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Virus diseases of *Ribes* and *Rubus* in Europe and approaches to their control

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Abstract: There are more than 30 viruses and virus diseases reported to occur in *Rubus* and *Ribes* crops worldwide and several of those occurring in Europe can damage crop production and/or quality severely. As with all crops, planting virus-tested certified material away from likely sources of infection is essential for control but, because of the longevity of these crops (more than 10 years), satisfactory control of incoming virus infection over the life of the crop is often difficult to achieve.

In *Ribes* the major pathological problem is Blackcurrant reversion disease and its gall mite vector, both of which cause serious damage. The only other virus disease of significance is Gooseberry vein banding, the causal agent of which is transmitted by aphids. Rapid and sensitive detection assays have recently been developed at the Scottish Crop Research Institute (SCRI) for the viruses involved in these two virus diseases. Effective control of reversion disease and of the gall mite vector seems very promising using resistance genes to these organisms in cultivars. However, the species structure, ecology and virus vector capabilities of other eriophyid mites found on *Ribes* species needs further study to assess the likely stability of currently deployed gall mite resistance genes.

In *Rubus*, the most widespread virus and the most difficult to control is the pollen-borne *Raspberry bushy dwarf virus* (RBDV). Cultivars with resistance genes to the common isolates of this virus are effective in preventing infection but the occurrence of resistance-breaking isolates of this virus pose problems for control. Aphid-borne viruses are also widespread and can be very damaging. Very effective control of most of these viruses has been achieved through the use of cultivars carrying genes for resistance to the main aphid vector, *Amphorophora idaei*. However, these viruses have been increasing in incidence in crops due to the ability of the aphid to overcome these resistance genes. Viruses causing Raspberry leaf spot mosaic disease are lethal in sensitive raspberry cultivars and their spread is unlikely to be controlled effectively by the application of aphicides. *Raspberry vein chlorosis virus* transmitted by the small raspberry aphid, *Aphis idaei*, is common in several crops posing problems for control. Soil-borne viruses, although very damaging when they occur in crops, are usually only localised in occurrence, but they may become of more widespread significance following the withdrawal of soil sterilants used commonly in horticulture to control nematodes.

Key words: virus transmission, aphids, nematodes, eriophyid mites, pollen transmission, virus resistance, virus tolerance, virus vector resistance, *Ribes, Rubus*

Introduction

More than 30 virus or virus-like diseases are reported in *Ribes* and *Rubus* crops worldwide (Jones, 1992) and several of those occurring in Europe can damage crop production and/or quality severely. Most viruses are transmitted between plants by plant-feeding vectors and the vector species associated with the transmission of the most important viruses and virus complexes in *Ribes* and *Rubus* crops in Europe are given in Table 1 together with the virus acronyms used throughout this paper.

Table 1. Important virus vector species on cane fruit crops in Europe and the viruses or virus complexes they transmit.

Virus vector species	Virus/virus disease and acronym
Aphid:	
Amphorophora idaei (large raspberry aphid)	Black raspberry necrosis virus (BRNV) Raspberry leaf mottle virus (RLMV) Raspberry leaf spot virus (RLSV) *Rubus* yellow net virus (RYNV) Raspberry veinbanding mosaic disease
Aphis idaei (small raspberry aphid)	Raspberry vein chlorosis virus (RVCV)
Nasonovia ribisnigri, Hyperomyzus lactucae, Myzus persicae, Cryptomyzus ribis	Gooseberry vein banding disease (GVBD)
Nematode:	
Longidorus species	Raspberry ringspot virus (RpRSV) Tomato black ring virus (TBRV)
Xiphinema diversicaudatum	Arabis mosaic virus (ArMV) Strawberry latent ringspot virus (SLRSV)
Eriophyid mite:	
Cecidophyopsis species	Blackcurrant reversion virus (BRV)
Pollen-borne:	
No known vector	Raspberry bushy dwarf virus (RBDV)

Because viruses are obligate parasites that use the host plant biochemical machinary for their own replication and spread within plants, control of viruses in commerce is currently possible only by indirect means. These include the production of healthy planting material, the control of vectors to minimise spread within such initially healthy crops, and the use of host plant resistance to virus infection, replication and spread within plants. This paper briefly summarises and evaluates these methods to control the important viruses and virus diseases in Table 1, identifies the current problems in using these different approaches to control, and assesses the future prospects of overcoming these difficulties.

