will facilitate their mobility and distribution, we calculated a patch proximity index distribution (Gustafson & Parker, 1992). Then we calculated a Tree Patch Proximity Index (TPPI) to test the hypothesis of high biocontrol in areas where trees are connected and a Millet Crop Patch Proximity Index (MPPI) to test the assumption of a low biocontrol level in areas where millet crops are dominant.

### Statistical analysis

To study the effect of landscape attributes on BSI values, a generalized linear model was performed at 9 different spatial scales spacing of 250 m, from 0.250 to 2.250 km around the 45 millet fields. Then, we used the corrected Akaike Information Criterion (AICc) to identify the best statistical model.

### **Results and discussion**

For each scale radius around the 45 millet fields, nine best model are calculated (Table 1). Then among these models and according to AICc values (AICc = -167), results show that the BSI can be explained as a function of three landscape variables calculated for a 1750 m buffer size (Table 2). Tree Density Index (TDI), the Millet Patch Abundance Index (MPAI) and the Shannon's Diversity Index (SHDI) are the most appropriate variables explaining the greatest proportion of the variation of biological control of the millet head miner.

Buffer ring (m)	Model	Parameter (nb)	AICc	Diff AICc
1750	TDI+MPAI+SHDI	3	-14.96	0.00
2000	TDI+MPAI+SHDI	3	-14.06	0.90
2250	TDI	1	-13.87	1.08
500	MPAI+SHDI	2	-13.84	1.12
1500	TDI+MPAI+MPPI+SHDI	4	-13.69	1.26
1250	TDI+MPPI+SHDI	3	-12.42	2.54
1000	MPPI+SHDI	2	-11.46	3.50
250	MPPI+SHDI	2	-10.90	4.06
750	TDI+MPPI+SHDI	3	-10.87	4.09

Table 1. Table of the nine best models according to their buffer size of the BSI in the Bambey area, Senegal. (Models are ordered from best to worst).

Table 2. Parameters of the best model explaining biocontrol of MHN.

_	Estimate	Std. Error	T value	Pr (>/t/)
Intercept	-1.650e-01	2.937e-01	-0.562	0.5764
TDI-1750	8.379e-05	3.252e-05	2.576	0.0126 *
MPAI-1750	-2.793e-04	1.243e-04	-2.248	0.0284 *
SHDI-1750	5.114e-01	2.235e-01	2.289	0.0258 *

BSI values increased with the SHDI and the tree density index (TDI). Conversely, the index decreased in landscapes dominated by millet fields (MPAI). Tree vegetation and landscape diversity may offer favourable habitats for natural enemies of the millet head miner. Furthermore, this suggests that enhancing landscape diversity has the potential to stabilize or increase biocontrol services. To improve landscape management, tree species and their function as habitats for natural enemies of this pest should be taken into account.

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**Session III** 

# **Plants on karst that attract predatory insects**

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**Abstract:** One of the main problems in vineyards and olive orchards on karst is very scarce source of plants useful for conservation biological control which can survive high temperatures and lack of water during the summer months. The aim of this research was to choose plants as potential elements of ecological infrastructure which could play important role in attracting of beneficial insects in particular predators. This research was conducted on the island of Pašman (Zadar County – Croatia) from 2015 to 2016. The following plants were selected; *Arbutus unedo* L., *Dittrichia viscosa* (L.) Greuter, *Dorycnium hirsutum* (L.) Ser, *Helichrysum italicum* (Roth) G. Don, *Salvia officinalis* L., *Spartium junceum* L., *Origanum vulgare* L. Between predator species *Deraeocoris scach* Fabricius, *Deraeocoris ruber* L., *Geocoris erytrocephalus* Lepeletier & Serville, *Scymnus* spp. and *Rhynocoris rubricus* Germar were found.

Key words: karst, plants, predators, insects, ecological infrastructure

# Introduction

Biological control in organic and integrated production is partly based on functional biodiversity a key element of ecological compensation areas or ecological infrastructure. Many authors (Burgio et al., 2004; Burgio et al., 2006; Franin et al., 2016) reported positive effects of ecological infrastructure on attraction and accumulation of beneficial organisms in agriculture. Non-cropped areas provide shelter, food resources, alternative hosts and hibernation sites for beneficial organisms (Zurbrügg and Frank, 2006). According to data of Caballero-López (2016) weeds as elements of ecological infrastructure play a key role in pest suppression providing them food and alternative prey. Although carnivores, some insects require pollen and nectar during their adult stage (Van Rijn, 2012). In olive orchards and vineyards, in particular those founded on ameliorated karst low biodiversity was noticed. Karst is a type of landscape created from the dissolution of solvable rocks. According to Čustović et al. (2014) the most important characteristics of karst areas are lack of surface water, shallow soils and scarce vegetation. Furthermore, other problems in mediterranean region are high temperatures and lack of water during summer months. Such conditions often limitate plant survival, richness and abundance and have indirect effects on beneficial fauna. The main goal of this research was to select appropriate mediterranean plants as hosts for beneficial insects.

## Material and methods

Our research was conducted from 2015 to 2016 on the island of Pašman (Zadar County – Croatia). This island has a Mediterranean climate (type Csa). Samples were taken during the growing season, from the beginning of May till the end of September, during the sunny weather (between 10am and 3pm). For insect collection two methods were used: sweep netting and visual inspection of plants. Shrubs were sampled by mechanical knockdown every fifteen days. Each plant was random selected and insects were taken by branch beating. Other plants like weeds were monitored by visual inspection. Samples were preserved in 70% ethanol until identification. For plant identification Croatian flora (Rogošić, 2011) was used. All material was identified to genus or species level. Insects were identified with help of entomological books. Identification was done in Department of Ecology, Agronomy and Aquaculture (University of Zadar).

### **Results and discussion**

During this research 101 individuals belonging to 2 orders, 4 families, 1 genus and 4 species were found and identified. As we can see in Figure 1, true bugs (Heteroptera) were the most abundant order (82%) presented with 4 species. On the other hand only one genus belonged to Coleoptera (family Coccinellidae).

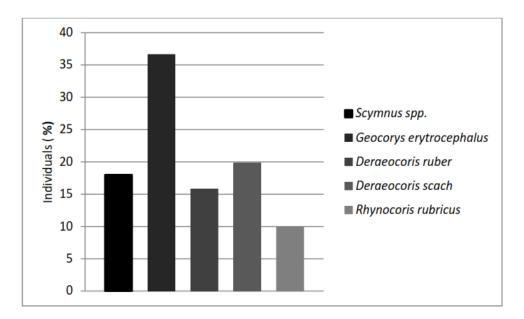


Figure 1. Composition of predatory insects (%)

Scymnus spp. was found only on kitchen sage (Salvia officinalis L.). It is interesting that after blooming period we still found lady bugs in dry calyx of kitchen sage. One reason for that may lay in fact that colour, size and shape of salvia seeds is similar to this coccinellid (biomimicry). Therefore, these parts of plants maybe serve as shelter. True bugs Deraeocoris ruber L. and Deraeocoris scach Fabricius were found on strawberry tree (Arbutus unedo L.). in May and June. Aphids on A. unedo obviously attract Deraeocoris species. Geocoris

erytrocephalus Lepeletier & Serville was noticed on oregano (Origanum vulgare L.) during blooming period (from June to August). *Rhynocoris rubricus* Germar was collected on curry plant [*Helichrysum italicum* (Roth) G. Don] as well as on hairy Canaryflower (*Dorycnium hirsutum* L.). Yellow and white flowers of those plants maybe attract smaller insects which are prey for assassin bugs. A similar situation occured on Spanish broom (*Spartium junceum* L.) which grows as shrub. This plant serve as host for aphids and thanks to their yellow flowers also attract natural enemies (*D. ruber* and *D. scach*). Among all species *G. erytrocephalus* was the most abundant (36.6%). False yellowhead (*Dittrichia viscosa* L. Greuter) hosted only few individuals of *D. scach*. Some of this species have a long flowering period. Preveously mentioned species are important natural enemies in vineyards (Lozzia *et al.*, 2000). Except as habitats for beneficial organisms some of these plants produce essential oils and pollen in huge amounts and therefore attract pollinators. However, we could conclude that plants on karst with suitable root system show good adaptable abilities and can attract natural enemies. Our results highlight the importance of some Mediterranean plants on predatory insects' fauna.

	Coccinellidae	Lygaeidae	Mir	Reduviidae	
Plant species	Scymnus spp.	Geocoris erytrocephalus	Deraeocoris ruber	Deraeocoris scach	Rhynocoris rubricus
Arbutus unedo	-	-	+	+	-
Dittrichia viscosa	-	-	_	+	-
Dorycnium hirsutum	-	+	-	-	+
Helichrysum italicum	-	-	-	-	+
Origanum vulgare	-	+	_	+	-
Salvia officinalis	+	-	_	-	-
Spartium junceum	_	-	+	+	-

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# Structure of the epigeic beetle community in a suburban hilly area in Northern Italy

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**Abstract:** The research has been developed in a hilly area in Northern Italy characterized by different semi-natural habitats. The proximity to crowded and industrialized cities causes it to be vulnerable. We studied the community structure of beetle pedofauna from April to September from 2010 to 2012 in two localities, where five and two sites were selected, representing both woodland and meadow. Two sampling plots, each with five pitfall traps that were replaced fortnightly were located at each site. A total of 7,672 specimens belonging to 14 families were obtained. Staphylinidae appeared to be the richest family covering more than 77% of total captures, followed by Carabidae, Silphidae and Curculionidae. These four families pooled together covered more than 98% of all the specimens collected. The analysis of the variability distribution estimated the components of variations among the considered spatial scales, showed a biodiversity locally high as the plot size well reflected the homogeneity of the structure of the chosen habitats. The limited variability suggested that community composition changed gradually, reflecting a relative homogeneus area without large differences within the habitats of the research areas.

Key words: Carabidae, Staphylinidae, ground beetles, rove beetles, anthropic areas

# Introduction

Woodlands and meadows play an important role to guarantee the equilibrium in natural environment. However, the increase in world population, urbanization and the reduction of natural pristine habitats makes impossible to think of any ecosystem not influenced by humans (Vitousek *et al.*, 1997; Berkes & Folke, 1998). Mosaic landscapes, composed by farmlands, cities, forestry and natural environments can greatly contribute to biodiversity, especially in suburban areas (Duelli, 1997), and natural areas in the proximity of crowded and industrialized cities are considered a great resource (Myers *et al.*, 2000). However, according to McArthur & Wilson (1967), a habitat is able to maintain its stability in relation to its surface and to the distance with a similar one; hence if it is too small or too isolated it could be very fragile.

Recently different studies have been focused on the effect of urbanization gradient on biodiversity (Niemelä *et al.*, 2000; Elek & Lövei, 2007). They revealed that forested urban habitat fragments can retain several endemic species, but the abundance and species richness decrease with the increasing level of anthropogenic disturbance.

The present research has the purpose to compare the community structure of meadow and woodland areas in strict relation to anthropic area, characterized by different resources support, and to give evidence if any differences or gradient in the community structure can be evidenced.

# Material and methods

### Study area

The research has been developed in a hilly area in Northern Italy characterized by different semi natural habitats. However, the proximity to crowded and industrialized cities causes it to be vulnerable. In the last decade a rapid destruction of meadows and natural woodlands (mainly composed by *Castanea sativa*) occurred in the area, due to the presence of plant species as *Robinia pseudoacacia*, and *Sambucus nigra*, characterized by strong competitive abilities that allow them to colonize wood and invade meadows (Kleinbauer *et al.*, 2010; D'Abrosca *et al.*, 2001). We studied the composition of epigeic beetle community from 2010 to 2012. In detail our objective was to partition the spatial variability in the species composition according to a hierarchical, mixed experimental design, based on different spatial scales of sampling.

#### Experimental design

We considered two localities, at a distance of about 10 km from each other, where five and two sites respectively were selected, representing both woodlands (young and mature, W1 to W4) and meadow (natural and dry meadows, M1 to M3). Two sampling plots with five pitfall traps were located at each site to cover the habitat structure variability. Traps were settled at a distance of about 15-20 m to each other and were replaced fortnightly, from April to September. Data of the three-year captures have been pooled together in the statistical analysis (no year factor) in order to manage a more representative list of the experimental area, as in this case we were not interested in evaluating the time fluctuations of the populations.

The differences or similarities in the population sampled have been preliminarily visualized adopting Non-Metric Multidimensional Scaling (NMDS) ordination technique (Clarke & Warwick, 2001). Significant variability among localities, sites and plots in the species composition of pedofauna assemblages was then determined. Null hypothesis, that beetle communities showed the same level of variability was tested by performing a permutational multivariate analysis of variance (Permanova), based on Bray-Curtis similarity coefficient resemble matrix. A full spatial hierarchical mixed design was adopted, with locality and site as fixed factors and plot as a random factor. The homogeneity of the withingroup dispersion among groups was performed by a test of homogeneity dispersion (Permdisp) (Anderson *et al.*, 2008).

### **Results and discussion**

A total of 7,672 specimens belonging to 14 families were obtained. Staphylinidae resulted the most abundant covering more than 77% of total captures, followed by Carabidae, Silphidae and Curculionidae. All these families pooled together covered more than 98% of all the specimens captured. Most of the species detected are generalistic and very common in Italy. This allowed us to relate the community composition to local urban disturbances and to their

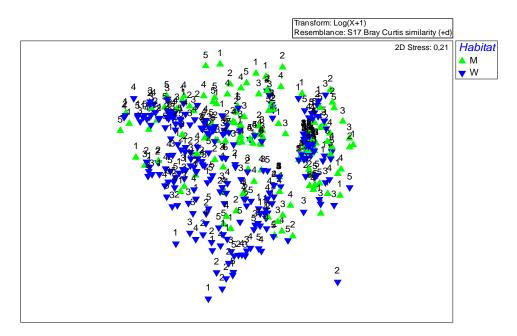
ability to colonize very different environments. We found four dominant species *Atheta trinotata* (Kraatz, 1856) and *Omalium rivulare* (Paykull, 1789) (Coleoptera, Staphylinidae) with 4,527 and 639 specimens, respectively, *Silpha carinata* Herbst 1783 (Coleoptera, Silphidae) with 616 specimens, *Abax parallelepipedus* subsp. *lombardus* Fiori, 1896 (Coleoptera, Carabidae) with 557. The NMDS representation did not produce a marked separation of the samples of the habitat considered (woodland *vs* meadows) (Figure 1A), and many samples of different sites grouped in same area of the graphic, underlying a similar composition of the same habitat [e.g.: mature woodland (W1) *vs* (W4)] (Figure 1B). This is also stated by Permanova analysis, which discriminated samples with statistical significance both for the district, as well as for the locality (site) factors (Table 1). Actually, the analysis of the dispersion of the samples produced significant differences, resulting in a strong influence on the Permanova results for both factor (deviation from centroid for locality factor: F: 5,4167, df1: 6, df2: 420, P (perm) = 0.0001; for district factor: F: 10,145, df1: 1, df2: 425, P (perm) = 0.0035).

Perm	nanova	table of re	sults	Estimated components of variation		
Source	df	Р	Unique perm.	Estimate	Sq. root	
Locality	1	0.0009	9931	320.62	17.906	
Site	5	0.0001	9922	389.11	19.726	
Plot	7	0.522	9864	-1.7791	-1.3338	
Residual	413			1861.2	43.142	

Table 1. Result of Permanova analysis for three different spatial levels.

The main contribution to variation among the samples has given by the residuals, in our case the traps level (Table 1), meaning that each trap captured only a reduced component of the habitat community (only around 43% of the species are shared by two contiguous traps). It could be consistently associated to the reduced movement range of the Staphylinids and Carabids, which well falls in the distance range of the trap positioning. For instance, it is well known that many carabids spend the majority of their life span within a radius of 5 m (Thiele, 2012).

The chosen plots well reflected the homogeneity of the structure of the chosen habitats and their community did not increase at all the variance among the samples. The size of the plots represents an area suited to supply an established ground beetles community in the observed environment. The variance of the community increases again at the site and locality spatial scale in a similar way. The contribution to the variance at the site and locality level suggested that community changes their composition only gradually, reflecting few changes within the habitats structures of the research area. Obviously further and different analyses have to be performed in order to obtain more useful information.



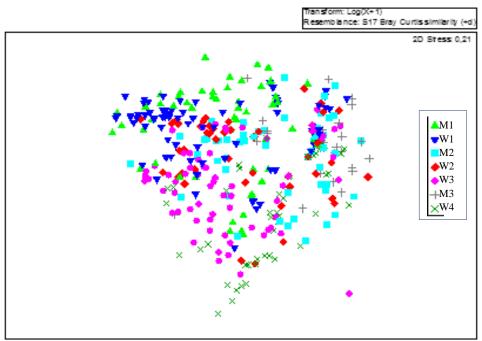


Figure 1 NMDS - 1A. Sites are labelled on the base of the main habitat (M = meadow, W = wood). 1B. Sites are labelled on the base of the sites factor. (The closer the positions of the samples are, the more similar is their community composition.)

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# The community composition of ladybirds (Coccinellidae) occurring in three vineyards in Tuscany (Italy)

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**Abstract:** The ladybird communities were investigated in three differently managed vineyards of the province of Pisa (Tuscany, Italy), two of which were organic. A total of 4,057 specimens were captured in 2012 and 866 in 2013, all belonging to the subfamilies of Scymninae, Coccinellinae, Coccidulinae and Chilocorinae. The ladybird community was analysed on the base of the simple species composition as well as of the trophic guild to whom they belonged. Scymninae and Coccinellinae were the most represented subfamilies. Aphidophagous components of the community were the highest everywhere, followed by the species feeding on coccids, fungi and mites. The analysis based on the community functional biodiversity was better in discriminating different vineyards then the one based on species diversity. The two organic vineyards showed a higher potentiality to host a high number of species and a complex community structure. Coccid predators were predominant in the conventional vineyard.

Key words: biodiversity, bioindicators, Coleoptera, conventional vineyards, organic vineyards

# Introduction

The intensification of agricultural practises results in loss of habitats, landscape simplification and biodiversity reduction. The knowledge of the arthropod community and the ecological structure in an agroecosystem is crucial to a rational use of chemicals. This is paramount in the vineyard, which represents one of the most economically important agroecosystems in the Mediterranean basin. Among beneficials occurring in vineyards, Coleoptera, Coccinellidae (ladybirds) include important and effective predator species. Coccinellids can colonize many habitats, occur in all terrestrial ecosystems and agroecosystems, but show strong sensitivity to the anthropic perturbations (Zahoor et al., 2003). A rich and diversified coccinellid community can play a crucial role in the containment of pests associated with vineyards and, at the same time, can indicate an established and balanced ecological structure. Hence, coccinellids were adopted as useful bioindicators, providing information on the habitats where they occur (Iperti, 1999; Andersen, 1999). More than 90% of all ladybirds are predators (Iperti, 1999), feeding on a wide range of insect orders (Obrycki et al., 2009; Evans, 2009; Hodek and Honek, 2009) as well as on mites (Biddinger et al., 2009). The knowledge of coccinellids occurring in Italian vineyards is scarce, and only in recent years' contributions to this topic were made (Canovai et al., 2014, Burgio et al., 2016). In this paper we investigated coccinellid communities in three differently managed vineyards of Tuscany for two consecutive years.

# Material and methods

### Sites

Surveys were carried out in 2012 and 2013 in three vineyards (labelled as C, Md and O) in the Province of Pisa (Tuscany, Central Italy).

In each vineyard different management systems for controlling the main insect pest, the grape vine moth *Lobesia botrana*, were adopted during the last seven years. In vineyard C a conventional strategy based on organophosphate insecticides was adopted, in Md only pheromone mating disruption with Isonet L dispensers was used and in O no insecticide treatments were made at all. The agronomic characteristics (density per hectare, cultivated varieties, age of the vineyards) were similar among the three vineyards.

### Coccinellids' sampling

Insects have been sampled by two Malaise traps in each vineyard. They were set up approximately in the centre and near to the edge of the agroecosystem, maintaining a minimum distance of 60 meters between both sites. Malaise traps were in place continuously from the half of May to the beginning of October both in 2012 than 2013. Trap containers were changed bi-weekly. Species were classified and attributed to their trophic guild such as predators of aphids (Ap), of coccids (Co), of mites (Mi), of aleyrodids (Al), mycophages (My) and others (O).

### Data analysis

Samples were organised in a similarity matrix on which we applied the ordination technique Non- Metric Multidimensional Scaling (NMDS), obtaining a graphical representation of the rank similarity among all the couples of samples. We tested the null hypothesis that all the samples, expressing the coccinellid communities, had the same level of similarity across different vineyards by performing a permutational analysis of variance (Permanova) (Anderson *et al.*, 2008). We repeated the analysis now grouping the species on the basis of their trophic guild (functional biodiversity). The percentage contribution of each species to the average (dis-)similarity among the samples was tested by performing a Simper analysis (similarities percentage). This analysis lists in decreasing order such contributions, identifying the species that can be considered typical of the habitat of the samples. Non-parametric estimators Chao1 and Chao2 have been used to compare the estimates of species richness among data sets from the different vineyards.

# **Results and discussion**

### Total captures

In the two years, a total of 4,923 coccinellids were captured, 4057 in 2012 and 866 in 2013, including 40 species in 2012 and 38 in 2013. These specimens belong to the subfamilies Scymninae, Coccinellinae, Coccidulinae and Chilocorinae. The most represented species across the two years were the aphidophagous *Hippodamia variegata*, *Propylea quatuordecipunctata*, *Scymnus frontalis*, *S. interruptus* and *Platynaspis luteorubra*. Noteworthy is the trapping of the exotic species *Harmonia axyridis*, recently recorded in Tuscan vineyards (Canovai & Lucchi, 2011). *Nephus bisignatus*, predator of *Planococcus citri* (Kontodimas *et al.*, 2007), *Scymnus flagellisiphonatus* and *Chilocorus bipustulatus* were the most represented species among the coccid predators. *Tytthaspis sedecimpunctata* and *Psyllobora vigintiduopunctata*, were the only mycophagous species, feeding on fungi of the

family Erysiphaceae, *Stethorus pusillus* and *S. gilvifrons*, represented the only predators of spider mites (Tetranychidae). Aphidophagous species of ladybirds were dominant in both years across all vineyards, but particularly in Md (80% in 2102 and 91% in 2013). Mycophagous ladybirds are in the second position in O (14% in 2012 and 27% in 2013). Predators of mites resulted over 5% only in C (5% in 2012 and 6% in 2013).

#### Multivariate analysis

NMDS analysis performed on the species matrix in both years, showed two main group containing samples of the vineyards Md and O together, and the vineyard C at a similarity level of 52% and 36% in 2012 and 2013 respectively. Such separation showed to be statistically significant by permanova analysis in 2012 (p = 0.0001, df = 2, permutations: 9935) and 2013 (p = 0.0001, df = 2, permutations 9921). Similar results emerged by NMDS performed on the matrix, where the species were grouped on the base of the trophic guild, but at a higher level of similarity among the samples (72% in 2012 and 65% in 2013) (Figure, 1). Again, permanova showed the statistical significance of the graphical grouping of NMDS (p = 0.0012, df = 2, permutations: 9945, in 2012 and p = 0.0007, df = 2, permutations: 9954, in 2013).

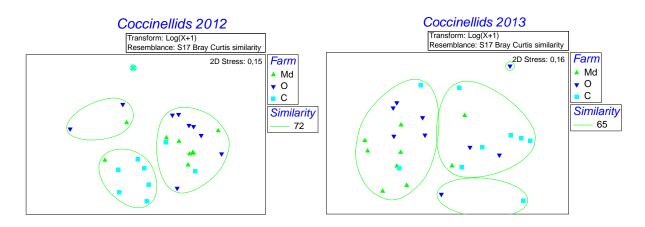


Figure 1. NMDS representation, based on the relative similarity level ranks among all the samples. Species are grouped according their trophic guild. Different symbols represent different vineyards. The more close the symbols are, the more similar is the composition of the samples they represent.

The higher level of similarity with which NMDS has separated the samples from different orchards on the basis of the trophic guilds of the species, suggested that a functional analysis of coccinellid biodiversity produced a better characterization of the sites than the analysis simply based on the species diversity.

The Simper analysis indicates *Scymnus interruptus*, *S. frontalis* and *Propylea quatuordecimpunctata* as species typical of the vineyard Md in both years. *P. quatuordecimpunctata* and *Hippodamia variegata* emerged as typical of C, while in the case of the vineyard O the typical species changed each year, resulting *S. flagellisiphonatus*, *H. variegata*, *Nephus bisignatus* in 2012 and *S. frontalis* and *Platynaspis luteorubra* in 2013.

Non-parametric estimators Chao1 and Chao 2 showed an increasing tendency or a stabilization in vineyards Md and O suggesting that more species (ranging from 5 to 10 in 2012 and 2 to 5 in 2013) could be found in addition to the number of the observed species. In

vineyard C estimators showed a decreasing trend, suggesting that no more species could be found both in 2012 (Figure 2) and 2013.

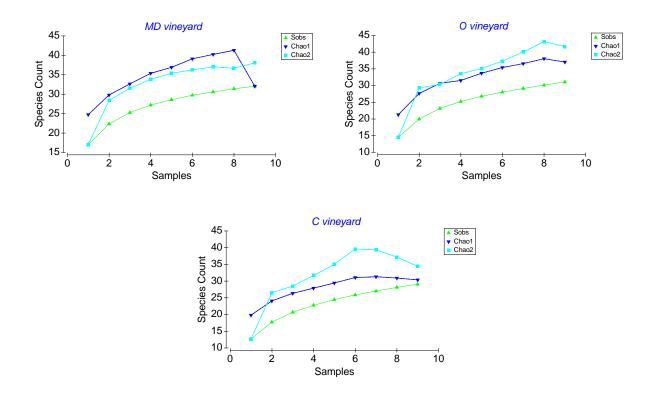


Figure 2. (2012) Non-parametric species richness estimators Chao-1 (triangles, dotted line) and Chao-2 (squares, dotted line) compared to the number of species observed (Sobs, circles, dotted line). Samples are added chronologically.

The two organic vineyards (Md and O) showed a higher potential to host a high number of species and a complex community structure than the conventionally managed vineyard C. The population of the coccid predators was higher in the conventional vineyard, consistent with the hypothesis that treatments with organophosphates stimulate the increase of the phytophagous mite population. The composition of the species changed with time, particularly in O, but that did not influence the relative percentages of the different functional components of the trophic guilds. This observation is consistent with a high resilience of the ecosystem, where lost or reduction of one species can be compensated by the arrival or the population growth of other species that have similar functions.

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# The genus *Paragus* Latreille (Diptera Syrphidae), a useful bioindicator in the vineyards

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**Abstract:** We studied for two consecutive years the community structure of the Syrphid populations in three vineyards, observing the suitability of this family in discriminating the different sites. We also focused our attention on the community structure of the species belonging to the genus *Paragus* Latreille, which is the dominant taxon in the Hoverflies' population. The community structure of the *Paragus* species complex was able to discriminate the different environmental situations, while all the Syrphid species complex did not it. Many species of Syrphids show a high flight ability, which allow them to cover vast areas, rapidly colonizing or recolonizing stressed habitats from surrounding, as well as from long distances and this probably explain the homogeneity among the population occurring in the three different vineyards. As far as we know, the species of the genus *Paragus* are associated to aphid populations living on roots and on wood plants, so that they are more strictly linked to structural vegetal components of the habitat.

Key words: biodiversity, hoverflies, community structure

# Introduction

Vineyard represents one of the most important agro-ecosystem across the Mediterranean basin. It is a perennial crop, which offers very useful conditions for studies regarding the biodiversity of insect communities. The conservation and enhancement of the most important ecosystem services represent the main target of sustainability as they are increasingly being targeted by manifactures and appreciated by wine consumers around the world.

The choice of the biota and sampling methods, the representation of the communities, the discrimination of the sites and of the experimental conditions, and the linking of the changes in communities to some environmental variables, represent the four main stages in biodiversity investigations. The reliability of a bioindicator group in the discrimination of different sites is the keystone step from which descends the possibility to obtain useful information on the environment and on the community structures (Clarke & Warwick, 2001).

