Anthocorid bugs (Hemiptera, Anthocoridae) as natural predators of apple aphids and pear psyllids using spatial and temporal scales

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Preface

This master thesis was written as a part of a research project on integrated pest management of apple aphids and pear psyllids "Integrert bekjempelse av eplebladlus og pæresuger", funded by the Norwegian Research Council (project nr. 245455). The study was carried out at NIBIO (Norwegian Institute for Bioeconomy Research) at Ullensvang and Ås. "Foregangsfylke for frukt og bær" also supported my study financially.

Fieldwork in Lofthus and Balestrand was done together with co-student Elena Therese Langeland Næss. Back home in Ås we went our separate ways and did laboratory work, statistics and writing. Elena's thesis "Molecular analysis of predation by anthocorid bugs (Hemiptera, Anthocoridae) on the pear psyllid *Cacopsylla pyri*" can be found on the department for ecology and natural resources management at NMBU's pages in the brage.bibsys database.

I would like to thank my supervisors Bjørn Arild Hatteland and Nina Trandem for help in setting up the trials, writing and statistics. Thank you, Jostein Lutro and Jon Asbjørn Målsnes for letting me use your pear and apple orchards for research. Also thanks to Toril Sagen Eklo, Torstein Kvamme, Knut Helge Jensen and Lene Sigsgaard for help with species identification of aphids and ants, statistics and general tips.

Thanks to Sverre Storberget, Elena Therese Langeland Næss, Marte Olsen and Charlotte Norseng for just being who you are. Also a great thanks to everyone in "Soerhellingen vin og konditorlaug". The certain knowledge of cake every Friday, kept me going strong all semester!

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Place and date Signature

Abstract

Farmers in Norway are facing challenges like reduced access to pesticides, climate change and problems associated with increased homogeneity of crops in farming. Biological control may provide a solution to these challenges, by increasing the biodiversity and robustness in the surrounding ecosystems. The anthocorid bugs *Anthocoris nemorum* and *A. nemoralis* are considered to be effective predators on three important pest species in Norwegian fruit production: the pear psyllid *Cacopsylla pyri* in pear, and the green apple aphid *Aphis pomi* and the rosy apple aphid *Dysaphis plantaginea* in apple. The aim of this study was to investigate the potential of *A. nemorum* and *A. nemoralis* as biological control agents of these pest species in Norway.

Field surveys and semi-field experiments were carried out in apple and pear orchards in western Norway. The predation potential was tested by performing sleeve-cage experiments. Information about spatial and temporal patterns of natural populations of *A. nemorum* and *A. nemoralis* and their prey was obtained by sampling orchards and analyzing the data with generalized linear mixed models (GLMM) incorporating the spatial structure.

The results from the sleeve cage experiments showed that *A. nemorum* has the potential to control both *C. pyri* populations in pear and aphid populations in apple. Surprisingly, commercially bought *A. nemoralis* did not manage to reduce *C. pyri* populations, when compared to the control. Differences between commercially reared and natural populations of *A. nemoralis* should be researched. Results from the orchard survey in pear supported the hypothesis that natural populations of *A. nemoralis* are more specialized towards finding *C. pyri* nymphs and eggs than *A. nemorum.* However, *A. nemorum* was present in the orchard earlier, and in larger abundances than other studies has suggested. This highlights that *A. nemorum*'s role in controlling pear psyllids may be more important in Norway than previously assumed. The presence of ants was negatively related to number of *Anthocorid* nymphs in pear, and to imago *A. nemorum* in apple. Ants should therefore be taken into account when biocontrol systems in orchards are planned.

Sammendrag

Norske bønder står foran utfordringer som redusert tilgang på kjemiske plantevernmidler, klimaendringer og problemer som oppstår når homogenitet i avlinger øker i jordbruket. Disse utfordringene kan møtes ved bruk av biologisk kontroll, fordi økt biodiversitet i jordbruksområder gjør habitatet mer robust mot endringer. Nebbtegene *Anthocoris nemorum* og *A. nemoralis* er ansett å være effektive predatorer på tre viktige skadedyr i eple og pære i Norge: Vanlig pæresuger *Cacopsylla pyri* er skadedyr i pære. Grønn eplebladlus *Aphis pomi* og rød eplebladlus *Dysaphis plantaginea* er skadedyr i eple. Målet med dette studiet var å undersøke om *A. nemorum* og *A. nemoralis* har potensiale som nyttedyr for biologisk kontroll i eple- og pærefelt.

Informasjon om *A. nemorum* og *A. nemoralis* ble samlet inn gjennom feltundersøkelser og kontrollerte feltforsøk. Predasjonspotensialet til nebbtegene ble undersøkt gjennom et nettposeforsøk. For å analysere hvordan nebbtegene beveger seg i tid og rom i pære- eller eplefeltet ble en *generell lineær blandet modell* (GLMM) brukt. Feltundersøkelsene og felteksperimentene ble utført i både eple og pære.

Nettposeforsøket viste at *A. nemorum* kan kontrollere populasjoner av *C. pyri* i pære, og populasjoner av *A. pomi* i eple. *A. nemoralis* påvirket ikke antallet *C. pyri* i nettposeforsøket, noe som var overraskende. Forskjeller mellom naturlige norske populasjoner og kommersielt oppfostrede *A. nemoralis* kunne med fordel undersøkes mer. I feltundersøkelsen støttes hypotesen at *A. nemoralis* er mer spesialisert og bedre til å finne *C. pyri* enn *A. nemorum* i pære. *A. nemorum* var likevel også viktig, fordi det var flere individer av den i pærefeltet, noe som ikke var forventet. I eple har *A. nemorum* potensialet til å kontrollere populasjoner av eplebladlus hvis antallet nebbteger øker. Det var et negativ forhold mellom maur og *anthocoris* nymfer i pære og imago *A. nemorum* i eple. De kan derfor være en viktig faktor i skadedyrkontroll, og burde tas hensyn til når bruken av biologisk kontroll planlegges i eple- og pærefelt.

Contents

Introduction

Farmers in Norway face big and small challenges every day. Some of the larger challenges involve, pesticide use, climate change and increased homogeneity of farmland. Problems with pests may increase in farms with larger homogenous fields of crops. This is because homogenous crop can sustain a larger population of pest species than natural vegetation (Altieri 1999; Driesche & Bellows 1996). Farms in Norway have for several years grown in size (Rognstad 2009). When increasing farm sizes are combined with government goals that Norwegian farms should have larger yields (Norwegian Ministry of Agriculture and Food 2015), the result may be an increase in homogeneity in the farming areas. Creating natural vegetation surrounding the crop fields, which could attract natural enemies of the pests, may mitigate this.

Historically, problems with pests in crops were addressed by using natural enemies as biological control agents (Lacey et al. 2001). However, after the Second World War chemical pesticides became mass-produced (Gaugler et al. 1997; van Lenteren 2012). Since then, the world's use of pesticides has increased 12 folds in all types of crops (Driesche & Bellows 1996). Pesticides can be harmful to humans and other non-target taxa, including natural beneficial predators (McCauley et al. 2006). They have also turned out to be inefficient in controlling the target insects in the long run, and the number one reason for pest control failure is resistance to pesticides (Driesche & Bellows 1996). Although pesticides were not yet commonly used, an arthropod species in the United States was reported to have evolved a pesticide-resistance already in 1914 (Melander 1914). Pesticide resistance in arthropods has since then increased in importance, and from the 1950s the number of arthropod species evolving resistance to pesticides went from almost zero, to 533 species in 2000 (Mota-Sanchez et al. 2002). Governments in Norway as well as in the EU now try to mitigate the problems caused by the over-use of pesticides. Action plans concerning goals to reduce the overall use of pesticides in farming has been released, and causes new challenges for farmers in Europe (Hillocks 2012; Norwegian Ministry of Agriculture and Food 2009).

Three of the most important pests in Norwegian pome fruits are the pear psyllid *Cacopsylla pyri* (L.), the rosy apple aphid *Dysaphis plantaginea* (Passerini) and the green apple aphid *Aphis pomi* (De Geer), (Hemiptera: Psyllidae, Aphididae) (Edland 2004). Pear psyllids have evolved resistance to pesticides; this includes Norwegian populations of *C. pyri* (Civolani et al. 2015; Edland 2004; Sigsgaard et al. 2006a). The nymph of *C. pyri* causes damage in pear, sucking plant-juice from leaves and fruit buds, causing small and miss-shaped fruits (Stamenkovic et al. 2001). In Norway, *C. pyri* usually has two generations a year, where the second generation causes most damage (Edland 2004). *A. pomi,* together with *D. plantaginea,* are the most damaging aphid species in apple in Norway. Because of the parthenogenetic reproduction in parts of the life cycle, their numbers can increase rapidly (Edland 2004). *C. pyri, D. plantaginea* and *A. pomi* all secrete honeydew that can cause damage to the leaves. The honeydew creates a good habitat for sooty molds, which can cover and damage leaves and fruits (Edland 2004). The honeydew also attracts tending ants, like *Lasius niger* and *Formica fusca* (Linnaeus) (Cushman & Addicott 1989; Novak 1994). The ants collect the honeydew and in turn protect the plant sucking insects against predators (Novgorodova 2005).

In Norway, climate change can have an effect on agricultural production, through increased temperatures, earlier onset of spring and also more flooding (Intergovernmental Panel On Climate Change 2001). Increased temperatures can affect the life-cycle of pear psyllids and aphids (Arbab et al. 2006; Birch 1957; Kapatos & Stratopoulou 1999). The Norwegian populations of *C. pyri* has two generations per year, but three generations have also been observed in warm summers on the south west coast (Edland 2004). More flooding may increase runoff and possibly also increase nutrient-flow to crops in the agricultural fields (Wetzel 2001). This may increase problems concerning psyllids in pear, because a relationship between increasing *C. pyri* populations and nitrogen abundance in pear trees has been observed (Daugherty et al. 2007).

Use of biological control instead of pesticides will usually prevent or slow the development of pesticide resistance; because natural enemies normally exert other selection pressures on the pest than pesticides (Davies et al. 2012; Georghiou 1972).