Results and Discussion

Planting of virus-tested stock plants

Planting virus-infected material provides an infection source within the crop at the earliest possible time and when plants are most vulnerable to, and suffer the greatest effect from, infection; it allows the maximum time for virus spread within that crop. Planting virus-free stocks is therefore vital for satisfactory control of all viruses in Rubus and Ribes plantings. Nevertheless, until the 1960s, growers were commonly planting material derived from diseased stock, so that viruses became widely distributed in planting stocks, often together with their vector. In Scotland, the development and introduction of a Certification Scheme for planting stock, based on material derived from plants tested and found free from important viruses and other diseases and pests, was the first formal scheme to provide growers with the opportunity to break the cycle of perpetual infection (Jones, 1991). Subsequently, similar cane fruit stocks schemes, based on the Scottish model, were introduced in other countries. These have been a major contribution to improved plant health and crop yield.

Control of nematode-borne viruses

The four nematode-transmitted viruses listed in Table 1 occur commonly throughout Europe, especially Northern Europe, and can cause serious damage and yield loss. However, they are usually restricted in distribution to patches within crops. Each of these viruses, and their respective vector, has a very wide natural host range including many weed species. The viruses are transmitted through the seed of many of these species, often to a high incidence (Murant et al., 1996). *Rubus* species are commonly infected with these viruses but infection in *Ribes* species is rare (Jones & McGavin, 1996). Although immunity to these viruses occurs in raspberry (Jones & McGavin, 1998), this is highly strain specific and is not effective in the field where several different strains can occur at a single site (Jones et al., 1989).

Effective virus control in several different crop species has been achieved using soil fumigants prior to planting to kill viruliferous nematodes. Nematodes that occur beneath the fumigated zone will in time re-colonise this zone, therefore effective weed control is necessary to prevent seedlings from virus-infected weeds germinating and then providing a source from which nematodes can re-acquire virus to transmit to crop plants. Whilst these combined control measures have been effective for controlling RpRSV and TBRV, control of ArMV and SLRSV, especially in raspberry, has been less effective because after acquiring virus, *X. diversicaudatum* retains and transmits it for 12 months or more. These nematodes are therefore still viruliferous when they colonise the initially sterile zone even in the absence of virus-infected weed sources.

The proposed withdrawal of many soil sterilants from commercial use in the near future may limit the continued use of this approach for virus control. One possible future approach to control these viruses in perennial crops is to engineer resistance using genes derived from the viral genome. In a preliminary assessment of this approach, effective resistance to nematode inoculation with SLRSV was obtained in tobacco plants transgenic for some of the coat protein gene sequence of SLRSV (Kreiah et al., 1996).

Control of aphid-borne viruses

The large raspberry aphid, *Amphorophora idaei,* transmits four of the five important aphid-borne viruses infecting *Rubus* (Table 1). Some insecticides are very effective in controlling *A. idaei* numbers on *Rubus*, but not in decreasing significantly the four viruses it transmits (Taylor & Chambers, 1969; Jones, 1986).

No immunity to the four *A. idaei*-transmitted viruses (Table 1) has been detected in *Rubus* germplasm (Jones & Jennings, 1980), but several sources of strong resistance to *A. idaei* are known (Knight et al., 1959; Keep & Knight, 1967). Such resistance has been introduced into commercial raspberry cultivars with a dramatic effect on the incidence of the four viruses it transmits (Jones, 1976; 1979; 1988). Indeed, observations in the UK for over 20 years has shown that crops of commercial *A. idaei*-resistant raspberry cultivars contained less than 10 % infection with some *A. idaei*-transmitted viruses up to 16 years after planting, without the use of aphicides (Jones, 1988; A.T. Jones, unpublished data). Consequently, a major requirement for most raspberry breeding programmes is resistance to *A. idaei*. However, the widespread and prolonged cultivation of such resistant raspberry cultivars has created a strong selection pressure on the aphid to overcome such resistance. Five biotypes of *A. idaei* are now recognised in the UK and one or more of these biotypes are capable of colonising plants containing the *A. idaei*-resistance genes A_1 and A_{10} (Birch & Jones, 1988; Jones et al., 2000). Biotypes able to overcome gene A_1 are now prevalent in raspberry crops in the UK, resulting in an increase in the incidence of the viruses transmitted by this aphid (Jones, 2004, this volume).