Syrphids are one of the most interesting and representative family in the Diptera order. They occur in a very large variety of habitats, occupying all the trophic levels of the ecological pyramid (Stubbs & Falk, 1983; van Veen, 2010). Widespread across all over the zoogeographical regions, about 1,590 species have been identified for the Palaearctic region (Sommaggio, 1999). The use of Syrphids as bioindicators has been investigated by different authors. They occur in a wide variety of habitats, reflecting the very different habits and life

histories of this group (Speight & Castella, 2011; Burgio & Sommaggio, 2002, 2007; Duelli & Obrist, 2003).

We studied the community structure of the Syrphid populations in three different Tuscan vineyards for two consecutive years. We observed the suitability of this group in discriminating the different vineyards, analyzing the populations at the species level.

We also focused our attention on the community structure of the species belonging to the genus *Paragus* Laitrelle, testing their suitability in performing discrimination of different environmental context.

### Material and methods

#### Sites

The studied area is located in the district of Crespina, (Pisa province, Tuscany, Central Italy). Three vineyards, labelled as C (Conventional), MD (Mating disruption) and O (Organic), were chosen within an area of 3 kilometers in diameter to reduce the variability of topographical and weather conditions.

The vineyard sizes were 40 hectares C, 30 hectares MD, and 2 hectares O, respectively.

In each vineyard, different management systems were adopted against the main insect pest, the grapevine moth *Lobesia botrana*, in the last seven years: C adopted a conventional strategy based on organophosphate insecticides, MD only pheromone mating disruption with Isonet L dispensers and O no insecticide treatments at all. Plantations of all the vineyards were more than 9 years old, supporting a well established environmental structure, and their agronomic characteristics (density per hectare, cultivated varieties, age of the vineyards) were similar.

### Syrphids sampling

Two Malaise traps were settled, one in a central position and the other in one side of the vineyard to assemble a more representative local population (Loni & Lucchi, 2014; Sommaggio & Burgio, 2014). Traps captured from the end of May to the first week of October in 2012 and from half May to the first week of October in 2013. Malaise containers were changed every two weeks, so obtaining twenty samples per vineyard.

All Syrphids were separated from all the other insects and stored in 70% ethanol at the Department of Agriculture, Food and Environment of Pisa University. They were classified at the species level according to Van Veen (2010).

### Data analysis

Samples were organized in a raw data matrix reporting an array of rows (Syrphid species) and columns (all the Malaise trap samples).

Data regarding the species of the genus *Paragus* were elaborated selecting only the males, because of the difficulty to attribute the females to the correct species. We preliminarily visualized differences or similarities in Syrphid population adopting the ordination technique Non-Metric Multidimensional Scaling (MDS), then we tested the null hypothesis that Syrphid species communities showed the same level of similarity across different vineyards by performing a permutational multivariate analysis of variance (Permanova), based on Bray-Curtis similarity coefficient resemble matrix.

A P value < 0.05 was adopted. We tested the homogeneity of the within-group dispersion among groups by performing a test of homogeneity dispersion (Permdisp) (Anderson *et al.*, 2008).

# **Results and discussion**

### Total captures

In 2012 and 2013, 4,404 Syrphids distributed on 21 genera and 2,367 distributed on 25 genera were respectively captured. The Site O showed the highest number of specimens in 2012, followed by sites C and Md. In 2013 the vineyard C showed the highest number of specimens, followed by O and Md (Table 1). The most represented genera were *Spaerophoria* (3,561), *Paragus* (685) and *Melanostoma* (334) in 2012, *Paragus* (710), *Spaerophoria* (334), *Cheilosia* (316), *Eupeodes* (302) and *Melanostoma* (284) in 2013. We are going to publish the complete list of the species in a future, more exhaustive, paper. Seven species of *Paragus* were found in 2012 and eight in 2013 (Table 1).

Species (2012)	0	С	MD	Total	Species (2013)	0	С	MD	Total
albifrons	3			3	albifrons	7	5	1	13
bicolor	8	131	8	147	bicolor	7	45	28	80
haemorrous	5		7	12	haemorrous	20	13	26	59
oltenicus					oltenicus		1		1
pecchioli	2		2	4	pecchioli		1		1
quadrifasciatus	2	5	12	19	quadrifasciatus	5	25	7	37
strigatus		25		25	strigatus		4		4
tibialis	16	21	29	66	tibialis	7	35	30	72
Total	36	182	58	276	Total	46	129	92	267

Table 1. Paragus species captured in the three vineyards. Figures refer only to the males.

# Multivariate analysis

# Syrphid community similarity

The NMDS of the 2012 data, based on the "Site" factor, showed a random distribution of the samples, producing no grouping of those of the same site. Such observation apparently does not match with the results of the Permanova analysis, which found significant differences among the "Sites" (p = 0.0004, df 2, permutations 9927). This apparent discrepancy is due to the significant differences underlined by the Permdisp analysis, characterizing their no-homogeneity of the within-group dispersion (F = 4.2517, df 2, p 0.0393). Also in 2013 the NMDS produced a random distribution of the "Site" samples. This was confirmed by of the Permanova analysis, which did not find significant differences.

# Paragus species community similarity

NMDS produced a good discrimination of the sites (Figure 1), consistently with the Permanova results, which provided statistically significant differences both in 2012 (p = 0.0002, df 2, permutation 9951), and in 2013 (p = 0.0055, df 2, perm. 9948). Also the within-group dispersion of the "Site" factor samples resulted homogeneously distributed.

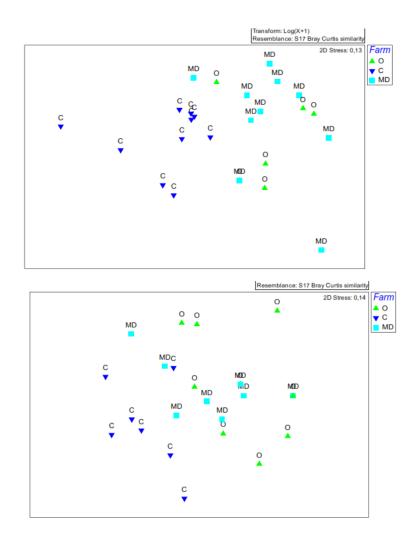


Figure 1. *Paragus* population. NMDS representation, based on the relative similarity level among all the samples. A 2012, B 2013. Different symbols represent different vineyards.

The absence of statistical differences emerged by analyzing the distribution of the Syrphids' community structure are linked to two main factors: the Syrphid biology and the structure of the Tuscan vineyard landscape. Many species of Hoverflies are excellent fliers, able to cover long distances, so that they are often defined as migratory species (Aubert & Goeldlin. 1981). The spatial proximity of the three vineyards probably represented the spatial range of many species of Syrphids which have mixed their population among the vineyards. Moreover, the Tuscan vineyard landscape shows a highly fragmented distribution of the habitats, where each plot is surrounded by many uncultivated areas, rich in wild trees, shrubs and herbs. In this context Syrphids can easily move and perform spill-over in case of phytosanitary treatments or other disturbing actions. On the contrary, the community structure of the Paragus species showed a very interesting distribution, suitable to discriminate among the different sites. Specimens of Paragus are moderately small sized and are not supposed performing migrations. As far as known about their biology, all the *Paragus* species prey on aphids feeding on roots of herbs and on wood species of trees and shrubs. These represent structural components of vegetation of the habitat, allowing the species depending on them, to establish in a more restricted area, where they can found all the resources they need. Though Syrphid community is known to play an important role as bioindicator in large landscapes

(Sommaggio, 1999), our study suggests a potential use of the genus *Paragus* as bioindicator for small spatial scale areas, where it can show a higher sensitiveness to the local environmental factors.

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## Overwintering arthropod assemblages across and within habitats of a Swiss agricultural landscape

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**Abstract:** Semi-natural habitats (SNHs) are known to support high levels of biodiversity within agricultural ecosystems. Management of SNHs in agricultural landscapes should seek to increase arthropod diversity and optimize ecosystem functioning and services, such as natural pest control through high predator abundances. Emergence traps were used to sample overwintering arthropods in Swiss lowlands in order to determine which habitat types are important for promoting high diversity and abundance of overwintering arthropods. Woody habitats proved to be valuable in supporting high predator abundance, herbaceous habitats supported lower predator and higher herbivore abundances, while crops supported the lowest density of overwintering arthropods. Woody habitats should be promoted in agroecosystems in an effort to maximize arthropod diversity and predator abundance. By supporting high abundance of predators they can fulfil multiple objectives. It is therefore recommended that existing hedgerows are conserved and their value enhanced by targeted planting of new hedgerows to connect existing overwintering habitats.

**Key words:** Agroecosystem, arthropod diversity, emergence trap, overwinter, habitat type, habitat trait, Carabidae, Araneae

## Introduction

Agricultural land use accounts for roughly 40% of the earth's land surface, therefore research on sustainable land management of agroecosystems is a major objective for global biodiversity conservation. Increasing homogenization of agricultural landscapes (Purtauf, Dauber & Wolters, 2005) as well as intensification of agricultural practices (Billeter *et al.*, 2007) over the past century has led to a rapid decline of arthropod diversity. Management of agricultural land is often geared to consider only the effects of practices on crop yield, disregarding impacts on biodiversity. However, fostering biodiversity in agricultural landscapes is not only beneficial through a conservation perspective but also through provision of ecosystem services (Bommarco, Kleijn & Potts, 2013). Diverse functional groups of arthropods may provide increased ecosystem services such as pest control in crops. However, due to a reduction in diversity of arthropods, crops have experienced a reduced ability to autonomously regulate pests (Altieri, 1999).

Agroecosystems exist as a mosaic of crops and semi-natural habitats (SNHs), and it is known that predators of pests migrate from SNHs to crops during spring and summer months (Smith *et al.*, 2014) contributing to pest control. Until now research has often focused on food availability and structural traits of habitats and crops during the major activity period of service providing organisms. However, finding shelter during winter and experiencing

optimal conditions for early population development in spring may be crucial parameters for sustaining strong populations sufficient in providing adequate pest control in crops. Understanding which habitat types influence overwintering of functional arthropod groups is a critical component in maximizing the potential of pest control in agricultural landscapes. In order to provide insight into how agricultural land can be more efficiently managed to promote arthropod diversity, as well as high predator densities, we investigated overwintering of ground dwelling arthropods among different types of SNHs in comparison to crop fields.

## Material and methods

## Study area

This study was conducted in the northern part of the central Swiss plateau (cantons Zurich and Aargau, N: 47°36', S: 47°21', W: 8°17', E: 8°38'), a region characterized by a small-scaled mosaic of arable crops, grasslands and forest fragments. Eight sectors, with a gradient of landscape complexity, were established, each encompassing the five investigated habitat types within a one km radius from a randomly selected midpoint. SNHs consisted of sown perennial (3-4 years old) flower strips, meadows, hedgerows associated with a narrow grassy margin, and forests. Four sampling points were established within each SNH, with two situated on the habitat's edge and two within the interior (128 sampling points). Crop fields (winter sown oilseed rape *Brassica napus*) were also sampled but not regarded as SNHs (16 sampling points).

## Sampling method

Emergence traps designed to capture arthropods emerging from the top soil and leaf litter layer, were used to sample overwintering arthropods. Trap frames were 50 x 50 cm and utilized mesh as a semi-permeable barrier, allowing air flow but restricting dispersal of surfacing arthropods. One pitfall trap was inserted into a single corner of each trap for collecting epigeous arthropods. At the top, a plastic jar with a narrow entrance opened upwards into a chamber, also partially filled with propylene glycol solution to catch flying arthropods. Samples were collected every two weeks from March-June, resulting in six subsamples per trap. Taxonomic groups were sorted into trophic groups when possible, however many groups exhibit great variability in diet and therefore were not assigned to a particular trophic group. Specimens were sorted and identified at least to order level, with Araneae, Asilidae, Carabidae, Ichneumonidae, Reduviidae and Staphylinidae grouped as predators, whereas Aphididae, Cicadellidae, *Meligethes* spp. and *Oulema melanopus* were grouped as herbivores.

## Statistical analysis

To test whether overwintering abundance of all arthropods, predators and herbivores differed among and within habitats, generalised linear regression models were used. Differences between edge and interior parts of SNHs (hereafter referred to as "distance") as well as potential interactive effects with habitat type, were detected by including type, distance and their interaction as fixed factors and site ID nested in landscape sector ID as random effects. Models were checked to make sure that assumptions were met.

## Results

#### Overwintering of arthropod groups among and within different habitat types

Habitat type was found to affect overwintering arthropod and predator abundance. Trap distance as well as the interaction of distance and habitat type were found to significantly influence all response variables within SNHs (Table 1).

Table 1. Results for loglikelihood ratio tests for the effects of habitat type and distance on total arthropod, predator, and herbivore abundance.

	SNH Habitat		SNH Distance		SNH Habitat * Distance		All habitats Habitat					
Response group	df	$\chi^2$	р	df	$\chi^2$	р	df	$\chi^2$	р	df	$\chi^2$	p
arthropod abundance	3	5.12	0.16	1	7.59	0.01	3	14.19	< 0.01	4	16.1	< 0.01
predators	3	10.11	0.02	1	9.5	< 0.01	3	15.93	< 0.01	4	10.75	0.03
herbivores	3	7.53	0.06	1	5.22	0.02	3	26.15	< 0.01	4	6.37	0.17

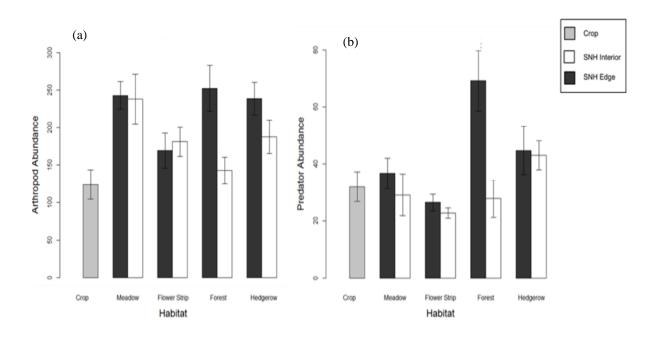


Figure 1. Mean ( $\pm$  1 standard error) (a) number of total arthropod individuals, (b) number of predators in edges (< 2 m from adjacent arable crop) and interior (2-12.5 m from adjacent crops for hedgerows and flower strips; 12.5 m for forests and meadows) of SNHs compared to arable crops (winter oil-seed rape) (N = 8).

Crop fields supported low abundances of total overwintering arthropods compared to SNHs (Figure 1). Similar levels of predators and herbivores were found overwintering in crops compared to SNHs. Flower strips supported the lowest density of overwintering

predators, differing from forests, where the highest density was found. Few differences were found between habitat types compared to analysis of SNHs including distance. Large differences between forest edges and interiors were found regarding all response groups, where edges were always more productive. In addition, forest interiors often had higher numbers than other habitat interiors and edges.

## Discussion

Our results highlight distinct variability in overwintering arthropod assemblages across different habitat types in agroecosystems. In our study, woody habitats (forests and hedgerows) predominantly supported the highest levels of overwintering arthropods followed by herbaceous habitats (meadows and flower strips). Crops supported relatively low densities of overwintering arthropods compared to the SNHs. Intense land management practices are major factors contributing to low arthropod diversity in crops (Tscharntke et al., 2005). Arable fields in our study had been tilled and planted before winter, leaving large patches of bare soil present, reducing insolation properties provided by vegetation. In addition to homogenization of habitat and diminishment of favourable microclimatic conditions required to support healthy arthropod assemblages, also pesticides impose a negative impact on arthropod communities (Tscharntke et al., 2005). The cumulative effects of management on arable fields lead to low abundance of overwintering arthropods in agroecosystems. Presence of SNHs in agroecosystems provides refuges for fauna which cannot survive the intense management regimes of arable fields. Woody habitats in particular supported relatively high predator density, and only low herbivore density, which are important factors in agricultural land management. Woody and protected habitats often contain more biodiversity than habitats with stronger anthropogenic disturbances (Diekötter, Billeter & Crist, 2008). Tall, diverse vegetation with patches of bare ground in hedgerows may provide more opportunities for overwintering arthropods (Pywell et al., 2005). Herbaceous habitats incorporated into arable field systems are often rotated with crops on a multiple year cycle. Changes in botanical structure associated with age such as increases in forb cover, soil organic content matter, as well as height and diversity of vegetation may be a reason for differences in overwintering arthropod abundance between different herbaceous habitats observed within our study (Pywell et al., 2005). Management of SNHs in agricultural landscapes should seek to increase arthropod diversity and optimize ecosystem functioning and services, such as natural pest control through high predator abundances. Woody habitats, especially forest edges, were productive in promoting high densities of overwintering predators. Herbaceous non-crop habitats supported lower predator abundances, and higher herbivore abundances while crops supported the lowest densities of overwintering arthropods. Woody habitats should be promoted in agroecosystems in an effort to maximize arthropod diversity and predator abundance.

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## The abundance of overwintered predatory arthropods in agricultural landscape elements

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**Abstract:** Predatory arthropods are important natural enemies that can play a major role in reducing pest populations. Various types of landscape elements in the agricultural landscape offer hibernation sites for predatory arthropods, and thus contribute to the occurrence of predatory arthropods in agroecosystems, which in turn enhances biological control.

The abundance of different overwintered predatory arthropod groups was measured and compared in different agricultural landscape elements adjacent to arable fields, to determine whether these groups have any overwintering site preferences. Five different landscape elements were studied in five one-km radius landscape circles. Each circle contained all five types of landscape elements. Emerging ground dwelling arthropods were sampled with pitfall traps installed inside emergence traps. Four major predatory arthropod groups were found: ground beetles (Carabidae), rove beetles (Staphylinidae), spiders (Araneae) and harvestmen (Opiliones).

According to the results, landscape element types influenced the abundance of overwintered predatory arthropods. The abundance of predatory arthropods was significantly higher in linear grassy field margins and their abundance was also high in woody linear elements. Significantly fewer arthropods emerged from semi-natural grasslands, woodland edges and cover crop edges, than from grassy field margins.

Key words: ecosystem services, carabids, rove beetles, spiders, harvestmen, pest control

## Introduction

Agricultural intensification has led to the degradation of landscape complexity, a decrease in crop diversity and to an increase in agricultural pesticide use. This has placed substantial pressure on biodiversity, which could greatly affect the delivery of ecosystem services. Predatory arthropods play a major role as ecosystem service providers through natural pest control. Natural enemies of pests are biological control agents, who have the ability to keep pest populations under control through predation, parasitism and pathogenicity (Landis *et al.*, 2000). Land use intensification and development of homogenized landscapes, with fewer types of landscape elements in them have reduced the self-regulating ability – such as natural pest control – of agroecosystems. One way to restore the functionality of ecosystem services is to diversify the landscape by creating semi-natural habitats (SNH) (Tscharntke *et al.*, 2007; Martin *et al.*, 2015), however, the value of different SNHs in pest control is highly variable (Holland *et al.*, 2016). Several authors find that conserving the existing natural habitats and creating semi-natural habitats is the most effective way to restore and increase biodiversity in agricultural landscapes (Duelli & Obrist, 2003; Schweiger *et al.*, 2005). Various types of landscape elements in the agricultural landscape offer hibernation sites for predatory

arthropods, and thus contribute to the occurrence of predatory arthropods in agroecosystems, which in turn enhances biological control (Pfiffner & Luka, 2000).

Other studies on overwintering arthropods have included examination of species diversity and densities of predatory arthropods (Geiger *et al.*, 2009) and the effect of sown or unsown field margin strips on over-wintering arthropod populations (Thomas & Marshall, 1999). In this study, we compared the effect of different types of semi-natural landscape elements on overwintering predatory arthropod populations. Our objective was to identify suitable hibernation sites, adding to the existing understanding of the habitat requirements of predatory arthropods, and to demonstrate to farmers the value of SNHs in their agricultural landscapes.

## Material and methods

#### Study site

We studied five different landscape element types bordering crops in five one km radius landscape circles (25 study sites in total) all located in Tartu County, Estonia.

The studied landscape element types were: woodland edges, woody linear elements (line of trees or shrubs, hedgerows), semi-natural grassland edges, grassy field margins, and cover/intercrop edges. Each landscape circle contained all five types of landscape element, all of which bordered conventionally managed agricultural fields. Landscape circles did not overlap and were at least 200 m apart.

#### Sampling of arthropods

The sampling took place during a four-month period, April-July, in 2014. Pitfall traps (Marc *et al.*, 1999) were used for catching emerging ground dwelling arthropods inside tent-like emergence traps (Schmidt *et al.*, 2008) with the surface area of 0.25 m<sup>2</sup>. In this way the movement between the plot and surrounding environment were eliminated (Thorbek & Bilde, 2004). Pitfall traps (100 ml plastic cup with diameter 66 mm and height 70 mm) were filled with a 1:3 mixture of propylene-glycol and water, and were emptied every two weeks, between April 8 and July 1, six times in total. Trapped arthropods were sorted to order level in the laboratory. The major orders were ground beetles (Carabidae), rove beetles (Staphylinidae), spiders (Araneae) and harvestmen (Opiliones).

#### Statistical methods

Statistical analysis was carried out using Statistica 13 (StatSoft Inc. USA). For the analysis of the impact of different SNHs on the abundance of arthropods, the number of collected arthropods from each collection date was pooled to avoid pseudoreplication. To test the impact of SNH on arthropod abundance generalised linear models were fitted to the pooled pitfall count data assuming a Poisson distribution and log link function and the significance of SNH tested using the Wald test. Differences between the landscape element types of the SNH variable were analysed with the Tukey HSD post-hoc test (p < 0.05).

## Results

## The abundance and taxonomic assemblage of predatory arthropods and effect of landscape element type on overwintering

Carabidae, Staphylinidae, Araneae and Opiliones were the most abundant taxonomic groups of natural enemies with 5817 individuals in total. The most numerous group was carabids with 1913 individuals followed by rove beetles with 1575, spiders 1350 and harvestmen 979 individuals. The average number of predatory arthropods per trap was  $58.17 \pm 4.20$  (233 specimens/m<sup>2</sup>).

Landscape element type had a significant effect on the total abundance of predatory arthropods ( $\chi^2 = 795.24$ ; p < 0.001). There was a higher abundance recorded in grassy field margins in comparison to cover crop edges (p < 0.001), woodland edges (p < 0.001) and grassland edges (p = 0.01). There were more predatory arthropods found in woody linear elements than in cover crop edges (p = 0.019) and woodland edges (p = 0.017). However, there was no significant difference between grassy field margins and woody linear elements (p > 0.05). Also, there were no significant differences between predatory arthropod abundance per pitfall trap in woody linear elements and grassland edges, nor between woodland edges, cover crop edges and grassland edges (p > 0.05).

#### SNHs as overwintering sites for carabids and rove beetles

According to our results, SNH type had an effect on the abundance of ground beetles ( $\chi^2 = 25.36$ ; p < 0.0001). The abundance of carabids was most supported by grassy field margins where the average number of carabids per pitfall trap was significantly higher in comparison to grassland edges (p < 0.001), woodland edges (p = 0.00017) and cover crop edges (p = 0.027). The abundance of carabids in grassland edges, cover crop edges and woodland edges was similar (p > 0.05). Carabid abundance in woody linear elements did not differ from any other landscape element type (p > 0.05; Figure 1).

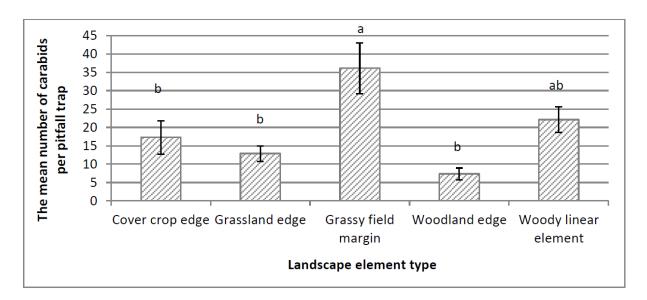


Figure 1. The mean number of carabids per pitfall trap (n = 100) in different landscape elements, Tartu County 2014. Different letters indicates significant differences between landscape elements (Tukey post-hoc: p < 0.05).

Similar to ground beetles, landscape element type affected the abundance of rove beetles ( $\chi^2 = 201.82$ ; p < 0.0001). Significantly more rove beetles were collected from grassy field margins (p < 0.001) and woody linear elements (p = 0.0007) compared to cover crop edges. No differences appeared in comparison to other landscape element types (p > 0.05).

#### SNHs as overwintering sites for spiders and harvestmen

Similarly to ground beetles and rove beetles, the landscape element had a significant impact on the abundance of spiders ( $\chi^2 = 256.17$ ; p < 0.0001). Significantly more spiders emerged from grassy field margins in comparison to cover crop edges (p < 0.001), woodland edges (p < 0.001) and woody linear elements (p < 0.05). No differences were found in comparison of cover crop edges, woodland edges, grassland edges and woody linear elements (p > 0.05).

Landscape element type also affected the number of overwintered harvestmen ( $\chi^2 = 185.66$ ; p < 0.05). There were more harvestmen in woody linear elements (p = 0.014) and grassland edges (p = 0.008) compared to cover crop edges. No differences were found between other landscape element types (p > 0.05).

#### **Discussion and conclusion**

This study demonstrated that the landscape element type influenced the abundance of overwintered predatory arthropods, as there were more arthropods found in grassy field margins and woody linear elements. This result concurs those observed by Thomas and Marshall (1999) who found the highest number of overwintering carabid and staphylinid adults in hedgerows compared to field and sown margin. In their study, the abundance of spiders was also highest in hedgerows but had no significant difference with the number collected from sown margins. Furthermore, Geiger *et al.* (2009) compared grassy field margin, herbaceous field margin under a tree line, forest and ditch edge and found that the abundance of predatory arthropods (Staphylindae, Carabidae, Araneae) were most supported by herbaceous field margin.

Surprisingly, woodland edges were not the most preferred hibernation sites for the studied beneficials, which demonstrates that the common argument by local farmers, that Estonia's relatively high overall forest cover ensures the dispersal of natural enemies in the agricultural landscapes, does not fully apply; even though those ecotones could offer habitat for both open landscape and forest species (Thomas *et al.*, 2002).

Grassland and cover crop edges were not amongst the most preferred landscape element types for overwintering in our study. The lower number of emerging arthropods in cover crop edges could be related to the survival rate of arthropods in the uppermost soil layer (Pfiffner & Luka, 2000). The reason for modest overall numbers from grassland edges with better temperature buffering properties could be related to the spatial aspects: arthropods had more space for dispersal and the area was not limited compared to grassy field margins and woody linear elements. The dispersal of predatory arthropods is also affected by the field margin management practise that in turn can affect the humidity, an important factor for ground dwelling arthropods (Varchola & Dunn, 1999). Cardarelli and Bogliani (2014) studied the effect of cutting frequency on the abundance of different functional groups and found that more specialized ground beetles, such as predatory and short-winged ones, were associated with an absence or low levels of human disturbance. Most grassy field margins located in our study sites had the final cut in the middle of August and thus the vegetation could recover by the time arthropods were looking for their hibernation sites.

In our study, the mean density of overwintered predatory arthropods was high – on average, we trapped 233 specimens per m<sup>2</sup>. The density was the highest in grassy field margins (373 specimens/m<sup>2</sup>), followed by woody linear elements (289 specimens/m<sup>2</sup>), grassland edges (215), cover crop edges (145) and woody edges (142). In comparison, Geiger *et al.* (2009) found 214 predatory arthropods (mostly Staphylinidae, Araneae, Carabidae, Coccinellidae and Dermaptera) per m<sup>2</sup> on average from soil samples, in open agricultural landscape (290 per m<sup>2</sup>) and forested landscape (137 per m<sup>2</sup>). Pfiffner and Luka (2000) found a wide range of overwintering arthropods (Carabidae, Staphylinidae, Coccinellidae, Araneae, Chilopoda, Isopoda) in the semi-natural habitats (134-1163 specimens/m<sup>2</sup>) from soil samples. However, the variations in mentioned studies and our study may be caused by the different sampling methods used and different arthropod groups measured.

