Developing IPM Tools for Greenhouse Cucumber Production in Sweden – A Participatory Action Research Approach

Control of the European Tarnished Plant Bug and Cucurbit Powdery Mildew

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Licentiate Thesis
Swedish University of Agricultural Sciences
Alnarp 2016

Cover: Cucurbit powdery mildew and *Lygus rugulipennis* in cucumber greenhouses. (Photos: *Upper left:* Mira Rur (Powdery mildew infected cucumber plant in greenhouse experiment), *Upper middle:* Thilda Håkansson (Adult *L. rugulipennis* on cucumber plant), *Upper right*: Barbro Nedstam (damage on cucumber crop caused by *L. rugulipennis*), *lower left*: Sara Johansson (damage on cucumber shoots by *L. rugulipennis, Lower right:* Mira Rur (Sunflower as trap crop in commercial greenhouse experiment).

ISBN (print version) 978-91-576-9428-7
ISBN (electronic version) 978-91-576-9429-4
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Print: SLU Service/Repro, Uppsala/Alnarp 2016

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Abstract

Two of the most important plant protection problems in Swedish cucumber production are the European Tarnished Plant Bug (ETPB), and Cucurbit Powdery mildew (CPM). The control of the ETPB relies on Imidacloprid, a pesticide, which breaks down slowly and is harmful to beneficial insects. CPM fungi has begun to develop resistance to the commonly used fungicide (Imazalil) rendering its current use less effective.

The main objective of this thesis was to evaluate alternative control methods for future incorporation into IPM strategies against ETPB and CPM using a participatory action research (PAR) approach. Towards this aim, two projects, tackling both of these problems were conducted in consultation between growers, researchers and advisors.

The specific objectives of the ETPB project were to 1) compare responses of the ETPB to cucumber and candidate trap crops in olfactometer assays, 2) examine the responses of the ETPB to headspace volatile collections of candidate trap crops, 3) identify the attracting chemical compounds and 4) examine if sunflower could serve as a trap crop for ETPB in commercial cucumber greenhouses. The specific objectives of the CPM project were to 1) screen for effective alternative products against CPM and 2) to evaluate these, alone and in combination with Imazalil at different application intervals and in cultivars with different levels of resistance.

The ETPB study showed that sunflower was more attractive than cucumber in greenhouse experiments but did not provide a sufficient level of control. In olfactometer assays, adults were more attracted to odours from flowering sunflower or lucerne than odours from flowering cucumber. Chemical analysis of plant odours showed a distinct differentiation between sunflower and cucumber. Sunflower exclusively released a number of monoterpenes and had an overall emission rate almost four times higher than cucumber. Therefore, it may be possible to use synthetic sunflower volatiles to attract ETPBs in the future.

In semi-commercial CPM experiments, Sakalia, based on *Reynoutria sachaliensis* combined with Yuccah, a wetting agent, based on *Yucca Schidigera,* applied at 7-day intervals, consistently had the most suppressive effect, on CPM disease severity in two commercial cucumber cultivars. Further testing of this combination in commercial greenhouses is proposed to enable evaluation of the potential effects on yield and beneficial insects. The PAR approach took advantage of the knowledge and experience of researchers, advisors and growers and was seen as highly rewarding by all participants. Based on the results from this thesis, a potential future IPM strategy to control the major insect and fungal pests of cucumber is proposed.

Keywords: integrated pest management, cucurbit powdery mildew, *Podosphaera xanthii*, *Lygus rugulipennis,* participatory action research

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Dedication

To Misha

Att våga är att förlora fotfästet en stund. Att inte våga är att förlora sig själv.

Søren Kierkegaard

Contents

List of Publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

I Ondiaka S, Migiro L, Rur M, Birgersson G, Porcel M, Rämert B, Tasin M. 2015. Sunflower as a trap crop for the European tarnished plant bug (*Lygus rugulipennis*), Journal of Applied Entomology 140 (6), 453-461.

II Rur M., Rämert B., Hökeberg M., Vetukuri R. R., Grenville-Briggs L., Liljeroth E. Screening of alternative products for integrated pest management of cucumber powdery mildew in Sweden (manuscript).

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The contribution of Mira Rur to the papers included in this thesis was as follows:

I Planned and performed all greenhouse experiments and some laboratory experiments together with co-authors. Continuously presented results to the working group and co-authors.

II Planned and performed all greenhouse experiments supervised by co-authors. Analysed data and wrote the paper together with co-authors.

Abbreviations

CPM	Cucurbit powdery mildew
IPM	Integrated Pest Management
AUDPC	Area under disease progress curve
rAUDPC	Relative area under disease progress curve
ETPB	the European tarnished plant bug
VPD	Vapour pressure deficit
RH	Relative humidity
AR	Action research
PAR	Participatory action research

1 Introduction

One of the biggest challenges today is to produce high quality food for an increasing population in a sustainable way. Greenhouse production of crops is considered the most intensive form of vegetal production. However, the advanced technology available today offers the possibility to grow greenhouse crops in a highly resource efficient way (Garcia-Mier *et al.*, 2014; Dorais *et al.*, 2013).

In 2013, worldwide production of cucumbers and gherkins reached 71.3 million tonnes ranking it at place 24 of the top 50 list of food and agricultural commodities produced in the world. In Sweden, cucumber is the most widely grown greenhouse crop (Persson, 2015a; *FAOSTAT*, 2015).

Advances made in the last decades in the use of biological control against pests and disease, breeding of resistant cultivars and the possibility of recycling greenhouse waste water have also contributed greatly to improving the sustainability of greenhouse cropping systems. Nonetheless, cucumber growers today still struggle with many pests and disease problems. Although the climate of the greenhouses can be controlled by environmental computers, the balance between maintaining plant vigour and high production at the same time as avoiding favourable conditions for pests and pathogens throughout the season is difficult. The humid greenhouse environment with highly productive plants most often offers an excellent breeding ground for various pests and pathogens.

The EU framework directive 2009/128/EC on the sustainable use of chemical pesticides, made it compulsory for farmers to follow integrated pest management principles from January 1, 2014. This has further stressed the need to find efficient alternatives to chemical pesticides and examining how they can work together in an integrated pest management (IPM) system.

Two of the most important plant protection problems of Swedish cucumber production are the European tarnished plant bug (ETPB) and Cucurbit powdery mildew (CPM). The control of the ETPB in conventional cucumber production in Sweden today mainly relies on a chemical (Imidacloprid) which breaks down slowly and is harmful to biological control agents such as predatory mites. As for CPM, the fungicide Fungazil 100 (Imazalil), which is available in Sweden and has been most effective up till now, recently began to loose its effect as the pathogen has developed resistance.

In this thesis, alternative methods aimed for incorporation into IPM strategies against the ETPB and CPM have been tested and evaluated. Two projects have been performed using the methodology of participatory action research. Researchers together with growers and advisors have collaborated in order to find highly applicable and practical solutions for the cucumber production industry. Hopefully, the findings of these studies can contribute to a more sustainable way of controlling the ETPB and CPM in the future and thus impose less of a risk for the environment, for the health of the greenhouse workers and for development of fungicide resistance.

Using action research methodology means taking advantage of the different experiences and the knowledge of all group members in order to reach higher learning outcomes and highly relevant and applicable results. In the process of working with these projects, researchers, advisors and growers have worked together to find new solutions to highly relevant problems but also learned from each other and gained more understanding of each other's professional roles. Furthermore, it seems that collaboration between stakeholders can facilitate in the development and implementation of IPM strategies by bringing attention to potential bottlenecks or gaps of knowledge thus enabling more focused efforts.

2 Background

2.1 Cucumber

2.1.1 Origin and botany

Cucumber, *Cucumis sativa*, belongs to the very diverse *Cucurbitaceace* plant family including about 115 genera (Pitrat *et al.*, 1999). Other cultivated cucurbits include melon, watermelon, squash, pumpkin, chayote, citron melon, gherkin, gourds, horned cucumber and wild type cucumber. Cucurbits are grown in many different climate zones and landscapes, in both field and greenhouse settings (Zitter *et al.*, 1996).

Different cucurbits originate from different parts of the world. Cucumber, which along with melon is one of the most well-known members of the *Cucumis* genus, most likely originates from India and was domesticated around 1,500 BC (Pitrat *et al.*, 1999; Zitter *et al.*, 1996). The strongest evidence for this is the occurrence of the wild predecessor *C. sativus* var. hardwickii which is fully cross compatible with cucumber (Sebastian *et al.*, 2010; Pitrat *et al.*, 1999). Cucumber is known from descriptions in Iraq dating back to 600 BC and from the Mediterranean region in 200 BC. The roman emperor Tiberius was allegedly very fond of cucumber (Pitrat *et al.*, 1999).

Cucurbits are either monoecious, andromonoecious or gynoecious depending on species. Depending on the sexual system, different flower types are produced during different phases. The flower types can be hermaphroditic, pistillate or staminate. Monoecious and gynoecious F1 hybrid cultivars have been developed by manipulating sex expression (Zitter *et al.*, 1996).

Greenhouse cucumber cultivars grown today for fresh consumption are usually parthenocarpic. This means that they have the ability to produce fruits without pollination and fertilisation. They are also gynoecious otherwise they would still set seed when being pollinated which would reduce the eating quality (Zitter *et al.*, 1996).

2.1.2 Cucumber production

In 2013, worldwide production of cucumbers and gherkins reached 71.3 million tonnes placing it on the top 50 list of produced amount of food and agricultural commodities of the world. With a production of 54.3 million tons, China is by far the world's biggest producer of cucumber and gherkins followed by Turkey and Iran which produced 1.7 and 1.5 million tons, respectively, in 2013 (*FAOSTAT*, 2015).

In Sweden, cucumber is the most widely grown greenhouse crop. For example, 67.2 ha of greenhouse space across Sweden were used for cucumber production in 2013. This yielded a total of 28 000 tonnes or 41.7 kilos per square meter. The second most grown greenhouse crop in Sweden is tomato (Persson, 2015a).

Figure 1. Cucumber flower in commercial greenhouse (Photo: Mira Rur).

2.1.3 Plant protection

Over the last three decades, the use of biological control of pest insects has increased greatly in greenhouse vegetable production in Sweden and in other countries (Jordbruksverket, 2001; Jarvis, 1992). However, in Sweden, there is still a lack of efficient chemical and biological control agents for several pest insects and some of them have developed resistance to commonly used chemical pesticides. Because of the extensive use of biological control insects there is also a demand for more selective chemical control agents without harmful effects on beneficial organisms. Biological control of pathogens is not as widely used and there are fewer products available than for pest insects (Jansson, 2016; Jordbruksverket, 2001). As reported by growers participating in this study, cucumber growers still have to rely a great deal on chemical pesticides in order to secure production, especially for control of fungal pathogens.

Greenhouse workers, in particular the persons in charge of mixing and applying chemical pesticides, are facing the greatest risk of exposure. As reported by Bolognesi (2003), the enclosed spaces, high temperature and humidity of the greenhouse environment increase the risk of exposure. Even if proper re-entering intervals are employed after chemical pesticide application, there is a prevailing risk from prolonged low level exposure. Residues of chemical pesticides on plants can be absorbed through skin if unprotected (Bolognesi, 2003).

2.2 Modern greenhouse cropping systems

Greenhouse production of crops is a highly intensive form of vegetal production by which the growing season in temperate climates is extended and large yields can be obtained from a relatively small area. The advanced greenhouse technology available today has also opened up the possibility to grow greenhouse crops in a more sustainable and resource efficient way.

Examples of greenhouse technologies which contribute to increasing the sustainability and the production per cultivated unit area are:

- Optimization of greenhouse structures and coverings for better light transmissivity.
- Hydroponic cropping with recycling of greenhouse waste water.
- Use of high-performance culture media improving the root environment.
- Enrichment of the greenhouse atmosphere with carbon dioxide to increase productivity.
- Use of lighting in winter to secure year-round production.
- Computational management of climate (temperature, light, moisture, CO_2), irrigation and fertilization.
- Use of rootstocks resistant to pathogens and with higher water use efficiency.
- Use of biological control against pests and diseases.
- Genetically improved new varieties.
- Mechanisation of procedures to reduce labour.
- Improved energy efficiency and the use of renewable energy.
  (Dorais *et al.*, 2013)

In temperate climate zones, where heating is needed to prolong the season, the level of sustainability is of course very much dependent on the fuel source. In recent years, a major transition from fossil fuels, such as oil and natural gas, to renewable biofuel alternatives, such as wood chips and pellets, has been made by Swedish greenhouse companies. For example, in 2014, cucumber companies used 50 136 MWh of fossil fuels compared to 73 668 MWh of biofuels. In 2002, the same figures were 183 436 MWh for fossil fuels compared to 10 993 MWh for biofuels (Persson, 2015b). According to growers, the transition has been mainly driven by taxes levied on carbon dioxide emissions.