All raspberry cultivars are infectible with each of the four *A. idaei*-transmitted viruses but only some develop severe disease symptoms when infected with some of these viruses. For example, infection with RLMV or RLSV in raspberry cultivars sensitive to these viruses causes leaf spot mosaic, a severe disease that makes plants unproductive within 3-4 years of infection. The basis for this sensitivity to infection is determined by single dominant genes termed *Lm* and *Ls* for reaction to RLMV and RLSV respectively (Jones & Jennings, 1980). It is possible therefore for breeders to select progeny lacking these genes to produce plants tolerant to infection with these viruses.

The small raspberry aphid, *Aphis idaei,* transmits only RVCV that is very common in crops in Europe. No sources of resistance to this aphid have been identified in *Rubus* germplasm but resistance to the virus occurs in some *R. strigosus var. idaeus* material. Crosses between RVCV-infectible and –resistant material produced a significant number of progeny with high resistance/immunity to RVCV (Jennings & Jones, 1986). Although the precise mechanism of inheritance of this resistance remains undetermined, it seems that selecting RVCV-resistant material might not be difficult to achieve (Jennings & Jones, 1986).

GVBD has been little studied, so no information is available on the control of this, or other, aphid-borne viruses of *Ribes* by insecticide applications, nor is resistance to GVBD reported in germplasm. Control of GVBD by breeding for resistance to its aphid vectors seems remote because several different aphid species are reported as vectors (Table 1). However, as with other virus diseases, the planting of healthy stock away from sources of infection, roguing out infected plants, and the control of possible vector aphids are sensible precautions to minimise virus spread (Jones, 2002).

Control of eriophyid mite-transmitted virus

Reversion of blackcurrant is the most important disease of blackcurrant worldwide. The causal virus (BRV) is transmitted by the blackcurrant gall mite, *Cecidophyopsis ribis,* which is itself the most important pest problem in blackcurrant (Brennan, 1990; Jones, 2000). In the past, control of both of these organisms has been through roguing affected bushes and the control of mite numbers using acaricides. However, satisfactory chemical control of mites is difficult to achieve due to the fact that applications must coincide with mite dispersal from blackcurrant buds. This dispersal is determined by environmental conditions and by the position of the bud on the cane. When there are more than 5% galled plants in crops, further chemical control of spread is regarded as uneconomic. Breeding for resistance to *C. ribis* and/or BRV is therefore a major priority in blackcurrant breeding programmes (Brennan, 1990; Brennan et al., 1993).

Resistance, possibly immunity in many instances, to each of these organisms has been identified in *Ribes* germplasm. Gene *Ce* from gooseberry has been introduced into blackcurrant to give gall mite resistant plants (Knight et al., 1974). Additionally, *R. nigrum* var. *sibiricum*, *R. pauciflorum*, *R. petiolare* and *R. ussuriense* have also been used as donors for gall mite resistance, which, in these species, is determined by gene *P* (Anderson, 1971). The resistance conferred by gene *Ce* is much stronger than that of gene *P*, virtually preventing mite infestation altogether. In gene *P*-containing plants however, gall mites are initially able to infest buds that become necrotic thus inhibiting further survival and reproduction of mites. In the field, BRV is found in plants containing gene *P* (Pavlova, 1964; Jones et al., 1998a), suggesting that this resistance source is not very satisfactory for controlling BRV. By contrast, gene *Ce*-containing blackcurrant cultivars remained uninfected with BRV in field experiments using high inoculum pressure from viruliferous gall mites (Jones et al., 1998a). Resistance, possibly immunity, to BRV has been identified in *R. cereum, R. dikushka* and *R. pauciflorum* (Pavlova, 1964; Brennan, 1990; Brennan et al., 1993). In field trials under high inoculum pressure of BRV, reversion-resistant blackcurrant cultivars failed to become infected even though such plants contained many galls (Jones et al., 1998a). Despite this, and because of the serious damage caused by gall mites as pests, the use of gene *Ce* that confers strong resistance to these mites and consequently also to BRV, seems the best means of control.