Other advantages of biological control are: a reduction in pesticide residue in food, it is safer for humans to apply and lastly it increases the biodiversity in the affected areas, because pesticide residue does not hit the non-target organisms (Lacey et al. 2001; McCauley et al. 2006) . The economic cost of multiple applications of pesticide can also be higher than the loss from pests in untreated fields (Symondson et al. 2002). Effective pesticides can be hard to attain, and an increasing popularity in organic farming (Paull 2011; Rognstad 2009) limits the pesticides legal for farmers to use. The use of biological control world wide has therefore become more popular (Symondson et al. 2002). The backside is, that successfully applying biological control can be difficult, mostly because of nature's complexity. A high knowledge of species, agriculture and ecology of plants and beneficial organisms is needed, as factors may play different roles than expected (Waage et al. 2012). When introducing new species to an ecosystem through biological control, it can also cause more harm than good and decline the biodiversity in surrounding areas (Clavero et al. 2005; Simberloff & Stiling 1996).

There are three main types of biological control in agriculture: classical biological control (importation), augmentation, and conservation (Driesche & Bellows 1996). Classical control is when a beneficial organism is released into the area with a pest problem, usually an introduced pest species. The beneficial is expected to survive, spread and reproduce in the new habitat (Driesche & Bellows 1996; Waage et al. 2012). Augmentation is the mass release of mass-reared beneficial organisms. The release have a short time effect (no longer than one season), because of inhospitable climatic conditions or because the host-plants are not suitable for the beneficial organism (Waage et al. 2012). Another type of augmentation is inoculative release. This is the release of a small number of natural enemies early in the season, where the beneficial is expected to survive and reproduce in order to hold populations of pest species down (Driesche & Bellows 1996). Biological control through conservation is maintaining healthy populations of beneficial organisms already present in or around the crop (Driesche & Bellows 1996). This can be done by manipulation of habitat, where both predator friendly and pest hostile plants can be planted, or by not applying pesticides harmful to the beneficials. Conservation alone may keep the pest species at a lower abundance than the economic threshold (Fitzgerald & Solomon 2004).

If the economic threshold is reached nonetheless, mass release of predatory insects (augmentation) may provide a solution (Sigsgaard et al. 2006b). Optimal foraging theory tells us that a predator will choose its prey based on an optimal trade-off between the cost of foraging and the benefit of the energy attained from the prey (Drickamer & Jakob 2002). When releasing insects into agricultural areas to control pest species, you can use generalist or specialist predators. Parasitoids are usually more specific than specialist predators, and have been preferred biocontrol agents because the selections of hosts are species specific (Symondson et al. 2002). The downside to using a specialized parasitoid, in addition to general problems concerning biological control, is that a high knowledge of taxonomy of pest species as well as the species of parasitoid is needed for the control to be successful (Waage & Hassell 1982). Among predators, specialist species have traditionally been regarded to be the best control agent of pests, but generalists as well as specialists can be effective predators (Hawkins et al. 1999; Novak & Achtziger 1995; Sigsgaard et al. 2006a; Symondson et al. 2002). Generalist predators will not have a functional response to increasing pest numbers in the same way as specialist predators will; but because generalists may switch their preference of prey when the abundance reaches a certain level, a composition of different species of generalists may be more important in keeping the pest species abundance down than previously assumed (Symondson et al. 2002). Biological control, and mostly the conservation type of biological control, will not deteriorate the robustness or biodiversity of habitats surrounding agricultural areas. This could be an advantage when facing challenges like climate change, because biodiversity in agricultural ecosystems are important also when it comes to performing other ecological services than biological control (Altieri 1999).

Anthocoris nemorum (L.) and *A. nemoralis* (Fabricius) are polyphagous predators of the Anthocoridae family (Hemiptera), which are naturally occurring in Norway. *A. nemorum* and *A. nemoralis* have been shown to respond to plant volatiles released by pear trees when attacked by pear psyllids (Drukker et al. 1995; Scutareanu et al. 1996; Scutareanu et al. 1999). They have both shown the potential to be effective predators of psylla and

the Aphididae family (Fig. 1) (Novak & Achtziger 1995; Shaltiel & Coll 2004; Sigsgaard et al. 2006a; Solomon et al. 2000; Symondson et al. 2002).

A. nemorum is common in fruit orchards in Europe, some parts of Asia and Africa, and in Norway (Butler 1923; Edland 2004). In Norway *A. nemorum* is univoltine, it overwinters as an imago, and lays eggs in spring. The nymphs hatching in spring are fully grown into imagos in late summer (August, September) (Edland 2004; Våge 1991). It has been recorded to be the most abundant predatory bug in fruit orchards in Sogn in western Norway, with *A. nemorum* consisting of more than 80% of the predatory bugs in the orchard (Sørum 1977). In a study in Ås, Norway on apple trees by Austreng and Sømme (1980) the relative abundance was more modest, with *A. nemorum* only making up 6.5% of the total predatory bug population. Of these, 35% of the specimens from the Anthocoridae family were *A. nemorum*. It has also been observed on several different species of trees and shrubs in Norway (Taksdal 1965). In the rest of Europe it is one of the most abundant species in both apple and pear orchards (Solomon et al. 2000). It has been documented to be an effective predator on different pests in apple and pear, including aphids and pear psyllids (Campbell 1977; Solomon et al. 2000). Being a generalist, *A. nemorum* has a quick response time to build-up of pest species, because it can quickly switch between different prey if one species becomes abundant (Novak & Achtziger 1995).

A. nemoralis has a life cycle and distribution similar to *A. nemorum*, but has a higher preference to eating pear psyllids (Sigsgaard 2010). *A. nemoralis* can affect the population of pear psyllids, but is usually too slow to respond to pest build-up to prevent economic loss (Artigues et al. 1995; Novak & Achtziger 1995; Shaltiel & Coll 2004; Sigsgaard et al. 2006a).

In Europe, and the rest of the world, biocontrol companies sell and distribute insects used as biological control agents. *A. nemoralis* is one species that is sold. None of the companies that sell *A. nemoralis* are located in Norway, or use Norwegian populations for further breeding. To import and sell macro-organisms into Norway it has to be approved by the Norwegian agricultural agency, and an application for registration of *A.*

nemoralis has been sent (Sundbye & Björkman 2014). There are restrictions on how and where the organisms can be used; most of the approved organisms can only be released in greenhouses or polytunnels. In total, 20 species of macro-organisms are currently approved for use in Norway, but so far none are registered for use in orchards against *C. pyri, D. plantaginea* or *A. pomi* (Norwegian agricultural agency).
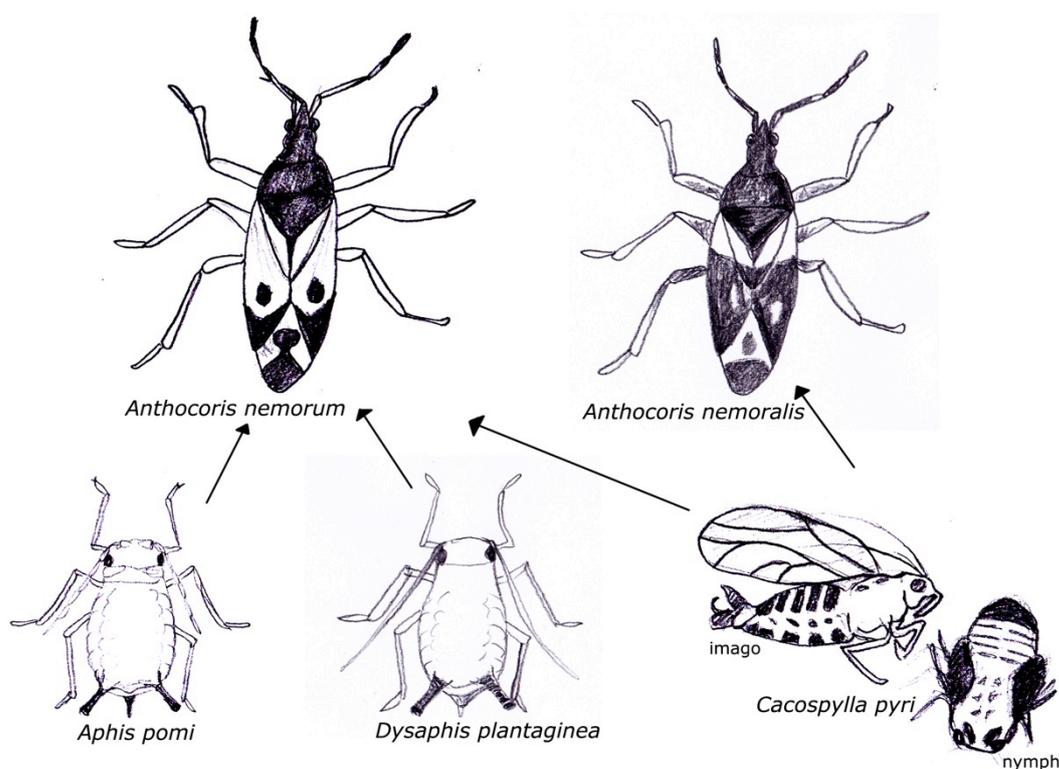
Food web

Figure 1: A simple food web describing the interactions of the two predators and three pest species looked at in my study .

The overall aim of this study was to investigate the potential of the anthocorid bugs *A. nemorum* and *A. nemoralis* as biological control agents in Norwegian pear and apple orchards.

In order to achieve this aim, the objectives were to:

(1) Investigate if *A. nemorum* and *A. nemoralis* affects the abundance of *C. pyri* in pear, using exclusion cage experiments.

I predicted to find that *A. nemorum* and *A. nemoralis* will affect the *C. pyri* numbers when other potential prey and ants are excluded, but that *A. nemoralis* will have a larger effect than *A. nemorum*.

(2) Similarly, investigate if *A. nemorum* affects the abundance of *A. pomi* in apple.

I predicted to find that *A. nemorum* will affect *A. pomi* numbers when other potential prey and ants are excluded.

(3) Analyse spatial and temporal patterns of *A. nemorum* and *A. nemoralis* in a pear orchard, including how they are affected by prey species and ants.

I predicted to find that *A. nemoralis* is more specialized and has a stronger positive temporal and spatial relationship with the population of *C. pyri*, than *A. nemorum*. The presence of ants will have a negative effect on the spatial and temporal relationship *A. nemorum* and *A. nemoralis* have with prey species in pear.