The identification of carabids and staphylinids collected in this study are currently being identified to species level. The data on species composition will allow us to have more precise knowledge on distribution of specialists, generalists, predatory and non-predatory, brachypterous and more mobile, macropterous beetles which in turn help the researchers in the dialogue with farmers.

## Acknowledgements

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## Effects of organic farming at different spatial scales on natural enemies of crop pests and pest predation levels

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**Abstract:** Biological control of pests by their natural enemies is considered a key process to reduce pesticide use in modern agricultural systems. Diversity of natural enemies and pest control levels have been shown to be enhanced in organic farming systems and in landscapes with high amount of semi-natural habitats, but the role of organic farming at the landscape scale remains little explored, especially on pest control levels. We investigated the effects of organic farming at the field and landscape scales on the diversity of predatory arthropods and on pest predation levels in 20 pairs of cereal crops located in bocage landscapes with varying proportion of area covered by organic farming in western France. Our results confirmed a strong effect of farming system at the field scale on arthropod diversity but not on pest predation levels. Arthropod diversity and pest predation were little or not influenced by organic farming at the landscape scale, but in some cases, by land-use diversity, grassland area and hedgerow densities. Our results suggest that the promotion of biological control in bocage landscapes might rely on both the local adoption of organic practices and on the maintenance of hedgerow habitats.

**Key words:** farming systems, landscape context, carabid beetles, ladybirds, aphid and weed predation, hedgerows

## Introduction

Enhancing biological control of crop pests by their natural enemies is considered a promising strategy to increase the sustainability of agricultural production systems. Diversity of natural enemies and pest regulation processes were found to be enhanced in fields or farms under organic farming (OF) compared to conventional ones (CF) (Bengtsson *et al.*, 2005). Landscape heterogeneity or complexity, expressed in most cases as the amount of semi-natural habitats (SNH), is another important driver of diversity of natural enemies and pest control levels (Bengtsson *et al.*, 2005; Bianchi *et al.*, 2006).

Recently, it has been suggested that landscape heterogeneity related to farming systems might also impact biodiversity and ecological processes (Vasseur *et al.*, 2013). The few studies addressing this issue have found a higher diversity of natural enemies and pests in landscapes with high proportion of area covered by OF (e.g. Gabriel *et al.*, 2010). However, the effect of both proportion and configuration of OF remains little explored, especially on pest control levels.

The aim of the present study was to investigate the effects of OF at the field and landscape scales on diversity of predatory arthropods (carabid beetles and ladybirds) and prey predation levels (aphids and weeds) in cereal crops. This question was addressed using data from two surveys conducted on a network of organic and conventional farms in northwestern France.

## Material and methods

## Study area and field selection

Our study took place in 'bocage' landscapes in Brittany, northwestern France, characterized by a high density of hedgerows and mixed crop-livestock farming. The main crops are grassland (40%), maize (30%) and wheat (20%). Sampling was conducted in 20 pairs of OF and CF cereal fields, distributed along a landscape gradient characterized by a high variability in the proportion of OF (from 1 to 44%, calculated in 1000m diameter circles centered around studied fields), and by a lower variability in the amount of SNH (hedge density from 1 to 2%) (Puech *et al.*, 2015).

## **Biological sampling**

Arthropods were sampled in each field in 2012 and 2013, at least 10 m away from field edges to avoid edge effects. Carabid beetles were collected using four pitfall traps during six sampling periods (one week each). Adult ladybirds were caught with sweep nets (with 500 sweeps per field) during four sampling sessions. Data were pooled over sampling sessions to work out total species richness and abundances/activity-densities of arthropods in each field. Pest predation levels were measured using two types of sentinel preys – pea aphids (*Acyrthosiphon pisum*) and seeds of field pansy (*Viola arvensis*) – exposed during three sessions in 2016 in each field. For each session, four cards with 5 aphids were exposed during 24 h at the top of cereal plants, and four cards with 10 seeds were exposed during 5 days at the bottom of the same plants. The number of predated and exposed prey items were counted to estimate predation rates of aphids and seeds.

## Landscape description

Aerial ortho-photographs and field surveys were combined to digitize land-uses (grassland, annual crops, woodland, urban areas), hedgerows, and type of farming system (OF vs. CF) for each agricultural field in a 1 km diameter circle centered on each sampled field. Several composition and configuration metrics were calculated at three spatial scales (in 250 m, 500 m and 1000 m diameter circles) to describe the heterogeneity (i) related to SNH and other land-uses, and (ii) related to farming systems (Table 1).

Table 1. Composition and configuration metrics calculated to describe landscape heterogeneity related to SNH and land-uses, and to farming systems.

Type of	Heterogeneity component					
heterogeneity	Composition	Configuration				
SNH and land-uses	Proportion (%) of land-	Hedge density (%),				
SINT and fand-uses	uses, land-use diversity	mean patch size (ha)				
Farming system	Proportion (%) of OF	Edge length between OF & CF (m)				

#### Statistical analysis

Generalized Linear Mixed Models (GLMm) were used to test, in a first step, the effects of local farming type (OF *vs*. CF) and landscape metrics on arthropod diversity and predation rates. Separate models were built for each type of landscape heterogeneity (SNH + land-uses *vs*. farming systems) and each spatial scale (250 m, 500 m, 1000 m). In a second step, analyses were performed separately for OF and CF fields to account for possible interacting or confounding effects between local farming type and landscape heterogeneity. For each analysis, multi-model inference and model averaging were used to build all possible combinations of explanatory variables and to determine the average of models presenting similar relevance ( $\Delta AICc < 2$ ).

## **Results and discussion**

#### Effects of farming systems at the field scale

Our results confirmed a strong positive effect of field scale OF on species richness and abundances or activity-densities of predatory arthropods in cereal fields (Figure 1), probably because of higher resource quality and absence of pesticide use in OF fields. On the contrary, predation rates of aphids and seeds were similar in OF and CF cereal fields, which contrasts with earlier studies reporting higher pest control levels in OF systems (e.g. Winqvist *et al.*, 2011). The lower prey availability for arthropods in CF fields might have led to higher predation of exposed preys in these fields.

#### Effects of landscape heterogeneity related to farming systems and SNH

Overall, farming system heterogeneity had little effect on arthropod diversity and no significant impact on prey predation levels (Table 2). Complex configuration of OF and CF (high edge length) resulted in increased ladybird abundances in OF fields, but reduced ladybird abundances in CF fields (Table 2). These contrasting findings could reflect different processes related to the high attractivity of OF fields for arthropods: colonization of the studied OF fields by surrounding OF crops (source effect) *vs.* dispersal from the studied CF fields to surrounding, more suitable OF fields (dilution processes). The lack of effect of farming heterogeneity on pest predation might be due to similar prey predation levels in studied OF and CF fields.

In addition, heterogeneity related to SNH and other land-uses influenced predatory arthropods and prey predation in specific cases. Ladybird abundances increased in CF fields surrounded by a high hedgerow density (250 m), suggesting a positive role as refuge or overwintering site of hedgerows. By contrast, arthropod abundances and seed predation were reduced in fields located in landscapes with high land-use diversity, which contrasts with existing literature (e.g. Gardiner *et al.*, 2009). Seed predation was further reduced in CF fields surrounded by large area of grassland. In the study area, landscapes with diverse land-uses are characterized by more grassland fields and relatively litte cereal cultivation. This lower availability of crop habitats might explain the lower arthropod abundances and lower seed predation levels.

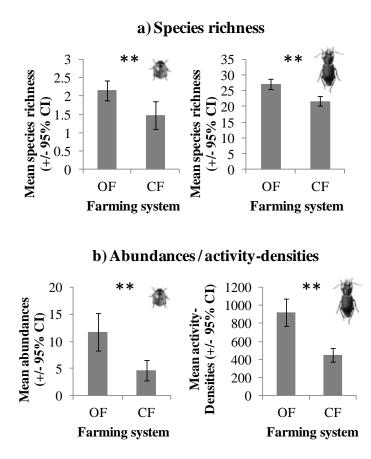


Figure 1. Average abundances/activity-densities ( $\pm$  95% CI) of ladybirds and carabid beetles in OF and CF fields. \*\*: significant effect identified in GLMm.

Table 2. Overview of effects of landscape metrics (in 250 m, 500 m, 1000 m diameter circles), identified in averaged GLMm on arthropod abundances/activity-densities and prey predation rates in OF and CF fields. (+) or (-) indicates positive or negative effect; *n.s.*: no significant effect. LUD: land-use diversity, HD: hedge density, GR: grassland, OF-CF: edge length between OF and CF.

Type of	Fields	Abundances / act	ivity-densities	<b>Predation rates</b>		
heterogeneity	Fleids	Ladybirds	Carabids	Aphids	Seed	
Farming	OF	OF-CF 500m (+)	n.s.	n.s.	<i>n.s.</i>	
systems	CF	OF-CF 1000m (-)	n.s.	n.s.	n.s.	
SNH & land- uses	OF	n.s.	LUD 250m (-)	n.s.	LUD 250m (-)	
	CF	HD 250m (+)			% GR 250m (-)	
		LUD 500m (-)	n.s.	n.s.	% GR 500m (-)	

#### Conclusion and perspectives

Our study suggests that the promotion of predatory arthropods in bocage landscapes will rely on both the local adoption of organic practices by farmers, and the maintenance of hedgerows on farms. However, further investigation is needed to better understand the interactions between farming systems and landscape heterogeneity on pest predation levels. Synchronic analyses of the relationships between arthropod diversity and predation levels measurements are especially needed.

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## Identifying a suitable annual floral mixture and its relative attractiveness to pollinators in Central Spain

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**Abstract:** Wildflower strips in crop margins can contribute to the conservation and enhance of pollinators in the crop area. We have tested during 2 years in Central Spain, a mixture of several wild species of different botanical families well adapted to the area. By assessing floral coverage, precocity and duration of bloom and pollinator visits, we conclude that *Coriandrum sativum, Diplotaxis virgata, Borago officinalis* and *Calendula officinalis* are the best candidates for a seed mixture for the study area.

Key words: field margins, pollinators, annual flower strips, attractiveness

## Introduction

The degradation and fragmentation of natural habitats are among the most significant factors that negatively affect the abundance and diversity of pollinators (Winfree *et al.*, 2009). The decline in plant diversity in agroecosystems prevents a continuous supply of resources to insect pollinators (pollen, nectar and shelter) beyond the duration of the crop blossom, contributing to their scarce presence in these areas. The implementation of wildflowers strips in the crop margins contributes to the conservation of these beneficial arthropods by enlarging the availability of the essential resources needed for their growth and reproduction (Morandin & Kremen, 2013).

The aims of this study were the selection of plant species with a good adaptation to the study area to be included in seed mixtures for wildflower strips and the determination of their relative attractiveness to pollinators.

## Material and methods

A 2-year study was carried out at the experimental farm La Poveda (Arganda del Rey, Madrid, Central Spain; 40°19'N and 3°29'W, elevation 536 m.s.l.). The study area is characterized by continental Mediterranean climate with cold winters, hot summers and scant rainfall ( $\approx$  400 mm per year). The 2-year experiment consisted of a randomized block design of 3 blocks with 2 drip irrigated melon plots each (10 x 10 m<sup>2</sup>) and flower margins of 1 m

wide on both sides. Based on our previous experience (Viñuela *et al.*, 2012) the seed mixture broadcast-sown in winter, was initially composed of *Coriandrum sativum* L. (Apiaceae), *Calendula officinalis* L. (Asteraceae), *Diplotaxis* sp. and *Lobularia maritima* L. (Brassicaceae), *Borago officinalis* L. (Boraginaceae), *Silene vulgaris* Moench. (Caryophyllaceae), *Medicago sativa* L. and *Vicia sativa* L. (Fabaceae), *Salvia verbenaca* L. (Lamiaceae), *Nigella damascena* L. (Ranunculaceae).

The floral coverage and the number of visits of pollinators (3 observation frames 1 x 1 m<sup>2</sup> per margin, 3 minutes) were weekly recorded during the bloom period in the margins. The number of beetles, bees and hoverflies was recorded. For analysis, bees were grouped in long-tongue (L-T) bees (Apidae and Megachilidae) and small ( $\leq 1$  cm) or large (> 1 cm) body size short-tongue (S-T) bees (Halictidae, Andrenidae and Colletidae) (Michener, 2007). A linear mixed-effect model was used to analyze the frequency (visits per min) of each pollinator group in every plant species. The mean number of insect visits per min within the observation area was the dependent variable; the pollinator group in every plant species [*D. virgata* and *S. vulgaris* (year 1), *C. sativum*, *B. officinalis*, *C. officinalis*, *M. sativa* (years 1-2); *L. maritima* (year 2)] was the fixed factor; the block and the observation frames were the random factors; and the sampling dates, the repeated measures factor.

## **Results and discussion**

Year 1 had a longer and cooler winter and was rainier than year 2.

The floral coverage of the species visited by the different pollinator groups, their precocity and duration of the bloom period varied between years (Figure 1). Bloom period of year one was delayed compared to year 2. Floral bloom was staggered. In spring, the species that more contributed to the floral coverage were *C. sativum* and *D. virgata* in year 1 and *C. sativum* and *L. maritima* in year 2 because by error, *D. catholica* was supplied instead of *D. virgata* in the seed mixture and it has a lower height and number of flowers. From June onwards, the highest floral coverage was that of *B. officinalis* and *C. officinalis* the two years. Even though the other species of the flower strip had a very low floral coverage, some received pollinator visits (*M. sativa* and *S. vulgaris* in year 1 and *M. sativa* and *L. maritima* in year 2) contributing to enhance the potential attractiveness of the wildflower strip. However, *V. sativa, N. damascene* and *S. verbenaca* were not visited by any pollinator in year 1 and were removed from the mixture. In year 2, *S. vulgaris* and *D. catholica* were not visited either.

As expected since some plants are more attractive than others (Hogg *et al.*, 2011; Barbir *et al.*, 2015, 2016), the pollinator groups varied significantly according to the year with plant species in the wildflower strip (year1:  $F_{29, 1488.12} = 93.30$ , P < 0.001; year 2:  $F_{24, 2114.30} = 48.63$ , P < 0.001) (Figure 2). In general, the small S-T bee group was the most frequently recorded irrespective of the plant species.

Concerning pollinator visits, some plants behaved differently between years (*C. sativum*, *B. officinalis*). The pollinator visits to *C. sativum* were higher in year 2, when bloom was more precocious. Beetles and hoverflies were very abundant because they usually appear early in the season. Moreover the absence of *D. virgata* this year prevented the possible competition. *Apis mellifera* L. was the species that contributes more to the high number of visits of L-T bees to *B. officinalis* in year 2 and no differences were observed with small S-T bees. The pollinator groups visiting the remaining species were similar in the two years. The principal visitors of *C. officinalis* were the small S-T and the large S-T bees and of *M. sativa* the L-T bees [especially *Megachile rotundata* (F.)] and the small S-T bees.

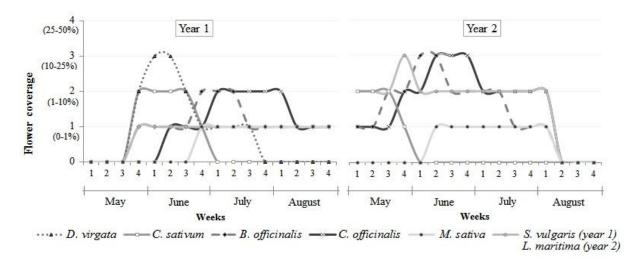


Figure 1. Flower coverage yearly evolution in the plant species visited by pollinators.

In general the species with high floral coverage are the most attractive to pollinators, but *L. maritima* was an exception in our trials. Even though it is recognized as highly attractive to pollinators (Colley & Luna, 2000) it was barely visited, probably because it was not visible for its lower height in respect to other species in the mixture

In conclusion, *C. sativum*, *D. virgata*, *B. officinalis* and *C. officinalis* were the best candidates for a seed mixture for the study area. Out of the rest species studied, only *M. sativa* could complement the potential attractiveness of the flower margin.

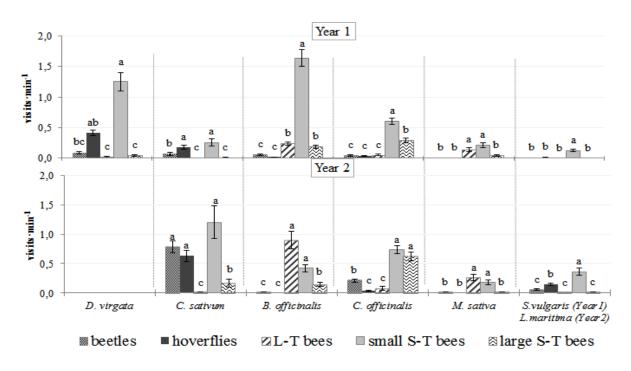


Figure 2. Pollinators mean number of visits per min ( $\pm$  SE) in the different plant species of the wildflower margins. Means with different letters are significantly different (LSD pairwise comparison test; *P* < 0.05).

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## The importance of the qualitative composition of floral margins to the maintenance of rich communities of bees

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**Abstract:** The transformation of natural environments due to the intensification of agriculture has had negative effects on the communities of bees, with negative side-effects on pollination, in both farmed and natural systems. The addition of herbaceous plant edges to crop fields may improve the abundance and diversity of pollinators. The present study emphasises the importance of the right choice of plant species in floral margins to improve the abundance and diversity of bees.

Key words: pollinators, bees, biodiversity, field margins, edges, agriculture

## Introduction

In recent decades, we have witnessed a progressive decline in the diversity and abundance of pollinators (Potts *et al.*, 2010). The intensification of agriculture is one of the main factors responsible for this decline. Bees constitute one of the groups of pollinators that have suffered most from the intensification of agricultural practices, especially because of the destruction of nesting sites, the reduction of pollen and nectar resources and the negative effects of pesticides (Ortiz-Sánchez & Belda, 1994; Klein *et al.*, 2007). This global biodiversity crisis underlines the need for sustainable agriculture and the adoption of appropriate landscape management practices, to restore or preserve biodiversity in agricultural areas. The addition of floral margins to crop fields has generally resulted in benefits to pollinators (Sanchez *et al.*, 2014; Willmer, 2011). This work has the purpose to state the importance of the composition of floral margins to the maintenance of flourishing bee communities.

## Material and methods

The present study was conducted in four localities characterized by intensive agriculture in the Region of Murcia (SE Spain). In each locality, a strip of approximately 100 m<sup>2</sup> in the margin of a vegetable crop was sown in autumn using nine plant species: *Borago officinalis, Brassica oleracea, Chrysanthemum coronarium, Coriandrum sativum, Diplotaxis catholica, Echium vulgare, Salvia verbenaca, Silene vulgaris* and *Vicia sativa*. Enough seeds were used to achieve densities of 5 or 10 plants per square meter for the species having medium-sized or small plants (Pérez-Marcos *et al., 2017*), respectively, considering the germination and

survival rates obtained in laboratory and field trials. Pollinator sampling was carried out fortnightly, from February to July 2014. The abundance of pollinators was estimated counting the number of specimens visiting each plant species within a 2 x 2 m square during 5 minutes. This procedure was performed, placing the square randomly within the floral strip, three times per locality and sampling. Bees were identified to the genus and the rest of the pollinators to the order level. Sampled specimens were collected to confirm the identity of the taxa in the laboratory.

## **Results and discussion**

Bees were the most-abundant group of pollinators (*Apis mellifera*, 49.7%, and wild bees, 41.4%); the rest of the groups (Syrphidae, Diptera, other Hymenoptera, Coleoptera and Lepidoptera) represented 8.9% of the pollinators (Figure 1a).

A total of 21 bee genera were registered during the sampling period. *Apis* (50.27%) was the most-abundant genus, followed by *Andrena* (13.83%), *Eucera* (9.94%), *Lassioglossum* (6.35%) and *Hoplitis* (6.26%). The rest of the 16 genera represented less than 5% of the observations.

There was great variation in the number of bee genera among plant species (Figure 1b). Echium vulgare registered the highest number of genera (19), followed by B. officinalis (17), C. sativum (10), Ch. coronarium (9), Diplotaxis spp. (8, including D. catholica and D. erucoides that grew naturally), B. oleracea (7), S. vulgaris (7), S. verbenaca (4) and V. sativa (4). Besides, there were specific differences in the communities of bees visiting each plant species. A. mellifera represented more than half of the bees observed on B. officinalis, C. sativum, Diplotaxis spp. and V. sativa, whereas wild bees were dominant on the rest of the plant species. Andrena was the most-abundant bee on B. oleracea, Eucera on Ch. coronarium, Lassioglossum on S. vulgaris and Hoplitis on E. vulgare. The differences in the number of bee genera among plant species could be explained by bee preferences and the length of the blossoming period of the plants. For example, E. vulgare had a long blossoming period while S. vulgaris and V. sativa had short ones (Sanchez et al., 2014). Differences were also patent regarding the host-pollen specialisation of the bee genera. For instance, Apis, Andrena, Lasioglossum and Eucera were recorded on six or more plant species (polylectic species), while Hoplitis was observed on only three (oligolectic species). This specialisation could be due to the ability of bee species to exploit plant resources depending, for example, on bee morphology and floral structure. The plants with the highest bee richness had, in general, the greatest bee abundance.

In this work, we show how the richness and abundance of bees varied according to the plant species. *E. vulgare*, *B. officinalis* and *C. sativum* were the plants with the highest richness and abundance scores. The floral margins were also highly used by *Apis mellifera*; thus, the revegetation of edges in crop fields may be beneficial also to apiculture.

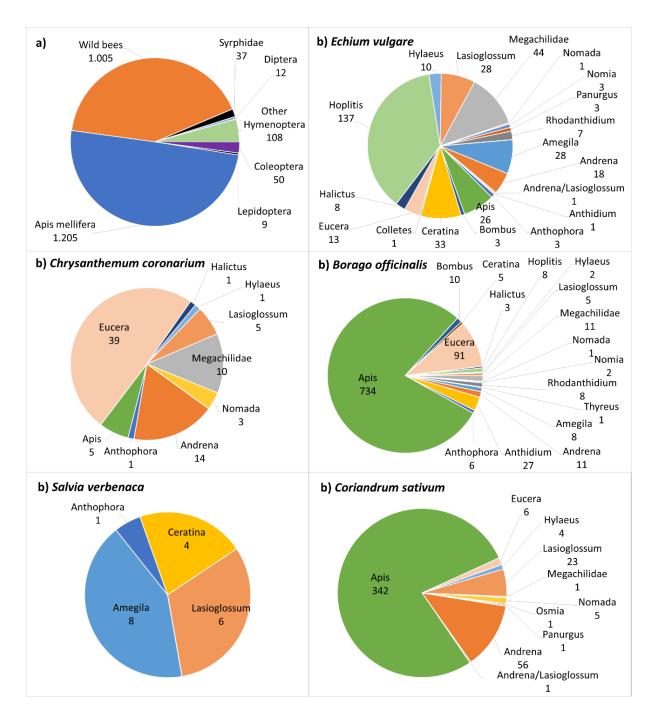


Figure 1. a) Abundance of the different groups of pollinators in the floral margins. b) Abundance of the bee genera registered on different plant species in the floral margins. The numbers indicate the total number of observations for each taxon.

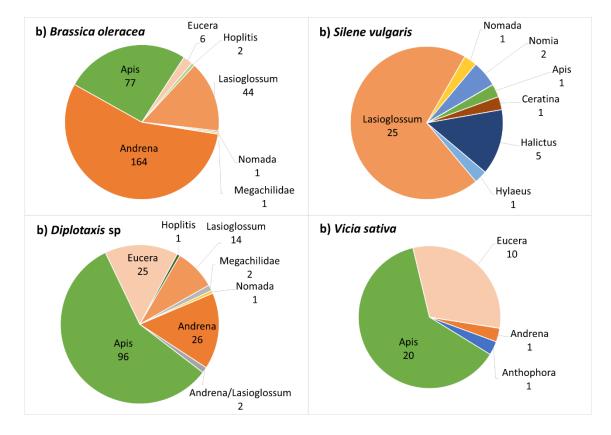


Figure 1. b) Continued from previous page. Abundance of the bee genera registered on different plant species in the floral margins. The numbers indicate the total number of observations for each taxon.

This work outlines the importance of the composition of the floral margin to the maintenance of diverse and flourishing communities of bees. It has also to be taken into account that the use of different plant species with different blossoming periods increases the availability of pollen and nectar through extended periods and, thus, could cover the needs of bees to a greater extent.

#### Acknowledgements

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## **B-Lines**

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**Abstract:** The significant loss of wildflower and grasslands meadows has left many populations of pollinating insect fragmented and isolated and unable to move across the countryside. Buglife's B-Lines are an imaginative and beautiful solution to this problem. The B-Lines are a series of 'insect pathways' running through our countryside and towns, along which we and partners are restoring and creating a series of wildflower-rich habitat stepping stones. They link existing wildlife areas together, creating a network, like a railway, that will weave across the British landscape. This will provide large areas of brand new habitat benefiting bees and butterflies – but also a host of other wildlife. B-Lines have been mapped across large areas of England, Scotland and Wales. Along with conservation partners, land managers, businesses, local authorities and the general public, we are helping to fill the mapped areas with restored and new wildflower-rich areas.

Key words: pollinating insects, wildflower, grassland, meadow, restored, insect pathways, network

**Session IV** 

## Aphid infestations and biocontrol in cereal crops in Scotland: effects of winter cropping across three regions

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**Abstract:** A study was conducted to determine whether i) winter cereals can 'escape' aphid pest infestations due to more advanced crop maturity and ii) whether insect pests on winter cereals can escape biocontrol compared to spring cereals. Summer dynamics of aphids and their natural enemies were investigated in spring and winter cereals at six farm sites across three geographic regions in eastern Scotland. Variation in aphid and natural enemy abundance was assessed in relation to crop type and farm location.

**Key words:** cereal aphids, habitat composition, natural enemies, spring barley, winter barley

## Introduction

Aphids are successful herbivores in crop and non-crop vegetation, forming a significant component of invertebrate biodiversity in many agroecosystems. Aphid infestations in arable crops in spring and summer can lead to crop damage through feeding and disease transmission. Aphid abundance and success are regulated by temporal variation in biotic factors, including plant developmental changes in quality and in the abundance and activity of natural enemies (Karley *et al.*, 2004), and in abiotic factors such as temperature (Brabec *et al.*, 2014). Further, research in recent years has recognized that abundance of aphids and other insect pests can also be regulated by large-scale factors such as habitat composition in the surrounding landscape (Caballero- López *et al.*, 2012).

Cereal crops are a dominant feature of arable-grass production systems in the UK and Europe, and represent approximately 35% of the agricultural area in Scotland. Risk of aphid infestation is potentially high for winter-sown cereals, as plants are available for colonisation by early-arriving aphids in spring, and for spring-sown cereals, as young plants provide a high quality resource for aphids infesting in spring and early summer. Cereal plant quality for aphids declines post-anthesis (Watt, 1979), suggesting that spring and winter cereals grown within the same landscape offer different qualities of resource for summer aphid populations. As a consequence, winter cereal crops might 'escape' heavy aphid infestations compared to spring cereals due to more advanced maturity, which could lead to differences between spring and winter cereal fields in the abundance and activity of natural enemies attacking insect pests. Such an effect might be exacerbated at northern latitudes due to different thermal requirements of aphids and their host plants (Brabec *et al.*, 2014).

Focussing on cereal aphids on spring and winter barley, the aim of this study was to investigate the hypotheses that i) winter cereal crops can escape heavy aphid infestations and ii) that any resulting differences in aphid abundance in spring and winter cereal crops leads to changes in natural enemy pressure and aphid biocontrol when compared to spring cereals. To

take into account any large-scale effects of landscape composition on aphid population dynamics, we compared winter and cereal fields in three geographic regions of eastern Scotland.

## Material and methods

#### Field sites

Six farms in eastern Scotland were visited in June and July 2014: two in Angus (A, B); two in Fife (C, D); and two in Perthshire (E, F). All these farms were located in landscapes dominated by arable cropping, with some woodland and urban areas. At each farm, two fields located < 1 km apart were chosen, one of spring barley and one of winter barley, for field observations. In each field, three adjacent transects, running the length of the field from the field margin and separated by tramlines, were selected at the central part of the field. Seven loci were marked with flexicanes along each transect, the first at a distance of 20 m from the field margin, and each subsequent locus separated by a distance of *ca*. 30 m, giving a total of 21 loci per field.