Recent studies on the molecular ecology of eriophyid mites on *Ribes* species in Europe indicate the occurrence of a larger number of *Cecidophyopsis* species than that known previously, and that some mite species are able to colonise *Ribes* species assumed previously to support only one species (Fenton et al., 1995; 1996). These findings may have implications for the durability of the mite-resistant genes currently being deployed.

Pollen-transmitted virus

RBDV causes Yellows disease of *Rubus,* can induce severe crumbliness of fruit and, in association with aphid-borne viruses, causes 'raspberry bushy dwarf disease' leading to degeneration in plant vigour (Jones et al., 1996). RBDV is currently regarded as the most widespread and important virus of *Rubus.* It is transmitted between *Rubus* plants in the field only in association with flowers; no animal vector is reported (Jones et al., 1996). Because of its mode of spread, control is only possible either by growing healthy *Rubus* stocks away from infection sources, or by growing RBDV-resistant cultivars. Resistance to the common isolate of RBDV, that is prevalent wherever *Rubus* is grown, is present in many commercial raspberry cultivars and this resistance is determined by a single dominant gene, *Bu* (Jones et al., 1982; 1998b). This has provided effective control of such virus isolates for more than 40 years even under strong inoculum pressure. However, in more recent years, isolates of RBDV able to overcome gene *Bu* (termed resistance-breaking isolates, RB) have been identified in commercial raspberry crops in England and Wales, Northern Europe, and Russia. Such isolates are able to infect almost all *Rubus* germplasm tested (Jones et al., 1996), and control of these RB isolates is therefore a serious problem.

A possible approach to control RB isolates is to engineer resistance using transgenes derived from the RBDV viral genome. Initial experiments using a tobacco model suggest that such an approach might be successful (Angel-Diaz et al., 1997). Attempts to produce and evaluate the resistance to RBDV of transgenic raspberry are being made currently in Scotland (Angel-Diaz et al., 1997) and in the USA (Martin & Mathews, 2001). More recently, identification of possible somatic mutants of an RBDV-susceptible cultivar with immunity or very strong resistance to RB isolates has been observed (A.T. Jones, unpublished data).

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Exploiting natural enemies in Integrated Pest Management in blackcurrant crops

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Abstract: A 3 year research project developed Integrated Pest Management (IPM) approaches for mites, blackcurrant leaf midge and aphids on blackcurrant. An experimental plantation was planted at HRI-East Malling in September 1999 for the purposes of the project. It comprised a 5x5 Latin square of 25 plots. Each plot contained three varieties in a 3-way split plot design. The varieties were Ben Gairn (reversion resistant but gall mite susceptible), Ben Hope (gall mite resistant but reversion susceptible) and Ben Alder (susceptible to both gall mite and reversion). Five different spray programmes were applied annually as treatments as follows: A) two sprays of sulphur, one at the start and one at the peak of gall mite migration B) two sprays of sulphur (as in A)) plus a pre-flowering spray of the OP chlorpyrifos C) two sprays of sulphur (as in A)) plus a pre-flowering spray of pirimicarb D) three sprays of fenpropathrin (positive control = current grower practice) E) untreated (negative control).

The work showed that combining host plant resistance with early season sprays of sulphur is an effective strategy for preventing gall mite infestation, far superior to using programmes of sprays of fenpropathrin with susceptible varieties. The fenpropathrin spray programme, an established industry standard for many years, was shown to be less effective than early sprays of sulphur. The work showed Ben Gairn to be resistant to gall mite and Ben Hope to be highly, though not fully, resistant. The need to protect the resistance with sprays of sulphur was demonstrated.

It was shown that predatory phytoseiid mites, mainly the orchard predatory mite *Typhlodromus pyri*, which is resistant to OP insecticides, establishes in blackcurrant plantations and populations are sustained even in the apparent absence of spider mite prey. *T. pyri* is likely to be important in naturally regulating spider mite populations in blackcurrant plantations. The predator was badly affected by the 3x fenpropathrin spray programme and was somewhat reduced by the 2x sulphur programmes.

The parasitic wasp *Platygaster demades* was found to be an important natural enemy of blackcurrant leaf midge. It is likely that the parasite is highly susceptible to insecticides, especially in the adult stage. Sprays of broad-spectrum insecticides need to be avoided to allow the parasite to establish.