(4) Analyse spatial and temporal patterns of *A. nemorum* in an apple orchard, including how the prey species and ants affects its movement

I predicted to find that *A. nemorum* has a positive spatial and temporal relationship to the aphid populations in apple. The presence of ants will have a negative effect on the spatial and temporal relationship *A. nemorum* has with prey species in apple.

(5) Observe how edge vegetation affects *A. nemorum* and *A. nemoralis'* distribution in both apple and pear orchards.

I predicted to find that edge vegetation would have a positive effect on the abundance of *A. nemorum* and *A. nemoralis* in the pear and apple orchard.

Material and Methods

The data for this thesis was collected in two semi-field experiments called "the sleeve-cage experiments" and two field surveys called "the orchard surveys". Both the sleeve-cage experiments and field surveys were performed in the same apple and pear orchards in Western Norway.

Study sites

The pear orchard was in Lofthus, in Hordaland county (Coordinates WGS84 60°22'0.1"N 6°40'40.5"E) and consisted of 1.8 ha conventionally grown 'Ingeborg' pear trees planted in 2001 (Fig. 2). The part of the orchard used in the survey was in a western facing slope surrounded by other pear trees in the west and north. The vegetation towards south and east consisted of birch (*Betula pubescens*), pine (*Pinus sylvestris*), goat willow (*Salix caprea)* and different graminoids. The northeastern edges of the pear orchard had different species of nettles (*Urtica*). The mean temperature in Lofthus in 2015 were 12.1, 14.4 and 15.1°C in June, July and August respectively (Norwegian Meteorological Institute 2014-2015). The temperatures were measured at a weather station 7 km south of the study area. The normal temperature for Lofthus from 1961-1990 are 13.5, 14.4, and 13.4 in June, July and August respectively (Norwegian Meteorological Institute 1961-1990).

The apple orchard was in Balestrand in Sogn og Fjordane county (coordinates VGS84 61°8'18.6"N 6°32'18.4"E) and consisted of 0.37 ha organically grown 'Aroma' apple trees, planted in 1995 (Fig. 3). The orchard was in a southeastern facing slope and surrounded by birch and pine in the northeast, newly planted apple trees in the west and commercially farmed raspberries on the other side of an asphalt road to the south. The mean temperature for the area in 2015 in June, July, August and September was 11.8, 14.2, 15.5 and 12.4, respectively (Norwegian Meteorological Institute 2014-2015). The temperatures were measured at a weather station 8 km north of the study area. The normal temperature for Balestrand from 1961-1990 are 13.7, 14.8, 14.2 and 10.3 in June, July, August and September respectively (Norwegian Meteorological Institute 1961-1990).

Figure 2: Aerial photo (from www.gulesider.no) of the pear orchard in Lofthus. The red area is where the orchard survey was conducted. The green area is where the sleeve-cage experiment was conducted and the blue area was used as a buffer zone for pesticide-drift from the rest of the conventionally grown pear field.

Figure 3: Aerial photo of the organic apple orchard in Balestrand (from www.gulesider.no). The red area is where the orchard survey was conducted. The green area is where the sleeve-cage experiment was conducted and the blue area was a buffer zone for pesticide-drift from conventionally grown apple field west of the study area.

The sleeve-cage experiments

We did the sleeve-cage experiments in the rows marked with green in the pear and apple orchards (Fig. 2 and 3). Sleeve cages around branches (Fig. 4) were used to contain the study species and at the same time exclude other prey, ants or predators. Before the sleeve-cages were put on, we beat and visually observed the branches to remove all insects. The pest species (*C. pyri* in pear and *A. pomi* in apple) was then introduced in all sleeve-cages; predators were introduced one hour later.

In pear, the following three treatments were applied between the 08 – 26th June 2015:

-Ten *C. pyri* in nymphal stage 4-5 + two imago *A. nemorum*

-Ten *C. pyri* in nymphal stage 4-5 + two imago *A. nemoralis*

-Ten *C. pyri* in nymphal stage 4-5 (control).

In apple, the following two treatments were applied between the 17 – 30th August 2015:

-Ten imago *A. pomi* + two imago *A. nemorum*

-Ten imago *A. pomi* (control).

Each treatment consisted of one branch, repeated 10 times. This added up to 30 sleeve-cages in pear and 20 in apple. Because the orchards were in steep hills, the control and treatment cages were placed alternately on every tree along the row to offset the difference in hours with sun exposure from top to bottom of the row.

The insects used in the sleeve-cage experiments were obtained in different ways: The *A. nemoralis* were bought from a company (Borregaard Bioplant ApS) specializing in biological control, localized in Denmark. The *A. nemorum* were all collected by beat sampling in Lofthus, outside of the buffer field surrounding the 30 plots, or in another pear orchard in the same area; same with the *C. pyri*. The *A. pomi* were collected by hand in a conventional apple orchard next to where the field experiments were done in Balestrand.

After approximately two weeks we emptied the cages containing *A. nemorum* and the control, cleaning the branches of insects in the same way as at the start, by beating and visual observation. The branches containing *A. nemoralis* were cut off without removing the sleeve cage and frozen before opened to minimize the risk of spreading the commercial biotype.

Figure 4: Picture of sleeve-cage on a pear branch. The sleeve cages were funnels with zippers and made up by a fine mesh, letting sun, wind and water go through but not letting insects go in or out. They were tied to the branch at both ends with string.

Figure 5: A Styrofoam covered stick was used to tap one branch three times each. This treatment dislodged insects into the funnel that had a paper bag connected at the end.

The orchard surveys

The aim of the orchard surveys were to see how the anthocorid species were affected by the prey species and ant abundances in both space and time; and to see if edge vegetation was an important factor on their abundance and distribution in the orchard.

In pear, an area of 0.2 ha in the southeastern corner of the orchard was used (Fig. 2). In apple, the whole orchard was used, with the exception of a few rows that did not fit into the plot setup plan. Three rows of buffer-trees to the west were also excluded from the survey (Fig. 3).

Each orchard was divided into 30 plots consisting of seven trees each (Fig. 6 and 7). Four trees in each row separated the plots. The study area was not sprayed with insecticides between the collection periods. To hinder insecticide used on the pear trees in the rest of the orchard to stray, my study area was surrounded by 10 "buffer" trees (Moreby et al. 1997). Arthropods were collected by the beating funnel method and by visual examination of leaf samples (Southwood & Henderson 2009). The two sampling methods were intended to complement each other and collect different taxa and life stages. Three branches on each tree were beaten, resulting in a total of 21 branches sampled from each plot. Two of the beaten branches were distributed on each side of the tree; the third branch was localized high on the tree. Each beat consisted of three rapid taps on the branch. The beating stick was covered with Styrofoam to minimize damage

to the trees. A rectangular funnel (45x64 cm) with a paper bag at the narrow end was placed below the branch during beating to collect the dislodged arthropods (Fig. 5). The paper bags with the collected arthropods were returned to the laboratory and frozen in -80°C. For the leaf sampling, seven leaves from each tree were collected, from high, middle and low branches from all sides of the tree, giving a total of 49 leaves from each plot. The sampled leaves were contained in paper bags, and frozen in -22°C when returned to the laboratory.

The beating and leaf sampling was repeated three times in pear (5. June, 1. July and 18. August) and four times in apple (5. -6. June, 3. -4. July, 18. August and 12. September) during the research period. All the sampling was done in sunny weather, on dry vegetation, as the beating funnel method will not collect specimens evenly when the vegetation is wet.