#### Field observations and sampling

The number of insects (aphids and their natural enemies) was counted on ten cereal stems at each locus on two occasions in 2014, during  $20^{\text{th}} - 25^{\text{th}}$  of June and during  $9^{\text{th}} - 11^{\text{th}}$  of July. Crop development stage and crop and weed ground cover were also assessed. At the latter visit, each set of ten stems was inserted into an open-ended net sleeve; half of these nets, selected at random, were closed at either end using cane ties (after removing any natural enemies present), while the remaining nets were left open-ended. At each locus with a 'closed' net, two yellow sticky traps were attached to the flexicane, one situated above the canopy and one at ground level. After a further five days, the number of insects was counted in each bag (open and closed) and all bags were sealed (top and bottom) around the monitored plant stems (after removing any natural enemies present). These bagged plants were dug out of the field and transferred into 1 l pots. Sticky traps were removed from the flexicanes. The plants were transported to the institute and maintained with regular watering at 20 °C (16:8 h day:night) for 10-12 days, after which the number of mummies and emerged parasitoid wasps was counted. Sticky traps were stored at -20 °C prior to insect identification and counting.

#### Statistical analysis

Analysis of (co)variance was applied to data confirmed to satisfy the assumptions of parametric analysis (normal distribution and with homogeneous variance) using Genstat ( $18^{th}$  Edition; VSN International Ltd, UK). Insect variables were transformed ( $log_{10}[1+x]$ ) prior to analysis.

## **Results and discussion**

#### Plant development

Plant development was more advanced in mid-July than late-June, and in winter barley compared to spring barley, and varied significantly between farms, being less advanced at the two Perthshire farms and one Fife farm (D). Differences between farms might have related to crop variety and/or timing of sowing.

#### Aphid abundance

The aphid species identified in barley fields were *Rhopalosiphum padi*, *Sitobion avenae* and *Metopolophium dirhodum*. Figure 1A shows mean values of aphid abundance per ten cereal stems (averaged across the late-June and early-July visits) for each field visited. Aphid abundance increased between late-June and mid-July and varied between farms, with highest abundance at Farm E (in Perthshire) and lowest at Farm D (in Fife). Aphid abundance was higher on spring barley than winter barley, as would be predicted by our first hypothesis, although this difference narrowed in mid-July. This difference between crop types was observed at all farms with the exception of Farm E, where aphid abundance was higher on winter barley than spring barley plants. The significant effect of crop type on aphid abundance disappeared when crop development stage was included as a covariate in the analysis (not shown), confirming that developmental changes in plant suitability are likely to be a key factor driving temporal and spatial variation in aphid populations.

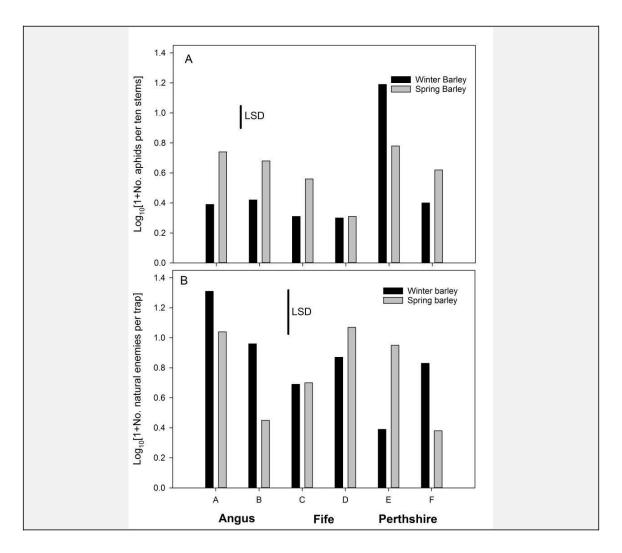


Figure 1. Log<sub>10</sub>-transformed mean abundance values of (A) aphids per ten cereal stems and (B) aphid natural enemies per sticky trap in winter and spring barley fields at six farm sites (A-F) in eastern Scotland. Bars show least significant differences (LSD) for the Farm\*Crop type interaction.

#### Natural enemy abundance and activity

When plant stems were completely enclosed in net bags for five days in mid-July, aphid abundance was c. two-fold higher than on stems contained in 'open' bags, indicating a decrease in aphid abundance when exposed to the external environment. The most abundant natural enemies identified on traps were parasitoid wasps, with smaller numbers of spiders, ladybird larvae and beetles, hoverfly larvae and other predators. Figure 1B shows the mean number of natural enemies per sticky trap for each field collected during the five-day sampling period in mid-July. Natural enemy abundance on sticky traps varied significantly between farms and between crop types at each farm, although there was no overall difference in abundance between spring and winter barley, which is contrary to the prediction of our second hypothesis. In general, there was a negative relation between natural enemy abundance and aphid abundance, particularly in winter barley, indicating a role for natural enemy suppression of aphid numbers. Further analysis is needed to confirm whether the differences between farms in insect abundance relates to differences in farm management and landscape factors. The study findings could be used to inform new measures for conservation biological control to suppress regional aphid populations (Begg et al., 2017), for example by manipulating the spatial location of winter and spring cereal crops.

## Acknowledgements

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## Food webs in barley fields: implications for biological control

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Abstract: The feeding patterns of generalist predators have important implications for biological control, but in the past it has been difficult to get a comprehensive picture of predator food webs due to difficulties in quantifying feeding links. However, recent developments in molecular methodology provide opportunities to parameterize predator-prey food webs with a high level of spatio-temporal resolution. We have constructed food webs of generalist carabid beetles and spiders and their prey in Swedish spring-sown barley fields using multiplex molecular gut content analysis, and explored implications for cereal aphid biological control. We investigated the presence of DNA of 13 different intraguild and extraguild prey types in the guts of > 3500 generalist predators in 10 barley fields during the aphid colonization and population growth phase. We found that although carabid beetles and spiders frequently fed on a range of different prey types, they maintained high levels of aphid pest predation during both phases of aphid population dynamics. Neither feeding on intraguild prey or alternative extraguild prey diverted the predators from aphid feeding. Overall we found strikingly low levels of specialization in the food webs, suggesting a high level of functional redundancy in the predator communities. However, we still detected certain changes in prey choice over time with small carabids and spiders preferring to feed on aphids early in the season, while larger carabids preferred to feed on aphids later on. In a newly started project we will explore whether the high redundancy observed in these generalist predator communities can provide resilience against climate change and environmental variability.

Key words: *Rhopalosiphum padi*, carabid beetles, spiders, redundancy, prey choice, specialization

## Introduction

Generalist predators such as carabid beetles and spiders can significantly reduce aphid populations in cereal crops (Östman *et al.*, 2003), but until recently, prey choice and feeding patterns of generalist predators in the field have been difficult to assess. This has made it difficult to determine the importance of different generalist predators as biological control agents and how the availability of alternative prey, and intraguild predation affects their ability to reduce pest populations. It has also limited our ability to assess the importance of predator diversity for biological control under open field conditions. Recent developments in molecular methodology, now provides opportunities to overcome these problems and enables parameterization of predator-prey food webs with a high level of spatio-temporal resolution (Traugott *et al.*, 2013). In the research presented here we have compared generalist predator food webs using molecular gut content analysis (MGCA) during different times of the aphid

population cycle in Swedish spring-sown barley fields. We investigated how intraguild, and extraguild predation depended on availability of different prey types and explored the level of specialization in the food webs.

## Material and methods

Predation (assessed through MGCA), and abundances of pests (*Rhopalosiphum padi*), generalist predators (carabids and spiders) and alternative prey (springtails, earthworms, flies and midges, thrips) were studied in five organically managed and five conventionally managed spring-sown barley fields located close to Uppsala, Sweden in 2011. Two time periods were considered: the colonization phase of the aphids in early June, and the population growth phase of the aphids in late June - early July.

We collected live arthropods for MGCA weekly following practices recommended by King *et al.* (2008). We used dry pitfall traps containing clay balls to decrease the likelihood of predation in the traps (King *et al.*, 2008). Within each sampling period, 12-35 traps (number depending on predator abundances) were open for 24 h each week, in each field. All predators were individually collected in 1.5 ml microtubes and frozen on dry ice, then stored at -80 °C until identification and DNA extraction. A total of 3680 individuals were processed, belonging to the eight most common generalist predator species: *Pterostichus melanarius, Poecilus cupreus, Harpalus rufipes, Bembidion lampros, Trechus se calis* (all Carabid beetles), *Pardosa agrestis* (Lycosid spider), *Oedothorax apicatus*, and *Agyneta rurestris* (Linyphiid spiders). After species identification each sample was subjected to whole -body DNA extraction and screened with diagnostic multiplex PCR assays (Staudacher *et al.*, 2016).

Abundances of *R. padi* were estimated with weekly tiller counts (100 tillers per field), carabid and spider activity density through weekly catches in wet pit fall traps (6 traps per field), flies and midges and thrips through weekly sweep netting sessions (100 sweeps per field), and springtails and earthworms through soil sampling once every sampling period (10 soil samples per field). Weed abundance was estimated weekly with quadrat sampling (6 per field).

To analyse our data, we first tested if differences between farming systems and sampling periods affected the composition of detected herbivore and decomposer prey in the guts of the predators using PERMANOVA. Using distance-based linear models, we then tested if detection frequencies for intraguild and extraguild prey reflected the availability of prey in each field and if DNA detection frequencies were affected by weed cover.

We next built food webs parameterised by the frequency of predation found in the MGCA. We estimated the level of specialization of the food webs at the network as well as species level and analysed the potential randomness of predator diets in relation to a null model. Deviation from expectations in the null model indicates active prey choice or avoidance. To assess level of specialisation we used quantitative, specialization metrics: web specialization,  $H_2$ ', and degree of predator specialization, d' (Blüthgen, 2010).

For more details on sampling, MGCA, network and statistical analyses see Roubinet (2016), Staudacher *et al.* (2016) and Roubinet *et al.* (2017).

## **Results and discussion**

We found that aphid pests and springtails were equally important prey for generalist predators early in the season during the aphid colonization phase (Roubinet *et al.*, 2017). Later in the

season, the importance of aphid prey increased with increasing aphid densities while springtail predation rates were positively correlated to abundance of springtails at both the aphid colonization and population growth phase. Intraguild predation occurred primarily in one direction: carabids fed on spiders, whereas spiders rarely fed on carabids. Carabids had higher predation rates on the two most common spider families in organically than in conventionally managed fields (Roubinet *et al.*, 2017).

We found very low levels of food-web specialization (Roubinet, 2016), indicating high levels of functional redundancy (e.g., lower specialisation than recently found in parasitoid-herbivore networks, Rand *et al.*, 2012). Predator-prey interactions often deviated from random. For example small carabid beetles and spiders preferred to feed on aphids early during the aphid colonization phase whereas larger beetle species preferred aphids later during the population growth phase (Roubinet, 2016).

Our study showed that predation by generalist predator communities on aphid pests in Swedish barley fields increases with pest numbers independently of the generally widespread consumption of alternative, non-pest prey. Therefore, conservation strategies in agricultural fields could support biological control by promoting high levels of alternative prey for generalist predators. Our findings furthermore highlight a high overall functional redundancy of the predator community but also suggest temporally complementary prey choice. This indicates that even if a high diversity of generalist predators may not be needed to ensure high levels of predation at a specific point in time, high predator diversity is likely to enhance the stability of predation over time in this system. In a new project we will investigate if the high functional redundancy observed in this system can provide an insurance against changes in land-use and climate.

## Acknowledgements

I'm grateful to all my collaborators in this project. First and foremost big thanks to former PhD-student Eve Roubinet upon whose work this presentation is based. Also big thanks to Michael Traugott, Barbara Ekbom, Tomas Jonsson, Klaus Birkhofer, Gerard Malsher, Karin Staudacher, Nico Blüthgen and Cory Straub. Grants from Formas and BiodivERsA funded this work. Thanks also to the farmers that allowed us to work on their farms.

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# Measuring biological control using surrogate prey items in winter wheat

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**Abstract:** Surrogate prey (insects or seeds) forming sentinels were used to measure the effects of semi-natural habitats at field to landscape scales on levels of biological control in winter wheat in the UK. Sentinels were located adjacent to three boundary types: grassy margin, hedgerow s and woodland in landscapes of varying heterogeneity. The mean levels of predation were higher for most insect prey (up to 57%) compared to seeds (up to 8%). Semi-natural habitats had both positive and negative effects at field and landscape scales but the response varied with the sentinel type.

Key words: biological control, sentinels, agroecology, landscape ecology, field margins

## Introduction

The measurement of biological control posses many challenges because of the sparodic nature of pest infestations whilst they are typically heterogeneously distributed requiring appropriate sampling strategies (Alexander *et al.*, 2005). To overcome some of these difficulties sentinel systems based upon surrogate prey items have the advantage that their numbers, spatial and temporal location both within field and on the plant can be chosen to represent particular pest infestions. A range of predators can be selectively allowed access using exclusion cages.

Starting in 2013 the QuESSA (Quantification of Ecological Services for Sustainable Agriculture) project designed and used a range of sentinel systems to test whether the type and proportion of semi-natural habitats (SNH) from field to landscape scales was influencing levels of biological control. Here we report on the findings for winter wheat based upon up to two years data and for seven different sentinel systems applied in the UK.

## Material and methods

The study was conducted in 18 landscape sectors (LS) each of 1 km radius in Dorset and Hampshire, UK in 2014 and 2015. The standard QuESSA protocol was followed with sentinels placed at 2, 25, 48 and 71 m from the nearest SNH along two transects 10 m apart in winter wheat. The studied field was located in the centre of the LS. The maximum distance into the field was chosen to ensure that other boundaries were not closer than the one selected. The impact of different boundary types was compared by having six replicate fields with transects leading out from either a grass margin alone between fields (control), woody linear habitat (hedgerow) or woodland. The project wide sentinels were deployed in 2014 and 2015 consisting of *Calliphora vomitoria* larvae, *Ephestia kuehniella* eggs, *Poa trivialis* and *Chenopdium album* seeds on the ground, and *Ephestia* eggs on the crop. The same

experimental design was repeated in each year, although the fields were different because of the crop rotation. In addition, sentinels of *Drosophila melanogaster* pupae and on the ground were used in 2014 which represented dipteran pests such as *Sitodiplosis mosellana* and in 2015 *Lucilia sericata* (Green blue bottle fly) larvae which are smaller than *Calliphora* and consequently vulnerable to a wider range of predators. The cereal aphid *Sitobion avenae* attached to the crop was also tested in both years. Sentinels were attached to dry stick card that was then coated with fine sand to allow predatory insects to walk across the surface. All sentinels placed on the ground were covered with a metal cage (1 cm mesh) to prevent access by birds and rodents. In 2015, fields were chosen close to the ones used in 2014. All types of SNH of > 2 m width within a 1 km radius were mapped and entered into a GIS. Predation was measured after 24 h for animal prey and 7 d for seeds. Partially or totally consumed prey items were recorded as predated.

Analysis was conducted in R v.3.2.0. Count data (e.g. Poa) was analysed using the glmer function and continuous response variables (e.g. Ephestia) analyses used the lmer function from the package lme4. For glmer models the family type specified varied depending on the type response variable which had either a Poisson (no. predated items) or Binomial (no. times predation event happened/didn't happen) distribution. For each sentinel two models were constructed, one containing total SNH as a response variable and a second composed of the SNH sub-categories – herbaceous linear (HL), herbaceous areal (HA), woody areal (WA), and woody linear (WL). These SNH area measurements were rescaled prior to analysis using the centring function in the arm package. LS was included as a random factor in all models to account for potential variation associated with farm scale factors. Where appropriate round and year were included as fixed effects. All models used Pearson residual plots with 95% pointwise confidence intervals to highlight potential outliers and non-linear relationships in the habitat area data. Non-linear relationships were then tested with a Generalised Additive Model (GAM). If the GAM showed a relationship was significantly non-linear the model was rerun as a Generalised Additive Mixed effects Model (GAMM) with a smoother term added to the variable in question. We also tested for overdispersion in models and if overdispersed a random level intercept was added to the equation.

#### **Results and discussion**

Predation levels and distribution of the data varied considerably between the different sentinel systems with higher mean predation for *Calliphora* larvae (53%), *Lucilia* larvae (57%), *Ephestia* eggs (63%) and *Drosophila* pupae (37%) on the ground, but low levels for *Sitobion* (12%) and *Ephestia* eggs (22%) on the crop, with even lower levels for the seeds, *Chenopodium* (8%) and *Poa* (4%) on the ground (Figure 1). We can conclude that biological control was occurring and there is considerable potential to improve this because in all cases predation levels were sometimes 100%.

The proportion of HA habitat in the LS had a positive effect on the level of *Calliphora* larvae and *Chenopodium* predation, but was also found to have a significant negative effect on natural cereal aphid infestations and a non-linear effect on *Ephestia* ground predation (Table 1). On the other hand, the proportion of WL had a negative effect on *Chenopodium*, *Sitobion* and *Calliphora* predation, alongside a positive effect on natural cereal aphid infestations. Similarly, WA habitats had a negative effect on *Calliphora* predation and a non-linear effect on *Chenopodium* predation. The total area of SNH in the LS had a negative effect on natural cereal aphid infestations, in other words LS with more SNH led to fewer aphid pests. Total SNH had an additional non-linear impact on *Ephestia* (ground) predation. Models

would not converge for *Ephestia* on the crop. Our findings indicate that what was measured were background levels of control by predators that reside within fields, rather than by those relying on resources provided by SNH, such as floral or alternative prey. Few effects of the adjacent SNH type were found except that predation of *Calliphora* (individual SNH model), *Poa* seeds and *Lucilia* larvae (total SNH models) were lower in fields bordered by WL than HL habitats. *Chenopodium* predation (total SNH model) was higher with adjacent WA compared to HL habitats.

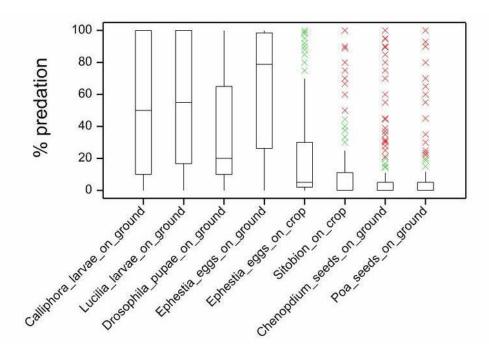


Figure 1. Sentinel percentage predation showing median (horizontal line), interquartile range (box), solid whisker extending to the upper quartile plus 1.5 times the interquartile range or maximum value if smaller.

Predation of *Calliphora* and *Lucilia* larvae, and seed predation for both species in total SNH and individual SNH models increased with distance from the SNH. In contrast, predation of the sentinel *Sitobion* (total and individual SNH models) decreased with distance from SNH, probably because pest natural enemy abundance is higher around field edges (Holland *et al.*, 2009). For seeds this may be because alternative foraging resources were not available. Weed levels and therefore weed seeds are usually higher in field headlands. Predation of the fly larvae was probably higher further from the field edges because the larger carabid beetles capable of attacking them overwinter within fields and densities are higher in field centres than the edge (Holland *et al.*, 2005).

In both sets of models there was variation in predation levels for most sentinels between sampling occasions and years, indicating that biological control is unreliable to some extent and greater resilience needs to be built to ensure more consistent control. Table 1. Summary of effects for predation of sentinels and in winter wheat.

( $\uparrow$  = increase,  $\downarrow$  = decrease in relation to variable and significance of effect indicated by \*= p < 0.05, \*\* = p < 0.01, \*\*\* = p < 0.001), N.S = Non-significant, N.A. = Not applicable)

Method	Model	SNH types in landscape	SNH type in adjacent boundary	Distance from SNH	Sampling occasion	Year	
<i>Calliphora</i> on ground	Individual SNH	$\begin{array}{c} HA^{\uparrow **} \\ WL \downarrow ** \\ WA \downarrow * \end{array}$	WL↓*	↑*	Not tested	Not tested	
	Total SNH	N.S	Not tested	↑ *	Not tested	Not tested	
<i>Lucilia</i> on ground	Individual SNH	N.S	N.S	↑*			
	Total SNH	N.S	WL↓*	↑ *			
Drosophila pupae on ground	Individual SNH	N.S	N.S	N.S	N.A	N.A	
	Total SNH	N.S	N.S	N.S	N.A	N.A	
<i>Ephestia</i> on ground	Individual SNH	HA non- linear effect	N.S	N.S	1 > 2**	2015>2014***	
	Total SNH	Total SNH non-linear effect	N.S	N.S	1 > 2**	2015>2014***	
	Total SNH	Model would not converge					
Sitobion on crop	Individual SNH	WL↓* HL non- linear effect	N.S	↓ *	2 > 1**	2014>2015***	
	Total SNH	N.S	N.S	$\downarrow *$	2 > 1**	2014>2015***	
Poa on ground	Individual SNH	N.S	N.S	<b>^</b> **	N.S	2015>2014***	
	Total SNH	N.S	WL↓*	<b>^</b> **	N.S	2015>2014***	
<i>Chenopodium</i> on ground	Individual SNH	HA↑*, WL↓* WA non- linear effect	N.S	<b>^</b> **	1 > 2*	2015>2014***	
	Total SNH	N.S	WA†*	<b>^</b> **	1 > 2*	2015>2014***	
Natural cereal aphid infestations	Individual SNH	WL↑* HA↓* HL non- linear effect	N.S	N.S	2>1**	N.S	
	Total SNH	$\downarrow^*$ with total SNH		N.S	2>1**	N.S	

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# Video monitoring of brown planthopper in rice shows importance of frogs for biological pest control

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**Abstract:** The contribution of different predator guilds to biological pest control is usually inferred from the abundance of these guilds or from selective exclusion, but direct observation is rarely used. However, the best evidence for predation is obtained by catching the predator in the act. We used direct observation by video recording to identify the most important predators of brown plant hopper (BPH), Nilaparvata lugens, a pest of major importance in rice. In the first experiment, we used dead BPH and demonstrate that long-horned grasshoppers (Tettigoniidae: Conocephalus longipennis), which are primarily herbivorous, but scavenging if given opportunity, were responsible for most removals of dead prey. Wolf spiders (Lycosidae) were the second most important. In a second experiment we compared the consumer guilds removing (i) dead BPH, (ii) live, immobilized BPH, and (iii) live, mobile BPH. Long-horned grasshoppers (C. longipennis) were again the main removal agents of dead BPH, ground beetles (Carabidae) of live, immobilized BPH, and frogs (Ranidae: Rana limnocharis) of live, mobile BPH. This study highlights for the first time the important contribution of frogs to predation on BPH in rice. Furthermore, we show that removal of immobilized sentinel prev is not representative for predation of live mobile prev, underlining the need for a critical assessment of commonly used sentinel methods.

Key words: *Conocephalus longipennis*; direct observation, frogs, immobilized prey, *Nilaparvata lugens*, validation, *Rana limnocharis* 

## Introduction

Natural enemies have the potential to suppress herbivore populations in crops, but quantification of predation is challenging. Predation rates can be measured by placement of immobile prey in the field (referred to as "sentinel") and quantifying the prey removal rate by comparing the number of prey before and after a known exposure period. Sentinel methods are widely used because they are quick, cheap and easy to carry out when compared to alternative methods, such as predator exclusion.

While eggs and pupae are immobile by nature, juvenile and adult insects may be immobilized to facilitate placement and retrieval in the field, for instance by fixing them on cards. Here we focus on the assessment of predation using immobilized insect stages. Although this methodology is often used, it has some potential biases. We studied the removal of brown planthopper (BPH) *Nilaparvata lugens* Stål (Homoptera: Delphacidae) in rice using video cameras. BPH is an important rice pest in Asia (Heong *et al.*, 2015a). Varietal resistance and chemical spraying are the main strategies for BPH control in Asia, but outbreaks may still occur as a result of breakdown of varietal resistance to BPH, resistance development in BPH against the insecticides used, and destruction of natural enemy populations by pesticides (Cheng, 2009). Biological control offers an alternative control method to prevent outbreaks and reduce the side effects of pesticides. Our research objective was twofold: (i) to assess the identity and relative importance of the agents responsible for removal of dead, immobilized BPH, and (ii) to assess whether the removal of immobilized BPH is representative of BPH predation under unmanipulated conditions. This contribution is based on Zou *et al.* (2017), for full details we refer to this study.

#### Material and methods

The study was conducted in irrigated rice fields at the Jiangxi Agricultural University, Jiangxi Province, China. Hoppers were reared on rice plants in cages in a greenhouse.

In the first experiment, we recorded removal events of immobilized, dead BPH. BPH obtained from the rearing cages were killed by placing them in a freezer at -18 °C for 24 hours. Five 20 cm-long sections of rice plant stem with a total of 25 adult female BPH (5 per stem section) were placed in the field next to a rice plant, at least 3 meters from the field edge. BPH were fixed 5 cm from the top of the stem, on a 1 cm<sup>2</sup> double-sided tape. After placement in the field, the BPH sentinels were monitored over a 72- hour period. A new set of BPH exposures were replaced when fewer than three of the original BPH were left on the bait stems. In total, over the three replicate exposure trials, we monitored the fate of 250 BPH using a digital video camera.

In the second experiment, we included three treatments: (1) dead, immobilized BPH, (2) live, immobilized BPH, and (3) live, mobile BPH. In treatment (1) BPH were attached to the rice leaf using double sided sticky tape. In treatment (2), BPH were temporarily (reversibly) immobilized by cooling them at -18 °C for five minutes, and then fixing them on rice stems using double-sided sticky tape. While all BPH were alive when introduced to the field, they eventually died. For treatment (3), the initial BPH were transferred to a tube containing a 20 cm section of rice stem. After 20 minutes, when BPH had settled on the rice stem, the stem pieces were placed in the field, pressed against a rice plant in front of the camera, and the tube was carefully removed. After placement in the field, the three treatments were monitored over a 24-hour time period. The above protocol was replicated six times, at 32, 47, 59, 67, 80 and 88 days after transplanting of the rice in the experimental field, respectively. We used three surveillance cameras to monitor BPH predation during day and night. For further details we refer to Zou *et al.* (2017).

#### **Results and discussion**

#### **Experiment** 1

In the first experiment, 168 full removals (the removal of the entire body of a BPH) and 42 partial removals (the removal of only a part of the BPH body) of dead, immobilized BPH were recorded out of 250 BPH exposed. Long-horned grasshoppers (Tettigoniidae: *Conocephalus longipennis*) were responsible for 85% of the full removals, while the

contribution of known BPH predators such as wolf spiders (Lycosidae) and ground beetles (Carabidae) accounted for only 9% of the full removals. Marsh flies (Sciomyzidae: *Sepedon* spp.) and long-horned grasshoppers (Tettigoniidae: *C. longipennis*) were the main agents responsible for partial removal of BPH, with again a minor role for spiders (Linyphiidae, Lycosidae, Salticidae) and predatory beetles (Staphylinidae, Carabidae).

#### **Experiment 2**

In the second experiment, a total of 44 full removals and 28 partial removals of dead immobilized BPH were observed out of 150 BPH. For live, immobile BPH we recorded 66 full removals and 25 partial removals, also out of 150 BPH. For live, mobile BPH, we observed 93 full removals out of 634 exposed and freely moving BPH, while 493 left the rice stems.

There was a variety of predator species groups responsible for removal of prey in the three treatments (dead immobilized prey, live immobilized prey, and live mobile prey). Long-horned grasshoppers (*C. longipennis*) removed 20 dead immobilized BPH (45%), rain was responsible for 30% of the removals, while ground beetles and marsh flies (Sciomyzidae: *Sepedon* spp.) caused a major part of partial removals, 68% and 21%, respectively. For removal of live immobilized BPH, ground beetles (82%) and wolf spiders (Lycosidae, 8%) were the key predators. In addition, BPH escaped occasionally from the sticky tape. The major predators for partial removals of live immobilized prey were again ground beetles (60%) and marsh flies (27%). The major species group removing live mobile prey were frogs (Ranidae: *Rana limnocharis*), which removed 75% of the BPH. The warbler bird *Locustella ochotensis* (Locustellidae) was responsible for the removal of 9% of live, mobile BPH.