The blackcurrant aphid and currant sowthistle aphid were the most abundant aphid species and that there are many natural enemies of aphids on blackcurrants of which ladybirds are often the most common. Fenpropathrin sprays, which had little affect on aphids, adversely affected ladybird populations. Other predatory insects included anthocorids, syrphids and lacewings. In 2002, a season when very wet conditions occurred during May and June when aphid populations were high, parasitic wasps and entomopathogenic fungi (mainly *Entomophthora planchoniana*) caused very high mortality of aphids, causing populations of aphids to collapse naturally. In none of the three years of the project were natural enemies able to control aphid populations before they caused significant plant damage.

Key words: blackcurrant gall mite, *Cecidophyopsis ribis*, blackcurrant leaf midge, *Dasineura tetensi, Platygaster demades Typhlodromus pyri* ,currant sowthistle aphid *Hyperomyzus lactuca,* blackcurrant aphid, *Cryptomyzus galeopsidis,* host plant resistance, biocontrol, parasitoid, *Entomophthora,* chlorpyrifos, pirimicarb, fenpropathrin

Introduction

For many years all commercial blackcurrant crops in the UK have been sprayed at least three times per annum with the broad-spectrum insecticides endosulfan and/or fenpropathrin to control the blackcurrant gall mite, *Cecidophyopsis ribis*, the vector of reversion virus disease. The disease causes sterility in blackcurrant bushes and is the principle factor limiting the life of blackcurrant plantations. Its gall mite vector is difficult to control even with the intensive spray programmes of the insecticides. However, an important recent development has been the release of two new blackcurrant varieties, one (Ben Gairn) resistant to the virus, the other (Ben Hope) resistant to the gall mite. These varieties, which are only available to UK growers, are being planted on a rapidly increasing scale. At the outset of this project it was estimated that they would comprise over 50% of the UK acreage in 5 years time. The intensive use of broad-spectrum insecticides to control the gall mite has hitherto effectively prevented the development of Integrated Pest Management approaches for the crop. The advent of these new resistant varieties means that for the first time it may be possible to grow blackcurrants commercially without routine use of broad-spectrum insecticides, providing excellent opportunity to develop Integrated Pest Management approaches which will result in substantially reduced use of pesticides.

The overall aim of this project was to develop effective Integrated Pest Management (IPM) approaches for mites, blackcurrant leaf midge and aphids on blackcurrant. Specific objectives were:

1. To determine whether the use of host plant resistance to the gall mite (in cv Ben Hope) or to reversion virus disease (in cv Ben Gairn) combined with two sprays of sulphur (at the start and peak of migration of the gall mite) can adequately prevent gall mite and /or reversion disease infection under conditions of severe pest pressure. To determine the effects of such approaches on the natural enemies of other important pests.
2. To determine the species composition of the predatory phytoseiid mite fauna on blackcurrant and how phytoseiid and spider mite populations are affected by the IPM strategies for gall mite control in objective 1 above.
3. To determine whether the parasitoid *Platygaster demades* can be exploited as a natural enemy of blackcurrant leaf midge.
4. To characterise the natural enemy complex of currant sowthistle aphid and to quantify levels of natural enemy attack in an IPM programme.

First results of this work were reported by Cross & Harris (2003).

Material and methods

Experimental plantation

A new experimental blackcurrant plantation was planted at HRI-East Malling in September 1999 for the purposes of the work. It comprises a 5x5 Latin square of 25 plots. Each plot consists of 6 rows of 6 blackcurrant bushes; two rows of each of three varieties in a 3-way split plot design. The varieties are Ben Gairn (reversion resistant but gall mite susceptible), Ben Hope (gall mite resistant but reversion susceptible) and Ben Alder (susceptible to both gall mite and reversion). Five different spray programmes were applied annually as treatments in a Latin square experiment design with five replicate plots of each treatment. These were

A) two sprays of sulphur, one at the start and one at the peak of gall mite migration
B) two sprays of sulphur (as in A)) plus a pre-flowering spray of the OP chlorpyrifos
C) two sprays of sulphur (as in A)) plus a pre-flowering spray of pirimicarb

D) three sprays of fenpropathrin (positive control = current grower practice)
E) untreated (negative control). The new experimental planting is close to other blackcurrant plots, which are severely infested with gall mite and reversion virus disease.

In order to seed the experimental plot with gall mite infestation, in the first year branches bearing 10 galls were placed in the middle of every row at the start of the migration period.