Arthropods were counted and specified to species or family in a laboratory in Lofthus or Ås, using a stereomicroscope. I was not able to identify *Anthocoris* nymphs to species. *A. nemorum* and *A. nemoralis* nymphs in this study are therefore grouped together. Nymphs of *C. pyri* were divided in two; nymphs in nymphal stages 1-3 and nymphs in nymphal stages 4-5. By looking at how developed the wing buds on the nymphs were, the different nymphal stages were separated.
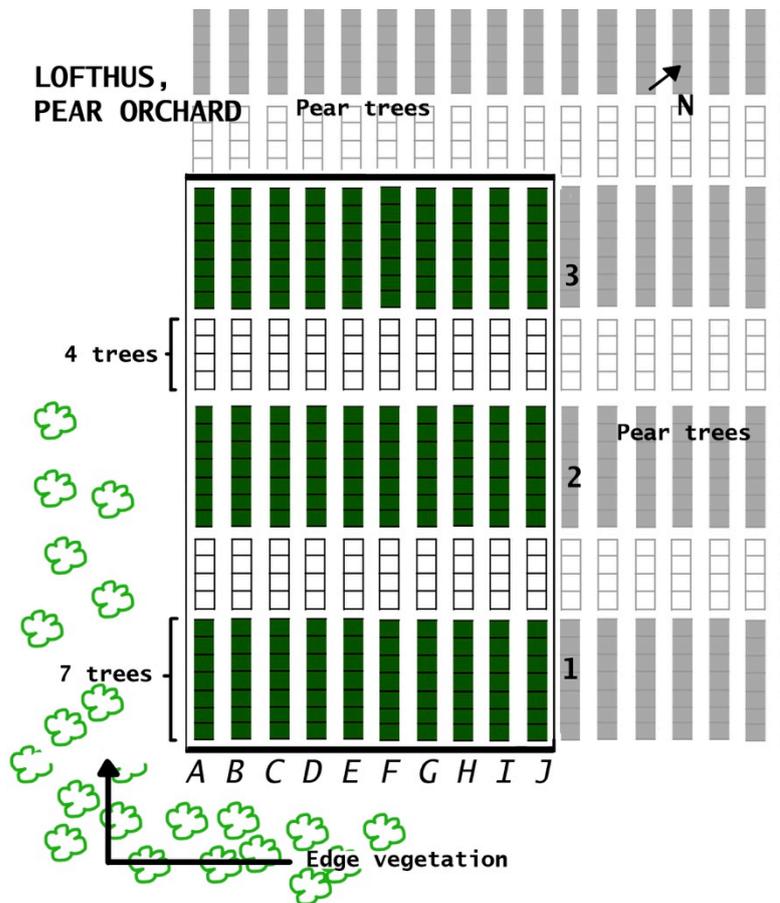
Figure 6: Map of plot set-up in commercially grown 'Ingeborg' pear. Each plot (green filled rectangles) consisted of 7 trees in a row, with 4 trees between the plots. Each plot was 31.5 m². Edge vegetation in west and east consisted of birch (*Betula pubescens*), pine (*Pinus sylvestris*), goat willow (*Salix caprea)* and different graminoids. Vegetation in north and west consisted of other 'Ingeborg' pears. Past the pear orchard to the north, there were different species of nettle (*urtica*) in the edge vegetation.
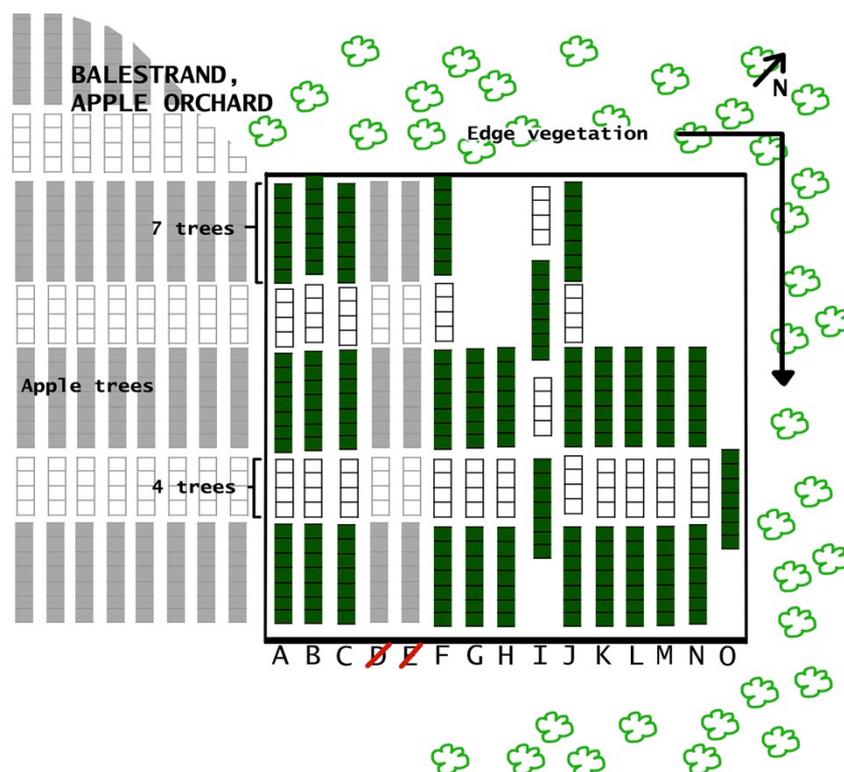
Figure 7: Map of plot set-up in organically grown 'Aroma' apple. Each plot (filled green rectangles) consisted of 7 trees in a row, with 4 trees between the plots. Each plot was on average 47.8 m². Edge vegetation in northeast and northwest consisted of birch (*Betula pubescens*) and pine (*Pinus sylvestris*) trees, with typical species connected with this type of forest. Vegetation in southwest was newly planted apple trees and in southeast commercially grown raspberries. Rows D and E were not included because of other cultivar than 'Aroma'.

Statistical methods

I used RStudio version 0.99.489 for all statistical analyses in this study (RStudio Team 2015).

The sleeve-cage experiments

Because the data was over-dispersed, the sleeve-cage experiments were analyzed using a Kruskal Wallis H-test on both pear and apple (Kruskal & Wallis 1952). On the pear branches a Dunn's test was carried out as a post hoc test to check for differences between the three treatments (Dunn 1964)

The orchard surveys

The data from the orchard collections were not normally distributed and had an over-dispersion; so to analyze the spatial relationship a generalized linear mixed-effects regression (GLMM) with a quasi-poisson distribution was selected. For the temporal

relationship a negative-binomial regression was selected, so the robustness of the model by using Akaike information criterion (AIC) could be determined (Crawley 2012). The best model was found using model selector in the MuMIn package (Bartoń 2013). I performed the statistical analyses as described in Hatteland et al. (2013).

Spatial maps showing the collected number of specimens in each plot were made of the pear and apple orchards. I made the spatial maps by creating a grid (x, y) from measured distances (in meters) between the plots and the distances to the edge vegetation. This was done using the "ppp" function from the "spatstat" package (Baddeley 2009). Generalized linear mixed-effect models (GLMMs) were used to test if the spatial patterns of the anthocorid bug were affected by the density of prey species and ants. In the pear orchard the response variables were either imago *A. nemorum,* imago *A. nemoralis* or nymphs. The added explanatory values were *C. pyri* nymphs, psylla eggs*,* imago *C. pyri* and *L. niger.* In the apple orchard the response variables were either imago *A. nemorum* or anthocorid nymphs. The added explanatory variables were *A. pomi*, *D. plantaginea, F. fusca* and *L. niger.* The grid made for the spatial maps were also included in the model to make it a spatial GLMM. The spatial maps was added as a random effect to adjust for spatial dependence (auto-correlation) between the samples (Dormann et al. 2007). The grid was added to the GLMM using the functions "corSpatial" and "glmmPQL" available in the packages "nlme" (Pinheiro et al. 2016) and "MASS" (Venables & Ripley 2002) in RStudio, respectively.

Analyses for the spatial patterns were carried out separately for June, July and August in pear and June, July, August and September in apple. The temporal patterns were analyzed from June to August in pear and only from June to July in apple; this was because of an unscheduled spot spraying for aphids in the apple orchard. The number of *A. pomi* and *D. plantaginea* in apple were merged to create an aphid dataset with fewer zeroes, assuming that *A. nemorum* does not have a large preference for one of the aphid species. A merge of collected *C. pyri* nymphs and psylla eggs was also done in pear. *F. fusca* and *L. niger* were excluded from some of the GLMM formulas in apple for spatial analyses (Table 4). The factors were taken out because the complexity was too high to calculate.

A Pearson product-moment correlation coefficient was generated for the factors that proved to be significant in the GLMMs (one response variable and one explanatory variable). This was done on the data from both pear and apple. The numbers produced show a correlation between 1 and -1 where 1 = total positive correlation, 0 = no correlation and -1= total negative correlation.

Results

The sleeve-cage experiments

There was a significantly lower *A. pomi* abundance on apple branches in the *A. nemorum* treatment compared to the control (*P*< 0.001, Kruskal Wallis H test) (Fig. 8). Similarly, on the pear branches, there was a significantly lower abundance of *C. pyri* in the *A. nemorum* treatment than in the control (*P*= 0.002, Dunn's test), while the *A. nemoralis* treatment did not differ significantly when compared to the control (*P*= 0.12, Dunn's test). The *A. nemorum* treatment also had significantly lower *C. pyri* abundance than the *A. nemoralis* treatment (*P*= 0.049, Dunn's test) (Fig. 9).
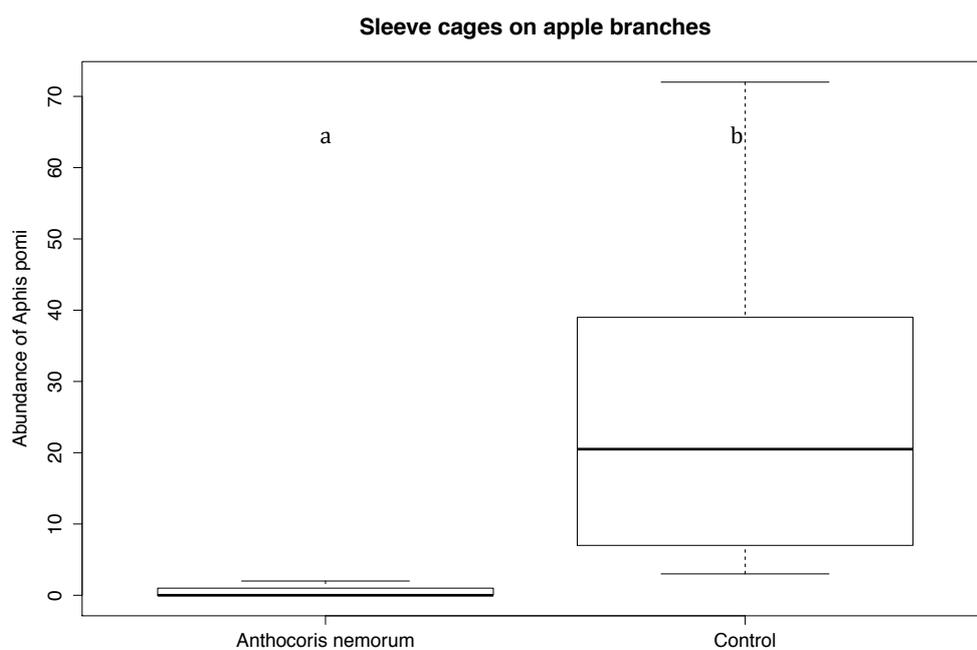

Figure 8: Sleeve- cage experiment in apple. Boxplot showing the abundance of *Aphis pomi* in the two treatments. Significant difference (*P*<0.05) is marked with different letters.
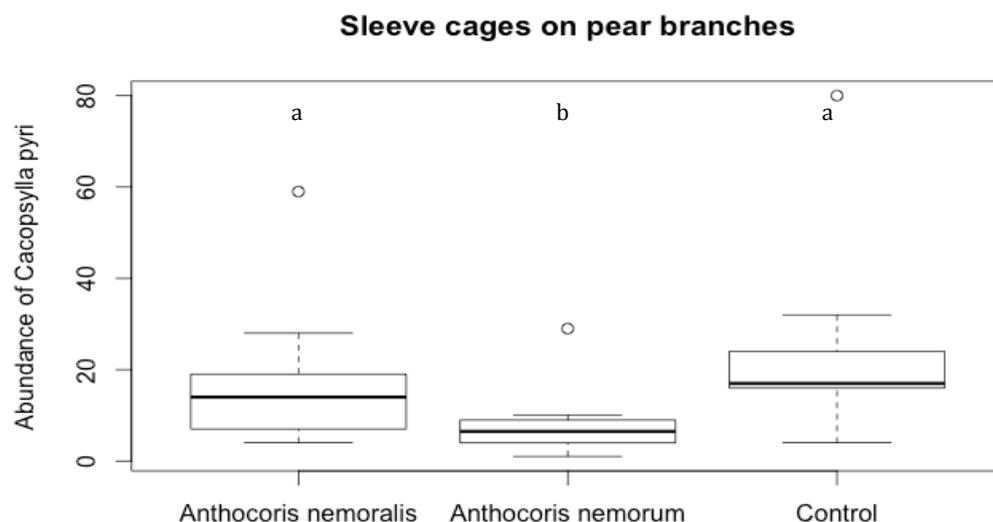
Figure 9: Sleeve-cage experiment in pear. Boxplot showing the abundance of *Cacopsylla pyri* in the three treatments. Significant difference (*P*<0.05, type of test) is marked with different letters.