There was little similarity in the predator species responsible for BPH predation in the three treatments. Ground beetles (Carabidae) and wolf spiders (Lycosidae) were the only two species groups that removed BPH in all treatments. Long-horned grasshoppers (Tettigoniidae) and crickets (Gryllidae) were only observed to remove dead, immobilized BPH, while frogs (Ranidae), warbler birds (Locustellidae), rove beetles (Staphylinidae) and dwarf spiders (Linyphiidae) were only observed to remove live, mobile BPH.

#### **Implications**

The study has two key findings, both previously unreported: (1) frogs contribute substantially to predation on brown plant hopper, and (2) consumer guilds removing live mobile prey (i.e. true predators) are not the same as those removing immobilized prey. Therefore, the use of immobilized prey for measuring predation is questionable.

The use of direct observation to validate sentinel methods has not been conducted before, most likely because direct observation is laborious and difficult to carry out. However, with the current availability of video technologies with night vision, direct observation has become much more feasible. Video observations can provide essential data to complement data acquired by sentinel methods.

Our study has two important implications. First, we show that caution must be taken in the interpretation of results from experiments using immobilized sentinels. Our findings provide a strong argument for validation of sentinel methods. The second implication of our study is that frogs are a major group of predators of live, mobile BPH. While current strategies to enhance natural suppression of BPH in rice focus on arthropod natural enemies (Heong et al. 2015a), the role of frogs deserves further investigation and scrutiny.

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# Natural pest control requires a complete landscape

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Abstract: In many studies on landscape and functional biodiversity it is assumed that pestregulating insects require other habitats than only agricultural fields. Other habitats can e.g. be important as hibernation site, as source of alternative prey or as source of floral resources. The relative contribution of the different habitats to the performance of the beneficial insects and to pest-regulating services they provide is very difficult to study empirically. Landscapebased population-dynamical modelling can then come at hand. Such models can be used to predict mechanisms and patterns that can be tested experimentally. As an example, I created population models of aphid-feeding hoverflies moving between different habitats in an arable landscape. The models are designed and parameterised on the basis of field observations on temporal and habitat-related availability of resources. Habitat-structured models can indicate the optimal composition of the landscape with respect to pest control. The models e.g. indicate that the amount of woody elements, which are particularly important for hoverflies as spring habitat, are a likely a bottleneck for effective pest regulation in Dutch arable landscapes. They also show that various arable crops can enhance each other's pest control when their aphid populations peek at different moments in time (such as winter wheat and potato). These and other habitats are complementary in their function for the predators, and several of them are needed to create a 'complete landscape'.

**Key words:** natural pest control, complementary habitats, aphid-feeding hoverflies, population-dynamical modelling, landscape complementation

## Introduction

The importance of various landscape elements for ecosystem services (ES) such as pest control and pollination are typically studied by aggregating landscape elements in larger classes (e.g. merging all 'woody habitats') and correlating the amount or the diversity of these habitat types with measured levels of ES or (more often) of ES providers (the natural enemies or pollinators). Among other limitations, this approach will generally not reveal important interactions between the qualitatively different habitats, nor which landscape measures are most effective to enhance the ES.

A life-cycle approach, checking the match between the requirements and the habitats of the ES providers year-round, indicates that many species are dependent on more than one habitat to be maintained year-round. These species require more than one resource and the resources are not all available in one habitat, or only during part of the season. Such theory and the patterns that emerge from it have been labelled 'landscape complementation' (Dunnings *et al.*, 1992; Pope *et al.*, 2000). Examples are aphid-feeding hoverflies in an arable landscape. Whereas the larvae require aphids for their development, the adults require nectar and pollen from flowers for their survival and activity and for egg maturation respectively

(Van Rijn & Wäckers, 2016). They may find these resources in spring on early-flowering woody plants, where aphids hibernate and develop their first generations (Van Rijn, 2014). Later they can move to crops when aphids have emerged here as well, but at the same time they need to visit habitats with suitable flowers, such as field margins.

This dependency on multiple 'complementary' habitats can best be studied by modelling the dynamics of habitat-structured populations, where each habitat temporarily provides certain resources, as observed in the field. These models can be used to predict the population patterns emerging from these dependencies, and to design specific field studies to test these predictions.

### **Model formulation**

The population-dynamical model is framed in a set of ordinary (ODE) and delay differential equations (DDE), some representing the number of predators in various stages and various habitats, others representing the number of aphids in various habitats. Three predator stages are distinguished: non-consuming non-reproductive (eggs and pupae), aphid-consuming non-reproductive (larval) and flower-consuming reproductive (adult) predators. Within each habitat these stages are connected by development and reproduction, which are decelerating functions of resource density.

The sub-populations in various habitats are connected by dispersal of the adults. Dispersal occurs at two different spatial and temporal scales. At the field scale the adult insects are assumed to alternate between foraging for oviposition sites (*i.e.* aphid patches in crop fields) and for feeding sites (*i.e.* flowers in adjacent field margins) depending on its feeding state. At the landscape scale the crop-flower combination is considered as one unit from which dispersal to other habitats occur when local resource (aphid and flower) densities decline.

Resources are seasonally driven. Aphids have an intrinsic growth rate that varies with time, being positive only in periods of extensive phloem transports. In addition, the growth rate is reduced at higher densities (logistic growth) and due to consumption by larval predators. Floral resources are modelled directly based on field observations, being present in spring in woody habitats and in summer in flowering field margins (see Figure 1).

Population densities and distributions are simulated for seasons running from early April until end of October. Adult predators are emerging from hibernation during April and May, and are going into hibernation again during September and October. A fixed proportion of predators and tree aphids is assumed to survive to the next season. Aphids in annual habitats are assumed to migrate into the fields at fixed numbers and moments early in plant life. The simulations are run for four (identical) seasons, starting with initial values that allow quick convergence to a stable seasonal pattern. The results presented are taken from the last season.

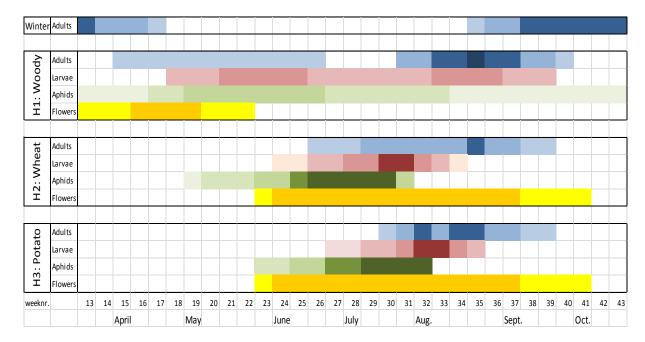


Figure 1. Seasonal population dynamics according three-habitat model including woody habitat (H1), winter wheat crop (H2) and potato crop (H3) for flowers (yellow), aphids (green), hoverfly larvae (red) and hoverfly adults (blue). Top row represents adult hoverflies in hibernating stage. Each darker shade represents a 10-fold increase in density.

#### Results

In the current example I based the parameters on the biology of the predacious hoverfly *Episyrphus balteatus*. I assumed a very simple arable landscape with only two crops (winter wheat and potato), both with flower-rich field margins, and a non-crop habitat with spring-flowering shrubs making up 5% of the landscape. All three habitats have habitat-specific aphids. The results in figure 1 show that in this specific example the resources in the three habitats are available at different periods in the season. To study the impact of this type of complementarity, each habitat can be taken out one by one. Obviously all habitats have a positive impact on the hoverfly population and on aphid control, since taking away any habitat will reduce the predator population and increases the aphid densities in some other habitats, as shown in Figure 2.

Removing all woody habitat strongly reduces the predator population and its suppression of aphids in both crops. Taking away the flower strips along the wheat fields also causes a strong increase in aphid numbers; not only in the wheat, but to a lesser extend also in the potato crop. Obviously, the wheat-flower combination is a source for predators that later helps to suppress the aphids in potato. On the other hand, flower strips next to potato fields have no impact on the wheat crop. Taking away the potato crop itself strongly reduces the overwintering hoverfly population. This however allows the aphids in the woody habitat to reach much higher numbers next spring, which then serve as a food source allowing hoverflies to largely restore their original numbers again.

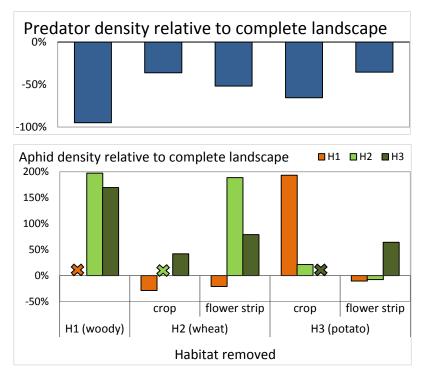


Figure 2. The impact of removing habitats from the complete landscape on mean hoverfly density (upper panel) and mean aphid densities in the other habitats (indicated by colour, lower panel). A cross indicates this habitat is absent.

Apart from taking out habitats completely we can also vary the relative amount of habitats. Figure 3 shows that increasing the proportion of woody habitat in the landscape further decreases the aphid densities in both crops.

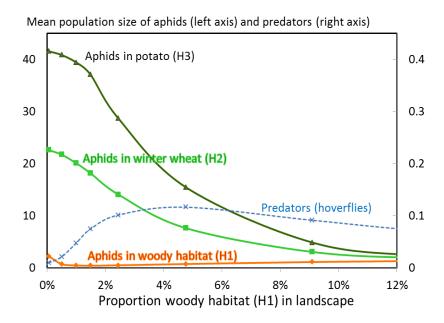


Figure 3. The effect of the amount of woody habitat on mean hoverfly and aphid population sizes, according to the three-habitat model.

#### **Conclusions and discussion**

Although the model has been formulated for aphid-feeding hoverflies, similar results are expected for other species that shift from prey to sugar/pollen feeding after their pupation, such as green lacewings and parasitoid wasps.

The analysis gives insight in the importance of non-crop habitats and shows that the difference in timing of resource availability in woody and herbaceous habitats are essential for sustaining effective densities of hoverflies throughout the growing season. This means that one cannot replace one habitat with the other when more convenient. Given that in an area like the Hoeksche Waard where only 2-3% of the habitat is woody and only one out of 10 crop fields has flower strips nearby, there is still a lot to gain.

The analysis also shows that crop fields that provide resources at different times can also be important in supporting natural enemies and natural pest control. Potato fields e.g. can benefit from nearby winter wheat fields (when bordered by flower strips and not sprayed with insecticides) by being a timely source for natural enemies, even when wheat aphid numbers remain below damage levels.

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# **Tipping the system: the difficult transition from chemical towards natural pest control in agriculture**

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**Abstract:** Insecticides are used intensively for agricultural pest management, despite decadeslong efforts to diminish pesticide dependency. Pest management can be characterized as a dynamic system with alternative states – a biocontrol-dependent state and a pesticide dependent state. A pesticide dependent state is characterized by a positive feedback between insecticide use, reduction of natural enemies, release of pests from top-down natural enemy control, and need for more insecticides. A biocontrol-dependent state is characterized by effective pest suppression by natural enemies, no or limited need to use insecticides, and limited disturbance of natural enemy populations. Each stable state is self-reinforcing due to positive feedback, and the transition from one state to the other could be characterized by "tipping points": critical points in the management intensity where the system switches from one stable state to the other. More insight is needed on these pest control system dynamics, and the trajectory of pesticide dependency to biocontrol dependency. This will contribute to a better understanding under which set of conditions a coordinated effort between stakeholders has potential to "tip" the pest control system to an insecticide-independent state.

Key words: pest control system dynamics; biocontrol; pesticides; pesticide dependency

## Introduction

In undisturbed ecosystems biological pest control by natural enemies plays a central role in limiting herbivory by pests. However, agroecosystems may be subject to frequent and intense disturbances such as pesticide use. The lack of specificity of broad-spectrum insecticides can have detrimental effects on natural enemy populations, disrupt biological control and create an increasing dependence on insecticides, ultimately reducing natural enemy populations even further. This phenomenon is known as the pesticide treadmill and involves a positive feedback between the use of pesticides, the decimation of natural enemy populations, and releasing pests from top-down natural enemy control (DeBach, 1951; Hansen, 1988; Thrupp, 1990; Turnbull & Hector, 2010).

The regime shift from biocontrol to insecticide control and vice versa (via so-called tipping points) may take different forms. Insecticide use-induced declines of natural enemy populations could be continuous, shifting smoothly from one stable state to another (Figure 1A). Reversing the change in insecticide use will then allow a smooth return to the

original stable state (biocontrol). Alternatively, the trajectory could involve sudden changes in biocontrol services (i.e. hysteresis; Figure 1B). In this case, reaching the original state will require a major change in the control variable (Figure 1B), i.e. the input of insecticides would have to be reduced considerably to enable natural enemy populations to recover and to reestablish the stable state of biocontrol. The shape of the biocontrol – insecticide use relationship also has consequences for the resilience of the system; i.e. the capacity of the system to recover from a disturbance. Resilient agroecosystems will be able to return quickly to the original state, while systems with low resilience will need extended periods to reach the pre-disturbance state. The transition from insecticide dependence to greater reliance on biocontrol may entail a restoration phase in which natural enemy populations are still low and biocontrol has not reached its full potential, hence the incentive for pesticide use remains high (Bianchi *et al.*, 2013). Thus, the pathway out of pesticide dependency would differ depending on the shape of the biocontrol – insecticide use relationship.

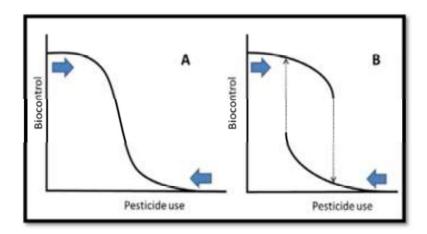


Figure 1: Two alternative hypotheses concerning tipping points and systems dynamics. The vertical axis denotes the stable state of biocontrol as influenced by the pesticide loading on the horizontal axis. Panel A shows a continuous trajectory from one stable state to the other as pesticide loading is changed. Panel B shows that for the intermediate range of pesticide loading (between the dotted lines); there are two alternative stable states possible. The upper branch of the drawn line denotes the pesticide-independent stable state. Pesticides may be used, but biocontrol remains effective. The lower branch denotes the pesticide-dependent state where pesticides are indispensable.

#### Lock-in mechanisms

The concept of lock-in stresses the nature of interactions and feedbacks that have a low potential to change and refers to systems that follow specific trajectories which are difficult to change. Systems that are subject to positive feedbacks tend to show path dependence and inflexibility, as well as lock-in effects and interdependencies that impede the development of alternative techniques and strategies (Vanloqueren & Baret, 2009). In the specific case of pesticide use, lock-in mechanisms are influenced by environmental, technological and sociopolitical factors that prevent transition to more sustainable pest management strategies. Here we will briefly discuss various examples of lock-in associated with pest management.

#### Technological lock-in

Technology developments and adoption of technologies are important factors determining a technological lock-in. Positive feedbacks are created through technological externalities (e.g. market share), learning and user experience, as well as uncertainty reduction that enforce the preference of using a specific technology. Consequently this makes a technology more valuable and enforces its use. Technical lock-in happened in pest management based on the use of synthetic pesticide use. Since the rapid increase of the use of these pesticides after the Second World War, the use of other pest control strategies declined. As chemical pest control was subject to increasing returns, pesticide dependency was enforced through research & development, learning curves in chemical development, and through increasing returns to scale in their manufacture. Nowadays, the pest control development has become inflexible in a way that alternatives are hardly offered (Cowan & Gunby, 1996) – a technological lock-in.

#### Societal lock-in

Parsa *et al.* (2014) discuss the collective action dilemma where payoffs from adopting a technology depends on whether others adopt the technology as well. Especially when introducing an alternative pest control strategy, like biocontrol or integrated pest management (IPM), this is an important issue. Biological control is based on preventative measures to control pests and insecticide-use disrupts the potential for biocontrol (Bianchi *et al.*, 2013), which emphasizes the need for collective action. Transitioning from pest management based on insecticides to biocontrol could include large costs due to new management practices, skills and patterns of behaviour, but also network coordination and technological interdependencies. This can create an inertia of a technology development or implementation (Cowan & Gunby, 1996; Hammond Wagner *et al.*, 2016). Another example of a societal lock-in is the standard of quality imposed by the market. Marketing standards for agricultural products, such as the size and appearances of crops, can greatly influence farm management and pest management strategies. Quality standards driven by consumers that want perfect and regular products can lock farmers into a certain pest management strategy (Lamine, 2011).

#### Landscape lock-in

It is widely recognized that landscape context can influence pest-natural enemy interactions. For instance, landscape elements such as semi-natural habitats may provide pollen, nectar, overwintering sites, refuge from disturbances, and alternative prey for natural enemies (Rusch et al., 2016). However, if more land is converted to agriculture, and less semi-natural habitat is available, natural enemy populations may decline and a threshold for effective pest control may be reached. To illustrate this point, landscapes with large proportions of agricultural land are generally dominated by only a few species of the predator community, while complex landscapes can harbour a diverse range of natural enemies (Bianchi et al., 2006). Other landscape-specific factors are: 1) the size of habitats, 2) the spatial arrangement of habitats, and 3) habitat connectivity. One can expect higher biocontrol rates in landscapes consisting of diverse habitats (i.e. smaller patches), resulting in shorter distances between source habitats and crop fields, which allows natural enemies to colonize crop fields earlier and thus more effectively suppress pests (Bianchi et al., 2006). Agricultural landscapes dominated by large arable fields and few semi-natural habitats may support only relatively low natural enemy populations, enhancing insect pest pressure, and resulting in an increased need for insecticides (Meehan et al., 2011), possibly reinforcing the aforementioned pesticide treadmill.

### Discussion

Industrial agriculture has been locked into place (IPES-Food, 2016). This lock-in needs to be broken to allow a transition towards a new state. Understanding the dynamic interactions between pesticide use and environmental, technological and societal developments which have evolved from past agricultural practices, will give us better insight in the barriers for changing practices. The pesticide treadmill involving an increase in pesticide use, decreasing populations of natural enemies, evolving of pesticide resistance in pest populations, and increasing pest pressure has been observed across the globe. Switching to a different pest control strategy is possible, but requires fundamental changes in social, political or technological factors. By learning from the past we can critically evaluate current pest management strategies to tip the system to an insecticide-independent state.

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# Designing multifunctional agro-landscapes: a multi-objective optimization approach

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**Abstract:** To counteract the pressures of intensive agricultural production systems on the environment there is increasing interest in habitat management to strengthen ecosystem services. By creation of flower strips and management of semi-natural habitats, farmers and other stakeholders in agricultural landscapes try to enhance populations of ecosystem service providers' habitats, such as natural enemies of pests and pollinators. However, the redesign of landscapes for multiple ecosystem services needs to take the trade-offs and synergies between ecosystem services and agricultural profitability into account. We used the modelling framework Landscape IMAGES to explore options for reconfiguring and managing habitats for multiple ecosystem services in the Hoeksche Waard, The Netherlands. A wide variety of alternative landscape configurations and management practices were evaluated using a Pareto-optimizing algorithm. The analysis shows that there are both trade-offs and synergies between crop production, floral resources for flying natural enemies, water quality, and the management costs allocated to different stakeholders (private and public) in the landscape. These findings can help stakeholders to make informed decisions about their management actions.

**Key words:** agro-landscape; ecosystem services; trade-off; synergies; multifunctional; Pareto-optimization

## Introduction

Landscapes with specialized industrial agriculture are characterized by a low proportion of semi-natural habitats and highly disturbed fields. This has resulted in an overall loss of biodiversity across the globe (Foley *et al.*, 2005). As a consequence, several ecosystem services that support agricultural production have declined as well, such as soil regulation, pollination and natural pest control. There are inspiring examples of collaboration between farmers and other landscape managers to restore semi-natural habitats and the associated ecosystem services in agricultural landscapes (Geertsema *et al.*, 2016). Yet, such initiatives require understanding about the trade-offs and synergies between multiple ecosystem services when modifying management and spatial arrangement of semi-natural habitats in the landscape.

There is increasing attention for trade-offs and synergies among ecosystem services in agricultural landscapes (Power, 2010). In real landscapes, where stakeholders have the amibition to restore ecosystem services, there is a need for design-oriented approaches that allow stakeholders to make informed decisions about the expected outcomes of their

interventions. In this study we used the modelling framework LandscapeIMAGES that was designed to optimize landscapes for multiple objectives simultaneously (Groot & Rossing, 2011). This framework uses an evolutionary algorithm with Pareto-optimization, which allows the evaluation and ranking of a large set of alternative landscape configurations. In Pareto-optimal landscapes, the further improvement of one of the objectives will always result in loss of performance of other objectives.

The aim of the study was to explore trade-offs and synergies between different ecosystem services associated with semi-natural habitats (including sown field margins) in an intensively used agricultural landscape. The effect on associated costs for public and private stakeholders was explored as well.

## Material and methods

#### Study site

The case study area is the Hoeksche Waard, situated in the Southwest of the Netherlands. The soil consists of clay soils. The total region encompasses about 263 km<sup>2</sup> and arable farming is the main land use. Fields are seperated by ditches, and there is little woody habitat. Creeks with riparian zones and dikes are characteristic landscape features. Several farmers have created flower strips along their fields. We used a section of about 740 ha in the Southeastern part of the area for the modelling experiment. This consisted of potato (170 ha), wheat (114 ha), sugar beet (99 ha), vegetables (120 ha), orchard (92 ha) and grassland (51 ha). Seminatural habitats consisted of ditchbanks (15 ha), road verges (15 ha), riparian areas (13 ha), wetland (9 ha). Field margins were assigned to 60% of the fields (23 ha of sown field margins).

#### Model description and decision variables

Landscape IMAGES designs, evaluates and ranks alternative land use configurations in a spatially explicit environment using decision variables. The decision variables relate to the width of sown field margins (3.5 or 7 m), their vegetation composition (seed mixtures: annual flowers, perennial flowers, perennial grass), and management of other semi-natural habitats (such as ditch banks, road verges and riparian areas). The management of semi-natural habitats can vary from conventional to ecological, which affects the amount of floral resources for natural enemies and pollinators. Different management interventions have specific costs and impacts on ecosystem services. For instance, choice of width and seed mixture for a field margin has effects on natural enemies, pollinators and prevention of pesticide drift.

#### **Objectives and indicators**

The objectives for the optimization were based on a survey among different stakeholders in the Hoeksche Waard (Table 1). Evidence-based indicators were derived to quantify the extent to which objectives were met for any possible landscape configuration. Using Landscape IMAGES we identified landscape configurations that that maximised desired ecosystem services and minimized costs for public (water council, municipalities, landscape conservation organisation), and private stakeholders (farmers).

Objective	Optimization	Indicator		
-	direction*			
Crop production (€/ha)	+	Revenue minus costs; field margin subsidies		
		and pesticide costs are taken into account.		
Floral resources for	+	Area of flower cover in a radius of 150 m		
natural enemies (-)		around a field per crop area relying on		
		natural pest control. Only flowers accessible		
		for natural enemies included.		
Floral resources for	+	Area of flower cover in a radius of 150 m a		
pollinators (-)		field per crop area relying on pollination.		
		Only flowers accessible for pollinators		
		included.		
Water quality (-)	+	Fraction reduction of total pesticide drift to		
		surface water in landscape due to presence		
		of sown field margins.		
Management cost for	-	Management cost for private stakeholders		
private stakeholders		for ditch banks, production grassland and		
		flower -or grass field margins in the study		
		area.		
Management costs for	-	Management cost for public stakeholders for		
public stakeholders		road verges and riparian areas in the study		
		area.		

Table 1. Optimization objectives for Landscape IMAGES

\* +: maximize, -: minimize

## **Results and discussion**

We identified trade-offs and synergies for the indicators of the associated objectives (Figure 1). Compared to the original landscape, improvement of all indicators was possible.

Trade-offs were found between crop production and water quality (Figure 1A). This trade-off can be explained by the effect of field strips: establishment of wide field strips can enhance water quality by reducing drift, but at a cost of a smaller area available for production, which causes a decline in production.

Floral resources for natural enemies and production show synergy up to a crop production of about 4000  $\notin$ /ha, but beyond this production level a trade-off occurs (Figure 1B). This complex relation can be explained by an interaction of several management interventions in the landscape. The synergy at production levels up to  $\notin$  4000/ha is explained by replacement of wide grass strips in fields (low in floral resources, low subsidies, substantial loss of production area) by narrower flower strips (richer in floral resources, high subsidies, smaller loss of production area). The cluster of landscapes representing landscapes with production between  $\notin$  3500 and  $\notin$  5000/ha with low density of floral resources (< 0.015) is explained by absence of strips, but with floral resources in adjacent semi-natural habitats, which have no direct relation with crop production.

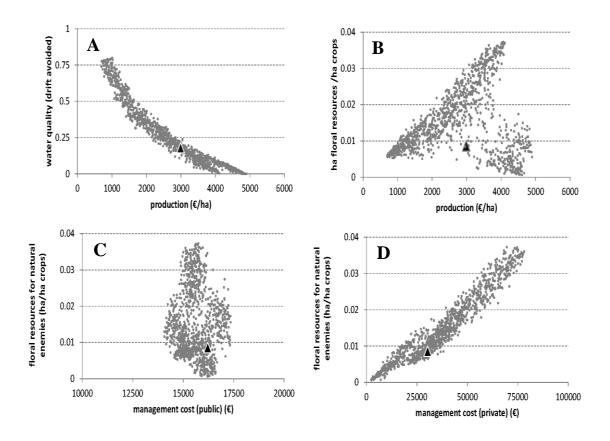


Figure 1. Relationship between indicators for desired ecosystem services and management costs. Each dot represents an alternative landscape configuration. Each landscape can be characterized as a point in a multidimensional space with one dimension for each indicator. A: Water quality x crop production; B: floral resources for natural enemies x crop production; C: floral resources for natural enemies x management cost for public stakeholders; D: floral resources for natural enemies x management cost for private stakeholders. The original landscape is represented by a  $\bigstar$  in each graph.

The relation between public management costs and floral resources for natural enemies does not show a trade-off or synergy (Figure 1C). This can be explained by the fact that the area of habitats managed at the cost of public stakeholders, such as municipalities and water board, are relatively small and hence contributes little to floral resources for natural enemies in the arable fields. These habitats also are often located outside the influence area of arable fields that could benefit from the natural pest control.

The relation between private management costs and floral resources shows a clear positive relation (Figure 1D). This can be explained by the fact that habitats that are closest to arable field (ditch banks in this case study area) that can benefit from the floral resources for natural enemies are also managed at the cost of the farmers (hence private costs).

### Implications

Our analysis shows how landscape performance in terms of multiple objectives can be improved compared to the original situation. The results highlight the presence of trade-offs and synergies between multiple ecosystem services in agricultural landscapes. The distinction between public and private management cost and options for management by different stakeholders make clear how financial costs and benefits are distributed for different landscape alternatives. Such insights inform discussions and negotiations on landscape development. The model results can be used to explore cost-effective options for the management of ecosystem services. The analysis of the decision variables (e.g. how many flower strips along which crops, type of management of semi-natural habitats) and the spatial distribution (clustered, random distribution) will provide valuable information when stakeholders and scientists collaborate in landscape design and management for multiple ecosystem services.

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Posters

# Does the association between fruit trees and vegetables promote functional biodiversity and biocontrol of vegetables pests?

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**Abstract:** Agroecological systems often rely on mixing crops to maximise production. In this study, we investigated if associating fruit trees and vegetables in plots increased predator arthropod abundance and pest control. For this purpose, we compared activity-densities of predator arthropods and predation of sentinel aphids in plots grown with cabbages within or outside of apple orchards. Sampling was performed six times during one season by pitfall trapping and visual observations. Our results indicate that there were overall few significant differences between the two types of plots and these were always in the direction of more predators in the control plots.