Results and discussion

Combining host plant resistance and early sulphur sprays in IPM of gall mite and reversion virus

Each year just before flowering when symptoms were readily visible, each bush was carefully inspected for symptoms of infection by reversion virus disease. The numbers of bushes infected in each plot was recorded. At the end of each year in the dormant period, the number of gall mite galls on each bush was counted. Treatment totals were calculated and, where appropriate, the data was subjected to analysis of variance.

In year 1, a total of 100 galls developed in the experimental plantation. Of these, 95 were on Ben Alder, 5 on the Ben Gairn and none on the Ben Hope (Table 1). The untreated and the fenpropathrin treatments had the most galls, approximately double the number on the sulphur treatments. No reversion virus infection was detected.

In year 2, the total numbers of galls had increased markedly but the same trends were apparent in the data. Most of the galls were found on Ben Alder, with small numbers on the fenpropathrin treated and the untreated controls of Ben Gairn but with no galls on Ben Hope. The fenpropathrin treatment did not reduce numbers of galls compared to the untreated control but the 3 treatments which included early sulphur sprays significantly reduced gall mite infestation by approximately 2 fold compared to the untreated control. No reversion virus infection was detected.

In the third year, the same trends were apparent in the data but small numbers of galls had developed on the Ben Hope, mainly on the untreated control plots (Table 1). The sulphur treatments were more effective than the fenpropathrin treatment, which was only partially effective. Reversion virus infection was detected in four bushes of Ben Alder just before flowering in the third year. There were two bushes in each of two plots that had received the sulphur+pirimicarb treatment (treatment C).

The results of this work lead to several conclusions. The relative susceptibility of the varieties to gall mite, as quantified by the number of galls present on them by the end of year 3, is as follows: Ben Alder : Ben Gairn : Ben Hope = 45 : 5.6 : 1. Thus Ben Gairn is partially resistant and Ben Hope highly, though not fully, resistant to gall mite in comparison with the susceptible Ben Alder standard. Varietal resistance needs to be protected by sprays of acaricides. The sulphur and the fenpropathrin treatments gave partial control of gall mite, but the standard 3x fenpropathrin treatment was less effective than the 2x sulphur treatments. Combining host plant resistance with use of early sulphur acaricide sprays gave a very high degree of gall mite control as follows: 2x sulphur + Ben Hope 99.5% control; 2x sulphur + Ben Gairn 99.85% control. It is too early to quantify susceptibility and the effects of treatments on reversion virus disease. The experiment is being continued for 4 more years.

Table 1. Total number of blackcurrant gall mite galls per 60 bushes in the dormant period at the end of each year.

Treatment	Ben Alder (susceptible)	Ben Gairn (reversion resistant)	Ben Hope (gall mite resistant)
Year 1 (2000)			
A. 2 sulphur	13	2	0
B. 2 sulphur+chlorpyrifos	12	1	0
C. 2 sulphur+pirimicarb	14	1	0
D. 3 fenpropathrin	30	0	0
E. untreated	26	1	0
Year 2 (2001)			
A. 2 sulphur	138	0	0
B. 2 sulphur+chlorpyrifos	126	0	0
C. 2 sulphur+pirimicarb	210	0	0
D. 3 fenpropathrin	360	9	0
E. untreated	456	21	0
Year 3 (2002)			
A. 2 sulphur	10	0	0
B. 2 sulphur+chlorpyrifos	6	2	1
C. 2 sulphur+pirimicarb	30	1	0
D. 3 fenpropathrin	50	4	2
E. untreated	195	62	15

Predatory phytoseiid mite fauna on blackcurrant and how phytoseiid and spider mite populations are affected by the IPM strategy

Each year, samples of 25 fully expanded leaves were taken from each plot on up to 3 occasions and the numbers of spider mites and predatory mites on each sample were counted. In years 1 and 2, samples were taken from each variety but in the third year sampling was concentrated onto the variety Ben Gairn. Microscope slide preparations were made of each adult predatory mite recorded so that they could be identified to species.

Small numbers of two-spotted spider mite were present in the samples taken in July 2000. Most were present on Ben Gairn with lower numbers on Ben Hope and only very small numbers on Ben Alder. No two-spotted spider mite individuals were found in any of the plots in 2001 or 2002.