The orchard surveys

Overview

In total 34 350 arthropod specimens (of these 2222 were psylla-eggs) were collected, counted and identified to species or family in the orchard surveys with the two sampling methods. In the pear orchard 9121 arthropods were collected. Of these 18% were anthocorid nymphs and imagos, 46% were *C. pyri* and 22% were *L. niger*. Collected eggs (24%) were all from Psyllidae. No other species was collected in large abundances in pear. In the apple orchard 25 209 arthropods were collected. Of these 0.5 % were anthocorid nymphs and imago. The relative abundances of other species used in statistical analyses were *D. plantaginea* (15%), *A. pomi* (2%), *L. niger (*6%) and *F. fusca (*4%). The most abundant species were the Psyllid *Cacopsylla mali* (Schmidberger) (28%) and the spider mite *Panonychus ulmi* (Koch) (30%).

Difference between sampling methods

Both the leaf- and beat sampling collected *D. plantaginea* and the 4-5-nymphal stage of *C. pyri*. To get the least amount of zeroes in the dataset and avoid pooling the two

methods, only the leaf samples were used for GLMM. For *D. plantaginea* the leaf samples accounted for the largest number of collected specimens in total (Fig. 10). For the *C. pyri*, only the leaf samples included the first three stages of nymphs (Fig. 11). Table 1 and 2 show the number of insects used further in statistical analysis.

Collected number of *Dysaphis plantaginea* using two different sampling methods

	D. plantaginea 15.06.2015	D. Plantaginea 03.07.2015	D. Plantaginea 18.08.2015	D. Plantaginea 12.09.2015
Leaf	3	2337	136	48
Beating	43	1245	4	2
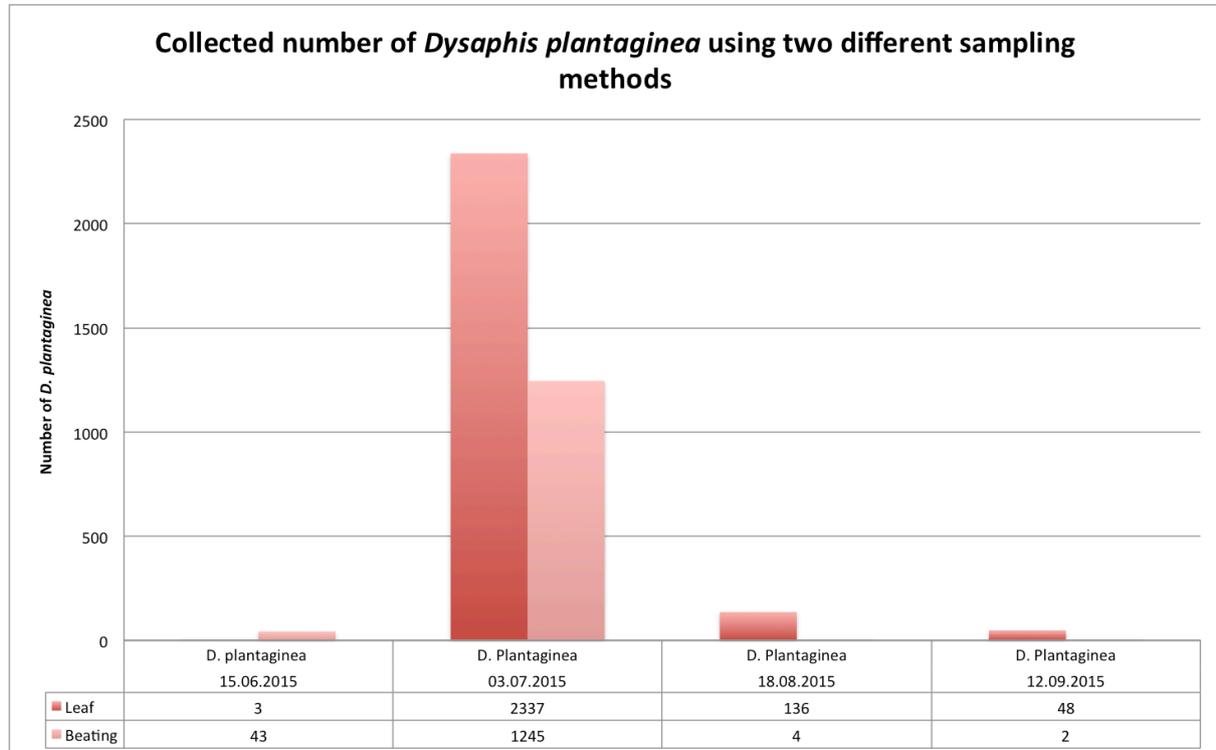
Figure 10: Bar graph shows the numbers of *Dysaphis plantaginea* (summing all mobile stages) in beating and leaf samples at the four sampling occasions in apple. All 30 plots are pooled.

Collected number of *Cacospylla pyri* using two different sampling methods

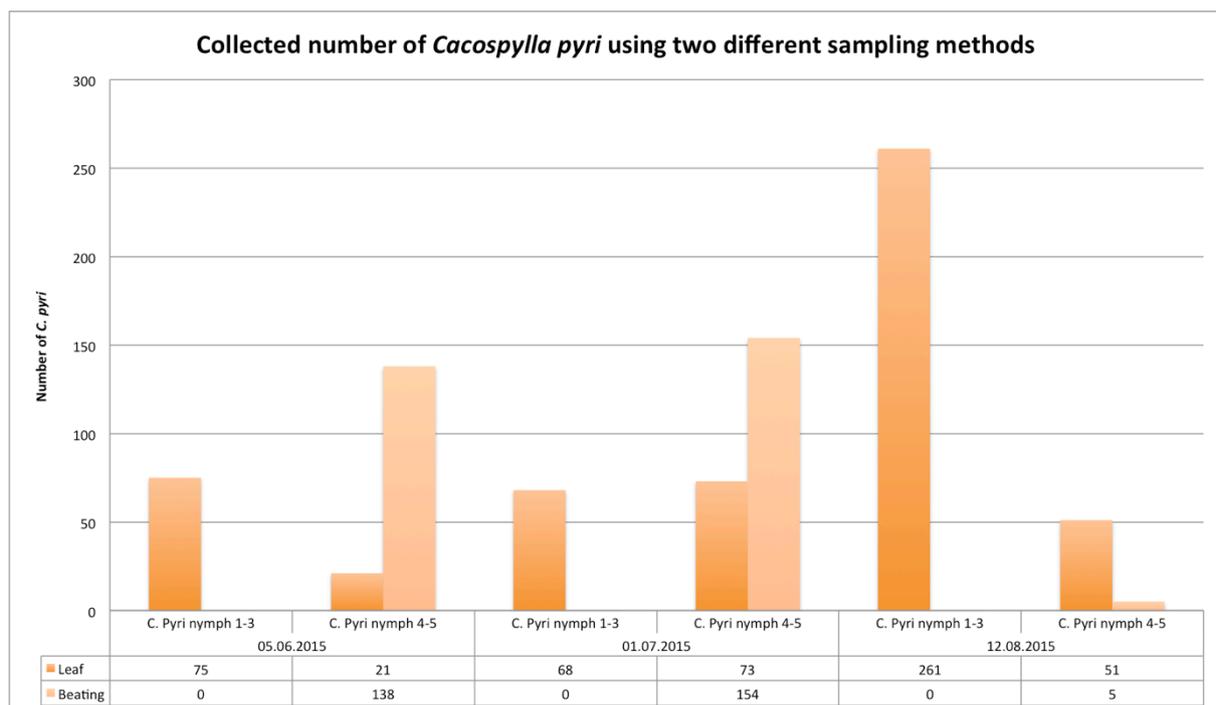|	C. Pyri nymph 1-3 05.06.2015	C. Pyri nymph 4-5 05.06.2015	C. Pyri nymph 1-3 01.07.2015	C. Pyri nymph 4-5 01.07.2015	C. Pyri nymph 1-3 12.08.2015	C. Pyri nymph 4-5 12.08.2015
Leaf	75	21	68	73	261	51
Beating	0	138	0	154	0	5

Figure 11: Bar graph shows the numbers of *Cacopsylla pyri* nymphs in beating and leaf samples at the three sampling occasions in pear. All 30 plots are pooled.

Table 1: Numbers of collected insects in pear used in further statistical analysis. "Insects per plot" is the average of the three sampling periods.

	June	July	August	Insects per plot	Total
Leaf sample					
Psyllid eggs	642	913	656	25 ± 29.4	**2211**
Cacopsylla pyri nymphs	96	141	312	6 ± 7.3	**549**
Beating sample					
Cacopsylla pyri imago	46	1107	1126	25 ± 24.7	**2279**
Anthocoris nemorum imago	24	40	213	3 ± 4.0	**278**
Anthocoris nemoralis imago	7	1	78	1 ± 1.8	**86**
Anthocoris nymphs	0	171	712	9 ± 13.5	**889**
Lasius niger	478	692	376	17 ±18.8	**1548**

Mean ± standard deviation.

Table 2: Number of collected insects in apple used in further statistical analyses. "Insects per plot" is the average of the four sampling periods.

		June	July	August	September	Insects per plot	Total
Leaf sample							
	Dysaphis plantaginea	3	2337	136	48	21 ± 87.8	**2524**
	Aphis pomi	0	0	542	38	5 ± 37.9	**580**
Beating sample							
	Anthocoris nemorum imago	24	13	22	19	0.7 ± 1.0	**78**
	Anthocoris nemoralis imago	1	2	1	0	0 ± 0.2	**4**
	Anthocoris nymphs	0	25	13	2	0.3 ± 0.8	**40**
	Lasius niger	874	606	16	8	13 ± 20.7	**1504**
	Formica fusca	473	415	52	21	8 ± 9.6	**961**

Mean ± standard deviation.