**Key words:** conservation biological control, fruit tree-vegetable association, functional biodiversity, sentinel prey

## Introduction

New agricultural systems are developed with the objective to ensure a high yield and to be eco-friendly (Schutter, 2010). These new systems tend towards the no-use of synthetic pesticides or synthetic fertilizers and a stronger reliance on ecosystems services such as biological control. They are often established on small farm areas and may thus mix crops and increase plant density to maximize production, in particular using crops occupying different strata such as fruit trees and vegetables. In these agroforestry systems, biological control may be enhanced because trees promote the predator guild by providing predators with 1) shelters to overwinter and against their own predators and 2) alternative prey (Ratnadass et al., 2012). Hence, we hypothesized that, compared to monocultures of vegetables, systems associating fruit trees and vegetables should be characterised by more predators and thus higher pest predation. However, the presence of trees can also have negative impacts on biocontrol because alternative prey can distract predators from consuming pests and because trees may support pest reproduction by generating a favourable microclimate. The present study focuses on the association between fruit trees and vegetables and investigates the impact of trees on the predator guild and in fine on biological control of vegetables pests. It is based on a comparison between experimental plots "vegetables + fruit trees" and the control "vegetables only".

## Material and methods

#### **Experimental plots**

Experiments were carried out in four experimental 50 m  $\times$  2 meter plots in Avignon, South-East France. Plots were either of type "fruit trees + vegetables", consisting in cabbages (*Brassica oleracea*) planted in the inter-row of an apple orchard (*Malus domestica*) (2 plots), or control plots "vegetables only" with only cabbages (2 plots). Each control plot was positioned less than 20 meter away from the "fruit trees + vegetables" plot to have a similar landscape on the two plots of the pair.

#### Sampling

To record all potential predator arthropods, each of the following measurements were repeated for 16 cabbages by plot (32 for each type of plots, differing from one session to the other) 6 times from April to June 2016. Terrestrial predators were sampled using pitfall traps, flying predators were monitored by 5 min-observations sessions and larvae were directly counted on cabbages. Predators were identified on the field. Predation rate was assessed by exposing sentinel prey on cabbages corresponding to 10 aphids *Myzus persicae* stuck on an adhesive paper card. It was computed as number of aphids eaten out of total stuck aphids.

#### Statistical analyses

Statistical analyses were carried out for each sampling session independently, using R software. We compared the occurrence of each predator group at each sampling cabbage between plots with fruit trees and control, using generalized linear models with a random plot effect. When predators were present, we compared their abundance or activity density between plots with fruit trees and control plots using Wilcoxon-Mann-Whitney non-parametric tests. We checked the effect of plot identity by comparing the two plots within each pair using the same test. We used the same procedure to compare predation rates between the two types of plots.

#### **Results**

#### Inventory of predator groups

Predators were classified according to taxonomy, sampling methods and stage in the following groups: Aranea spp., Opiliones spp., terrestrial Coleoptera spp., Formicidae, Chilopoda spp., Cecidomyiidae spp., Syrphidae spp. adults, flying Coleoptera spp., Coleoptera spp. larvae, Hemiptera spp. larvae, Acari spp., Thysanoptera spp., and Dermaptera spp.

#### Comparison of occurrence for each predator group

There were overall few significant differences between the two types of plots and these were always in the direction of a higher occurrence in the controls (Table 1).

For terrestrial predators, occurrence were significantly higher in controls for beetles for session 4 ( $\text{Chi}^2 = 3.61$ , p-value = 0.057), ants for sessions 1, 2 and 3 ( $\text{Chi}^2 = 20.49$ ; p-value = 56.10<sup>-6</sup>,  $\text{Chi}^2 = 7.98$ ; p-value = 0.0047;  $\text{Chi}^2 = 7.85$ ; p-value = 0.005, respectively), multipedes for session 5 ( $\text{Chi}^2 = 4.55$ ; p-value = 0.034) (Figure 1).

For flying predators, occurrence were higher in controls for Cecidomyiidae spp. only for session 1 (Chi<sup>2</sup> = 6.21; p-value = 0.013) and marginally for session 5 (Chi<sup>2</sup> = 3.39; p-value = 0.066).

For sedentary predators, occurrence were marginally higher in controls only for earwigs for session 4 and 6 (respectively  $\text{Chi}^2 = 3.52$ ; p-value = 0.061;  $\text{Chi}^2 = 3.11$ ; p-value = 0.078).

#### Comparison of abundances or activity densities for each predator group

A similar pattern was observed for the abundances and activity densities, with few significant results and higher values in control plots (Figure 1).

For terrestrial predators, activity-densities were higher in controls for spiders during session 5 (all plots: W = 488; p-value = 0,030 ; pair 1: W = 104.5, p-value = 0.55); for beetles for sessions 1 (all plots : W = 262; p-value = 0.002; pair 1: W = 56; p-value = 0.010 ) and 6 (all plots: W = 182; p-value = 0.011, pair 1: W = 42.5; p-value = 0.012) and almost significant for pair 2 during session 2 (W = 24, p-value = 0.060) and pair 1 in session 4 (W = 0.5; p-value = 0.054); ants during session 1 (all plots: W = 88; p-value = 0.0076 ; Pair 1: W = 36; p-value = 0.027 ), session 3 for pair 2 (W = 36; p-value = 0.05), session 5 for the two pairs (Pair 1: W = 63.5; p-value = 0.069; Pair 2 : W = 60; p-value = 0.48 ).

No difference was significant for flying and sedentary predators.

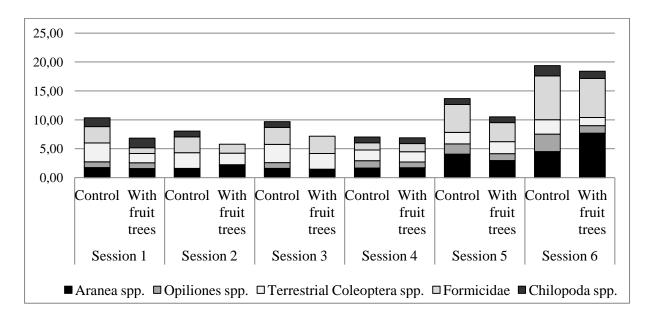


Figure 1. Mean abundance of terrestrial predators according sessions

#### Comparison of predation rate on sentinel preys

We did not notice any difference in predation rate for all sessions between the two types of plot except for the pair 2 for session 2 (W = 181; p-value = 0.029; Figure 2).

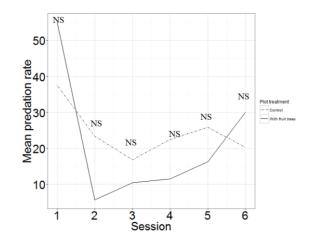


Figure 2. Mean predation rate for plots of types "vegetables + trees" (solid line) and control (dashed line) according to sessions.

#### Discussion

During this study, we did not notice strong differences between plots with fruit trees and controls, for the predator guild or for predation rate of sentinel aphids. Considering the predator guild, the few differences showed a higher occurrence or abundance of predators in controls than in plots with trees, which did not follow our assumptions. However, they are consistent with contradictory results found in the literature about agroforestry (Pumarino *et al.*, 2015). In plots associating vegetables and fruit trees, it is possible that generalist predators may share their time between the two kinds of plants, eating pests of vegetables and alternative prey from trees and thus be less present on the vegetables. However, predation rate was not higher for controls. It is possible that different predator groups consume sentinel prey in plots with trees and controls. A next step is to classify more precisely predator by refining taxonomic identification or grouping by functional traits and also to assess consequences on pests.

#### Acknowledgements

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# Soil matters: agronomic practices impacts on natural regulation of root pests of field vegetables

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Abstract: Natural regulation of crop pests by their antagonist community can help reduce crop losses and over-reliance on pesticides. Research has shownthat habitat manipulation at field and landscape scale can increase numbers of those antagonists over the growing season. Soil can also act as a reservoir for a wide range of natural enemies however manipulation of the soil habitat has often been overlooked. This study explores the impacts of agronomic practices on the soil and on the survival of the cabbage root fly Delia radicum, whose larvae cause serious damage to brassica crops. A wide range of naturally occurring organisms can help suppress the fly in a "top down" approach, such as predators, parasitoids and pathogens. Additionally, improved soil health has also been reported to contribute to pest suppression in a "bottom up" approach by fostering beneficial microorganism community, leading to stronger plant defenses. Two experimental long term rotations comparing organic and conventional practices were monitored over two years for fly eggs and pupae as well as their natural enemies. Field soils were characterized in the laboratory, revealing contrasting impacts of management on soil parameters. Further fly regulation experiments were carried out in growth room conditions with field soils. No clear differences in beneficial pest pathogens activity or bottom up control could be revealed between organic and conventional soil. Field monitoring showed however an overall positive impact of organic practices with a greater reduction of fly eggs and pupae compared to plots treated with pesticides, as well as an increase in activity density of their potential predators. Chemical crop protection inputs were shown to have a detrimental effect on those predators. Under the right management, soil can help foster functional biodiversity to help deliver ecosystem services such as pest regulation. Rebuilding sustainable and resilient agroecosystems will also need to include better soil husbandry.

Key words: natural regulation, *Delia radicum*, predators, soil management, organic vs conventional

# On the presence of Vespoidea on an Italian racecourse

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**Abstract:** Hymenoptera Vespoidea can represent a risk for humans in populated areas as they frequently nest near houses, within walls, or on trees in public parks. Moreover, their abundance can result in adverse encounters with people. In the present work we investigated the presence of Vespoidea in a racecourse in the city of Milan (northern Italy). In the past, the presence of nests of *Vespula germanica* under the ground has caused harm to humans and horses. The survey was conducted from May to October 2016 using 28 TAP TRAP<sup>®</sup> baited with beer and checked every 15 days. Specimens of only three species, *V. germanica, Vespa crabro* and *Polistes gallicus*, were captured in the presence of many burrows due to voles in the area that could facilitate the settlement of *V. germanica*, causing problems to horses and people, should be taken in account to limit the settlement of this species.

Key words: German yellowjacket, European hornets, urban pest, voles

## Introduction

Hymenoptera Vespoidea can represent a risk for humans in populated and urban areas. Wasps prefer to nest near houses, within walls, or at the bottom of trees in public parks. Frequently, wasp nests are difficult to localize as they can easily survive into small spaces, tunnelling into the ground or constructing their nests high into the trees. Wasps in great abundance near urban settings can result in adverse encounters with people and damages to economic activities (Beggs, 2001; D'Adamo & Lozada, 2007).

In the present work we investigated the presence of Vespoidea on a racecourse in the city of Milan (northern Italy), focusing on the three most abundant species *Vespa crabro* L. (European hornet), *Vespula germanica* (Fab.) (German yellowjacket) and *Polistes gallicus* L. The surveyed area is often highly frequented by people due to numerous events that take place throughout the years. In particular, in the past the presence of nests of the social vespid *V. germanica* under the ground has caused harm to humans and horses. This species is native to Eurasia and Northern Africa, where it is widespread (Archer, 1998). The species also invaded several regions, including New Zealand and Australia, South Africa, North America and Canada, arriving in Chile and Argentina (Edwards, 1976; MacDonald *et al.*, 1980; Tribe & Richardson, 1994; D'Adamo *et al.*, 2002). The German yellowjackets can interfere with other Vespoidea by preying on them (Beggs, 2001); some studies showed that, when population are numerous, *V. germanica* can threaten also birds and invertebrates (Farji-Brener & Corley, 1998; Moller & Tilley, 1989). Therefore, many studies have been conducted on the control of *V. germanica*, especially in the new colonized areas (Beggs, 2000; Harris & Etheridge, 2001; Sackmann *et al.*, 2001).

#### Material and methods

#### Study area

The racecourse of Milan is located in the western outskirts of the city (GPS: 45°29'09.0"N 9°06'40.5"E) and is currently used as a training track for horses. The area is confined on one side with a big city park, and on the other side with a densely populated quarter. In the area there are many ornamental trees and shrubs, as well as some old buildings for horses. The extension of the racecource is of about 2,000 sqm. The track, made of meadow and sand, includes an ellipse of 2,000 m long and a straight lane of about 1,000 m.

#### Wasp surveying

The presence of the Vespoidea within the area was surveyed from May to October 2016. A total of 28 traps were placed along the racetrack (24 along the elliptical lane and 4 on the straight one), with a distance of 80 m from each other. Traps were singularly georeferenced and data of the catches reported on a map with ArcView GIS<sup>®</sup>. TAP TRAP<sup>©</sup>, a cromothropic trap commonly used for mass-trapping a wide range of flying insects in different environments, was installed. Traps were baited with beer (200 ml) and hanged at 1 m height, hidden in the bush for not to annoy the horses. Beer was replaced every 2 weeks, and the insect content was stored in containers filled with 70% alcohol at the DeFENS laboratory of the University of Milan. In order to know the identity and composition of the samples specimen were separated, classified and counted. In addition, during each survey a visual inspection was made on the entire area looking for foraging wasps and nests.

#### **Results and discussion**

A total of 1,537 Vespoidea were captured including only the three species *Vespa crabro*, *Vespula germanica* and *Polistes gallicus*.

Figure 1 reports the mean number of insect collected at each sampling date. Specimen of Vespoidea were captured from mid-May to October. *V. crabro* and *V. germanica* were captured at all the dates of monitoring, whereas *P. gallicus* only in 6 out of 9 visits. The European hornet was always present in higher numbers than the other two species of Vespoidea at all sampling dates, with a mean of  $5.62 \pm 1.02$  per trap. A peak of its presence was registered at mid-July and at mid-August when the mean number per trap was  $9.93 \pm 1.28$  and  $9.86 \pm 1.42$ , respectively.

The German yellojacket was present at all sampling dates, but the mean capture of this species was very low (0.41  $\pm$  0.12), with a maximum of 1.04  $\pm$  0.38 specimen/trap at mid-August. *P. gallicus*, was found only from mid-June to mid-September with just 0.08  $\pm$  0.03 adults/trap.

Maps in Figure 2 show the spatial distribution and the species composition of the wasps into the traps. The European hornet was collected at the first sampling date in 28% of the traps and at the other dates in more than 80% of the traps. This species was therefore present on the whole area monitored. The European yellowjacket was present in 3% to 28% of the traps until the end of July, later it was captured in half the traps during August and early-September. Its occurrence started at the east-side of the track, later moving to the west. *P. gallicus* reached a maximum of 21% of the traps at the beginning of September. In this case the wasp was present only along the elliptical track, while no specimen was found on the straight track.

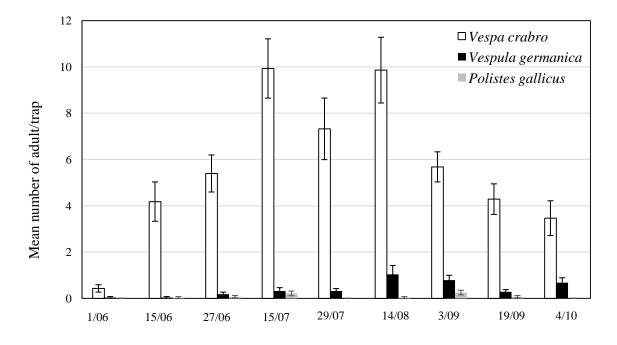


Figure 1. Mean number of the species sampled in the 9 surveys from May till October.



Figure 2. Maps showing the species composition of the catches in the traps for the 9 sampling dates.

The visual inspections conducted in the monitored area revealed the presence of burrows and tunnels due to voles that could facilitate the settlement of *V. germanica*. Moreover, numerous specimen of the European yellowjacket were seen exploring those burrows, expecially at the beginning of the season, and workers of this species were seen foraging the area throughout the period of the survey. However, no wasp nests were found during the monitoring season. A correct management of the racecource area should take in particular consideration the possible role of the voles in the settlement of *V. germanica*.

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# **Carabid composition in different agricultural landscapes**

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Abstract: Carabids are the most abundant and diverse group of beneficial insects inhabiting agricultural crops all over the world. They are known as bioindicators of ecosystem stability, since they can indicate the field quality caused by anthropogenic influence. In order to preserve carabid biodiversity in intensively managed agro-ecosystems, it is important to monitor their composition, abundance, dominance and diversity in agricultural landscapes. The carabid populations were investigated during the vegetation season 2015 in IPM perennial (orchard) and IPM annual (arable) agro-ecosystems. Altogether, 1612 individuals belonging to 38 species were collected. In perennial agro-ecosystem, 17 species were recorded, mostly belonging to genera Harpalus, Pterostichus and Calathus. In arable agroecosystem, 26 species were recorded with the most represented genera of Harpalus, Brachinus, Pterostichus and Amara. Only four species were registered in both agricultural landscapes. The orchard landscape can be characterized as having relatively low species richness in comparison with arable landscape. Presented results confirm that anthropogenically influenced agricultural landscapes generate different disturbance degrees in the carabid communities resulting in decreased biodiversity in long established landscape compared to variable annual landscape.

**Key words:** Carabidae, species composition, dominance, apple orchard, arable crops, intensive crop production

## Introduction

Ground beetles are considered to be one of the most importand group of ground dwelling insects in cultivated fields. Since they react sensitively to anthropogenical changes in habitat quality they serve as environmental bio-indicators (Thiele, 1977; Lövei & Sunderland, 1996; Kromp, 1999). Agricultural landscapes are highly influenced by anthropogenic disturbance and therefore represent unstable and devasteted biotope which has significant effect on carabids diversity (Miñarro & Dapena, 2003; Funayama, 2011; Baranova *et al.*, 2013).

The ground beetle assemblage in Croatian agricultural landscapes was recently explored in annual crops (Bažok *et al.*, 2007; Kos *et al.*, 2010, 2011, 2013; Gotlin Čuljak *et al.*, 2013; Drmić *et al.*, 2016) but there is a lack of such investigations in perennial crops. Different management types in perennial and annual agro-ecosystems could generate different disturbance degrees in the carabid communities resulting with changes in species biodiversity. In order to preserve carabids biodiversity in intensively managed agro-ecosystems, it is important to establish species composition, abundance and dominance in different agricultural landscapes.

#### Material and methods

Ground beetles were sampled in vegetation season 2015 in annual and perennial IPM landscapes by using epigeic covered pitfall traps (polythene pots:  $\emptyset = 12 \text{ cm}$ , h = 18 cm) half filled with salted water and addition of detergent. In annual (represented with arable crops) and perennial (represented with apple orchard) landscapes the carabids were collected from 1<sup>st</sup> May until 10<sup>th</sup> September. Traps were inspected on a weekly basis and the species were identified by standard keys (Freude *et al.*, 2006).

The dominance values of carabids presented in percentage shares of a particular species in community was calculated according to Tischler (1949) as follows: eudominant (10-100%), dominant (5-10%), subdominant (2-5%), recedent (1-2%) and subrecedent (< 1%). The data on carabid composition and abundance collected in different agricultural landscapes were analyzed and mutually compared.

#### **Results and discussion**

Altogether, 1612 individuals were collected 1429 in annual and 183 in perennial landscape. When determined, altogether 38 different species were distinguished (Table 1). The highest number of collected species belonged to the genus *Harpalus* (6 species), *Ophonus* (4 species) and *Pterostichus* (4 species). The most abundant species, with the share of 33% in the total catch was *Poecilus cupreus cupreus* Linné, 1758 followed by *Brachinus psophia* Audinet-Serville, 1821 (19%) and *Pterostichus melas melas* Creutzer, 1799 (15%). These three species account for 67% of the total catch and belong to the group of eudominant species. Two species were characterized as dominant (*Anchomenus dorsalis* Pontoppidan, 1763 and *Pterostichus melanarius melanarius* Illiger, 1798). Subdominant and recedent species were both represented with three species while 28 species were characterized as subrecedent.

When analyzed separately, in perennial agro-ecosystem 17 species were recorded and the most species belonged to genera *Harpalus*, *Pterostichus* and *Calathus*. Since there is a lack of ground beetle investigations in perennial agro-ecosystems in Croatia, the composition of recorded species was compared with ground beetle fauna in neighboring Hungary (Kutasi *et al.*, 2014), showing correspondences in findings. In arable agro-ecosystem, 26 species were recorded with the most represented genera of *Harpalus*, *Brachinus*, *Pterostichus* and *Amara*. The ground beetle community of arable agro-ecosystem was expected and corresponds with results of similar investigations in Croatia (Kos *et al.*, 2010, 2011, 2013; Bažok *et al.*, 2007; Drmić *et al.*, 2016) and abroad (Bukejs & Balalaikins, 2008; Woodcock *et al.*, 2010; Baranová *et al.*, 2013). Only four species were registered in both agricultural landscapes.

The species composition of ground beetle fauna in different agricultural landscapes can be characterized as the basic dominant ground beetle fauna which is uniform across Europe (Thiele, 1977). Usually, no more than 10 to 40 species are active in a habitat in the same season (Lövei & Sunderland, 1996) which corresponds with results of this study (38 species identified). The species richness in perennial landscape can be characterised as relatively low in the comparison with annual landscape which could be linked to more intensive pest protection measures than in perennial landscape. Presented results suggesst that anthropogenically influenced agricultural landscapes generate different disturbance degrees in the carabid communities.

Species name	Annual	Perennial
Calosoma (Campalita) auropunctatum auropunctatum Herbst, 1784	+	
Carabus coriaceus coriaceus Linné, 1758		+
Brachinus (Brachinus) crepitans Linné, 1758	+	
Brachinus (Brachinus) psophia Audinet-Serville 1821	+	
Brachinus (Brachynidius) explodens Duftschmid 1812	+	
Clivina fossor fossor Linné, 1758	+	
Asaphidion curtum curtum Heyden 1870	+	
Trechus (Trechus) quadristriatus Schrank, 1781	+	+
Chlaenius decipiens L. Dufour, 1820		+
Anisodactylus binotatus Fabricius, 1787		+
Anisodactylus (Pseudanisodactylus) signatus Panzer 1796	+	
Harpalus (Harpalus) affinis Schrank, 1781	+	
Harpalus atratus Latreille, 1804		+
Harpalus (Harpalus) dimidiatus P. Rossi, 1790	+	+
Harpalus (Harpalus) distinguendus distinguendus Duftschmid, 1812	+	
Harpalus rubripes Duftschmid, 1812		+
Harpalus (Pseudoophonus) rufipes DeGeer, 1774	+	+
Ophonus azureus Fabricius, 1775		+
Ophonus gammeli Schauberger, 1932		+
Ophonus parallelus Dejean, 1829		+
Ophonus sabulicola Panzer, 1796		+
Stenolophus (Stenolophus) teutonus Schrank, 1781	+	
Microlestes minutulus Goeze, 1777		+
Agonum (Amara) viridicupreum viridicupreum Goeze, 1777	+	
Anchomenus (Anchomenus) dorsalis Pontoppidan, 1763	+	
Abax (Abax) parallelepipedus parallelepipedus Piller & Mitterpacher, 1783	+	
Abax (Abax) exaratus Dejean, 1828	+	
Poecilus (Poecilus) cupreus cupreus Linné, 1758	+	
Pterostichus transversalis Duftschmid, 1812		+
Pterostichus (Feronidius) melas melas Creutzer, 1799	+	+
Pterostichus (Morphosoma) melanarius melanarius Illiger, 1798	+	
Pterostichus (Platysma) niger niger Schaller, 1783	+	
Calathus (Calathus) fuscipes fuscipes Goeze, 1777	+	+
Calathus (Neocalathus) ambiguus ambiguus Paykull, 1790	+	
Calathus melanocephalus melanocephalus Linné, 1758		+
Amara (Amara) aenea Degeer, 1774	+	
Amara (Amara) ovata Fabricius, 1792	+	
Amara (Amara) similata Gyllenhal, 1810	+	

Table 1. List of ground beetle species in annual and perennial agricultural landscape

Therefore, the results of study contribute to better understanding of ground beetle communities in intensive agricultural landscapes in Croatia and present valuable contribution for conservation programs.

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## Mites' functional biodiversity in terraced vineyards of Cinque Terre National Park

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**Abstract:** Cultural tendencies to simplify ecosystems caused the loss of the genetic diversity as represented by species but also as structural and functional biodiversity. Along the Ligurian Sea, in Cinque Terre National Park, terraced hills managed for viticulture are declared UNESCO Cultural Heritage site. As contribution to knowledge of the biodiversity, the present study on mites can be considered a key reading as bioindication in this agroecosystem to assess quality in viticulture, stability in soils and regulation of pests and diseases.

Mite groups, expressing high functional diversity, are usually the most abundant groups of microarthropods in soil and can highly colonize plant in temperate ecosystems. The study started by the evaluation of the acarofauna on grapevines in five vineyards, from 50 to 400m ASL (Manarola, Corniolo, Fossola, Groppo, Porciana). On the whole, grapevine pruned woods and leaves were sampled from autumn 2007 to autumn 2008. Five samplings of soil mite communities were taken from autumn 2008 to autumn 2010. For each site, the epigeic and edaphic acarofauna was determined at family or specific level. All abundances were determined and analyzed by Anova; the community structure was characterized on the basis of different functional mite groups: detritivorous/saprophagous, mycophagous, phytophagous, predators, others.

In all leaves sampled, very high presence of predator mites, mainly phytoseiids, was registered: they represented about 95% of the acarofauna. As regards the phytoseiids, the adults represented about 60% of population, the most frequent and abundant species was *Kampimodromus aberrans*, followed by *Typhlodromus pyri*, *T. finitimus* and *T. exhilaratus*. The presence of tydeiids, mites with less specialized feeding habits, was about 6%.

Soil mite community represented the main group (56.6%) of total microarthropods collected (> 6,700 specimens). The group of detritivorous/saprophagous was the most represented (> 70%), while mycophagous and predators ranged between 12 and 16%.

By considering soil mites, the group of oribatids was the most abundant and affected by sampling sites ( $F_{4,111} = 3.38$ ; P = 0.012). Twenty-seven families and 42 species of oribatids were identified, 20 families of Prostigmata and 9 of Mesostigmata with 29 species. Both density, diversity and functional diversity of Acari here registered were higher than in similarly managed agroecosystems.

Considering both aerial and edaphic levels, the analysis of functional mite groups performed in Cinque Terre Park, can be considered an efficient tool in the evaluation of quality and management of the ecosysytem. Key words: Soil mites, epigeic acarofauna, mite feeding habits, bioindication, ecosystem services

## Introduction

Viticulture in terraced coastal zone with particular climatic conditions can provide high levels of biodiversity, both at soil and plant levels, with benefit for ecosystem services and the aim at producing high quality products rather than at producing. Crucial study lines are finalized to analyze ecosystem services for growth and health of the grapevine such as soil fertility and stability, pest and weed control as well as grape quality in relation to biodiversity.

In the frame of the LABTER project (financed by MiPAAF Ministry of Agricultural, Food and Forestry Policies) that aimed to identify the main features for assessment of biodiversity in the protected area of Cinque Terre Park (Liguria, Italy), this study was focused on the acarofauna as key group for environmental quality assessment in vineyard ecosystem. Mesofauna comprises the middle links of soil food webs affecting nutrient cycling and soil fertility (Culliney, 2013). Furthermore, mites exert several roles on plant, both as pest and beneficial fauna. Biodiversity then doesn't regard only the species richness but goes "from genes to ecosystems" (Solbring, 1991). The richness and functionality of mite communities can be included among characteristics and traits useful to explain ecosystem properties. In the above-ground compartment, biodiversity and ecosystem processes are importantly influenced by intra- and interspecific competition and habitat exploitation; in soil, these processes are sensibly affected by dynamics and interactions in the soil communities and properties. The understanding of the nature of biodiversity-ecosystem services relationship and the possible effects of biodiversity loss on the delivery of ecosystem services is therefore critical and worth to deepen (Cardinale *et al.*, 2006).

## Material and methods

#### Study area

The Cinque Terre (Liguria, Italy  $-44^{\circ}06$ 'N 9°43'E) is a coastal zone with terraced hills managed for viticulture and declared UNESCO Cultural Heritage site. In this context, five different vineyards were selected in the areas of Manarola (45 m ASL), Corniolo (155 m ASL), Fossola (178 m ASL), Groppo (253 m ASL), Porciana (391 m ASL) with same exposition and management. In each study site, some chemical and physical parameters were analyzed for the characterization of the soils. Soils of the areas were classified according the USDA classification system: sandy-loam soils were in Corniolo, Fossola and Groppo; loam in Porciana and Manarola. The organic matter (SOC) was everywhere low, between 2 and 5%.