In greenhouse systems, solar radiation deficiencies are the most common factor limiting production. It cannot be compensated for with heating systems but rather by complementary lighting. Excess solar radiation in turn, can be redressed by ventilation to release hot air masses or by direct cooling systems.

Greenhouse constructions and growing systems including spacing and training systems have all been developed to maximise productivity of the crop, i.e. to maximise the photosynthetic area per unit of ground area. Cucumber plants are trained on strings attached to support wires at approximately 2.5 metres height (Jarvis, 1992). In Sweden, the renewal umbrella training system is most common and the standard average plant density is 1.5 plants per square meter. The renewal umbrella system is one of several training systems used to maximise productivity of the plants. After pinching out the growing point of the main stem as it reaches the top wire, only the two top lateral shoots are allowed to grow and are trained over the wire to hang down the sides of the main stem. In short, continuous pruning/pinching is then made of older shoots, to promote continuous generative growth.

An average weekly temperature of 21°C is optimal for cucumber production and is what the cucumber grower strives for. Temperature affects humidity by regulating the water vapour retaining capacity of the air masses. For optimal plant vigour and productivity, obtaining the right vapour pressure deficit (VPD) is more important than relative humidity. In short, for the plant, the VPD is the difference between the vapour pressure inside the leaf compared to the vapour pressure of the surrounding air and gives a more accurate description of how temperature and humidity immediate to the crop is affecting the plants and their transpiration. VPD is expressed in pressure or concentration units. This information helps growers to manage climate and plant transpiration as a strategy to maximise photosynthesis and thus productivity of their plants (Government of Alberta, 2003). The VPD unit commonly used by environmental computers in Sweden is g/m^3, where the optimal range is between 3 to 7 g/m^3 (Government of Alberta, 2003).

Irrigation levels and fertiliser composition are adjusted in relation to climatic conditions and growth stage of the plants. Fertilisation is made through trickle irrigation systems. Optimal pH and conductivity for cucumber is in the intervals 5.5-6.2 and 2.0-3.0 mS/cm, respectively (Badgery-Parker *et al.*, 2015). Analysis of the nutrient feed solution is usually made on a biweekly basis to enable adjustments of the nutrient composition.

Carbon dioxide enrichment of the greenhouse atmosphere is commonly used to increase crop productivity. Increasing the carbon dioxide concentration means increasing the efficiency with which light is converted to chemical energy during photosynthesis. Optimal CO_2 concentrations for the greenhouse atmosphere are in the range of 700 to 900 ppm (Government of Alberta, 2003).

The rooting media and substrates available to conventional cucumber growers today such as rock wool, perlite, and pumice and nutrient solutions are pathogen free from the start and most of these can be sterilized or pasteurized and reused. In Sweden, many cucumber growers use a hydroponic technique where plants are sown in rock wool cubes and placed on pumice gravel. The pumice is sterilised each year to avoid disease spread and reused for up to 8 years.

The greenhouse climate is monitored and controlled by environmental computers. As radiation, water relations, carbon dioxide exchange, and nutrition control are crucial factors for plant health, and thus crop production, as well as in pathogenesis, the greatest challenge of managing greenhouse cropping systems lies in obtaining the skills to manipulate these environmental factors. It is how these challenges are handled that determine both environmental and economic sustainability. An experienced grower can "read" the plant and make suitable environmental adjustments when necessary (Jarvis, 1992). By manipulating climate, particularly many fungal diseases can be avoided or kept at low levels. Nonetheless, to scout the greenhouse regularly and teach greenhouse workers to recognise pests, pathogens and symptoms of deficiencies is essential. Serious out-breaks can often be prevented as most control measures are more efficient when used in the early stages of an infection or infestation.

In spite of requiring high levels of economic investment, the overall sustainability of greenhouse systems has increased partly due to the possibilities to recirculate irrigation water as well as the shift from fossil fuels to biomass heating. Additionally, the environmental technology available to greenhouse growers, the advances in biological control, as well the possibility to use pathogen free substrates and resistant cultivars makes greenhouse cropping systems highly suitable for IPM solutions.

2.3 IPM of greenhouse systems

IPM was originally developed by entomologists who encountered problematic insect outbreaks due to elimination of natural enemies and pesticide resistance in relation to the use of broad-spectrum insecticides. Integrated control in greenhouses evolved in England and The Netherlands in the 1960s. Because of the positive experiences of these researchers and growers, the control system gradually became adopted throughout northern Europe during the 1970s and 1980s (Barzman *et al.*, 2015; Albajes *et al.*, 2006). Since then, the concept of IPM has evolved extensively to apply to all aspects of plant protection. IPM has recently gained renewed attention through the adoption of the EU framework directive 2009/128/EC on the sustainable use of chemical pesticides. The definition of IPM formulated in the directive is as follows: "*Integrated pest management means careful consideration of all available plant protection methods and subsequent integration of appropriate measures that discourage the development of populations of harmful organisms and keep the use of plant protection products and other forms of intervention to levels that are economically and ecologically justified and reduce or minimise risks to human health and the environment. Integrated pest management emphasises the growth of a healthy crop with the least possible disruption to agro-ecosystems and encourages natural pest control mechanisms*." (Barzman *et al.*, 2015).

Since the directive entered into force on 1 January 2014, all EU Member States are required to develop a National Action Plan which ensures that a set of eight general principles of IPM are implemented by all professional pesticide users. These eight principles of IPM mentioned in the directive are:

1. Prevention and suppression
2. Monitoring
3. Decision-making
4. Non-chemical methods
5. Pesticide selection
6. Reduced pesticide use
7. Anti-resistance strategies
8. Evaluation

(Barzman *et al.*, 2015)

For covered cropping systems more specifically, the most important measures to reach the goal of reducing the use of chemical pesticides is mentioned by Albajes et al. (2006):

- Improving the accuracy and speed of diagnosis.

- Extensive monitoring and improving diagnosis systems for determining the degree of infestation and economic thresholds of pathogens and pests to enable rational management decisions.
- Use of pest and pathogen-free material, and growing media disinfested with steam or naturally suppressive to soil borne pathogens.
- Use of resistant cultivars.
- Use of modern techniques for pesticide application.
- Use of biological control of diseases and pests.

Of these measures, use of resistant cultivars, modern application techniques for pesticides, use of pathogen free material and use of biological control of pests are already to a large extent practised in modern cucumber greenhouse systems. However, it seems that improvements could be achieved by focusing further on monitoring and development of diagnosis systems.

Well-developed damage and action thresholds are important tools in integrated control systems and knowledge of thresholds can reduce total control inputs (Albajes et al., 2006). Furthermore, as is evident in Sweden (Jansson, 2016; Jordbruksverket, 2001), there are currently fewer biological control agents available for pathogens than for pests. It appears that more research is needed to develop alternative control methods for pathogens, evaluate which ones are suitable for use in greenhouse environments and how to create favourable conditions for their survival.

According to the IPM principles of the EU framework directive mentioned above, pesticide users should base their strategies on prevention, suppression and monitoring. For greenhouse systems, the manipulation of climate using environmental computers provides an excellent, although challenging, opportunity to supress several pathogens and diseases.

Another highly important measure for prevention and suppression of pests and disease is the use of proper hygiene and thorough sanitation between cultures. Today, the fear of virus has led to improved sanitary measure among Swedish growers, e.g. it is common practise in many greenhouses to use disposable gloves and different pruning knives for each cucumber row to limit spread of pathogens. However, is seems that improvements can be made when cleaning up and sanitising between cultures.

In spite of the many mentioned benefits of modern protected cropping systems which facilitate the implementation of IPM practises, greenhouse growers face many challenges. One of the main challenges is that once pathogens or pests have entered, the humid climate and plants that are pushed into producing high yields as well as the labour intensive maintenance of plants creates favourable conditions and help them to spread (Jarvis, 1992).

Pathogens can enter greenhouses in many different ways, e.g. through ventilators, doorways, with windblown dust, visitors' footwear and machinery, seeds and planting material, through irrigation water from ponds, wells and ditches and via insect vectors (Albajes *et al.*, 2006; Jarvis, 1992).

Another challenge is the trade of plants that has enabled the introduction of exotic polyphagous new pests in many countries, e.g. whiteflies, spider mites, thrips and leaf miners which has led to increased use of chemical pesticides harmful to beneficial insects, and subsequent development of resistance (Albajes *et al.*, 2006). Resistance is also increasing due to lack of available chemical pesticides to alternate between.

Furthermore, in areas where continuous year-round cropping is employed, the lack of crop free periods greatly complicates pest control as pathogen and pest populations accumulate due to limited possibilities of cleaning and sanitation (Albajes *et al.*, 2006).

Altogether, there is an urgent need to develop IPM practices for pests and diseases in protected crops.

2.4 Terminology

Biopesticides and biological control agents are important tools for crop protection within IPM. There is no formally agreed collective term for pesticides derived from natural sources such as the ones tested in this study. In this thesis, the alternative pesticides used are referred to either as biological control agents for living organisms (natural enemies, microorganisms etc.) or biopesticides including all other reduced risk pesticides alternative to conventional synthetically derived chemical pesticides.

2.5 The European Tarnished Plant Bug, *Lygus rugulipennis*

2.5.1 Biology

The European tarnished plant bug (ETPB), *Lygus rugulipennis* Poppius is a highly polyphagous herbivore reported from 437 plant species from 57 families, for example cucumber, cereals, potato, sugar beet, brassicas, carrots, strawberries, pine and spruce. The most important host families are considered to be Brassicaceae, Asteraceae and Fabaceae. The ETPB is a member of the genus *Lygus* Hahn (Heteroptera: Miridae, tarnished plant bugs or grass bugs) which includes several other important pests (Holopainen & Varis, 1991).

In Sweden, the ETPB is the most dominant *Lygus* species. It feeds on a variety of cultivated crops and has become has become an increasing problem in greenhouse production of cucumber (Rämert *et al.*, 2007; Rämert *et al.*, 2005; Rämert *et al.*, 2001). Increasing problems have also been reported in England, particularly in cucumber cultures, with a similar pattern of damage to that which has been observed in Swedish cucumber greenhouses (Jacobson, 2002). In cucumber, it causes distorted foliage, dead growing points and malformed fruits. This damaged is caused by insertion of ETPB proboscis into various parts of the plant. Both adults and nymphs can cause damage. The damage is caused by a combination of mechanical wounds and a toxin injected into the tissue (Jacobson, 2002; Varis, 1972; Stewart, 1969).

The ETPB overwinters in Sweden as adults, preferably in a sheltered position within an evergreen forest (Varis, 1972). As reported by Varis (1972) it is mainly the adult overwintering tarnished plant bugs that give rise to the greatest problems. In many cultures, damage occurs in the spring and early summer (Varis, 1972). However, as seen in this study, adult ETPBs often migrate into Swedish greenhouses in June, with the population peaking in July and August. A study in England showed that the ETPB population had two clear peaks, one during the early summer and an even higher one later in July (Jacobson, 1999).

The greenhouse environment offers a highly advantageous breeding ground for the ETPB in comparison to outdoors, allowing several generations to develop and spread quickly.

Figure 2. The European tarnished plant bug, *L. rugulipennis* (Photo: Annika Wuolo).

2.5.2 Control methods

Available chemical control products for the ETPB in Sweden today include Confidor WG 70 and Warrant 700 WG (a.i. imidacloprid) and Mospilan SG (a.i. acetamiprid). Confidor WG 70 is only allowed for use one time per year and Warrant one time per culture whereas Mospilan, which is approved for cucumber "off-label", is allowed three times per year (Jansson, 2016).

Confidor WG 70 is the preferred product for Swedish growers even if they are reluctant to use products based on imidacloprid as they experience that is has a slow break-down process and effects biological control negatively for many weeks. Imidacloprid belongs to the chemical group of neonicotinoids which are considered to be partly to blame for the decrease in honey bee populations and is at risk of being permanently banned in the European Union in the near future.

The entomopathogenic fungus *Beauveria bassiana* (Balsamo) Vuillemin, has previously shown potential to control several species of the tarnished plant bug both in laboratory experiments as well as cage trials (Kapongo *et al.*, 2007; Fitzgerald, 2004; Liu *et al.*, 2002; Jacobson, 1999). In a cucumber greenhouse trial, *B. bassiana* was shown to decrease the number of *L. rugulipennis* by 78% compared with the control (Jacobson, 2002). However, an unpublished laboratory study indicated that *B. bassiana* kills adult *L. rugulipennis* too slow to control the pest and prevent crop damage in a sufficient way. Therefore, it should have limited potential for use in commercial cucumber production (B. Rämert, unpublished data).