Pear- spatial and temporal relationships

Imago *A. nemoralis* showed a positive spatial relationship with *C. pyri* nymphs combined with psyllid eggs in June and August. Imago *A. nemorum* had a positive spatial relationship with *L. niger* in June while *Anthocoris* nymphs had a negative spatial relationship with *L. niger* in July (Table 3).

Both imago *A. nemoralis* and the *Anthocoris* nymphs showed a positive temporal relationship with the *C. pyri* nymphs combined with the psyllid eggs. *Anthocoris* nymphs showed a negative temporal relationship with *L. niger* (Table 3).

The spatial maps show that imago *A. nemorum* was present in the June samples and was spread out into the entire orchard in June, July and August. Imago *A. nemoralis* however was not caught in abundance until August. It was then found in high occurrence in the southwestern edges of the orchard. *C. pyri* was also more abundant in this edge in August. *L. niger* kept to the same southwestern edge as *A. nemoralis* and *C. pyri* in all three collection periods (Fig. 12).
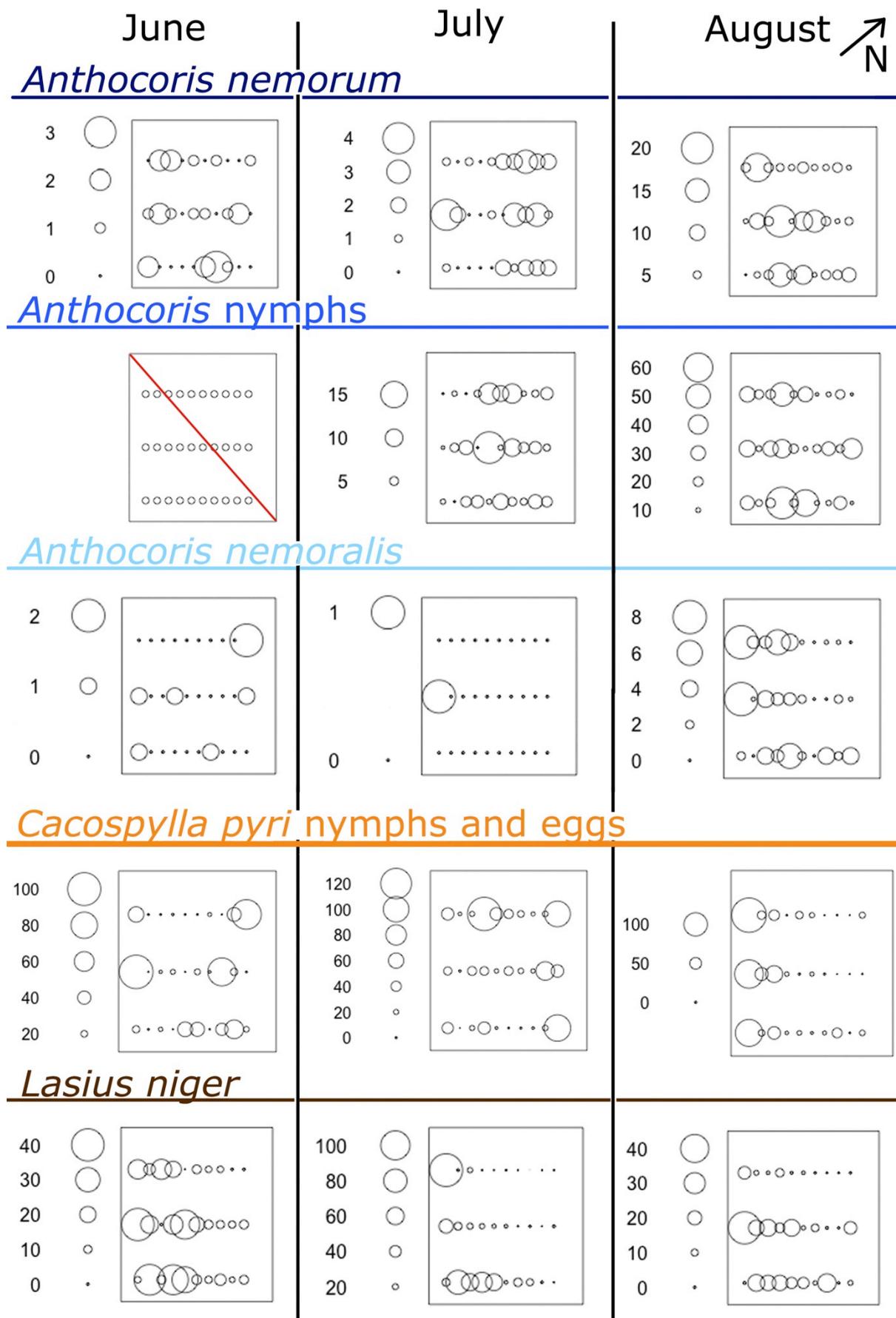
June   July   August

N

Anthocoris nemorum

Anthocoris nymphs

Anthocoris nemoralis

Cacospylla pyri nymphs and eggs

Lasius niger

Figure 12: Spatial map of the pear orchard, showing the number of caught specimens and their location in the different plots. Note that each map has a different value for abundance. Anthocoris nymphs were not found in June.

Anthocorid bugs as natural predators of pear psyllids and apple aphids

Table 3: Spatial relationship between imago *Anthocoris nemorum, imago A. nemoralis* or *Anthocoris* nymphs and the explanatory valuables; *Cacopsylla pyri* nymphs together with psyllid eggs, *C. pyri* imago and *Lasius niger* (GLMM, quasi-poisson family).
Temporal relationship between imago *A. nemorum*, imago *A. nemoralis* or *Anthocoris* nymphs and the same explanatory valuables as spatial (GLMM, negative binomial family). The temporal relationships were analyzed from June to August. Numbers shown are a Pearson product-moment correlation coefficient where 0 = no correlation, 1 = total positive correlation and -1= total negative correlation. All numbers shown are significant factors from GLMM.

		Cacopsylla pyri nymph and eggs	*Cacopsylla pyri* imago	*Lasius niger*
Spatial				
June				
	Anthocoris nemorum	N/S	N/S	**-0.14****
	Anthocoris nemoralis	**0.38***	N/S	N/S
	Anthocoris nymphs	-	-	-
July				
	Anthocoris nemorum	N/S	N/S	N/S
	Anthocoris nemoralis	-	-	-
	Anthocoris nymphs	N/S	N/S	**-0.39****
August				
	Anthocoris nemorum	N/S	N/S	N/S
	Anthocoris nemoralis	**0.60***	N/S	N/S
	Anthocoris nymphs	N/S	N/S	N/S
Temporal				
	Anthocoris nemorum	N/S	N/S	N/S
	Anthocoris nemoralis	**0.34***	N/S	N/S
	Anthocoris nymphs	N/S	**0.51****	**-0.09***

* = *P* < 0.05, **= *P* < 0.01. N/S = not significant

Apple- spatial and temporal relationships

Imago *A. nemorum* showed a negative spatial relationship with aphids *D. plantaginea* and *A. pomi* in August and September. They also showed a negative spatial relationship with *F. fusca* in July and *L. niger* in August. The *Anthocoris* nymphs did not have a significant spatial relationship with any of the explanatory variables (Table 4). Neither the imago *A. nemorum* nor the *Anthocoris* nymphs showed a significant temporal relationship with aphids, *L. niger* or *F. fusca* from June to July (Table 4).

The spatial maps show that *F. fusca* and *L. niger* appeared in plots closest to the pine forest in the eastern edge in all collection periods in the apple orchard. *A. pomi* and *D. plantaginea* populated the plots closer to the center of the orchard in July. In August and September the specimens mostly occurred along the eastern edge of the orchard. *A. nemorum* was in the southwestern corner in June and then spread out more or less to the whole orchard during July, August and September, except the eastern edge (Fig. 13).
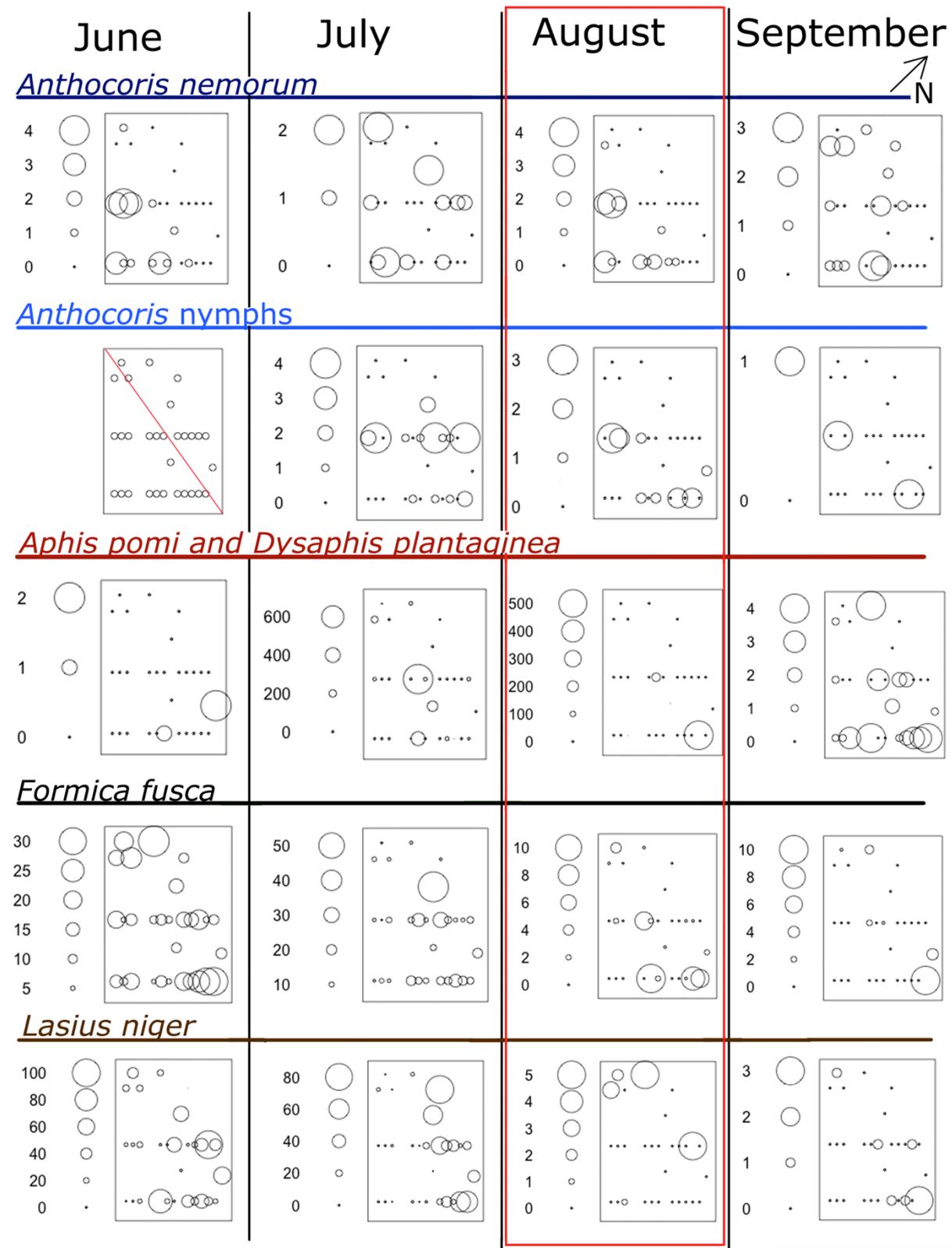
Figure 13: Spatial map over the apple orchard, showing the number of caught specimens and their locations in the different plots. Note each map has a different value for abundance. Anthocoris nymphs were not found in June. August is framed in red because of an unplanned spot spraying in that month.