#### Biodiversity and functional diversity of soil mites

Five samplings of soil mite communities were taken from autumn 2008 to autumn 2010. In each site, 5 soil samples (about 250 cm<sup>3</sup>) were intra-row collected; the mesofauna was extracted with Berlese-Tullgren funnels and, with the exception of Prostigmata, adult mite specimens were identified to species. The abundance of soil mites was determined and analyzed by Anova; biodiversity was evaluated by the main ecological indices – Shannon Wiener diversity index (H), the evenness index (J), species richness (R) and others – calculated with PAST software (Hammer *et al.*, 2001). As indicator of functional diversity, the mite species were attributed to predatory and non-predatory habits; non-predatory mites

were assigned, on the basis of their respective trophic role, as detritivores (macrophytophagous, microphytophagous), saprophagous, mycophagous and non-specialized feeders/unknown (others) based on Krantz and Walter (2009).

#### Mite communities on plant

On the whole, 30 grapevine pruned woods and 50 leaves were sampled in each of the cited sites from autumn 2007 to autumn 2008 (4 samplings). Simultaneously, control samplings were carried out in abandoned vineyards next to the selected ones. The mites collected were counted, identified and, on the basis of their main feeding source, assigned to the following groups: predators, phytophagous, not specialized feeders.

## **Results and discussion**

Mite community represented the main group (56.6%) of total soil microarthropods collected (> 6,700 specimens). Twenty-seven families of Oribatida (42 species), 20 families of Prostigmata and 9 of Mesostigmata (29 species) were determined. The feeding group of Acari, as functional identity and animal diversity, can be informative concerning the ecosystem functioning, below- and above-ground, better than species-based indices (Gagic *et al.*, 2015).

As shown in the Figure 1A, the detritivorous and mycophagous mites were the most representative in Porciana, predator species were less abundant in Fossola and Manarola sites while phytophagous mites were only in Groppo. All trophic categories were adequately represented by carrying on their functional ecosystem service and, eventually, recovering from the stress due to agronomic activities. In according to Wallwork (1983), the oribatids are the numerically dominant group of Acari (N = 1763) and the most important in decomposition processes. They were affected by sampling site ( $F_{4,111} = 3.38$ ; P = 0.012). On the whole, in Porciana, the highest abundance and the highest biodiversity were registered, in the community of oribatids (Figure 1B) and the Shannon index (H) maximum value (H = 2.1). Different micro-area of vineyards and small heterogeneous habitat can undoubtedly favor the development of a multispecies community. The terraced management implies also agronomic practices without mechanical disturbance and long time landscape conservation: species with low rate in population growth, usually frequent in relatively stable environments (i.e. Suctobelbidae and Brachychthoniidae) were present, unlike the opportunistic species which increase after disturbance (Maraun *et al.*, 2005).

On leaf, there are three functional groups mainly represented (Figure 2A); 95% of the acarofauna was represented by predatory mites. Tydeiids, mites with less specialized feeding habits, represented about 6%. While phytophagous mites were sporadic, in particular in the productive vineyards. The phytoseiids were the most abundant predators. Figure 2B shows the distribution of the phytoseiid species: *Kampimodromus aberrans* was dominant in productive vineyards while *Typhlodromus exhilaratus* in abandoned ones. (Figure 2B). On the pruned woods, the density of phytoseiids was  $0.5 \div 1.75$  specimens/10cm.

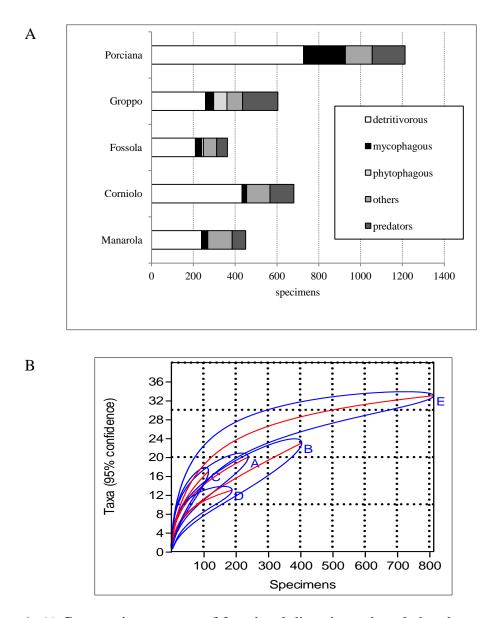


Figure 1. A) Community structure of functional diversity and total abundance of soil mites in study sites. B) Rarefaction curves of oribatid community diversity at different sampling sites: A = Manarola, B = Corniolo, C = Fossola, D = Groppo, E = Porciana (see Hammer*et al.*, 2001).

The study provides: insights into the mechanisms linking biodiversity and ecosystem functioning in animal communities; responses due to the identity and dominance patterns of the trait composition together with the richness of species *per se*. The ecosystem service by not-predator mites eventually can be preserved in the soil but the resilience and biodiversity structure changes.

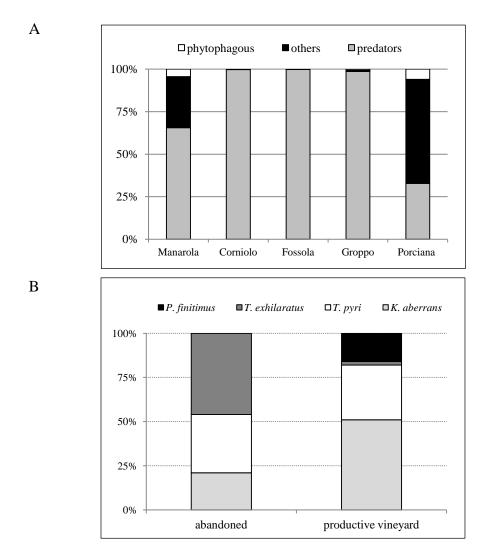


Figure 2. A) Community structure of functional diversity of mites on plant in the different sites. B). Frequency of phytoseiids in the productive and in the abandoned vineyards.

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## Landscape scale management of insect populations

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**Abstract:** Ecological approaches to pest management often depend on the maintenance and mixing of two or more insect populations. These systems frequently involve the establishment of favourable habitats for the populations that we wish to maintain in the landscape but little is known about how best to manage such habitats within the landscape. In this paper we describe a modelling approach to simulate the spatial dynamics of interacting populations and their response to landscape management strategies.

Key words: landscape management, pest regulation, resistance management, WCR, maize

## Introduction

Ecologically based pest management systems often depend on the maintenance and mixing of different insect populations. For example biological control systems rely on the mixing of pest and natural enemy populations, and insect resistance management (IRM) involves the mixing of susceptible and resistant pest populations. These systems frequently involve the establishment of favourable habitats to support the insect populations that we wish to maintain in the landscape. Using resistance management of Western Corn Rootworm (WCR), *Diabrotica v.v.*, on *Bt* maize (Gassmann *et al.*, 2011) as an example, we explore the role of landscape management in pest regulation.

## Material and methods

A spatially explicit population model of WCR was constructed using the AgBioscape modelling system which has been developed for the simulation of dynamic interactions between farming landscapes and multiple populations of natural and semi-natural species that inhabit them, including crop pests. AgBioscape couples a discrete time approach to modelling population dynamics with a landscape simulation model to simulate both landscape structure and composition, including the dynamics of crop sequences, crop management and other land use changes. In pursuing a spatially explicit approach, we assume the organisms inhabiting the system belong to a single regional population that is comprised of multiple local populations, defined spatially on a 2-dimensional lattice representing a regional landscape. In defining local populations we assume that they cover an area, defined by the lattice, that is at sufficiently small to permit the population to fully mixed and that this may vary between species according to the size and mobility of individuals, 10 m x 10 m in the case of WCR which encompasses the mean daily foraging distance of a WCR beetle. Many species inhabiting agricultural ecosystems have complex life-cycles with age or stage-specific differences in their response to abiotic conditions. In cropping systems the organisms are exposed to abrupt changes in the environment, often leading to discrete demographic events. The temporally discrete stage specific conditions that this invokes are well represented by a

stage/age structured matrix projection approach to modelling the local population dynamics. The population and landscape models are coupled by making vital rates of the life-history models dependent on habitat type. For example, in the model of WCR (the target pest), the survival and fecundity of insects exposed to *Bt* and non-*Bt* maize were selected to represent either susceptibility or resistance to the *Bt* toxin (Hoffmann *et al.*, 2015).

In addition to demographic processes that take place at a local level, regional populations and their dynamics are sensitive to dispersal and its effect on local immigration and emigration. Modelling dispersal and the connectivity between local populations is also key to determining the mixing of populations and their exposure to habitats within the landscape.

By managing the distribution of habitats within a landscape the exposure of insect populations to habitats of different type and quality may be affected. Using the WCR – Bt maize model we are able to conduct simulation experiments to investigate the effect of the landscape management on the development of insect resistance to Bt-maize through exposure to Bt crop and non-Bt refuge habitats.

#### Landscape model

For the simulations, spatially continuous landscapes, consisting of four square *Bt* maize fields were modelled. Each field contains a single refuge of non-*Bt* maize occupying 25% of the field area in which WCR escape exposure to *Bt*. Two different refuge shapes were modelled; a square block, and a rectangular strip with an aspect ratio of 1:4. The four fields that make up a landscape all have refuges of the same shape. However with the strip refuges, the orientation of the strips can vary (Figure 1). The orientation of any refuge strip remains fixed and does not change with time. The landscape was modelled as a torus, where WCR beetles leaving one side of the field are match by a similar number entering the opposite side.

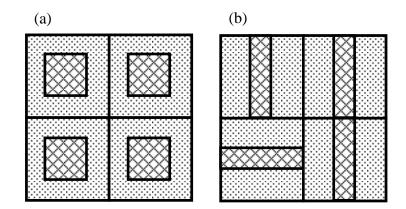


Figure 1. Examples of maize landscapes consisting of four fields; (a) with square refugia, (b) with strip refugia.

#### Pest management

The WCR population in each of the fields was managed independently. The abundance of WCR in the refuges was controlled by simulating the effect of a soil applied insecticide whenever an action threshold was reached. The abundance of adult WCR beetles was monitored weekly, and if the action threshold was reached, a larvae mortality event occurred before any density dependant mortality in the following year. Survival of this event was set to 30%. The action threshold value set to 4 adult beetles per square meter.

Pest management for the Bt maize fields was assumed to be by seasonal rotation; whenever root damage was detected in a continuous Bt maize field, the field was rotated to soybean. Root damage was assumed to occur when the density of WCR larvae is greater than 3 larvae per plant after the effects Bt toxin and density dependent mortality.

#### Simulations

Simulations were run for each strip and block refuge strategy for a range of field sizes (Table 1). Ten replicate simulations were run for every field size and each simulation was run for 50 years. Simulations were started at the beginning of the winter with an initial egg density on the fields of 300 eggs/m<sup>2</sup> resistance allele frequency of 0.001. The durability of each refuge strategy and field size to withstand the development of resistance was assessed in terms of the number of years for *Bt* resistance allele frequency to reach 25% or 50% within the regional WCR population, while the effect of this on the crop was assessed in terms of the time take for significant root damage (equivalent to 3 larvae/plant) to appear.

Field size (m x m)	Square (m x m)	Strip (m x m)
120 x 120	60 x 60	30 x 120
240 x 240	120 x 120	60 x 240
360 x 360	180 x 180	90 x 360
480 x 480	240 x 240	120 x 480
600 x 600	300 x 300	150 x 600
720 x 720	360 x 360	180 x 720
840 x 840	420 x 420	210 x 840
960 x 960	480 x 480	240 x 960

Table 1. Field and refuge sizes used to construct the landscapes.

#### **Results and discussion**

With a 25% refuge there is an optimum refuge size and associated separation distance between refugia for slowing the development of resistance to *Bt* maize, assuming 100% refuge compliance, which is achieved at field size of 24 ha (Figure 2a). Square refuges performed better than strip refuges in slowing the development of WCR resistance, although strip refuges were slightly less sensitive to changes in field size (Figure 2a). Square refuges were particularly effective at delaying the development of high levels of resistance (> 50% allele frequency) when combined with large field sizes (Figure 2b). However, it is clear that the onset of root damage by WCR is determined by the lower level of resistance (Figure 2a and 3). These results illustrate the importance of landscape in managing the bottom-up regulation of pest populations in a system reliant on a resistant crop variety for control. However, similar results have been obtained for systems using other control measures such as crop rotation and pesticide application, and also those that rely on supporting natural enemy populations to provide top-down regulation (Begg *et al.*, 2016).

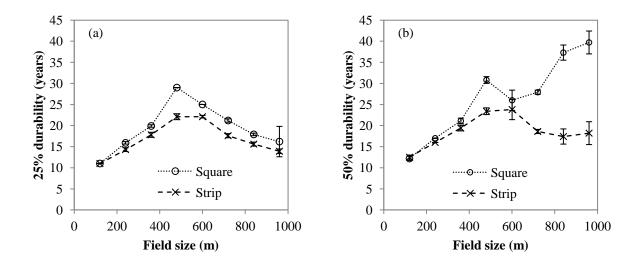


Figure 2. The durability of landscapes with square or strip refuge strategies at a range of field sizes assessed by the time taken to reach a resistant allele frequency in WCR of (a) 25% and (b) 50%. Error bars indicate standard deviation across replicate simulations.

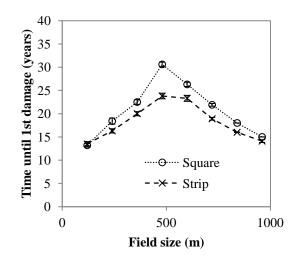


Figure 3. The effect of refuge strategy and field size on the time taken for significant root damage to appear. Error bars indicate standard deviation across replicate simulations.

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## EcoOrchard – collecting existing knowledge and generating new knowledge on functional biodiversity of organic orchards

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**Abstract:** Organic fruit growers often suffer economic losses due to insect damages. The available natural pest control products are not always effective; and most important: many organic fruit growers would prefer not to use any pesticide at all. EcoOrchard, a CORE Organic Plus project (2015-18) aims to collect existing knowledge and generate new knowledge in order to use Functional AgroBiodiversity (FAB) successfully in orchards. This includes experimental trials in seven countries on the potential of inter-row flower strips for control of key pests and natural enemy augmentation, the establishment of the EBIO-Network as a European-wide network of stakeholders for collecting, sharing and improving scientific and practical knowledge and experience in FAB management, the development and testing of simple FAB assessment tools for use on-farm by growers and advisors, and finally, using a participatory approach to learn about potential constraints that may hamper the adoption of innovative tools and how to solve these constraints by iterative reevaluation.

Key words: inter-row flower strip, natural enemies, Dysaphis plantaginea, Cydia pomonella

## Introduction

Organic fruit growers often suffer economic losses due to insect damages. The available natural pest control products are not always effective; and most important: many organic fruit growers would prefer not to use any pesticide at all. The project EcoOrchard "Innovative design and management to boost functional biodiversity of organic orchards" is a CORE Organic Plus project (2015-18) aiming to collect existing knowledge and generate new knowledge in order to use functional agrobiodiversity successfully in orchards (Sigsgaard, 2016; Sigsgaard *et al.*, 2016). Tracking innovative practices is one element of this including

interviews with growers and advisors and the establishment of the EBIO-Network as a European-wide network of stakeholders for collecting, sharing and improving scientific and practical knowledge and experience in functional agrobiodiversity (FAB) management. Further simple FAB assessment tools for use on-farm by growers and advisors have been developed and tested. Experimental trials have been established in seven partner countries on the potential of inter-row flower strips for control key pests and to augment natural enemies. Finally, using a participatory approach to learn about potential constraints that may hamper the adoption of innovative tools and how to solve these constraints are assessed through iterative reevaluation.

#### Tracking innovative FAB practices

More than 50 advisors and more than 100 growers (i.e. the EcoOrchard-stakeholder network) in the participating countries were interviewed for their knowledge and experiences on techniques for FAB management. The implementation of the ten most known or preferred methods ("Top- ten") was characterized for each country, revealing a high diversity between countries (Fernique *et al.*, 2016).

#### **EBIO-Network**

Literature collection from all partners resulted in 200 priority papers, reports and presentations on functional agro-biodiversity, available also on the EBIO-Network portal. Analysis of this literature is ongoing and will supplement the knowledge gained from the interviews. It is planned to summarize the analysis of this literature in a comprehensive review, especially on the results of the last five years and including the results of the practical knowledge analysis of the interview data. Online EBIO-network (European Biodiversity Orchard-Network) is available at http://ebionetwork.julius-kuehn.de/. The current content will be supplemented by material provided and developed by partners during the project period. The publishing of stakeholders' data still need their approval or their active registration at the thematic portal (Herz *et al.*, 2016).

## Common participatory methods for FAB assessment

In 2015, several methods to assess functional biodiversity have been chosen based on our previous experience with the methods and literature. We tested and set up in 1-2 orchards for each of the 3 countries concerned and compare them for different performance criteria (time, material or skill needed, information provided etc.). (i) visual observation, (ii) beating, (iii) sentinel preys or (iv) cardboards were considered as potentially feasible for farmers and presented to them during workshops (in France, Sweden, Denmark) or in the field directly.

At the beginning of the 2016 field season, interested and motivated farmers were asked to choose at least one method to use in their own orchard. Partners initially trained farmers at workshops or in the field, and provided all material needed. A preliminary booklet for partners and farmers was produced and translated to partner country languages. Based on experiences from 2016 they will be revised and made available on the EBIO-Network platform. The European network of involved farmers is getting established, with mostly organic farmers but also some integrated farmers. Winter 2016-17 a questionnaire is being completed about farmers' opinion of using the method(s), information obtained and any changes in farmers' practices.

A short didactic video was produced to inform the methods to farmers and advisors: https://www.youtube.com/watch?v=ahBsb-nA2AM&feature=youtu.be.

## New collaborative experimental trials and new orchards designing actions

We made two principal flower mixtures targeting our functional-agro biodiversity (FAB) criteria, a basic and complex mixture which will be tested in our field trials. Two types of field trials were set up considering botanical or entomological/botanical aspects of wild these flower strips (Wäckers & Van Rijn, 2012). Thus we will test both mixtures botanically (plant species richness, establishment, botanical successions) under the specific condition of orchards (nutrient rich soils, shaded by trees, greatly altered by machinery). We have installed these field trials in seven partnering countries, analysing the impact on natural pest control in relation of the botanical resources (with and without flower strips). Accordingly we have developed specific assessment field protocols and guidelines for entomology, botany and management practices.

The field trials data about the impact of flower strips on natural pest control of aphids and other pests of 2016 are currently being analysed. Spring recordings showed that most flower strips are still improving their floral qualities (number, quantity of sown plants). A good establishing of strips have been found in three countries (Be, CH and It) and moderate (poor to medium/good) in four countries (DK, D, PL and S). Up to now, we have got the impression that the establishment of strips in Northern-European countries could be more challenging due to the climatic conditions, and therefore it needs still more adapted mulching regimes (later dates and maybe fewer cutting dates). Thus we are anticipating these topics, e.g. using an indicator plant- list to optimise mulching dates.

#### Seed mixtures

Selection criteria of plant species for flower strips include good success in establishing under orchard conditions (adapted to orchard soils and shadowing by trees), durability and stability over 8-10 years of flowering species diversity, permanence and persistence of flowering key species (important for functional biodiversity) and significant impact to increase populations of key beneficials of apple pests, thus preferring species with short corolla blossoms and well accessible nectar and pollen sources.

The composition of the seed mixture aims to achieve a competitive and over 8-10 years perennial flower strip in the orchard alley way with a long flowering period with plants supporting key beneficials with respect to FAB. The strip must be compatible with 3-4 times cutting/mulching per year. This requires among other that the plant community is tolerant to regular mulching -3-4 times a year and is composed of bi-annuals and perennials, preferring ecotypes or wild form species and including some grass species typical for this species community which have to stabilize the plant community in mid-term.

## Learning from all sides and dissemination

Informal networks of growers, advisors and researchers have been built in France, Denmark and Sweden, through the workshops and the deep exchanges they fostered. A second workshop round with the same participants in winter 2016-2017 to share their experiences about the monitoring methods they decided to test in 2016. This should reinforce the community feeling and the installation of lasting exchanges, built through the EBIO-Network support.

Knowledge has been shared between the participants and the organizers of the workshops: about the different representations of what FAB is, about what are the practices to favor biodiversity and about FAB monitoring methods. Farmer's representations of FAB remains to be analyzed in details, but it already reveals large diversity, and some surprising

facts: – some conceptions are widely shared and rather expectable (FAB relates to beneficial insects, to ecological habitat, to natural elements like hedges or inter-row grass), some reveals a conception deeply linked to human-nature links (FAB relates to landscape planning, to local

Workshops reveal also a large diversity in the management styles of FAB. For some participants FAB management should partially and sometimes even totally be delegated to a specific person, for others the FAB management by the grower himself. If preliminary results are confirmed, we should foresee that FAB monitoring methods will not be the only way to invite farmers to use FAB, we will also have to work on on-farm and perhaps between farms organizational aspects of FAB monitoring.

Preliminary observations show that levers to control FAB and make it work for farmers are not all in the sole hand of the farmers, according to their own feelings as some require coordination among farmers, or between farmers and other landscape actors. If such different levels of *controls* are confirmed, we should foresee that FAB monitoring methods will not be the only way to invite farmers to use FAB, and some coordination actions should also be proposed. These conclusions might be tested during the next workshop round.

### Acknowledgements

political choices concerning land use etc.).

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## Key parameters for the management and design of field margins aiming to the conservation of beneficial insects

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**Abstract:** Edges of vegetation on crop fields may increase the abundance and diversity of natural enemies and pollinators. This study emphasizes some aspects on plant management. Germination, coverage and blossoming were registered on several plant species. Germination and plant emergence showed a great variation among species. High coverage plants (e.g. *Coriandrum sativum*) relegates small ones (e.g. *Salvia verbenaca* and *Silene vulgaris*) to the understory. *Borago officinalis* and *Echium vulgare* had extended blossoming periods, while *Coriandrum sativum* and *Vicia sativa* had short and peaked ones; *Diplotaxis catholica*, *S. verbenaca* and *S. vulgaris* showed an intermediate pattern. Guidelines for the choice and management of plant species are provided.

Key words: floral margins, beneficial insect, natural enemies, bees, biodiversity

## Introduction

The management of natural vegetation in agricultural land is utterly important because wild plants may serve both as eco-systemic service providers (e.g. pest control or pollination), and as reservoirs for crop pests and diseases (Willmer, 2011). The services plants provide vary with species, depending on factors such as their fitness and phenological state. In spite of the importance of plants as service providers, little information is available for their management and the design of vegetation margins. This work aims to emphasize some key parameters (e.g. germination rates, coverture and blossoming) to take into account when designing vegetation margins for the conservation of natural enemies and pollinators.

## Material and methods

The germination of eight plant species (Table 1) was assayed in laboratory and field conditions. This plant mix aimed to provide abundant floral resources, different floral structures and extended blossoming periods. In the laboratory assay, 30 seeds of each plant species were set up in eight Petri dishes with moist cotton and placed in a plant growth chamber (Binder KBWF-750, Tuttlingen, Germany) at 25 °C and 75% RH. The plates were checked every 3-4 days during 30 days and the emerged seeds were counted and removed. This experiment was repeated three times for each plant species. The assays of germination in field conditions were conducted in four localities in the Region of Murcia (SE Spain). In each locality, a strip of approximately 100 m<sup>2</sup> was sown manually in autumn using the eight plant species (Table 1). Enough seeds were used to achieve densities of about 5 plants per square

meter for the medium size species and 10 for the small size, using germination rates from preliminary trials (Table 1). The emergence of plants in each strip was estimated by counting the number of seedlings in a  $1 \times 1$  m square on the first week of January. This procedure was repeated three times randomly on each of the four margins. Three more assessments were carried out in every locality, from February to April, to assess the abundance of each plant species using the same procedure as for the plant emergence. The percentage of coverage and individuals in bloom of each plant species was estimated every one or two weeks from January to July within a 2 x 2 m square. The sampling was repeated three times for each margin at each date.

Table 1. Plant species assayed. Parameters used for the sowing of plant species to obtain the desired plant densities (Plants/m<sup>2</sup>). \*Experimental field emergence from preliminary trials.

Plant species	N seeds/g	Grams/m <sup>2</sup>	N Seeds/m <sup>2</sup>	%Emergence*	Plants/m <sup>2</sup>
Borago officinalis L.	51	0.230	12	42.3	5
Chrysanthemum coronarium L.	600	0.023	14	36.0	5
Coriandrum sativum L.	64	0.489	31	32.0	10
Diplotaxis catholica (L.) DC.	11,583	0.022	250	4.0	10
Echium vulgare L.	280	0.298	83	6.0	5
Salvia verbenaca L.	526	0.317	167	6.0	10
Silene vulgaris (Moench.) G.	1,353	0.064	86	11.6	10

## **Results and discussion**

All plant species showed a higher percentage of germination in the laboratory than of emergence in the field (Figure 1). In general, the emergence in the field was reduced from 16% to 95% in relation to the germination values registered in the laboratory. *Silene vulgaris, Coriandrum sativum, Vicia sativa, Salvia verbenaca* and *Borago officinalis* were the plants with the highest emergence. *Echium vulgare* and *Diplotaxis catholica* showed a low emergence both in the laboratory and in the field trials. The number of plants emerged in the field was similar to the predicted density according to the parameters in Table 1, with some extreme exceptions such as *S. vulgaris* that almost tripled the expected densities and *C. coronarium* that did not emerge in the field (Figure 1).

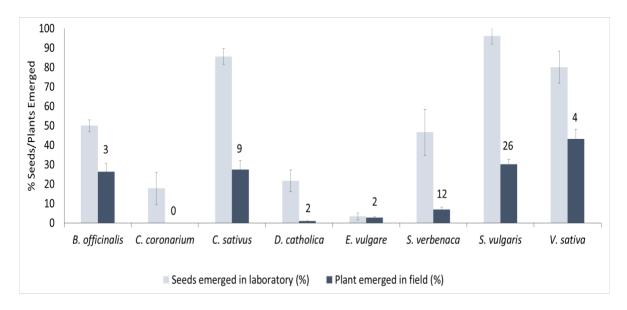


Figure 1. Percentage of seeds germinated in the laboratory and emerged in the field (Number of seeds emerged/Number of seeds sown). The figures on top of the bars represent the average number of plants per  $m^2$  emerged in the field.

During the field trial, the number of plants in mixed edges generally decreased over time, although in some species (i.e. *B. officinalis, E. vulgare* and *Salvia verbenaca*) it stayed almost constant (Figure 2). *Coriandrum sativum, B. officinalis* and *V. sativa* were the species with the highest coverage; in contrast, *D. catholica* and *E. vulgare* had the lowest values. *Silene vulgaris* was relegated to the understory by the bigger plants and its coverage did not increase until the rest of the species started to recede (Figure 3A).

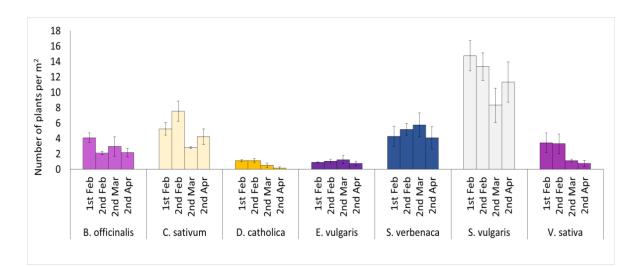


Figure 2. Abundance per  $m^2$  (Number of plants/SE) of each plant species on several sampling dates (1st and 2nd are first and second week of the month).

Some plant species such as *B. officinalis* and *E. vulgare* had extended blossoming periods, while others (i.e. *C. sativum* and *V. sativa*) had short and peaked ones. The percentage of individuals of *S. verbenaca* and *S. vulgaris* in bloom started to increase when the rest of the plants lowered their coverage (Figure 3B).