The common plant volatile compound phenylacetaldehyde which can attract both male and female ETPBs has been tested in field in Hungary (Koczor *et al.*, 2012). In spite of promising research results, further development is necessary before it possibly could be used for mass trapping.

The use of trap crops for managing tarnished plant bugs has been quite extensively tested in various field crops, e.g. lucerne (*Medicago sativa L.*), lettuce and cotton (Accinelli *et al.*, 2005; Godfrey & Leigh, 1994). The use of trap crops can help to protect the main crop from pest infestation, but it needs to be complemented by other management practises in order to be efficient. The most important supplementary measure is to eradicate the pests congregated on the trap crop in order to prevent them from returning to the main crop (Holden *et al.*, 2012; Shelton & Badenes-Perez, 2006).

A small-scale greenhouse experiment in the Netherlands, revealed that sunflower (*Helianthus annuus L.*) and white mustard (*Brassica rapa L.*) were both highly attractive to both *L. rugulipennis* and *Liocoris tripustulatus* Fabricius (van Steenpaal *et al.*, 2005).

Furthermore, the recent identification of the sex pheromone of ETPB has allowed the development of a trap to monitor and forecast immigration of the pest into the greenhouse (Fountain *et al.*, 2014; Fountain *et al.*, 2010; Innocenzi *et al.*, 2005; Innocenzi *et al.*, 2004; Innocenzi *et al.*, 1998).

2.6 Cucurbit powdery mildew

CPM (CPM) is one of the most important foliar diseases of greenhouse cucumber. It causes yield losses and crop quality reductions and often shortens the growing season (Cerkauskas & Ferguson, 2014; Nuñes-Palenius *et al.*, 2006; Sitterly, 1978). In worldwide cucumber production, the disease is considered to be the major cause of crop losses (Lebeda *et al.*, 2010).

Figure 3. Left: Powdery mildew infected cucumber leaf in experiment 2013. Right: Conidia of *P. xanthii* collected from cucumber leaves (Photos: Mira Rur).

2.6.1 Biology

CPM in the northern hemisphere is primarily caused by the two obligate biotrophic fungi *Podosphaera xanthii* (Castagne) U. Braun & Shishkoff (formerly *Sphaerotheca fuliginea* (Schlechend.:Fr) Pollacci) and *Golovinomyces cichoracearum* (syn. *Erysiphe cichoracearum*) (DC.) VP Heluta (Braun, 1995).

In spite of having morphological differences, the two species can be difficult to distinguish visually. *P. xanthii* conidia contain fibrosin bodies, produce forked germ tubes and lack appressoria whereas conidia of *G. cichorareum* lack fibrosin bodies, produce straight germ tubes and have unlobed appressoria (Zitter *et al.*, 1996; Björling *et al.*, 1991; Sitterly, 1978; Kapoor, 1967). However, polymerase chain reaction (PCR) methods have been developed for both species, which allow for more precise differentiation (Chen *et al.*, 2008).

Symptoms of CPM are characterised by white powdery spots which expand and develop on leaf surfaces, petioles, stems and sometimes also on fruits. Infected leaves gradually wilt and die and eventually the whole plant senesces prematurely (Zitter *et al.*, 1996). The symptoms usually first occur on older shaded leaves. As cuticle deposition is directly correlated with light intensity, shaded leaves have a thinner cuticle layer thus less of a barrier making them less resistant to infection. The more humid microclimate of the lower vegetation layer of the greenhouse crop could also explain why infection often starts there. (Dickinson, 2012; Nicot *et al.*, 2002; Zitter *et al.*, 1996; Braun, 1995).

Unlike many other fungal pathogens, CPM fungi are not dependent on free water on the plant surface for germination as their conidia contain water. High relative humidity in the air surrounding plants increases the survival of conidia but the CPM fungi can also infect in rather dry conditions. In fact, dry conditions favour colonisation, sporulation and spread of the fungi. When conditions are optimal, CPM conidia germinate on the host plant surface within two hours. The mycelia grow on the surface and penetrate the cells of the host plant by a penetration tube followed by establishment of the haustoria. The haustoria are specialised feeding organs used by the fungi to collect nutrition from its host. Conidiophores form approximately four days after infection and a life cycle takes five to six days in total (Sitterly, 1978).

As mentioned by Zitter et al. 1996, the primary inoculum source is considered to be conidia, which can be spread by the wind over long distances. Conidia from other greenhouses are believed to be the main source of infection. The infections usually start close to doors and windows, where there is a draught. Other likely sources of early infections are visitors and infected plant material (Zitter *et al.*, 1996; Schepers, 1984).

G. cichoracearum reportedly has a lower temperature optimum and therefore more often occurs in spring or early summer when temperatures are generally lower in the greenhouse (Aguiar *et al.*, 2012; Vakalounakis *et al.*, 1994; Sitterly, 1978). *P. xanthii*, on the other hand, mostly occurs from the height of summer and is known to cause more severe infections in greenhouse conditions than *G. cichorareum* (Zitter *et al.*, 1996; Braun, 1995; Sitterly, 1978). Both species can also be concomitant on the same plant (Sitterly, 1978). According to Zitter et al. 1996, *P. xanthii* is more commonly reported worldwide.

The host range of CPM fungi is wide and still not fully known. Different strains of the same species may vary greatly in their host range. Besides cucurbits, other plant species described as being susceptible to CPM are zinnia, phlox, aster, lettuce and sunflower. Some strains of CPM will cross infect and some will not (Sitterly, 1978).

As obligate biotrophs, CPM fungi are completely dependent on living host plant material for their survival. *G. cichoracearum* produces cleistothecia with ascospores on some plants which allow them to overwinter. However, cleistotecia are generally absent in CPM fungi. Thus, in tropical climate and supposedly also in greenhouses CPM fungi overwinter on living hosts as active mycelia or in conidial stages (Zitter *et al.*, 1996; Sitterly, 1978). For this reason, weeding and proper sanitation is highly important to avoid overwintering inoculum.

2.6.2 Control methods

Currently, there are two chemical fungicides registered for use against CPM in Sweden under, a so-called, off-label permit. These are Amistar® (a.i. azoxystrobin), a QoI fungicide and Fungazil 100® (a.i. imazalil), an imidazole fungicide (FRAC, 2016; Jansson, 2016). However, resistance of wheat powdery mildew to QoI fungicides was reported in 1998 and resistance of CPM soon after (Ishii *et al.*, 2001). Since then, resistance has been reported in CPM populations (mainly *P. xanthii*) to six groups of single-site inhibitors: benzimidazole, DMI, morpholine, hydroxypyrimidine, phosphorothiolate, QoI, and Pyridine carboxamides (Lebeda *et al.*, 2010; Miyamoto *et al.*, 2010).

Sulphur, which is categorised as a preventative fungicide, is registered for cucumber production (Jansson, 2016) but is commonly not used due to high risk of phytotoxicity, especially at high temperatures in the greenhouse (Cerkauskas & Ferguson, 2014); H. Hermans, Innocrop Consulting, pers. comm.).

In Sweden today, there are no alternatives to chemical fungicides, such as biopesticides or biological control products, available against CPM fungi (Jansson, 2016). There have been other alternative control products available in the past in Sweden, e.g. the anti-microbial biopesticide Enzicur®. In this particular case, the product was granted temporary exemption and the manufacturer chose not to continue the registration process (Swedish chemicals agency, pers. comm.12th of May, 2016).

New, partially resistant cucumber cultivars have been introduced. However, since they reportedly require more light that susceptible cultivars, they often produce smaller yields, particularly in spring and autumn. The negative relationship between disease resistance and yield has also been reported by (Staub & Grumet, 1993). Growers also report that the partially resistant cultivars are more prone to infection by gray mold (*Botrytis cinerea*) and gummy stem blight (*Didymella bryoniae*).

Breeding of cucurbit crops for powdery mildew resistance has been relatively successful. However, both CPM species are referred to by Lebeda et al., 2010 as having high evolutionary potential, making them more prone to overcome plant genetic resistance and/or to develop fungicide resistance (Lebeda *et al.*, 2010). Because of this pathogen adaptation, the degree of protection achieved with resistant cultivars is variable thus often not adequate as a sole management practice (Lebeda *et al.*, 2010).

Due to the lack of effective alternatives, Swedish conventional cucumber growers today mainly depend on Fungazil 100, for control of CPM. This of course creates a great risk of resistance development of the pathogen populations. Organic growers rely solely on climate control, proper hygiene and sanitation measures.

Several alternative products have been tested showing promising results. These include e.g. microbial products, botanicals and so called plant strengtheners.

Some of the microbial products available against CPM in different parts of the world are Polyversum® (Biopreparaty LtD./ Beta-Biologics Ltd.), containing the oomycete *Pythium oligandrum,* AQ10® (Ecogen Inc./Intrachem), containing the fungus *Ampelomyces quisqualis,* Cease® (Bioworks) and Serenade® (AgraQuest), based on *Bacillus subtilis* and Sonata® (AgraQuest), based on *Bacillus pumilus*.

A. quisqualis is a mycoparasite of CPM fungi which has been extensively tested. *P. oligandrum* is an antagonistic oomycete thought to produce substances inducing the plants own defense reactions as well as acting directly as a mycoparasite (Benhamou *et al.*, 2012). The mode of action of *B. subtilis* and *B. pumilus* is not known but it is likely that they work through competition, parasitism, antibiosis or by inducing plant defense reactions (Gilardi *et al.*, 2008; Paulitz & Bélanger, 2001). The use of mycoparasites does however require tolerance of a slight disease pressure as the parasite relies on CPM fungi to fulfill its life cycle (Kiss, 2003).

AQ10 and products based on *B. subtilis* are compatible with chemical fungicides and thus should be able to function as part of an IPM system (Gilardi *et al.*, 2008).

Products based on *Reynoutria* extracts have been successful in many trials and these work by inducing the plants own defense mechanisms (Giotis *et al.*, 2012; Kiss, 2003; Petsikos-Panayotarou *et al.*, 2002; Konstantinidou-Doltsinis & Schmitt, 1998; Daayf *et al.*, 1995). In a Dutch greenhouse experiment, it effectively controlled CPM in both a susceptible and a partially resistant cultivar (Dik & Vanderstaay, 1994). Other previously tested products are based on e.g. chitosan, plant extracts, silicon, mineral oil and potassium bicarbonate (Giotis *et al.*, 2012); (Benhamou *et al.*, 1999); (Gilardi *et al.*, 2008); (Cerkauskas & Ferguson, 2014); (McGrath & Shishkoff, 1999); (Su, 2012); (Wolff *et al.*, 2012). Many cucumber growers also add soluble silicon to the nutrient solution as it reportedly triggers plant defence reactions and subsequent incidence and spread of CPM. The effect is however highly variable and does not seem to work under all conditions (Belanger *et al.*, 1998).

As is evident, many products have been tested with successful outcome. However, very few of them have actually been adopted in practise. This of course has many different reasons, registration issues being one of them. Nonetheless, an important step towards implementation of new products and IPM strategies is testing in commercial settings. Furthermore, collaboration between growers, advisors and researches using action research approaches is one way of bridging the potential gap between stakeholders which could be beneficial for the implementation process.

2.7 Action Research

Action research (AR) is considered to be more of a research approach rather than a single academic discipline and it has evolved from several different fields over a long period of time. Its origin lies partly in practical experiences of developmental aid projects involving farming systems and anthropological studies conducted in developing countries (Eksvärd *et al.*, 2001; Udas, 1998).

Brydon-Miller *et al.* (2003) mention that elements and perspectives of AR can be found in the early labour-organising traditions in the US and in Europe, in the Catholic Action movement and in liberation theology. Furthermore, the spread of AR approaches to Sweden, Denmark and Germany can be linked to The Tavistock Institute for Human Relations who used AR perspectives to combine the work of participants from several different countries (Brydon-Miller *et al.*, 2003).

Specific persons highly influential in developing AR are Kurt Lewin, who introduced AR into social sciences, and Reg Revans, who introduced the concept of action learning (Udas, 1998).

The book "Farmer first" by Chambers et al. (1989) was important in introducing AR within agriculture, describing a model where the farmer's own capacity for innovation is central and which sought to develop more effective ways to serve diverse and risk-prone small farming systems (Chambers *et al.*, 1989).

Reason & Bradbury (2001) define action research in the following way: *"a participatory, democratic process concerned with developing practical knowing in the pursuit of worthwhile human purposes, grounded in a participatory worldview which we believe is emerging at this historical moment. It seeks to bring together action and reflection, theory and practise, in participation with others, in the pursuit of practical solutions to issues of pressing concern to people, and more generally the flourishing of individual persons and their communities"* (Brydon-Miller et al., 2003; Reason & Bradbury, 2001).

In short, the main goal is to improve the current situation based on biological, social and financial conditions and prerequisites (Eksvärd *et al.*, 2001).