Greatest numbers of phytoseiid predatory mites occurred in July-August each year. In the first year, small numbers appeared to be distributed erratically over the plots with greatest numbers on Ben Hope. The majority (75%) of the individuals were *Typhlodromus pyri* with small numbers (25%) of *Neoseiulus californicus* and one individual of *Neoseilus finlandicus* being recorded. In the second year, numbers of phytoseiids were very small only 3 individuals being found in total. They were all nymphs and hence were not identified to species. In the third year, only Ben Gairn was sampled. All the predatory mites recorded were *Typhlodromus pyri*. Greatest numbers were present on the untreated control plots. Only two individuals were found on the samples from the fenpropathrin treated plots. Intermediate numbers were recorded on the plots that were treated with sulphur.

Predatory phytoseiid mites established naturally in the first year. The orchard predatory mite *Typhlodromus pyri*, the species which is widely exploited in IPM in apple orchards, was the dominant species. It is likely that small numbers of two-spotted spider mite present mainly on Ben Gairn (a variety known to be susceptible to two-spotted spider mite) were controlled biologically by the predatory mites in the first year. It is likely that the two-spotted spider mites and the phytoseiid predators were present the original plant material supplied by the nursery. The predatory mites were able to survive in small numbers throughout the second and third years without detectable numbers of two-spotted spider mite being present. The counts in the third year show that the fenpropathrin treatment was very harmful to *Typhlodromus pyri* populations and that the sulphur treatments also has harmful effects. Overall, the results indicate that *Typhlodromus pyri* is likely to be important in IPM on blackcurrant and that avoiding the use of fenpropathrin is desirable.

Exploiting the parasitoid* Platygaster demades *to regulate leaf midge

In the years before this project started, blackcurrant leaf midge (*Dasineura tetensi*) was an abundant and damaging pest in other experimental plantations in the field at East Malling where the experimental plantation for this project was planted. However, in 2000, only 3 leaf midge galls occurred in the experimental plantation in 2000 (2 on bushes of Ben Alder sprayed which received the 2x sulphur only treatment and one on a Ben Gairn bush sprayed with sulphur + chlorpyrifos). Populations were thus too low to study parasitism on the experimental plantation. However, apple leaf midge galls containing parasitised apple leaf midge larvae were introduced into every plot in summer. A moderate infestation of blackcurrant leaf midge occurred on an old establish plantation of Ben Lomond and Ben Tirran nearby. Several thousand larvae were collected in June, July and August and were reared through to the adult stage to determine whether parasitoids were present. The parasitoid *Platygaster demades* was found in 1% of larvae. This confirmed that the parasitoid does use blackcurrant leaf midge as a host.

In 2001, populations of blackcurrant leaf midge in the experimental plots were zero and near zero throughout all the other blackcurrant plots at HRI-East Malling. However, several hundred mature larvae were collected from a commercial plantation near Taunton, Somerset and reared through to the adult stage. 18% were found to be parasitised by *Platygaster demades*.

In 2002, no leaf midge was again found in any of the plots at East Malling, either in the plantation used for this project of any of the other plantations (none had been sprayed with insecticides which would have controlled the pest. Samples of several hundred larvae were collected from the commercial plantation in Somerset and also from an 'in conversion' organic plantation near Yarmouth, Norfolk. 16% and 20% of larvae emerged as *Platygaster demades* from these two plantations respectively.

This work clearly showed that *Platygaster demades* is a significant natural enemy of blackcurrant leaf midge and that occurs in commercial plantations at moderate levels. Although these results are not conclusive proof, it is likely that the decline and near disappearance of blackcurrant leaf midge from the plots at HRI-East Malling was caused by the parasitoid. It is probable that the adult stage of the parasite is sensitive to insecticides.

Characterisation of the natural enemy complex of currant sowthistle and other aphids

The currant sowthistle aphid (*Hyperomyzus lactucae)* is the most damaging aphid pest of blackcurrant in the UK. However, there are several other common aphid pests of blackcurrants including the blackcurrant aphid (*Cryptomyzus galeopsidis*), a very abundant though less damaging species, the redcurrant blister aphid (*Cryptomyzus ribis*) and the permanent currant aphid (*Aphis schneideri*). These latter two species are also common but usually less numerous and only moderately damaging. Although the objective of the project was to investigate the natural enemy complex of the currant sowthistle aphid, the common natural enemies of all the aphid species, which often occurred in close proximity to each other and often in mixtures, were investigated.