Results

Table 4: Spatial relationship between imago *Anthocoris nemorum* or *Anthocoris* nymphs and the explanatory valuables; (1) Aphis pomi and *Dysaphis plantaginea*, (2) *Lasius niger* and (3) *Formica fusca* (GLMM, quasi-poisson family). Temporal relationship between imago *A. nemorum* or *Anthocoris* nymphs and the same explanatory valuables as in spatial formula (GLMM, negative binomial family). The temporal relationships were only analyzed from June to July, because of spot spraying in August. Numbers shown are a Pearson product-moment correlation coefficient where 0 = no correlation, 1 = total positive correlation and -1= total negative correlation. All numbers shown are significant factors from GLMM.

		Aphis pomi and *Dysaphis plantaginea*	*Lasius niger*	*Formica fusca*
Spatial				
June				
	*Anthocoris nemorum^*	N/S	-	-
	Anthocoris nymphs	-	-	-
July				
	Anthocoris nemorum	N/S	N/S	**0.21***
	Anthocoris nymphs	N/S	N/S	N/S
August				
	Anthocoris nemorum	**-0.16***	**-0.20***	N/S
	Anthocoris nymphs^	N/S	-	-
September				
	*Anthocoris nemorum^*	**-0.30***	-	-
	Anthocoris nymphs	N/S	N/S	N/S
Temporal				
	Anthocoris nemorum	N/S	N/S	N/S

* = $P < 0.05$, **= $P < 0.01$, ***= $P< 0.001$. ^= *L. niger* and *F. fusca* not added as explanatory values in formula.
\- = Specimens not present in samples. N/S = not significant

Discussion

In agreement with my predictions the predator *A. nemoralis* had a positive spatial and temporal relationship with its prey: *C. pyri* nymphs and psyllid egg abundances. This did not however fit with the results from the sleeve-cage experiment, where *A. nemoralis* did not affect the *C. pyri* numbers. In partial disagreement with my predictions *A. nemorum* had a negative relationship with not only the ant *F. fusca* but also with the prey species *A. pomi-* and *D. plantaginea* in apple. The result from the sleeve-cage experiment in apple indicates that the aphid population could be controlled by *A. nemorum,* at least if ants are absent from the trees.

The sleeve-cage experiments

Pear

Contrary to my predictions *A. nemorum* was significantly better at controlling *C. pyri* numbers than *A. nemoralis*. This contradicts previous studies where *A. nemoralis* is considered to be more specialized towards eating pear psyllids than *A. nemorum.* (Herard & Chen 1985; Scutareanu et al. 1996; Scutareanu et al. 1999; Shaltiel & Coll 2004; Sigsgaard et al. 2006a).

The *A. nemoralis* used in the sleeve-cage experiment was bought from a biocontrol company in Denmark. This could have affected the *A. nemoralis* individuals in many different ways. It is not known in which country the original population were collected. There could be genetic differences in the *A. nemoralis* compared to the Norwegian populations. They could also have been affected by a change in climate, or by genetic differences in the prey. The Danish reared population could also have been fed on different taxa of arthropods, and could then need time to adjust to *C. pyri* (Henaut et al. 2000). *A. nemoralis* used for the sleeve-cage experiment was sent with mail and stored at 8 °C a few days prior to release. Their overall condition can therefore have been influenced negatively by the shipping period from Denmark and the wait in cool storage temperature. *A. nemorum* used in the sleeve-cage experiment was collected from pear orchards one to two days before. It is therefore probable that they were used to eating

pear psyllids, and did not need a familiarization period with the prey or host plant species.

The number of *C. pyri* collected after the end of the experiment was in half of the sleeve-cages higher than the original 10 introduced individuals (16 of 30 sleeve-cages had more than 10 individuals). This indicates that the branches were not cleansed satisfactory of psyllid eggs before the sleeve-cages were put on.

Apple

As expected, *A. nemorum* significantly reduced the *A. pomi* abundances in the sleeve-cages. This result is in accordance with other studies (Sigsgaard 2010; Solomon et al. 2000). Feeding experiments using Petri-dishes in a previous study in Norway (Våge 1991) suggest that *A. nemorum* has a preference for aphids compared with psyllids typically found in an apple orchard. The average consumption per 24 hours was 3.9 aphids when no other prey was present. The *A. nemorum* in my study could potentially have cleaned the sleeve-cages of aphids in less than two days. The sleeve-cage experiments hindered the presence of ants and other potential prey species. These results indicate that *A. nemorum* could be a good biological control agent if the right conditions are present

The orchard surveys

Pear

A. nemoralis spatial and temporal patterns

There was a positive spatial and temporal relationship between imago *A. nemoralis* and *C. pyri* nymphs and psyllid eggs. Many other researchers have found a relationship between the two species in their studies (Drukker et al. 1995; Scutareanu et al. 1996; Shaltiel & Coll 2004; Sigsgaard 2005; Sigsgaard et al. 2006b; Solomon et al. 1989). For instance Novak and Achtziger (1995) found a positive correlation between the number of *A. nemoralis* and the size of the populations of three psyllid species found on hawthorn (*Crataegus*) in Germany. Scutareanu et al. (1999) reports that anthocorid bugs increases in numbers in pear orchard in the Netherlands when the density of the pear psyllid population increase; there is a numerical response of the predators when the

prey species increases. This corresponds to my own findings. The imago *A. nemoralis* population in my study increased late in summer, supporting studies in other parts of Europe (Artigues et al. 1995; Scutareanu et al. 1999; Solomon et al. 1989). It can be argued that *A. nemoralis* may come into the orchards too late to prevent fruit damage when there is a psylla outbreak (Shaltiel & Coll 2004).

The *C. pyri* population did not exceed the grower's damage threshold and hence no spray were targeted against it (pers. comm. pear grower Jostein Lutro). This could be because the anthocorid bugs kept the population down. This is supported by Næss (2016) where molecular analyzes of the anthocorids gut content showed that *A. nemorum* and *A. nemoralis* in the same researched field and summer as my study predated on *C. pyri.* The low numbers of *C. pyri* could also be because of other factors. The temperature in early summer was unusually cool, and probably affected the growing rate of the *C. pyri* population (Bale et al. 2002; Norwegian Meteorological Institute 2014-2015). The orchard was reported to have populations of *C. pyri* over the damage threshold the previous year (pers. comm. pear grower Jostein Lutro). According to Edland (2004) pear orchards are rarely affected by large pear psyllid populations several years in a row. The previous year was unusually warm and had three generations of *C. pyri* (pers. comm. pear grower Jostein Lutro) (Norwegian Meteorological Institute 2014-2015). This could have affected the anthocorid numbers next summer (Symondson et al. 2002). The food resources would have been abundant and late in the season, so more anthocorid individuals potentially survived the winter.

The spatial relationship between *A. nemoralis* and *C. pyri* nymphs and psylla eggs could also be the result of abiotic factors like wind, sun exposure, rain or other spatially heterogeneous factors like host plant defense, which influenced the pests to move to a particularly favorable spot in the orchard.

Anthocoris density

In the pear orchard there were on average 13 *Anthocoris* imagos or nymphs per plot and in apple 1 *Anthocoris* imago or nymph sampled per plot. When converted to hectare to make it easier to compare to other studies and recommended release rates; there would be 4121 anthocorids per ha in pear and 209 per ha in apple. The recommended release

rate of *A. nemoralis* per ha is 1200-2000 specimens, unrelated to how many anthocorids already inhabiting the orchard. In Beninato and Morella (2000) study in Italy, this release rate proved to be unsuccessful. The pear orchard in my study had natural abundances twice as high as the suggested release rate, and it was sufficient to keep the *C. pyri* population below the damage threshold. The farmer cultivating the orchard studied has reduced pesticide use in the last years, which may partly explain this high number of anthocorid bugs. If around 4000 *Anthocoris* are needed to keep psyllid damage low, a release of the recommended numbers of 1200 to 2000 specimens per ha would not have been sufficient if the anthocorid numbers were as low as in the apple orchard. The recommended release rate should be dependent on how many pre-existing anthocorids there are in the orchard.

The release rate should also be dependent on temperatures. High temperatures decreases the generation time of insects, and can increase the pest-population's potential to grow (Bale et al. 2002). Anthocorid bugs also responds to increasing temperatures (Simonsen et al. 2009). It is not known if one of the species responds more to high temperatures than the other.