A major concept within action research is the commitment to democratic social change and the respect for people's knowledge and the belief in people's own capacity to recognise and address the specific issues they and their communities are challenged with (Brydon-Miller *et al.*, 2003).

As AR is an interactive process, it is characterised by intervention rather than observation. There are several AR methodologies and PAR is considered a subdivision of AR. According to Udas (1998), participatory action research (PAR) implicates that an even higher level of participation than in AR takes place where the practitioners come to be both the research subjects and co-researchers. As with AR, the PAR approach is also characterised by striving towards practical solutions to pressing issues but the main focus is the change process and co-learning that occurs. Instead of being concerned with hypothesis testing with scientific objectivity and problem solving in a traditional sense, the process itself (which may well solve problems) has an inherent value (Udas, 1998). Additionally, PAR strives understand the world by trying to change it in a process based on co-operation and continuous evaluation and reflection (Reason & Bradbury, 2001).

There are different views of the level of participation required for authentic participation to occur. What can be said in general for PAR is that it is based on the concept that research should be made collectively "with" people instead of "on" or "for" people. (Reason & Bradbury, 2001; McTaggart, 1991).

Highly significant for PAR is the cyclical action learning process. The process is comprised by repeated cycles involving steps of planning, action, observation and reflection/evaluation. If participants have critical reflections of the result of the first action cycle the consequence may be that the initial problem/issue is redefined, leading up to adjustment of the action plan and succeeding action cycle (Udas, 1998).

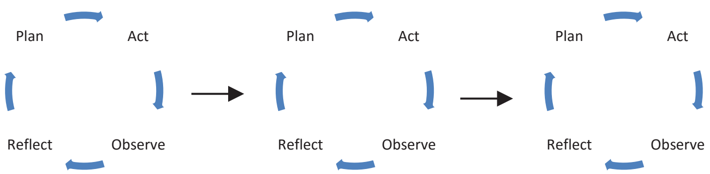

Figure 4. Illustration of the cyclical action learning process typical for PAR with repeated cycles of planning, action, observation and reflection.

The researcher's role within PAR is as a co-learner rather than an expert. Within PAR, all participants are seen as stakeholders (Checkland & Holwell, 1998; Udas, 1998; McTaggart, 1991).

The facilitator has a significant role in the co-learning process. Important functions of the facilitator are to provide tools, which help generate ideas and assist in prioritising, analysing and evaluating research. The facilitator should also make sure that every participant is heard. In doing so, different exercises can be used. These exercises may also for example be used to improve group dynamics (Eksvärd *et al.*, 2001). As mentioned by Cassara (1991), "*the facilitator should provide participants with knowledge, skills and resources but not with decision-making"* (Cassara, 1991).

Many different tools have been developed to assist in the PAR process. Some examples are triangulation and talking stick brainstorming. Triangulation is an established technique within social science and means that three different methods are employed to verify something, e.g. mapping, interviews and discussions. Talking stick brainstorming can be used to gain ideas from all participants to base further investigations on. In the process, an object is passed between participants and only the person holding the object is allowed to talk. No appraisal of ideas is allowed in this step as it can damper the flow and the object is passed between participants until no one has anything else to add (Eksvärd *et al.*, 2001).

All research methods of course have both strengths and weaknesses. The possible limitations of PAR differ depending on the research field and specific context of the project. General limitations of PAR that are mentioned are the lack of academic standardisation, the possibility of strong dependence on researchers, outside specialists not being accepted by the group/community, challenging group dynamics etc. There is also a discussion on the replicability of natural science projects versus social science projects and how views of replicability affect the perceived validity of experiments (Checkland & Holwell, 1998). Another possible limitation mentioned is the case-by-case nature of PAR projects. For example, even if many action research projects have successful outcomes in a local context, there may be difficulties in translating these results to a large-scale level enabling wider social change (Brydon-Miller *et al.*, 2003). Furthermore, as Brydon-Miller *et al.* (2003) points out, action research is a work in progress, which means that there are still many questions to try to answer and many different viewpoints to be discussed. Nonetheless, conducted in a professional way, collaboration using AR approaches have the potential to bridge gaps between stakeholders and through a collective learning process, possibly reaching higher learning outcomes compared to when working separately.

3 Objectives

The overall goal with this thesis was to, in collaboration with growers, identify the most important plant protection problems of the Swedish cucumber production system and to approach these problems collectively in order to develop applicable tools for IPM. The starting point was the shared knowledge and experience of the participants. The more specific objectives emerged through the collaboration process. These were to:

- Compare the behavioural responses of adult European tarnished plant bugs (ETPBs) to cucumber at different phenological stages with those of candidate trap crops, i.e. sunflower and lucerne, in olfactometer assays.

- Assess the response of the ETPB to headspace volatile collections of candidate trap plants and identify the attracting chemical compounds.

- Investigate if sunflower could serve as a trap crop for the ETPB in commercial greenhouses with cucumber as the main crop.

- Screen for effective alternative products against cucurbit powdery mildew (CPM).

- Evaluate the effect of selected products on CPM alone and in combination with the standard fungicide at different application intervals and in cucumber cultivars with different levels of resistance.

4 The Participatory Action Research process

4.1 Participants

The original PAR group of the “Lygus project” consisted of five conventional cucumber growers, and an organic cucumber grower, an experienced process facilitator with a background in extension services and participatory research from the Regional Board in Västmanland, a project leader with long experience as a researcher within plant protection of organic and integrated horticultural farming systems and PAR, an advisor from the Swedish Board of Agriculture specialised in greenhouse farming systems, a senior advisor with long experience of working with greenhouse farming systems, and me as a postgraduate student with previous horticultural education, specialised in plant protection.

During the first two years of the project, an experienced postdoctoral fellow also assisted in data collection and analysis of greenhouse trial data. Students and other advisors and researchers were also occasionally invited to group meetings during the project. During the project one of the growers sold her business and left the group.

The PAR group of the “mildew project” consisted of a majority of the growers of the “Lygus group” and me as a postgraduate student. The additional members of the group consisted of external resource persons who assisted with feed-back and advice. These were: an experienced Dutch extension officer active in Sweden among other countries, a researcher/advisor within greenhouse cultivation and the process facilitator from the “Lygus group”. Additionally, four researchers were linked to the project as supervisors.

4.2 PAR Methodology

The “Lygus project” was initiated by researchers specialised in plant protection at SLU who had contacted cucumber growers to survey their need for IPM solutions for the ETPB. The “powdery mildew project” was initiated by growers participating in the “Lygus project” who felt that CPM was a threat to their production and following on from the success of the “Lygus project” felt that a PAR project working with CPM would be highly beneficial to them.

Concerning the methodology of the projects, the main goal has been to listen to the cucumber growers, learning what problems are most relevant to them and to collaborate between stakeholders to find solutions. To assist this process we had an experienced facilitator with a background in farming extension services and PAR who was responsible for the PAR approach of the groups’ work.

Due to the different set up of the projects the level of participation of growers differed. The three-year long “Lygus project” was mainly based on trials performed in the commercial greenhouses of participants, which required a high level of participation from growers. The two-year long “powdery mildew project” was entirely initiated by growers during the process of working with the “Lygus project”. However, the growers decided that in order to reduce the risk-taking for them as business owners, trials should first be conducted in the university greenhouses and then tested under commercial conditions at later stage. Therefore, the level of participation was reduced to yearly field visits where the growers were able to discuss the ongoing trials on university grounds and give feedback. The collaboration with these growers was well established at this point and they served as an expertise resource group for the project along with advisors and researchers.

4.3 The research process

The research process timeline is displayed in Table 1. The cornerstones of the collaboration process were the yearly meetings. There were three meetings per year during the “Lygus project” and one meeting per year during the “powdery mildew project”. In the “Lygus project”, the meetings were focused on planning, updating and presentation and discussion of results in that order.

In each meeting, reflection/evaluation of the project and collaboration process was a set agenda item. Each summer meeting of the “Lygus project” also included a field visit at the greenhouses of one of the participating growers.

The yearly meetings of the "powdery mildew project" focused on discussion of ongoing experiments and receiving feedback from growers.

During 2013, which was the final year of the "Lygus project" and the first of the "powdery mildew project", a theme day was organised together with The Swedish Farmers' Organisation (LRF) with focus on plant protection in cucumber. The day included seminars by an experienced extension officer from England and a researcher within greenhouse plant protection from The Netherlands followed by discussions with growers and other stakeholders from the Swedish cucumber greenhouse industry. The theme day was followed by a field visit to the greenhouses of two growers of the PAR group. The invited speakers from the theme day were also invited to the field visit. During the field visit, the growers were able to discuss current plant protection issues with experts and researchers. During the evaluation following these days, growers stated that they found the seminars interesting but appreciated the field trip discussions even more.

In the first meeting of the "Lygus project", the participants were all asked to mention their expectations of the project (Figure 7). This was followed up at the final meeting to see how the far expectations of participants were met.

A group contract with so-called "game rules" was also formulated in the first meeting. Growers were asked what they felt was most important in order to facilitate collaboration during all phases of the project (Figure 6). Examples of "rules" which were set up were that growers should be allowed to keep their mobile phones on during meetings in case of urgent situations in greenhouses and that meeting times in the afternoon was preferred. These "rules" were evaluated in each meeting in order to inquire as to the need for possible adjustments.

All growers could not consistently come to all meetings due to urgent matters of their businesses, especially in summertime, but at most meetings everyone came. The atmosphere was positive and the collaboration between participants worked well. The growers enjoyed meeting colleagues and discussing common problems that they all face. There was also a common appreciation between participants of the perceived benefits of bringing together theoretical knowledge with practical knowledge and years of experience.

Researchers, in collaboration with growers, planned the greenhouse trials in the "Lygus project". However, researchers were entirely responsible for the scientific part of the process. During the trials growers made no changes to their normal management strategies except allowing space for sunflowers. It was important for growers that there was no risk-taking involved production wise.

The collection, compilation and analysis of experimental data was made by researchers. Meeting notes were taken by both the postgraduate student and the facilitator. The meeting notes were sent out to all participants for review after each meeting, they were also used to plan the next meeting.

4.3.1 Evaluations of the working group

During the final meeting of the "Lygus project" an extensive evaluation of the achievements of the project work was made. To begin with, the expectations written down in the very first meeting were run through, one by one, and the group reflected on the outcome of the project together. Before the first and before the final meeting of the "Lygus Project" a questionnaire was also sent to the growers. The motive was to map out the most important plant protection issues of each greenhouse business in order to provide a starting point for the project and enable a follow up at the end. At the final meeting, the answers from both occasions were compared and discussed. In relation to this meeting item, two open-ended questions were posed to growers. The answers from growers are listed here:

1) What knowledge and experiences have you gained by participating in the project?

- I have learnt more about the behaviour of the ETPB.
- I have to attend to problems instantly.
- Most of all an extended professional network, which is particularly important for growers geographically distant from others.
- We have gained more knowledge about the lifecycle of the ETPB and the time it enters greenhouses.
- We have gained more knowledge about other pests.
- I have learned more about how research is conducted and how you think.
- The structure and planning of the project was well made and I appreciated the follow up with presentation of results.
- The ETPB, is attracted to sunflower but is not over fond of them.

2) What changes have you made in your business in relation to the project?

- I'm more thorough with greenhouse hygiene.
- I have become more observant of pests and fungal diseases.
- We have continued catching ETPBs manually as before.
- None. I still want to avoid using chemical pesticides.
- None.

A SWOT-analysis was also made during the same final meeting in order for participants to evaluate how they perceived the PAR method used. SWOT in this case stands for strengths, weaknesses, opportunities and threats. The group mentioned that the strengths of the method are that is assists in keeping research relevant, the different approaches, experiences, and knowledge of participants, that the process of obtaining concrete results is faster, e.g. because we have been able to do parallel trials in different greenhouses, and further that it is a nice and fun way of working. Weaknesses mentioned were lack of continuity in funding which can create frustration and disappointment, and that the pressed financial reality for some growers may affect the research process and cause promising projects to end prematurely.

Reflections from growers on opportunities were that working with PAR could assists in creating an environmentally friendly profile/brand, which is important to them as business owners. The PAR method would in that sense serve as a form of quality assurance. Another possibility mentioned was that working with PAR methods diminishes the risk of lacking trust between growers and researches.

Threats that were mentioned were lack of resources and time and that possibly not enough people are interested in becoming involved in PAR groups.

To evaluate the results and how far we had come in finding solutions to the problem of the ETPB of cucumber greenhouses two techniques were used by the facilitator. The first one is called “the sun” where all growers were asked to elaborate on what they think characterises a good trap crop for the ETPB in cucumber (Figure 5).

By using another technique called “the force field”, the group collectively identified the driving forces and the hindering forces in order for the statement “Sunflower is a potential trap crop in cucumber plantings” to be accurate.