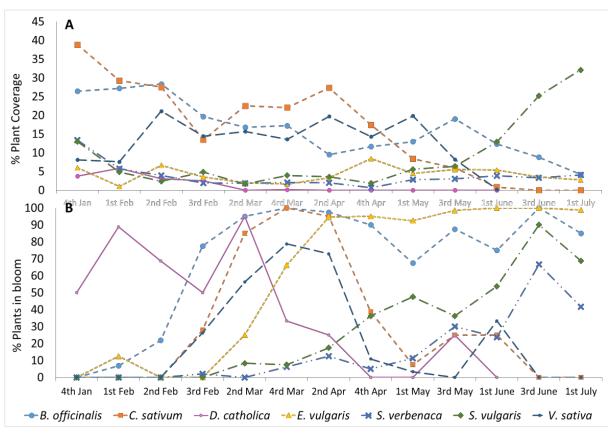


Figure 3. A) Percentage of plant coverage over time. B) Percentage of plant blossoming over time.

From the results of these assays, our own experience (Sanchez *et al.*, 2014) and the literature (e.g. Sheperd *et al.*, 2003), we conclude outlining some of the aspects that we consider should be taken into account in relation to the choice of plants when designing margins of vegetation for the conservation of beneficial insects and other arthropods:

- Take into account the viability of seeds and plant emergence in the field in order to predict the density of each plant species on the margin.
- Do not use excessively high plant densities (e.g. 10 individuals per square meter for small and 5 for medium size plants).
- Do not mix plants that are very different in size or growth rates because smaller and less vigorous plants will be displaced. For instance, *S. verbenaca* and *S. vulgaris* are relegated by bigger plant species.
- Use plants with extended periods of blossoming (e.g. B. officinalis and E. vulgare).
- Use plants with different floral structure to cover the requirements of a broad range of beneficial organisms (Willmer, 2011; Nichols & Altieri, 2012).
- Use a mixture of plants species that increase eco-systemic services but do not compete with the crop or serve as pest or disease reservoirs.

## Acknowledgements

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## Overwintering of *Encarsia tricolor* on the cabbage whitefly

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Abstract: Encarsia tricolor is the dominant parasitoid species of the cabbage whitefly, Aleyrodes proletella. The latter finds sufficient overwintering habitats to appear in masses on cabbage crops during cultivation periods, whereas habitats with suitable overwintering hosts for E. tricolor are hardly available. Therefore, specific management strategies are needed to facilitate parasitoid overwintering. As a first step, this study aimed to provide general knowledge about the overwintering stages, the overwintering period and the overwintering success of E. tricolor on its primary host A. proletella. Results show that Encarsia tricolor successfully survived winter as immature stages, but no adults were found during late winter months. Visual observations revealed that at least 2.4% of A. proletella nymphs actually enclosed vital parasitoid eggs/ larvae during winter (n = 1.603), because they started to turn dark (parasitoid pupation) between 13-20 April. The proportion of adult emergence from these subsequently developed parasitoid pupae was 41%. In contrast, only 1.1% of parasitoid pupae collected in January overwintered successfully (n = 356). First adult *E. tricolor* were found on yellow sticky traps in the field between 4-11 May. The gained insights on the overwintering of E. tricolor are compared with the population dynamics of A. proletella on cabbage crops and conclusions for additional management strategies are discussed.

Key words: Aleyrodes proletella, Aleyrodidae, Aphelinidae, biological control, parasitoid

## Introduction

One reason for the development of the cabbage whitefly, *Aleyrodes proletella*, (Hemiptera: Aleyrodidae) to a major cabbage pest in Central Europe is the increased cultivation of winter oilseed rape, which serves as optimal overwintering habitat for adult *A. proletella* (Richter & Hirthe, 2014, Ludwig & Meyhöfer, 2016). Its main parasitoid *Encarsia tricolor* (Hymenoptera: Aphelinidae) is supposed to overwinter as immature stages in whitefly nymphs, which are rarely present on winter oilseed rape (Stein, 1958). Since most cabbage crops are harvested before winter ends, there are hardly any overwintering habitats left for *E. tricolor*. This study aimed to gain deeper insight about the overwintering strategy of *E. tricolor* as basic requirement for the development of further management strategies to facilitate *E. tricolor* overwintering and increase biological control of *A. proletella*.

## Material and methods

## Experimental set up

Brussels sprouts plants (*Brassica oleracea* var. *gemmifera* 'Maximus F1') were planted in a 2.5 m wide and 88.5 m long strip on experimental fields of the Institute of Horticultural Production Systems, Leibniz Universität Hannover, Germany, on 26<sup>th</sup> May 2014.

#### Overwintering of eggs/ larvae and adults

Twenty-two plants were randomly selected in February 2015. One leaf with unparasitized *A. proletella* nymphs was marked on each plant, the number of nymphs counted and subsequent development of parasitoid pupae inside nymphs (turn dark) was recorded on a weekly manner until  $11^{\text{th}}$  May. Parasitoid pupae were further observed for adult emergence to evaluate the overwintering success of *E. tricolor* as egg/larva inside its whitefly host. Additionally, the numbers of living *E. tricolor* adults on the marked plants were determined with the same frequency to obtain information on the ability of the parasitoid to overwinter as adults. Local temperatures were recorded to allow explanations of observed events.

#### Overwintering of pupae

On  $20^{th}/21^{st}$  January 2015, 356 obviously parasitized *A. proletella* nymphs (contained visible dark parasitoid pupa) were randomly collected to investigate pupal *E. tricolor* overwintering. Therefore, parasitized nymphs were gently removed from old cabbage leaves with a dissection needle and transferred individually into gelatine capsules with a fine brush. Gelatine capsules were then placed on tissue paper in open plastic boxes in small groups to avoid overlapping of capsules. The boxes were deposited inside a gauze tent (2 m x 2 m x 2 m) with a transparent plastic roof (field insectary) and covered with lightproof mesh. This construction protected the capsules from rain and direct sun light and thus from softening and overheating, but kept temperatures inside the capsules similar to outdoor conditions. The temperature was recorded next to the capsules in one of the boxes. Parasitized whitefly nymphs were weekly checked for adult parasitoid emergence.

#### Adult emergence

Twelve randomly selected plants were equipped with a yellow sticky trap construction on  $16^{\text{th}}$  March 2015 to determine the time of first *E. tricolor* adult emergence in the field. Therefore, the undersides of two yellow cards (Horticoop B. V., Bleiswijk, The Netherlands) were coated with insect glue (Temmen GmbH, Hattersheim, Germany) (Hoelmer & Simmons, 2008). Cards were stuck together to one yellow sticky trap (190 mm x 220 mm) and a hole with a diameter of the respective plant stem was cut centrally in each trap. Constructed traps were then tightly fixed horizontally around the plant stems in 300 mm above ground, fixed with a clip and supported by a wire frame underneath (circle with a diameter of 200 mm and three wire feet pushed into the ground). All yellow sticky traps were replaced and trapped *E. tricolor* adults counted for each trap in a weekly interval.

## Start of whitefly reproduction

The amount of *A. proletella* egg batches was weekly recorded on 44 randomly selected plants from February to May 2015. The received information on the beginning of whitefly reproduction period enabled us to estimate the time when first hosts for *E. tricolor* (i.e. whitefly nymphs) were present on the overwintering plants (Stein, 1958).

## **Results and discussion**

#### **Overwintering period**

First whitefly nymphs that contained parasitoid pupae were observed on marked leaves between 13-20 April (Figure 1). These nymphs must have been parasitized already in 2014 and immature parasitoids inside must have overwintered as eggs or young larvae, because adult parasitoids were not yet present at that time. First *E. tricolor* adults were found on

yellow sticky traps between 4-11 May ( $0.5 \pm 0.3$  individuals per trap). This is a realistic period for the emergence of the first *E. tricolor* adults after overwintering based on respective average temperatures (12.5 °C) and calculations for pupal development times after Avilla & Copland (1988). Most likely the on average 1.2 °C higher temperatures caused an earlier adult emergence in the gauze tent (6<sup>th</sup>-13<sup>th</sup> April) compared to field observations.

First whitefly egg batches were observed between 6-13 April (Figure 1). Temperatures in this period exceeded the developmental threshold for *A. proletella* eggs (10 °C) for the first time of the year (Iheagwam, 1978). Eggs need about three weeks at respective average temperatures (Stein, 1958). Therefore, hosts were already available for the first generation of *E. tricolor* after overwintering.

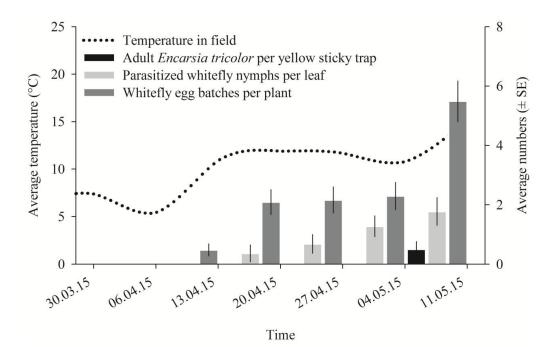


Figure 1. Average temperatures in field and average determined population parameters of *Encarsia tricolor* and *Aleyrodes proletella* during the experimental period (no activity observed before 30<sup>th</sup> March and therefore not shown here).

#### **Overwintering** success

The on average  $73 \pm 8$  *A. proletella* nymphs per marked leaf (mean  $\pm$  SE) showed a parasitism rate of 2.4% (i.e. turned dark due to parasitoid pupation). Adult *E. tricolor* females emerged from 41% of these subsequently pupated parasitoids. The parasitism rates are comparable to observations by Stein (1958), who reported 1-3% parasitism rates in February 1954 and 1955.

Only 1.1% of the collected *E. tricolor* pupae survived winter and emerged as adult females (n = 356). This high mortality could be explained by low temperatures between -2 °C and -7.5 °C that the overwintering *E. tricolor* pupae were exposed to for more than six hours on several days (Butler, 1938).

No *E. tricolor* adults were found on Brussels sprouts plants during the entire experimental period. Same was observed by Stein (1958), and Butler (1938) reported that adults as well as pupae do not survive temperatures under  $-2 \,^{\circ}C$  for six hours or more. This suggests that *E. tricolor* is not capable to overwinter as adults in the open field in Central Europe.

## Conclusion

Results revealed that *E. tricolor* can overwinter as eggs/larvae and pupae inside whitefly nymphs and the first adult generation appears in their overwintering habitat during May. However, the relatively low overwintering success, hardly available overwintering habitats with suitable hosts (i.e. whitefly nymphs) and the limited mobility of *E. tricolor* (Stein, 1958) may still cause a temporally and quantitatively insufficient migration into cabbage crops. Solutions are therefore needed to counteract this problem. For instance, offering overwintering habitats for *E. tricolor* near cabbage crops (e.g. perennial banker plants) or mass releasing of *E. tricolor* early in the year either manually or by annual banker plants may increase biological control services by *E. tricolor* or even promote the entire functional biodiversity of *A. proletella* natural enemies.

## Acknowledgements

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## Effects of landscape heterogeneity on crop colonization by natural predators of pests in protected horticultural cropping systems

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**Abstract:** In Mediterranean regions, colonization of protected horticultural crops by native predatory mirid bugs is frequent, but these processes remain highly heterogeneous among crops. Our study aimed at assessing the effects of crop management practices and local landscape heterogeneity (landscape composition and configuration within 300 m buffers around crops) on populations of *Macrolophus* and *Dicyphus* mirids in protected tomato crops in southern France. We found significant effects of landscape heterogeneity on mirid populations, but effects were similar for landscape composition and configuration. Tomato crops were colonized the most by *Macrolophus* mirids in landscapes with fallow, that seemed to act as source of mirids for crops. In contrast, crop colonization was reduced by nearby orchard, which reflected either sink or dilution effects. Mirid popuations were also reduced in crops with intensive management practices. Maintaining large areas of fallow is important to enhance native beneficial fauna, but adopting integrated plant management practices remains the most promising strategy to enhance mirid populations in protected horticultural crops.

**Key words:** conservation biological control, tomato colonization, landscape context, Miridae, *Macrolophus* spp., *Dicyphus* spp.

## Introduction

In protected horticultural systems, actual crop protection strategies rely on the use of chemical treatments or on periodical release of commercialized biological control agents. However, these methods have important limits regarding socio-technical, economic, health and environmental aspects. In Mediterranean systems, protected crops are open structures that can be colonized by indigenous natural enemies of pests from surrounding agro-ecosystems (Gabarra *et al.*, 2004). Among this native beneficial fauna, polyphagous predatory mirid bugs (Heteroptera: Miridae) of the genera *Macrolophus* and *Dicyphus* are considered major biological control agents against various Solanaceous crop pests (Perdikis *et al.*, 2008). Success of crop colonization and mirid population establishment in crops remains, however, highly variable from one crop to another. This variability has been partly explained by the intensity of crop management practices (Bonato and Ridray, 2007; Arno and Gabarra, 2011) and by the proximity of source and refuge host-plants of mirids in the close surroundings of crops (75 m) (Alomar *et al.*, 2002). However, the effects of host-plant availability and more generally of landscape heterogeneity on crop colonization remain unexplored at larger scales.

The goal of our study was to assess the variability in crop colonization by mirid bugs related to local crop management practices and surrounding landscape heterogeneity (described within 300 m buffers around crops). We focused on tomato crops under organic or conventional farming, in the French Mediterranean region.

## Material and methods

#### Study area and field selection

We sampled 34 tomato crops (26 under organic management and eight crops under conventional management) in 2010 and 2011 located in the Roussillon plain, in southern France. Studied landscapes are mosaics of annual (cereal, horticultural) and perennial (orchard and vineyard) crops, semi-natural elements (hedges, herbaceous elements, fallow), and urban areas. Tomato crops were planted from mid-March to mid-April in tunnels or plastic greenhouses without insect-proof nets, in soil.

#### **Biological sampling**

Abundance of *Macrolophus* spp. and *Dicyphus* spp. was estimated in crops by non-destructive weekly sampling from early March just after tomato planting until late July (during 12 to 17 weeks depending on the planting date). Adult mirids were counted on 6 leaves of 24 tomato plants in each crop. Abundances of the main pests of tomato crops (aphids, whiteflies, leafminers, and mites) were also assessed on the same leaves according to three classes: none to a few, medium, or high numbers of individuals or galleries. Occurrences of medium to high infestation classes were summed for all pests, leaves, plants, and weeks to estimate total pest infestation levels.

## Description of crop landscape context

Land-covers were digitized in a buffer area with a width of 300 m around the border of each sampled crop, using aerial ortho-photographs and field surveys. Eight land-cover types were mapped: fallow, woodland, grassland, protected (vegetable) crop, open field (vegetable) crop, grassy orchard, other grassy perennial crop (vineyard and olive grove), perennial crops with bare soil, water, and urban area. Landscape heterogeneity was described in 50 m, 100 m, 200 m, and 300 m buffers around crops by the percent cover of land-covers and land-cover diversity (compositional heterogeneity) and by land-cover connectivity (index from Hanski and Thomas, 1994) (configurational heterogeneity).

#### Description of crop management practices

Observations in crops and interviews of farmers were realized to describe the type of farming system (organic, conventional), greenhouse type and state, tomato management practices, and crop protection practices. Multiple correspondence analysis (MCA) was done on variables to build synthetic descriptors of crop management practices (crop coordinates along MCA axes). The three first axes (AG1, AG2 and AG3) described: (1) a gradient of crop protection strategies, from preventive and organic (release of biological control agents, *Bt* treatments) to curative and conventional ones (pesticides), (2) a gradient distinguishing specific cultural interventions and greenhouse condition, and (3) a gradient of increasing frequency of cultural interventions (crop protection and plant management) associated to greenhouse type (tunnel *vs*. plastic) and plantation date.

#### Statistical analysis

Random forests were used to pre-select important, uncorrelated landscape metrics. Generalized Linear Models (GLM) were used to analyze the effects of crop characteristics (pest infestation levels and agricultural gradients) and landscape metrics on mirid abundance. Separate analyses were conducted for each buffer size and class of landscape metrics (composition, configuration). Multi-model inference and model averaging were used to test all possible models and to determine the average of relevant models (with  $\Delta AICc < 2$ ).

## **Results and discussion**

## Effects of landscape heterogeneity on mirid abundances at different spatial scales

Landscape heterogeneity affected the abundance of both mirid groups. Models integrating composition or configuration metrics had similar effects on mirid abundance, as illustrated by the similar averaged AICc from GLMs (Figure 1). *Macrolophus* mirids responded the most to landscape heterogeneity at large scales (lowest averaged AICc within 200 m and 300 m buffers), whereas *Dicyphus* mirids responded the most at smaller scales (lowest averaged AICc within 100 m buffers) (Figure 1). Until now, these two mirid groups were assumed to disperse over short distances (up to 100 m) similarly to other mirid bugs (Alomar *et al.*, 2002). Our results suggest that *Macrolophus* species might disperse beyond these expected distances.

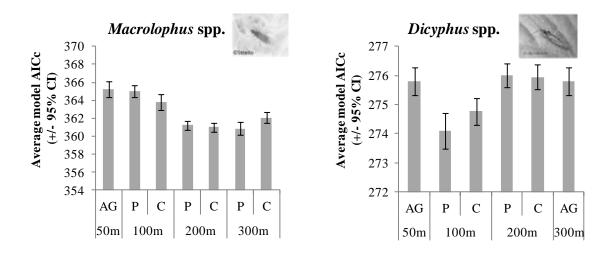


Figure 1. Mean AICc averaged over similar relevant models ( $\Delta$ AICc < 2) for *Macrolophus* spp. and *Dicyphus* spp. for each spatial scale (50 m, 100 m, 200 m, 300 m). AG: models with local agricultural practices. P/C: models with local agricultural variables and landscape (percent cover P or configuration C) metrics.

#### Relative effects of local crop characteristics and landscape heterogeneity

We collected 3010 and 696 individuals of *Macrolophus* spp. and *Dicyphus* spp. respectively. During the study, the average occurrence (over sampled plants on the same crop) of pests at medium or high infestation levels was 89 times  $\pm$  124 times.

Table 1 gives an overview of significant variables and their relative importance value (RIV) in the average GLMs. It shows that tomato crops were colonized the most by *Macrolophus* mirids in landscapes with fallow (100 m). This positive effect might be explained by the presence in fallow of host-plant species known to act as refuges in winter and as sources of mirids during their active period (Alomar *et al.*, 2002). In contrast, crop colonization by mirids was reduced by large areas of orchard (in 100 m, 200 m or 300 m buffers depending on mirid group). This negative effect might either reflect a sink effect for orchards that can be intensively managed, or, on the contrary, a dilution effect if the herbaceous strata of orchards contain attractive host-plants.

Variables	Macrolophus spp.			Dicyphus spp.				
	50 m	100 m	200 m	300 m	50 m	100 m	200 m	300 m
AG1	1.00 (-)	1.00 (-)	1.00 (-)	1.00 (-)	ns	ns	ns	ns
AG2	ns	ns	0.49 (+)	ns	ns	ns	ns	ns
AG3	1.00 (-)	1.00 (-)	1.00 (-)	1.00 (-)	0.86 (-)	1.00 (-)	0.87 (-)	0.86 (-)
Pest infest.	ns	ns	ns	0.14 (+)	0.70 (+)	0.86 (+)	0.73 (+)	0.70 (+)
P_fallow	ns	0.55 (+)	ns	ns	ns	ns	ns	ns
C_fallow	ns	0.79 (+)	ns	ns	ns	ns	ns	ns
P_Orchard	ns	ns	0.89 (-)	1.00 (-)	ns	0.87 (-)	ns	ns
C_Orchard	ns	ns	0.89 (-)	1.00 (-)	ns	ns	ns	ns

Table 1. Overview of variables having a significant effect on mirid abundance and their relative importance values (from 0 to 1) in average GLMs. (+) or (-): positive or negative effect; *ns*: not significant. AG: agricultural gradients. P: percent cover, C: connectivity.

Overall, the effects of landscape heterogeneity on mirid abundance were lower than those of local crop management practices (lower RIV, Table 1). Mirid abundance decreased from organic to conventional crop protection strategies (AG1). *Macrolophus* abundance further decreased with the intensification of cultural interventions on crops (AG3, in both organic and conventional systems). *Dicyphus* abundance was also positively related to pest infestation levels in crops.

Our study suggests that converting farms from conventional to organic production systems and adopting integrated plant management practices remain the most promising strategies to enhance mirid populations in protected horticultural crops. However, maintaining large areas of fallow seems also to be important to ensure colonization of protected crops by mirids. Specific field surveys are however needed to explain the negative impact of orchard on mirids.

## Acknowledgements

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## Relationship between landscape structure and abundance of airborne beneficial arthropods in an olive agro-ecosystem

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**Abstract:** Some of the groups of beneficial arthropods present in the olive grove canopy are hoverflies, hymenopteran parasitoids and bees. In this research we test the hypothesis that the abundance of these groups in olive groves increases with increasing landscape complexity. In spring 2015 and 2016 we determined the abundance of the beneficial insect groups in 15 olive groves by identification of insects captured on 4 yellow sticky traps in each olive grove. Indices of landscape structure were calculated in circular areas with radii ranging from 250 to 1500 m around the 15 olive groves selected. Preliminary results indicate that in both sampling dates, the abundance of hoverflies was negatively correlated with the patch size of the olive grove, but positively correlated with the total area of scrublands and oak forests and with the diversity index of land uses. The abundance of hymenopteran parasitoids and bees was also positively correlated with the total area of scrublands with oak, in both years, but negatively correlated with the patch size of olive groves in the case of bees. Not all these relationships were significant at all radii and years analysed. These results indicate that intermingling of spontaneous vegetation with olive groves favours the presence of beneficial arthropods.

**Key words:** apoidea, bees, olive, hoverflies, hymenopteran parasitoids, landscape, predators, pollinators, Syrphidae

## Introduction

Pollination and biological control are ecosystem services provided by arthropods in agriculture. Hoverflies (Diptera: Syrphidae), different families of hymenopteran parasitoids and bees (Hymenoptera: Apiformes) are groups providing these services. In the olive grove agro-ecosystem hoverflies and hymenopteran parasitoids play an important role as predators and parasitoids of important pests. Olive is wind pollinated and as such does not respond to insect pollination, but its pollen provides an important source of protein for wild and domesticated bee populations.

Abundance of these groups of beneficial arthropods varies depending on the presence of natural vegetation and landscape structure. This relationship has been studied in several agroecosystems, with an increasing number of works in Mediterranean climate and perennial crops (Burgio *et al.*, 2015, Paredes *et al.*, 2013, Thomson & Hoffmann, 2013, Tscheulin *et al.*, 2011). The general trend is a higher abundance of beneficials parallel to an increase in landscape complexity. This work aims to study the relationship between abundance of hymenopteran parasitoids, bees and hoverflies with landscape complexity in an olive growing area.

## Material and methods

Fifteen olive groves were selected in southeastern Madrid, Spain, to represent a gradient of landscape complexity. All olive groves were conventionally managed, and trees were planted in a 10 x 10 framework. Landscape structure was assessed in circular areas of 250, 500, 750, 1000 and 1500 m radii around the olive groves. Data taken from the Spanish Land Use and Land Cover Information System (SIOSE, 2005) map were used to assess the different types of land use and their proportion (area) within each circular area (minimum digitalization area was 0.5 ha). The software Patch Analyst for ArcGIS<sup>®</sup> 10.1 (ESRI, Redlands, CA, USA) was used to calculate the following landscape indices: Total area of scrubland with oaks (TASO), Shannon landscape diversity index (SHDI), edge density (ED) and patch size of olive grove (SOG). It is the size of the patch on which the olive grove is located

Airborne beneficial insects were sampled by 20x25 cm yellow sticky traps (Econex<sup>®</sup> Sanidad Agrícola, S.L). Four traps were positioned on a transect starting approximately 30-40 m from the edge to the centre of the olive grove in four consecutive trees, separated 10 m to each other. The area of the sampled olive groves farms ranged between 1 and 11 ha.Traps were in the field from  $20^{th}$  until  $27^{th}$  march in 2015 and from  $10^{th}$  until  $17^{th}$  march in 2016. Traps were taken to the laboratory and numbers of hymenopteran parasitoids, bees and hoverflies were counted. Abundance data were transformed by Log(x+1) and Pearson correlation with landscape indices was calculated.

## **Results and discussion**

From the indices used, only TASO, SOG and SHDI were related with the abundance of the beneficial arthropods studied (Figure 1). Significant relationships were more frequent for TASO, then for SOG. Relationships with SHDI were the less frequent. Different Mediterranean scrublands (garrigue in Italy or phygrana in Greece) have been shown to affect beneficial arthropods in the Mediterranean basin (Picchi *et al.*, 2016, Tscheulin *et al.*, 2011).

Abundance of hymenopteran parasitoids was consistently related to TASO (Figure 1a). However, landscape effect on parasitism rates of *B. oleae* was not found in this same area (González-Núñez *et al.*, 2015), although in Italy significant effects of landscape on parasitism rates were detected (Boccaccio & Petacchi, 2009).

Landscape indices had a stronger effect on bee abundance in 2016 than in 2015 (Figures 1a, b and c), although the trends were similar in both years. Neokosmidis *et al.* (2016) showed that natural forest also had a positive effect on bee abundance, while cultivated land had a negative effect.

Although trends were similar for the three groups studied, the relationship between abundance of hoverflies and landscape indices seems to be slightly stronger than that of the other groups (Figures 1a, b and c). Different studies have shown a positive influence of landscape complexity (Ricarte *et al.*, 2011) or natural elements (Miñarro & Prida, 2013; Sarthou *et al.*, 2005) on hoverflies.

Not all these relationships were significant at all radii. SHDI had an effect at 250 and 500 m radii. These results show the importance of buffer area in the analysis of landscape effect on beneficials in some cases.

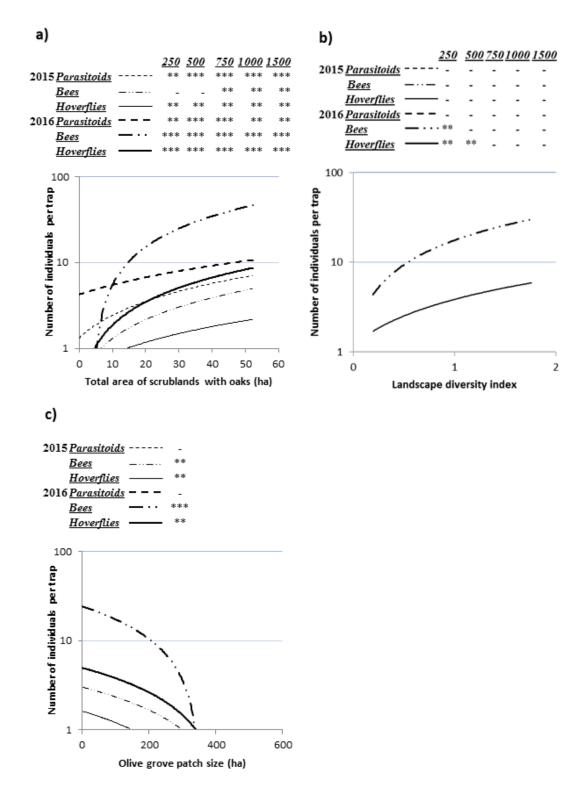


Figure 1. Linear relationships between abundance of hymenoptera parasitoids, bees and hoverflies. a) Total area of scrublands with oak TASO, b) Shannon landscape diversity index SHDI, c) Patch size of the olive grove SOG. Significance of the relationships for buffer areas of radii ranging from 250 to 500 m is indicated in the tables: \*: p < 0.10, \*\*: p < 0.05, \*\*\*: p < 0.01, -: not significant (n = 60). Graphs in a) are for a 750 m radius buffer area, graphs in b) are for a 250 m radius buffer area.

Our study shows that the abundance of the studied groups is related to specific land uses (scrublands with oaks), rather than to the diversity of land uses in the surrounding landscape. As abundance was lower for increasing areas of olive groves, it seems desirable to balance olive groves and scrubland areas to favour populations of these beneficial arthropods.

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