The density ratio of anthocorid nymphs to *C. pyri* and psylla eggs was three *C. pyri* or psylla eggs per anthocorid nymph per plot. According to Brunner and Burts (1975) *A. nemoralis* nymphs need between 5 and 10 small *Cacopsylla pyricola* nymphs or eggs every day to complete development to imago stage. The anthocorid population in pear in my study will therefore likely be smaller next year. In apple the density ratio was lower, with one anthocorid imago or nymph per 26 aphids per plot. There was also an abundance of other prey, like *C. mali* to sustain the population of anthocorids.

The high density of *A. nemorum* compared to *A. nemoralis* was not expected when compared to other studies. According to Sigsgaard (2004) Danish specimens of *A. nemorum* prefers to oviposition on apple leaves rather than pear. Scutareanu et al. (1999) found higher abundances of *A. nemoralis* compared to *A. nemorum* in pear orchards in the Netherlands. Norwegian populations of *A. nemorum* could be more flexible when it comes to selecting host plant when compared to populations in other European countries.

Differences between *A. nemorum* and *A. nemoralis*

Unlike *A. nemoralis*, imago *A. nemorum* had no statistically significant relationship with *C. pyri*; but imago *A. nemorum* was present in the pear orchard in larger abundances than imago *A. nemoralis*, and it was spread out in the entire orchard. There was no other abundant prey group in the orchard, indicating that imago *A. nemorum* mostly predated on *C. pyri*. Because imago *A. nemorum* did not have a statistically significant spatial relationship with *C. pyri*, it seems they are not as good at finding their prey as *A. nemoralis*. *A. nemoralis* is considered to be more specialized towards eating pear psyllids than *A. nemorum* (Sigsgaard 2010). Even though both *A. nemoralis* and *A. nemorum* has been shown to locate pear psyllids from volatiles sent by distressed pear trees (Drukker et al. 1995; Scutareanu et al. 1996; Scutareanu et al. 1999). *A. nemoralis* were better at finding pear psyllids, but both of the anthocorid species were probably important in this pear orchard when it came to keeping the *C. pyri* population under the damage threshold. Unlike imago *A. nemorum*, imago *A. nemoralis* arrived too late in the season to have an impact on the *C. pyri* population before damage to the pear trees was possible.

Ant- psyllid relationship

Anthocoris nymphs had a negative temporal relationship with *L. niger*. According to Novak (1994) who studied *L. niger* tending psyllid species on hawthorn; nymphs that were tended had a higher survival rate than the ones that were not. *L. niger* may repel the *Anthocoris* nymphs more than imagos because they are smaller in size. In accordance with this, *Anthocoris* nymphs had a spatial negative relationship with *L. niger* in July but not in August. *A. nemorum* and *A. nemoralis* imagos did not have a negative relationship with *L. niger* abundances.

The spatial maps of the pear orchard (Fig. 12) showed that *L. niger* and *C. pyri* nymphs and psylla eggs were in greater abundance close to the southwestern edge vegetation in August. *L. niger* may have protected the psylla against predation from *A. nemorum,* which had been abundant in the rest of the orchard the two previous collection periods.

Imago *C. pyri* as an explanatory variable

Because the different stages of the *C. pyri* life cycle were collected with two sampling methods, they were not summed and used as one explanatory variable in the GLMM formula for spatial or temporal analyzes. The two variables (1) imago and (2) psyllid eggs and *C. pyri* nymphs are expected to be correlated, because they represent the same species. In my study, *C. pyri* imagos did not have a significant relationship with *A. nemorum* or *A. nemoralis.* The same relationship was found by Scutareanu et al. (1999). This could be because *C. pyri* imago has an effective defense strategy where it can jump quickly away from danger. It is therefore hard to predate, making it a less profitable prey than *C. pyri* nymphs (Edland 2004).

Apple

Ant- aphid relationship

A. nemorum showed a negative spatial relationship with the abundances of *F. fusca* in July, indicating they deter the anthocorid's presence in the orchard. *F. fusca* are known to tend aphids and could also deter the anthocorid predation on *D. plantaginea* and *A. pomi* (Cushman & Addicott 1989; Novgorodova 2005; Stewart‑Jones et al. 2008).

A. nemorum showed a negative relationship towards apple aphids and *L. niger* in August. This could be because *L. niger* tends the aphids and protects them from predator attacks. Because *A. nemorum* is considered a polyphagous predator, it could easily switch from one prey to another if the handling cost of aphids increased too much in the presence of ants (Simonsen et al. 2009; Symondson et al. 2002). The negative relationship could therefore be because the *A. nemorum* switched to other available prey like *C. mali* and *P. ulmi*. According to Nagy et al. (2013) *L. niger* forms mutualistic bonds with both *A. pomi* and *D. plantaginea*. In Stewart‑Jones et al. (2008) study, the trees with a presence of *L. niger* had the highest number of predators in total, but the study also pointed out that the predator to aphid ratio was lower on trees with *L. niger* than on trees without.

Spot spraying of aphids in the organic orchard

Two days before the August sampling, the grower sprayed the trees infected by aphids with a pyrethrin compound, legal to use in organic farming. As a result the collected number of *D. plantaginea* from July to August dropped by 94%. This affected my research on spatial and in particular, temporal patterns. The relationship between the ants and apple aphids could have been affected by the spraying. Cushman and Addicott (1989) found that colonies of fireweed aphids (*Aphis varians*) had a significantly larger probability of growing after a decline in the previous week, if they were tended by three or more *F. fusca.* The spatial maps (Fig. 13) suggest a spatial connection between apple aphids and *F. fusca* in August, supporting Cushman and Addicotts research.

The spot spraying could explain the anthocorid negative spatial relationship with apple aphids in August. However the spatial pattern could also be a transitional phase. The anthocorids may not had time to redistribute after the major change in prey abundance two days prior. According to Souliotis and Moschos (2008), pyrethrin spraying also decreases the number of *A. nemoralis* drastically. The number of *A. nemorum* in this study did not statistically decline from July to August, but when compared to the data from the pear orchard, the lack of a strong increase is conspicuous.

There was a strong increase of *P. ulmi* specimens in the September samples. This corresponds with other research, showing that using pyrethrin compounds for insect control is not compatible with controlling populations of mite (Aliniazee & Cranham 1980).

Edge vegetation

Pear

In the pear orchard, imago *A. nemorum, A. nemoralis and L. niger,* as well as *C. pyri* nymphs and psyllid eggs were in greater abundance in the southwestern edges in August. The aggregated abundances of *C. pyri* nymphs could be because of favorable abiotic factors, or that the predation of *A. nemorum* in June and July reduced the population in the rest of the field, leaving only the southwestern edge. In August, *A. nemoralis* stayed close to the southwestern edges of the pear orchard. This could be because *A. nemoralis* did not venture far into the orchard, or because it had not been in

the orchard long enough to relocate when the sampling was done. Other studies suggest *A. nemoralis* stay in vegetation close to pear orchards until late summer, supporting my results.

A. nemoralis is known to stay in nettle (Herard & Chen 1985). There were nettles in the northeastern corner of the pear orchard, outside of my study area. There was no indication from the spatial maps (Fig.12) that *A. nemoralis* had stayed in the nettles before moving into the orchard, but because the edge closest to the nettles was not in in my study area it was not sampled.

Vegetation surrounding the southwestern edge was birch, pine, goat willow and different graminoids. According to Saulich and Musolin (2009) both *A. nemoralis* and *A. nemorum* stay on trees from the genus *Salix*. The high abundances of *A. nemoralis* in the southwestern corner in August could be explained by the presence of Goat willow; underlining the importance of maintaining vegetation in edges of orchards that are favorable for predator-species (Artigues et al. 1995; Shaltiel & Coll 2004).

Apple

I have not considered the edge vegetation as important in apple as in the pear orchard, because the apples in my study were organically grown and had much higher species diversity within the orchard itself. In the apple orchard the spatial maps (Fig. 13) show that both of the ant species were in greater abundance in the eastern corner, which is surrounded by pine forest. The aphids were also more abundant in this corner after the spot-spraying in August. *A. nemorum* was in the southern corner in June and spread to most of the apple orchard in July, August and September; except the eastern corner where ants and aphids were present. *A. nemorum* avoided this area in the orchard in all four sampling periods, further strengthening the earlier discussed results about ants deterring anthocorid bugs.

Further studies

There is still a need to further understand how the predator- pest interactions in apple and pear orchards work. Release experiments are often not satisfactorily and various

factors may affect the outcome (Beninato & Morella 2000; Waage et al. 2012). The important factors may vary from country to country; different climate changes the species composition and life cycles. Because of this it is important to do research in different countries or climate gradients.

Feeding experiments should be done to check what the *A. nemorum* and *A. nemoralis* prefer to eat when only presented with different species found in a typical Norwegian pear orchard. The Norwegian naturally occurring *A. nemoralis* should also be compared to the commercially available *A. nemoralis* from biological control companies abroad. The comparison should include feeding preferences, the differences in rearing conditions, activity under different temperatures, differences in oviposition temperature and at what time of year the oviposition occurs. Genetic differences should also be mapped.

More field experiments should be done around Europe, to find the *Anthocoris* density that will keep psyllid and aphid populations under control. The density will probably depend on the relative response of *Anthocoris* and prey species to temperature. More knowledge about how plant species, and vegetation structure surrounding orchards affects the anthocorid abundance is also needed.

Lastly, this survey should be repeated in a year closer to "normal" temperatures to observe patterns when psyllid densities are much higher.

Conclusion

Natural populations of *A. nemorum* and *A. nemoralis* can control *C. pyri* population in pear. In my study, the number of *A. nemoralis* was more related to abundances of *C. pyri* nymphs and eggs; while the more generalist *A. nemorum* was present in the orchard in larger abundances and earlier in the season. Because of this, *A. nemorum* may be more important in controlling pear psyllids in Norway than previously assumed. Commercially bought *A. nemoralis* did not manage to control *C. pyri* numbers in my exclusion experiments. Differences between commercially reared and natural populations of *A. nemoralis* should be researched. Natural populations of *A. nemorum* have the potential to control aphids if they are present in large abundances. The number of ants in this study had a negative relationship with the number of anthocorid bugs in both pear and apple; ants should therefore be taken into account when biocontrol systems in orchards are planned.

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