Driving forces mentioned were: “Works as an indicator for infestation”, “Can be used to kill ETPBs by using efficient systemic chemical pesticides”, “Is attractive to ETPB and other pests”, “Cheap plant”, and “Could work in brightest/warmest places of greenhouse where cucumbers do not thrive”.

Hindering forces mentioned were: “Doesn’t tolerate the climate”, Doesn’t tolerate the irrigation”, “Short-lived”, “Not realistic because of the number of plants needed”, “Have to be exchanged often”, “The cost of buying/growing/maintaining plants”, “Could shade and compete with cucumber crop”, “Too slow growth”.

After these exercises, an attempt was made to summarize the views of growers on sunflower as a trap crop for the ETPB. The views are listed below:

- Sunflowers did not thrive in the cucumber greenhouses.

- Keeping sunflowers in the greenhouse was difficult.
- The trap crop has to be much more attractive than the cucumber crop, the effect increases with the level of attractiveness.
- We had very few sunflower plants in relation to cucumber plants.
- Sunflowers in hanging baskets, hung above cucumber plants by ventilators, would be interesting to test. The sunflowers would have to be treated with a systemic pesticide to make sure that ETPBs are killed instantly and are not able to use sunflowers as a breeding ground.

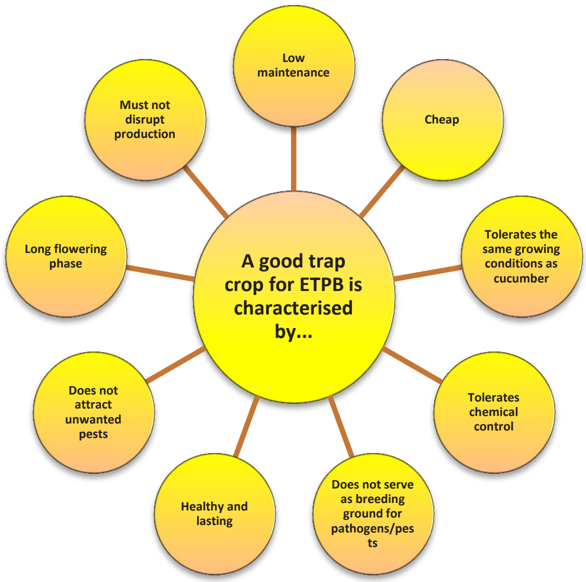

Figure 5. Characteristics of a good trap crop for the ETPB according to growers of the working group noted during the final meeting of the "Lygus project" (Modified from original notes by Elisabeth Ögren).

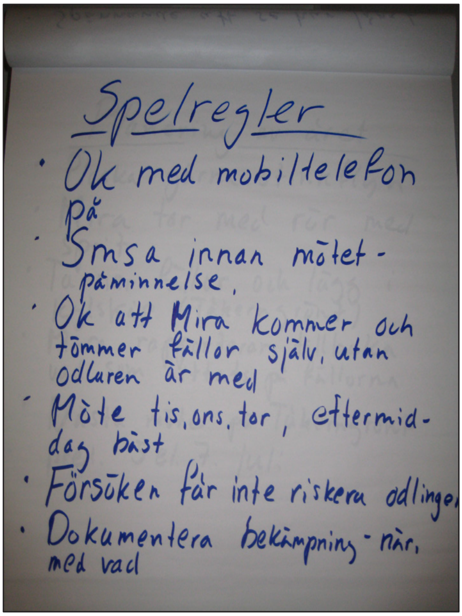

Figure 6. “Game rules” established at the first meeting of the “Lygus Project” (Notes taken by Elisabeth Ögren) (Photo: Mira Rur).

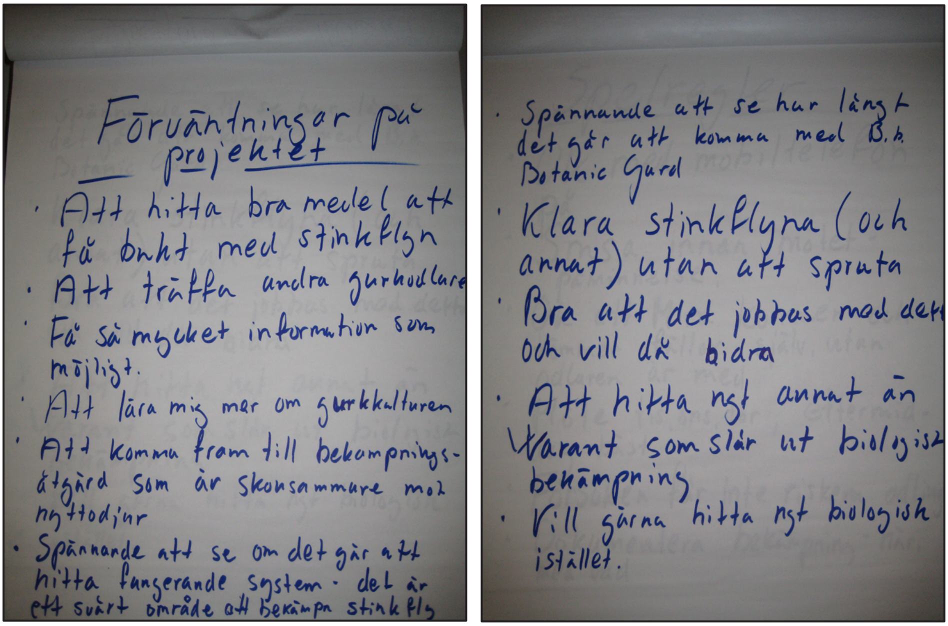

Figure 7. Expectations of participants written down in the first meeting of the “Lygus project” (Notes taken by Elisabeth Ögren) (Photo: Mira Rur).

Table 1. The timeline of the PAR process, including the type of activity, the purpose of the activity and participants.

Date	Type of Activity	Type of Information/Purpose	Participants
2011-01-15	Telephone contact with cucumber growers.	Establishing initial contact, examining grower interest in project.	PhD-students, cucumber growers.
2011-02-11	Initial visits to growers of the group, interviews about their greenhouse production.	Establishing contact with growers and becoming familiar with each greenhouse production system.	Researchers, Advisors, cucumber growers.
2011-02-11	Questionnaires to cucumber growers.	Mapping out the most important plant protection issues as a starting point and enable a follow up by the end of the project.	Cucumber growers.
2011-03-29	First PAR group meeting of the *"Lygus project"*.	Project presentation, expectations of participants, planning of greenhouse trials, going through questionnaire results, setting up ground rules for collaboration.	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers, facilitator.
2011-07-12	Second PAR group meeting of the *"Lygus project"*. Field visit.	Presentation of greenhouse trials so far, discussion, tour of organic cucumber greenhouse (Tåkerngrönt AB).	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers, facilitator.
2011-12-14	Third PAR group meeting of the *"Lygus project"*.	Presentation, discussion and evaluation of trials of year 1, run through collaboration ground rules, presentation of two coming Master's thesis projects on predatory mites and CPM.	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers, facilitator.
2012-02-29	Fourth PAR group meeting of the *"Lygus project"*.	Planning of greenhouse trials year 2, information and discussion about the two Master's thesis projects mentioned in previous meeting, presentation of CBC at SLU, discussing problems with CPM.	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers from SLU and CBC (Centre for Biological Control), facilitator, Master's students.
2012-05-29	Fifth PAR group meeting of the *"Lygus project"*. Field visit.	Tour of greenhouses (Sånnagården AB) and of trials of Master's project on CPM.	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers, facilitator,

			Master's students.
2012-12-18	Sixth PAR group meeting of the *"Lygus project"*.	Presentation, discussion and evaluation of trials of year 2, presentation of results of Master's project on CPM, planning and discussion CPM trials at SLU, planning of seminar on plant protection in cucumber.	Cucumber growers, advisors, researchers, facilitator, Master's students.
2013-02-28	Seventh PAR group meeting of the *"Lygus project"*.	Planning of greenhouse trials of year 3, summing up and evaluating previous trials with trap crops, discussion of lab experiments.	Cucumber growers, advisors from the Swedish Board of Agriculture, researchers, facilitator.
2013-04-16	Seminar: Plant protection in cucumber.	Invited advisors and researches gave talks on their work with plant protection in cucumber followed by discussion.	Cucumber growers, advisors, researchers, pest management companies, consultants from the Federation of Swedish Farmers (LRF).
2013-04-17	Field visit with experts.	Invited speakers from the seminar joined on a field visit to two growers of the PAR group. Current plant protection problems were discussed.	Cucumber growers, advisors, researchers.
2013-06-18	Eight PAR group meeting of the *"Lygus project"*. Field visit.	Information results from field trials so far, discussing an IPM model for *Lygus* in cucumber, discussion on CPM and resistant cultivars, tour of greenhouse (Ingemar Bengtssons Handelsträdgård AB).	Cucumber growers, advisors, researchers, facilitator.
2013-08-14	First field visit of the "CPM project".	Tour of the powdery mildew trials in university greenhouses followed by feed-back and discussion.	Cucumber growers, researcher.
2013-11-14	Questionnaires handed out to cucumber growers.	Same questionnaire as in initial phase of project used for follow-up.	Cucumber growers.
2013-12-17	Ninth PAR group meeting of the *"Lygus project"*.	Presentation of results, summary and conclusions, continued discussion on an	Cucumber growers, advisors, researchers, facilitator.

		IPM model for *Lygus* in cucumber, summary of participants' evaluation and follow up on initial expectations, evaluation of PAR methodology (SWOT-analysis).	
2015-08-16	Second field visit of the "CPM project".	Tour of the powdery mildew trials in university greenhouses followed by feed-back and discussion.	Cucumber growers, researcher.

5 Sunflower as a trap crop for the European tarnished plant bug (*Lygus rugulipennis)* (Paper I)

5.1 Results and discussion

5.1.1 Sunflower as trap crop in the greenhouse

The ETPB is an increasing problem in greenhouse production of cucumber (Rämert *et al.*, 2007). It causes significant damage to cucumber plants including distorted foliage, dead growing points and malformed fruits. The bugs enter greenhouses through the ventilation windows. In Sweden, the migration of the season's first generation of ETPB into the greenhouse unfortunately also coincides with the transplanting of the second cucumber crop, meaning these young plants are at a high risk of attack.

During 2011-2012 experiments were conducted where sunflower was tested as a trap crop for the ETPB in three different greenhouses. Aiming for maximal protection of the young delicate cucumber plants, the experiment was planned in a way that the placing of flowering sunflower plants was timed to coincide with cucumber crop transplanting.

The sunflower plants were placed individually at the end of cucumber rows (Figure 8), but the number of sunflowers at each grower varied depending on size and number of rows of each greenhouse. The average ratio of sunflower to cucumber plants was 5.9% (max 8.8, min 4.6%). The percentage of cucumber and sunflower plants infested with ETPB nymphs and adults was recorded on a weekly basis during the eight weeks following transplanting.

The results of greenhouse experiments showed that sunflowers were more attractive than the cucumber main crop during the complete period in which ETPB was active in the greenhouse. A significantly higher percentage of sunflower plants were infested by ETPB nymphs in both years, compared with cucumber plants (Paper I, Figure 2 a-d, page 6). The percentage of infested sunflower and cucumber plants increased gradually from July and peaked in August.

Unfortunately, this was not enough to avoid damage on the crop. In the following PAR group evaluation process, commercial growers claimed that the controlling effect of sunflowers as a trap crop, in this case, was not sufficient from an economic perspective.

As a single tarnished plant bug can damage a large number of cucumber plant tips (Varis, 1978), an efficient trap crop needs to be both highly attractive and immediately appeal to the pest upon entering. There is still a possibility that increasing the number of sunflowers and optimizing their placement in the greenhouse combined with a strategy to kill the bugs as they alight on the trap crop could increase the efficiency. The growers of the PAR group suggested placing plants of a suitable sunflower cultivar in hanging baskets above the cucumber crop, along the aeration vents of the house. The time to place these baskets could be determined using information gained from the capture in pheromone traps (www.agralan.co.uk).

Figure 8. Sunflower as trap crop for the ETPB in a commercial greenhouse 2012 (Photo: Mira Rur).

5.1.2 Attractiveness of ETPB to single and double plant odours in olfactometer assays

We tested the attractiveness of odours to ETPBs in y-tube olfactometer assays to determine the preferences of the ETPB.

In the control (no odour vs. no odour), there was no significant difference in ETPB choice to either of the Y-tube olfactometer arms (Paper I, Figure 1a, page 4). In the no odour vs. plant material situation test, a significant response to volatiles emanating from flowering sunflower was measured.

L. rugulipennis significantly preferred flowering sunflower to flowering cucumber, but showed no preference between flowering sunflower and non-flowering cucumber (Paper I, Figure 1a, page 4). No differences in preference were observed between sexes in these two trials. However, males and females responded differently when offered the choice between non-flowering cucumber and flowering lucerne. Female ETPBs significantly preferred non-flowering cucumber, and males significantly preferred flowering lucerne. Both sexes preferred flowering lucerne over flowering cucumber (Paper I, Figure 1b, page 4).

The response the ETPBs was the same for plant headspace odour as for the plant material. In both cases, the ETPB chose the collected odour from a flowering sunflower over the blank or that from a flowering sunflower over that from a flowering cucumber (Paper I, Figure 1c, page 4).

In summary, the ETPB was attracted both to the crop and to the trap crops. The olfactometer trials also showed that flowering sunflowers and non-flowering cucumber plants are equally attractive as an olfactory stimulus to ETPBs. In terms of visual stimuli, sunflower plants with their large yellow flowers supposedly offer a stronger visual stimulus than cucumber plants. As previous studies have shown that tarnished plant bugs use both types of stimuli to orient themselves to host plants (Williams *et al.*, 2010; Frati *et al.*, 2008), there is a possibility that the stronger visual stimuli of sunflower may be synergised with attraction to sunflowers olfactory cues.

In the beginning of the flowering phase of sunflower in the greenhouse trial, the cucumber plants extended greatly in height, making the sunflowers less distinguishable from the cucumber background. According to our olfactometer data, at that point the sunflowers were still more attractive in terms of olfactory stimulus and this may explain why they still continued to attract a higher number of tarnished plant bugs than the cucumber plants.

In addition, the low-growing lucerne plants with relatively small flowers also offer a weaker the visual stimulus than sunflower.

In a previous experiment, we found that females constitute 85–90% of the ETPB population in the greenhouses (B. Rämert, unpublished data). As female ETPBs significantly preferred non-flowering cucumber over flowering lucerne, and the latter also supposedly offers less visual stimulus, it would assumingly not be sufficient enough as trap crop for a commercial cucumber production system.

5.1.3 Chemical analysis of headspace collections

The chemical analysis of plant odours from non-flowering cucumber, flowering cucumber, flowering lucerne and flowering sunflower showed that the volatile profiles of the four plants were very different (Paper I, Fig 3, page 7). Furthermore, the phenological phase was a strong influence of the composition and release rate of cucumber volatiles. Compounds belonging to the ethyl benzenes were the most abundant before flowering while benzyl alcohol, benzaldehyde and decanal emerged during flowering (Paper I, Table S1).

Among volatiles found in flowering lucerne, the monoterpene (E)-b-ocimene was the major constituent followed by green leaf volatile (Z)-3-hexenyl acetate and toluene. Furthermore, the phenylpropanoid methyl cinnamate and the amine indole were exclusively detected in flowering lucerne at comparable release rates (Paper I, Table S1).

In sunflower headspace collections, terpenes were the dominating volatile group accounting for approximately 97% of the collections, with a-pinene and sabinene being the first and the second most dominant volatiles. Terpenes have previously been identified as sunflower constituents, but with sabinene as the dominant constituent (Schuh *et al.*, 1997; Etievant *et al.*, 1984). These monoterpenes were released exclusively by sunflower.

Volatiles found to be released by all four plants were, a green leaf volatile (Z3-hexenol), three benzenoids (benzaldehyde, benzyl alcohol and toluene) and three isoprenoids (a-pinene, limonene, and (E)-4,8-dimethyl-1,3,7-non-atriene) (Paper I, Table S1).

The overall release rate of volatiles showed a high variation among plant species. Cucumber emitted between 12.3 (flowering) and 14.2 (non-flowering) lg/h, while a higher release rate was measured from flowering lucerne (34.8 lg/h). Sunflower was shown to have the highest volatile release rate at 65.9 lg/h, which was almost four times that of cucumber. It is likely that the much higher release rate of sunflower compared to cucumber contributes to the higher attractiveness the ETPB to of sunflower.

6 Screening of alternative products for integrated pest management of Cucurbit Powdery Mildew *(Podosphaera xanthii)* in Sweden (Paper II)

6.1 Results and discussion

6.1.1 Inoculum and species identification

In both 2013 and 2015, conidia from sampled leaves were examined with light microscopy. Additionally, subsequent polymerase chain reaction (PCR) assays followed by sequence analysis was used to confirm *P. xanthii* as the species identified. The internal transcribed spacer (ITS) of nuclear ribosomal DNA regions was amplified using the powdery-mildew specific ITS universal primer pair PN23 (5'-CAC CGC CCG TCG CTA CTA CCG-3')/PN34 (5'-TTG CCG CTT CAC TCG CCG TT -3'). Pairs of primers specific to the ITS regions of *P. xanthii, G. cichoracearum,* and *Leveillula taurica* were used for PCR amplification. These were: S1 (5'- GGATCA TTA CTG AGC GCG AGG CCC CG -3')/S2 (5'- CGC CGC CCT GGC GCG AGA TAC A -3'), G1 (5'- TCC GTA GGT GAA CCT GCG GAA GGA T -3')/G2 (5'- CAA CAC CAA ACC ACA CAC ACG GCG -3'), and L1 (5'- CCC TCC CAC CCG TGT CGA CTC GTC TC -3')/L2 (5'- CTG CGT TTA AGA GCC GCC GCG CCG AA -3'), respectively (Chen et al. 2008).

The expected size of the PCR products of PN23/PN34, S1/S2, G1/G2 and L1/L2 are around ~740bp, 454bp, 391bp and 374bp respectively. Both PN23 and S1 primers were used for sequencing at the GATC biotech AG sequencing facility (Germany). Resulting sequences were searched using the National Center for Biotechnology Information (NCBI) GenBank non-redundant nucleotide database. Sequences mapping to known species were determined with coverage and identity and the best NCBI accession were recorded. *G. cichorareum* and *L. taurica* were not identified in the study (Figure 9).

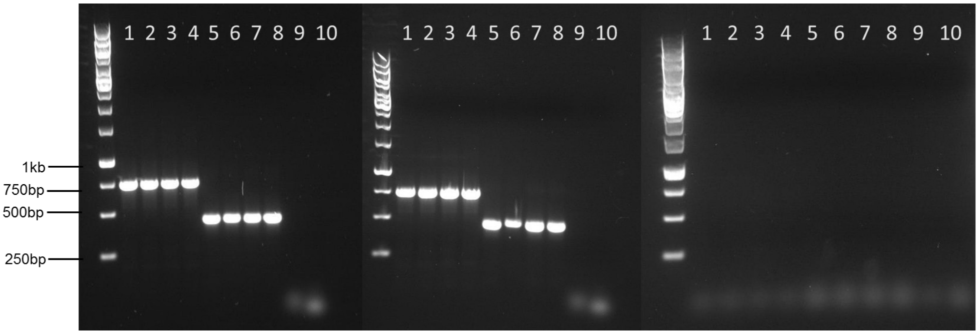

Figure 9. PCR detection of Cucurbit powdery mildew pathogens from infected leaf material using ITS primers. A) Samples from year 2013, Lane 1-4: using powdery mildew specific ITS universal primers PN23 – PN34 (~750 bp), Lane 5-8: primers specific to the ITS regions of *P. xanthii*, S1 – S2 (454bp), Lane 9-10: Negative controls. B) Samples 2015, Lane 1-4: powdery mildew specific ITS universal primers PN23 – PN34 (~750 bp), Lane 5-8: primers specific to the ITS regions of *P. xanthii*, S1 – S2 (454bp), Lane 9-10: Negative controls C) Primers specific to the ITS regions of *G. cichoracearum,* G1 – G2 (391bp), Lane 1-4: Samples 2013, Lane 5-8:Samples 2015, Lane 9-10: Negative controls for the year 2013 and 2015.

6.1.2 Effect of foliar treatments

In 2013 and 2015, infected leaves were collected from commercial greenhouses in the region with severe cucumber powdery mildew infections. The sampled cucumber leaves were used to infect plants of a susceptible cucumber cultivar in order to maintain the fungi until it was time for inoculation of the experimental plots. When inoculating experimental plots, leaves of infected plants were used to make a spore suspension. The plants of the experiment were inoculated at the four to five leaf stage and all leaves of the plants were sprayed.

During 2013 and 2015, the controlling effect of different alternative pesticides, including biological control agents, on CPM was examined in seven small-scale, semi-commercial greenhouse experiments. In 2015, the best treatments of the previous experiments were tested in different combinations and in different intervals (seven or fourteen days) both on a susceptible and a partially resistant cucumber cultivar. The treatments of each experiment are shown in Table 2. Consistently, all treatments, except Fungazil 100, were applied for the first time one day after CPM inoculation.

In all experiments, the plant extract based on *Reynoutria sachaliensis*, Sakalia, combined (tank mixed) with a wetting agent, Yuccah, from Yucca palm tree (*Yucca Schidigera*) applied at seven-day intervals, consistently had the most suppressive effect on powdery mildew disease severity in both cultivars tested. (Paper II, Table 4 and 5, Figure 1). Combining Sakalia and Yuccah with other products did, however, not further improve the disease control.

Sakalia is formulated from the plant extract of giant knotweed (*Reynoutria sachaliensis*). It was first formulated in the 1980s as Milsana® (Compo GmbH, KHH BioSci Inc.) then reformulated and introduced as Regalia® (Marrone Bio Innovations). Today it is named Sakalia® (Syngenta).

Experiments 2013

In these experiments (Paper II, Table 4), both Hortistar® and Kendal Cops® also showed good disease suppressing ability not significantly different from the best treatment with a combination of Sakalia and Yuccah. However, treatment with Kendal Cops resulted in chlorotic leaves indicating phytotoxic effects in spite of following recommendations to only do three applications.

Two fungal biocontrol agents, AQ10 (*Ampelomyces quiscalis*) and Polyversum (*Pythium oligandrum*) were also tested in 2013.

AQ10 combined with Bioglans®, was included in the short term experiment 1 (Table 2) and had a minor suppressive effect significantly different from the controls but less efficient than Fungazil 100. *A. quiscalis*, which is a well-known mycoparasite of CPM, is often found to co-occur at the same site as CPM fungi (Sedlakova & Lebeda, 2010; Kristkova *et al.*, 2009).

Polyversum was included in two experiments (Table 2) and did not significantly suppress powdery mildew compared to controls. The mechanism of *P. oligandrum* is not fully known but thought to rely on a combination of mycoparasitism, antibiosis and induction of basal plant immunity (Benhamou *et al.*, 2012).

Our results are supported by previous reports on the highly variable effect of *P. oligandrum* and *A. quiscalis* which is often explained by their demands for high humidity (Giotis *et al.*, 2012; Benhamou *et al.*, 1999). Furthermore, possible explanations could be different factors negatively affecting germination of resting spores e.g. relative humidity, adjuvants used and batch variations.

Even though the commercial product Polyversum seemed to have a low level of inoculum recovered, it is actually still possible that *P. oligandrum* may be an effective biological control agent, but new formulations of this organism need to be tested.

Additives are often used with biocontrol agents to obtain uniform coverage and ensure survival. However, these additives may also have an effect on powdery mildew (Dik *et al.*, 1998). For Polyversum, use of an additive was not recommended. For AQ10, the supplier recommended combining with Bioglans (paraffin oil). The latter treatment had little suppressive effect but was significantly different from control. Because we only tested the products combined it is unclear whether this small effect is can be ascribed to paraffin oil or to AQ10.

To estimate the controlling effect of each additive, they have to be tested separately. However, we decided to test all products in combination with the recommended additive so that the outcome of the study would be as relevant and applicable for commercial growers as possible. Testing the products in recommended combinations only also allowed us to screen more treatments.

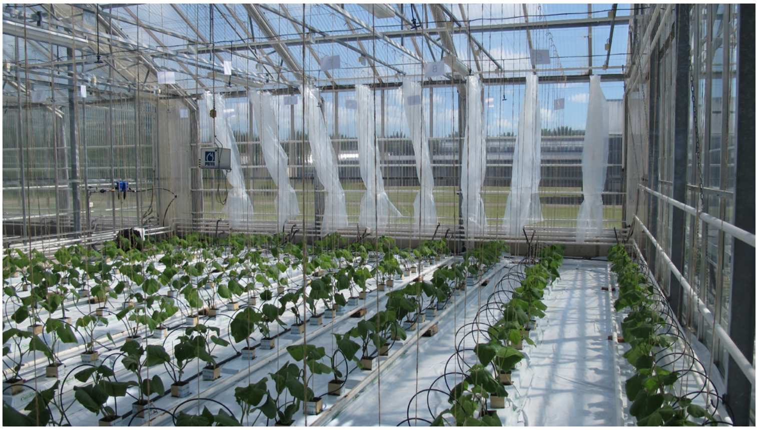

Figure 10. Newly planted cucumber plants in experiments 2013 (Photo: Mira Rur).

Experiments 2015

In these experiments (Paper II, Table 5), the previously most efficient treatments were tested in different combinations and intervals in two different cucumber cultivars (Table 2).

The two cultivars used were ‘Euphoria’, which is susceptible to CPM, and ‘Proloog’, which is partially resistant to CPM. The results displayed a difference in disease development between the two cucumber cultivars. In the partially resistant cultivar, ‘Proloog’, the first infection was delayed by 1-3 days compared to the susceptible cultivar ‘Euphoria’ (Paper II, Figure 2b). In general, treatments were slightly more effective in the partially resistant cucumber cultivar than in the susceptible. Specifically for treatments with *Reynoutria* extracts, this observation of cultivar effect was reported by Konstantinidou-Doltsinis and Schmitt (1998). It was also observed that CPM colonies sporulated more heavily in the susceptible cultivar.

The highly efficient combination of Sakalia and Yuccah, in seven-day intervals, significantly reduced the disease severity in both cultivars to a similar degree (Paper II, Figure 2b).

The treatments where we saw the biggest cultivar effect were Hortistar and Hortistar with Fungazil 100 treatments. These treatments both performed much better in the partially resistant cultivar. In spite of this, the disease severity was almost at the same level in both cultivars by the end of the experiments.

We did not see any synergistic effects when Sakalia and Yuccah were applied in combination with Fungazil 100. Some synergy was observed when Hortistar was combined with Fungazil 100 although this combination gave phytotoxic symptoms on leaves. In contrast, other reports suggest synergistic effects between *Reynoutria* extracts and chemical fungicides such as azoles, strobilurins, and sulfur in controlling powdery mildew and leaf spot diseases, copper in controlling bacterial diseases, and mancozeb and mefenoxam in controlling downy mildew (Su, 2012).

Furthermore, the results showed that a high level of control also could be obtained when Sakalia and Yuccah were applied at fourteen-day intervals. In the partially resistant cultivar 'Proloog', the suppressive effect was actually not significantly different from application at seven-day intervals. This is an important result as being able to reduce spraying intervals saves labour costs, time and reduces the impact on the environment as well as improving the work environment for growers.

Sakalia and Yuccah were also tested separately (Paper II, figure 2d). In those experiments, Yuccah had a moderate but significantly suppressive effect on powdery mildew. Nevertheless, Sakalia and Yuccah were more efficient when combined.

Spraying with Sakalia and Yuccah did, however result in brown spraying residues on plants. More residues developed when Sakalia was used alone than combined with wetting agent Yuccah. This is not described in previous experiments, which is why we assume it could be related to the spraying equipment used. High pressure sprayers are used in commercial settings with a pressure of 50-100 bar compared to 6 bar as in our case. It is possible that spraying with higher pressure would cause less residues but this is important to confirm by testing the treatments with commercial equipment.

6.1.3 General discussion

The results of this study on the effect of the *Reynoutria* extract are supported by previously reported experiments where the extract was tested against *P. xanthii* in greenhouse-grown cucumber in Germany, The Netherlands, Canada and Greece. Experiments in these countries have all proven *Reynoutria* extracts to be highly efficient against *P. xanthii*, often at a level comparable to the effect of the fungicide tested (Konstantinidou-Doltsinis & Schmitt, 1998). However, to our knowledge, this is the first time the effect of Sakalia in combination with wetting Yuccah has been tested scientifically in semi-commercial small-scale experiments.

The *Reynoutria* extract has been extensive tested in various crops, both in field and in greenhouse settings. Results have shown that it efficiently can control many fungal, oomycete and bacterial diseases e.g. powdery mildew of cucurbits, downy mildew of lettuce (*Bremia lactucae*), Botrytis of grapes and strawberries, bacterial spot of tomatoes and peppers (*Xanthomonas campestris* pv. *vesicatoria*), Cercospora on soybeans (*Cercospora kikuchii*) and bacterial canker on citrus (*Xanthomonas axonopodis* pv. *citri*) (Su 2012).

The extract seems to have a broad-spectrum effect which is explained by its ability to induce plant resistance. Studies have shown that it causes an increase of chalcone synthase and chalcone isomerase activity in the phenylpropanoid pathway and induces the production and accumulation of phytoalexins as well as well as different simple fungi-toxic phenolic compounds (Su 2012). After seeing a rapid haustorial collapse of CPM on cucumber and encasement by electron-dense substances stained blue by toluidine blue, the role of fungi-toxic phenolics was also suggested (Wurms et al. 1999). Furthermore, it has been shown to increase formation of papillae at pathogen penetration sites, contribute to lignification of plant cell walls as well as causing increased accumulation of pathogenesis-related proteins (PR-proteins) such as chitinase, glucanase, and peroxidase (Su 2012).

In cucumber specifically, different pathogen-induced defence reactions have also been studied. These reactions include lignification and suberisation of cell walls, cell wall reinforcement with cross-linked hydroxyproline residues, papillae formation, callose deposition and induced defence metabolites. Furthermore, McNally et al. 2003 specifically observed that a more rapid production of C-glycosyl flavonoid phytoalexins, within the epidermal tissues of CPM infected cucumber plants, was induced by treatments with a *Reynoutria* extract (McNally et al. 2003). Another observation made by Konstantinidou-Doltsinis and Schmitt (1998) was that treating the upper leaf surface with *Reynoutria* extracts also protected the lower leaf surface, indicating systemic effects of this treatment. No such indications were found in the experiments of this study. In contrast, leaves that were folded at the time of spraying, thus did not receive full coverage, had higher levels of infection than leaf surfaces evenly covered with the extract fluid, indicating that even dispersal of the extract is important.

Concerning the methodology of experiments, inoculating plants artificially creates an unusually high disease pressure. However, it also provides a homogenous disease pressure which is critical for a reliable and unbiased experiment. In commercial production systems, the disease will rather spread over an extended period of time.

Disease progression was slower in the final two experiments of 2015, and disease suppression greater in all treatments (Paper II, Table 4, Figure 1d). A potential explanation is that these experiments included the most efficient treatments and that the water control was removed. Removing the water control reduced inoculum levels and subsequently the disease pressure.

Since there experimental facilities did not allow adding carbon dioxide to the system and because an unconventional training system was used, we did not examine effects of the different treatments on yield in this study. Results of previous studies on the effect of the *Reynoutria* plant extract on cucumber yield are inconclusive. The different studies, using the Milsana formulation of the *Reynoutria* extract, either showed no effect on yield or an increase in yield (Wurms et al. 1999; Petsikos-Panayotarou et al. 2002; Konstantinidou-Doltsinis and Schmitt 1998).

In all experiments, Fungazil 100 had poor effect on disease severity compared to Sakalia and Yuccah combined. It delayed the disease progress slightly in some experiments but the disease severity was only slightly lower compared to untreated control (Paper II, Figure 2a). The poor effect of Fungazil 100 could be seen as an indication that the powdery mildew strain used in our experiments has developed some level of resistance towards Fungazil 100.

Figure 11. Experiments in 2015. On the left are plants treated with Sakalia and on the right are the water treated control plants (Photo: Mira Rur).

Table 2. Overview of experimental setup of the different experiments.

	2013			2015	
Planting Date	July 1	July 19	2 August	June 16	5 August
Duration	4 weeks	8 weeks	8 weeks	7 weeks	7 weeks
Cultivar	Euphoria	Euphoria	Euphoria	Euphoria, Proloog	Euphoria, Proloog
Plant Density*	1.95 plants/m^2	2.02 plants/m^2	2.02 plants/m^2	2.32 plants/m^2	2.32 plants/m^2
Treatment	**Experiment 1**	**Experiment 2**	**Experiment 3**	**Experiment 4 and 5**	**Experiment 6 and 7**
1.	Kendal Cops	Sakalia+Yuccah	Sakalia+Yuccah	Sakalia+Yuccah	Yuccah, 7 d
2.	AQ10+Bioglans	Polyversum	Polyversum	Hortistar	Sakalia, 7 d
3.	Fungazil	Hortifain	Hortifain	Hortistar+Fungazil	Sakalia+Yuccah+Fungazil 14 d
4.	Sakalia+Yuccah	Fungazil	Fungazil	Sakalia+Yuccah+Fungazil	Sakalia+Yuccah 14 d
5.	Hortistar	Resistim+Zence	Kendal Cops	Fungazil	Sakalia+Yuccah+Hortistar 14 d
6.	Untreated ctrl	Untreated ctrl	Untreated ctrl	Untreated ctrl	Untreated (Pos. ctrl).
7.	Water ctrl	Water ctrl	Water ctrl	Water ctrl	Sakalia+Yuccah 7 d (Neg. ctrl.)

*The standard plant density of Swedish commercial cucumber greenhouses is 1.5 plants/

7 Conclusions

7.1 The PAR approach

The "Lygus project" is a good example of both the advantages and the challenges of PAR. The great benefit of this methodology is the co-learning process and that views of participants with diverse backgrounds are equally valued. It is a great way of taking of advantage of peoples' individual experiences and competences and thereby increasing the chance of reaching higher learning outcomes. Furthermore, working directly with farmers/growers helps you to formulate relevant research questions. During the project we have also seen how working together increases the understanding between participants with different backgrounds and that preconceived ideas could be abandoned. Additionally, it seems as if the group's work also sparked an increased interest among growers for research and enabling of new research projects. For example, this led directly to the initiation of the CPM project.

The growers did find the research process of the project too slow but at the same time gained awareness of how costly, and sometimes slow, research projects can be. A general consensus between growers was that they wished for projects with larger budgets in order to obtain more relevant results in a shorter time.

The challenges of this specific project were mostly practical. Growers are business owners who face significant pressure to maintain high production levels and turnover and therefore have busy schedules. For this reason it was sometimes difficult to gather everybody at the same time.

Another challenge was to make everyone talk and contribute to discussions. This is where the facilitator comes in. A skilful facilitator is central to the co-learning process. By wisely selecting tools and techniques from the available and diverse toolbox created for co-learning processes, group communication and dynamics can greatly be improved. By timely asking key questions, the facilitator also helps to maintain the structure of the process, ensures that all participants are heard and that they are active in the continuous evaluation of the project.

In this study, combining expert research knowledge and practical knowledge and experiences of growers in these projects has been perceived as highly rewarding for all participants. Growers also expressed that the PAR approach in this case has helped assure that the research conducted was relevant, resource efficient and provided relatively fast answers.

In the final evaluation process of the “Lygus project” (section 4.3.1.), it was clear that several growers both felt that they had gained more knowledge of the biology of the ETPB and that they had made changes in their production in relation to the project. Changes such as becoming more observant and more thorough with greenhouse hygiene were mentioned, both of which are important in IPM systems.

As mentioned, collaboration between stakeholders could facilitate the development and implementation of IPM. By collaborating, the process of finding potential bottlenecks and knowledge gaps is shortened which subsequently leads more focused efforts.

7.2 Towards IPM strategies for the European Tarnished Plant Bug and Cucurbit powdery mildew

Investigation into if sunflower could serve as a trap crop for the ETPB in commercial greenhouses with cucumber as the main crop.

Flowering sunflower was more attractive than cucumber but was not sufficiently attractive or efficient as a trap crop to control the ETPB. In this experiment, there was still damage of the cucumber crop.

There is a possibility that the efficiency could be increased by increasing the number of sunflower plants in the greenhouse and optimizing their placement combined with a strategy to kill the ETPBs on the trap crop to avoid spread. However, according to growers of the PAR group, placing more sunflowers plants in their greenhouses is not practical or economically sustainable. Furthermore, the sunflower plants did not thrive under the same growing conditions as cucumber.

When using trap crops or traps the strategy must be thoroughly planned not to affect the spread of a pest in an unwanted manner or attracting the pest into the greenhouse.

Comparison of the behavioural responses of the adult European tarnished plant bugs (ETPBs) to cucumber with those of candidate trap crops, i.e. sunflower in olfactometer assays.
The growers of the PAR group considered the development of a trap combining both visual and olfactory stimuli to be of higher practical and economical relevance compared to using a trap crop. In relation to this, olfactometer experiments were conducted in order to test the behavioural response to volatiles of sunflower and cucumber. It was found that volatiles from flowering sunflower emanating from consistently attracted more bugs than those coming from the cucumber plant.

Assessment of the responses of the ETPBs to headspace volatile collections of candidate trap plants and identification of the attracting chemical compounds.
The chemical analysis of the plant odour collections showed a well-defined differentiation between sunflower and cucumber. Additionally, sunflower released several monoterpenes exclusively and was shown to have an overall release rate almost four times higher than cucumber. Based on this, sunflower is a highly interesting candidate for further testing of attractiveness to the ETPB within a cucumber background.

Screening for effective alternative products against cucurbit powdery mildew (CPM).
Sakalia and Yuccah combined (tank mixed) applied in seven-day intervals was the most efficient in controlling CPM in all experiments of this study.

Evaluation of the effect of selected products on CPM alone and in combination with the standard fungicide at different application intervals and in cucumber cultivars with different levels of resistance.
Sakalia and Yuccah combined and applied in fourteen-day intervals sufficiently controlled CPM in the partially resistant cultivar. The fourteen-day interval treatment did however not provide an equally high protection in the susceptible cultivar. Therefore, application timing needs to be optimised depending on the resistance level of commercially used cultivars. By optimising application timing, this treatment could provide sufficient control of CPM for commercial growers.

The main advantage of artificial inoculation is that it provides a timed and even disease pressure. A possible disadvantage is that it creates an unnaturally high disease pressure. We conclude that treatments which efficiently control the disease under these severe conditions must be considered highly efficient and that they could be even more efficient in less severe conditions, such as the natural infection levels of a commercial greenhouse.

7.2.1 Overall conclusion

Greenhouse systems are highly suitable for implementation of IPM practices. By testing and evaluating alternative pesticides for CPM and trap crops and odours for attracting the ETPB in semi-commercial, commercial greenhouse settings, and laboratory assays respectively, new knowledge has been gained which can be used to develop tools for potential future incorporation into IPM practises in cucumber production systems.

8 Future perspectives

Based on the knowledge gained from these experiments and previous studies made on visual and olfactory stimuli of tarnished plant bugs, traps with a combination of stimuli could be considered as an economically viable alternative. It is also the preferred method of growers.

Volatiles collected from sunflower were capable of attracting significant numbers of ETPBs when competing with volatiles collected from cucumber plants. However, additional research is necessary to identify the behaviourally active volatiles.

Based on previous studies, sunflower volatiles may also be tested in mixtures with other attractive volatiles such as phenyl acetaldehyde. Additionally, thorough testing in commercial cucumber greenhouses is necessary to evaluate their suitability as a new tool to reduce losses caused by the ETPB. In the future, this could hopefully lead to the development of traps for mass trapping. The traps could be placed at strategic positions in the greenhouse in order to catch and eliminate the bugs as soon they enter the greenhouse.

Before such traps are commercially available, hanging baskets with sunflower treated with a systemic pesticide above the cucumber crop along the aeration vents of the house, as suggested by growers, could be an alternative control method. The time to place these baskets could be determined using information gained from the capture in pheromone traps (www.agralan.co.uk).

As for the CPM study, the main objective was to screen for alternative products suitable for incorporation into an IPM program which subsequently could lead to a reduction in the use of chemical fungicides. The two years of experiments at the university were originally intended to lead up to a third year of implementation trials in a commercial greenhouse. Having seen the potential of the *Reynoutria* extract combined with wetting agent Yuccah, this is something that could be strongly proposed for future research.

Testing the most efficient treatment of our experiments in a large-scale commercial setting with high pressure spraying equipment would give the possibility to fully determine the potential of the treatment as part of an IPM program. It would also enable evaluation of possible controlling effects on other problematic diseases such as gray mold (*Botrytis cinerea*) and gummy stem blight (*Didymella bryoniae*) as well as effects on predatory mites commonly used for biological control. Furthermore, it would enable a highly relevant evaluation of treatment effect on cucumber yield.

If such studies show the efficiency of Sakalia with Yuccah in large scale commercial greenhouses, the treatment could function as an important part of an IPM program in cucumber. Due to its efficiency, it would strongly reduce the need for chemical fungicides.

Resistant cultivars are usually seen as an essential part of an IPM program. However, as reported by growers and advisors, the partially resistant cucumber cultivars available today do not produce sufficiently high yields under Swedish conditions as well as being more susceptible to gummy stem blight.

In this study, the *Reynoutria* extract combined with the wetting agent was highly efficient even in the susceptible cucumber cultivar. This means that there is a possibility that growers could stick to using the high yielding susceptible cultivars and thereby diminish the need for chemical fungicides for both CPM and gummy stem blight. As CPM, gummy stem blight and gray mold are among the most difficult diseases in cucumber greenhouses, finding alternative measures for all of them would enable a substantial reduction of the use of chemical fungicides. Furthermore, as biological control against pests is already used to a large extent in Swedish cucumber greenhouses, finding alternative measures also for the ETPB would enable cucumber cultivation with minimal use of chemical pesticides while retaining the same level of production.

Due to the new EU framework directive 2009/128/EC on the sustainable use of chemical pesticides and the reports on toxicity to beneficial insects, Imidacloprid is at risk of being banned in greenhouse cultivation the future. In light of this and the emerging resistance of CPM towards Fungazil (Imazalil) there is an urgent need to find alternative IPM strategies.

In this thesis, a general suggestion of how to incorporate the findings of the study into a potential future IPM program for cucumber is made (Figure 13). This IPM program would be based on education, and continuous prevention, monitoring and evaluation in order to minimise need for intervention. In the suggestion, the *Reynoutria* extract/wetting agent combination is proposed for prophylactic use. Since the treatment was highly efficient in experiments in spite of the unnaturally high disease pressure created by artificial inoculation, it is likely that prophylactic spraying in a susceptible cultivar could be reduced to fourteen-day intervals in commercial settings. Reduced spraying intervals is preferred not only for economical and practical reasons but because all spraying is potentially disruptive to biological control insects.

Among preventative measures, sanitation of greenhouse compartments is essential to avoid early CPM infestation due to overwintering mycelia.

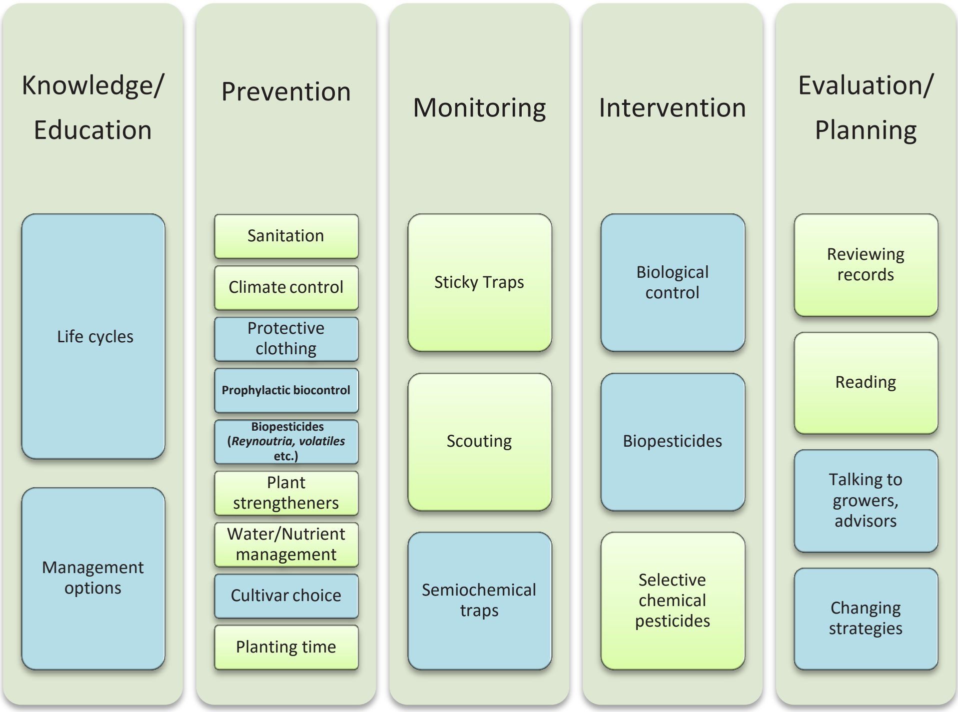

Figure 12. The cornerstones of a potential future IPM strategy for greenhouse cucumber with examples of suitable tools. Highlighted in blue are the tools used in this thesis.

In general, there seems to be a constant need for more applied studies where IPM strategies integrating different tools/control practises are compared and tested under various conditions. Since these strategies are in fact intended for use by growers, it is highly relevant to collaborate in some way to secure knowledge transfer. This thesis provides an example of how the use of a PAR approach can contribute to increased knowledge and understanding, to actual changes in management strategies and to facilitate the development of IPM strategies.

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Acknowledgements

I think I have to begin by going back to where this all really started for me, in my grandparents' allotment in Fredriksdal in Helsingborg. With them and in their allotment house and garden, which seemed quite huge to me then, I was in total peace and was allowed to pick as many flower bouquets as I wanted and only have to worry about which fruit to pick for making dessert on this particular day. I "helped" with taking up potatoes for lunch, picking fruit and berries, watched my grandmother collect seeds for next year, enjoyed the lovely scent of the sweet peas and listened to them talking about which their favourite apple cultivars were (they both liked 'James Grieve') and what their next gardening project would be. I also loved looking through their gardening books and trying to draw the flowers the best I could. *Karin and Lars*, wherever you are now, thank you for sparking my interest in plants and giving me the precious gift of childhood bliss!

Eventually this interest led me to the horticulture program at SLU in Alnarp… In Alnarp I met Birgitta Rämert as a teacher. She became a special teacher to me and to many in my year cohort mainly because of her ability to pass on her own enthusiasm and passion for learning and because of her commitment to us as students. I want to thank my main supervisor, *Birgitta Rämert*, for believing in me and giving me the opportunity to evolve within my field. The winding journey leading up to this thesis all began because of this. You always did your best to be available and gave invaluable advice based on your great experience.

I also want to thank my co-supervisor, *Margareta Hökeberg*, for your support and commitment. You were always enthusiastic and honest and gave me great advice along the way.

I very much want to thank *Erland Liljeroth*, my main supervisor during the final year of the project. You have given me great advice and input. While guiding me towards the finishing point you encouraged me to think independently and you always made sure to be available for feed-back.

I also want to thank my co-supervisor, *Laura Grenville-Briggs Didymus*, for always giving me great support in all phases of the project during the final year. You amazed me with your capacity and multitasking abilities.

Great thanks to my dear colleagues and co-authors *Mario Porcel Vilches* and *Marco Tasin* whose great contributions made the article on trap crops for the European tarnished plant bug what is today and who were wonderful to work with.

A special thanks to my colleagues and co-authors *Lorna Migiro* and *Sopher Natuluku Ondiaka,* whose amazing work ethics and professionalism, patience and good sense of humour was a joy and a privilege to be around.

To the group facilitator of the "Lygus project", *Elisabeth Ögren*, thank you so much for contributing with your great knowledge of participatory action research. Your experience, professionalism and humbleness made everyone feel at ease and greatly contributed to the great outcome of the group work. I also want to thank you for being so supportive of me.

Thanks to all the cucumber growers who took time out of their busy schedules to join the working group and who showed such dedication and interest in collaborating to try to find new solutions. *Ingemar Bengtsson, Göran Ektander, Tomas Isberg, Nils-Åke Månsson Boban Nikolic* and *Maria Adolfsson*, this wouldn't have been possible without you.

Thanks to *Barbro Nedstam,* senior advisor, for being so helpful and supportive and contributing with your great knowledge and experience during the "Lygus project".

Also great thanks to *Johanna Jansson* for your valuable contributions in your role as an advisor during the "Lygus project".

A big thanks to *Sven Hellqvist* for sampling insects in Umeå.

Rob Jacobson and Jantineke Hofland-Ziljstra, thank you so much, for contributing with your expertise advice in the beginning phase of the powdery mildew project.

Ramesh Vetukuri, thank you for your help and expert molecular biology support during the end of the project. You always managed to find time and your calm, honesty and sense of humour helped take my mind off worrying.

Big thanks to everyone in the Integrated Plant Protection unit. You have all contributed to a great working atmosphere offering professional advice as well as good laughs and made me really enjoy my years here.

My dear friend and colleague, *Linda-Marie Rännbäck*, thank you for being who you are. Smart, humble, fun, caring and the most balanced person I know.

Therèse Bengtsson and *Helena Hovmalm Persson* and *Malin Dörre*, thank you for always listening and being so supportive, you could always make me laugh when I needed it the most.

I also want to thank the Resistance Biology unit, for "taking me in" during the final year of the project.

To past and present undergraduate students, PhD students and postgraduate researchers, laboratory staff and all the lovely people that I met in Alnarp over the years, thanks for creating such a nice working atmosphere.

I also want to thank the staff at the Garden lab in Alnarp who assisted me in the greenhouse, who I could always count on and who made the heavy tasks much more fun.

Finally, I want to thank my extended family for all of their love and support and express my gratitude towards my loving little son *Misha* who is my greatest gift in life.

Last but not least, I want to thank the funding organizations SLF and Partnership Alnarp for their financial support which made this project possible.