In 2000, because the experimental plantation had only been planted the previous autumn, aphid populations were extremely small and all were the permanent currant aphid, *Aphis schneideri*. Beat sampling was done on 31 May but numbers were too small and erratic for statistical analysis. However, the data showed interesting trends, which were confirmed in subsequent years. The aphids were most abundant on the untreated controls and the 2x sulphur treatment where no insecticide spray had been applied. Interestingly, there were almost no coccinellids, anthocorids or spiders on the plots that received the 3x fenpropathrin treatment.

In 2001, assessments of aphid and natural enemy populations were made on 31 May and 11 June, when populations of aphids were comparatively high. The colonies of each species of aphid on 10 bushes of each variety in each plot were counted and the number of aphids in each colony estimated. Natural enemies in each colony were also identified, counted and recorded. The blackcurrant aphid was by far the most abundant species. Numbers of aphid colonies were generally greatest on Ben Gairn, the variety most susceptible to aphids. Mean numbers of colonies per bush were much smaller on the plots that received the 2x sulphur + chlorpyrifos or 2x sulphur + pirimicarb treatments. The 2x sulphur, 3x fenpropathrin and untreated control had similar numbers of aphids. Numbers of currant sowthistle aphid colonies were much (approximately 5x) smaller but followed the same trends. Coccinellids (especially *Calvia 14-punctata*) were the most abundant predators. They were most abundant on the 2x sulphur and untreated control plots where aphids were most numerous. They were absent from the 3x fenpropathrin treated plots. Other predators, of which anthocorids and hoverfly larvae were most numerous, followed the same trends, though numbers were smaller. Parasitoid mummies were also present in small numbers, including on the 3x fenpropathrin treated plots, where they were most numerous.

In 2002, aphids were far more abundant than in previous years and three assessments of aphid and natural enemy numbers were made, the first on the 30 May 2002 when aphid populations had increased to significant levels and then twice more at approximately fortnightly intervals on 11 and 24 June, by which date very few live aphids remained. Assessments were done mainly on Ben Gairn as it had been found to be the most susceptible to aphids. However, for the second assessment all three-blackcurrant varieties were assessed so that a comparison between varieties could be made. For each assessment, the numbers of aphid colonies and predators were determined in a random sample of four shoot tips from each of the 12 bushes in each plot. A single leaf was also sampled from each bush and the total numbers of each species of aphid, the number of aphid mummies parasitised by parasitoids or by entomopathogens and the numbers of predators were recorded. Predator populations were also assessed by beat sampling along one side of one row (6 bushes). The numbers and species of predators collected were recorded. Aphid numbers were much greater than in the previous 2 years. The weather was unusually wet with substantial rainfall on many of the days in May and early June. The blackcurrant aphid was the most numerous and present in large numbers on every bush with practically 100% of shoots infested with an average of 5 aphids per leaf on the 2x sulphur, 3x fenpropathrin and untreated controls at the first assessment on 30 May 2002. Currant sowthistle aphid was present in approximately 40% of shoots with an average of 1-2 aphids per leaf on the same plots. The 2x sulphur + chlorpyrifos and 2x sulphur + pirimicarb treatments reduced aphid numbers but were not 100% effective. Aphid numbers had declined considerably by the second assessment and were near zero by the time of the third assessment on 24 June 2002. However, the aphid infestations had caused substantial damage to the blackcurrant bushes, especially to the untreated control plots and to the plots that had received the 2x sulphur and the 3x fenpropathrin treatments. The heavy blackcurrant aphid infestations caused the bushes to be covered with honeydew and cast skins and the currant sowthistle aphid distorted the growing points of a high proportion of shoots. As in 2001, ladybird adults and larvae (especially *Coccinella 7-punctata*) were the most abundant predators. They were significantly more numerous on the 2x sulphur and untreated control plots and conspicuously absent from the 3x fenpropathrin treated plots even though aphids were very abundant on the latter. However, high proportions of aphids were parasitised by parasitoids and by the fungal entomopathogen *Entomophthora planchoniana* (Figure 1). These latter two natural enemies were largely responsible for the aphid population collapse.

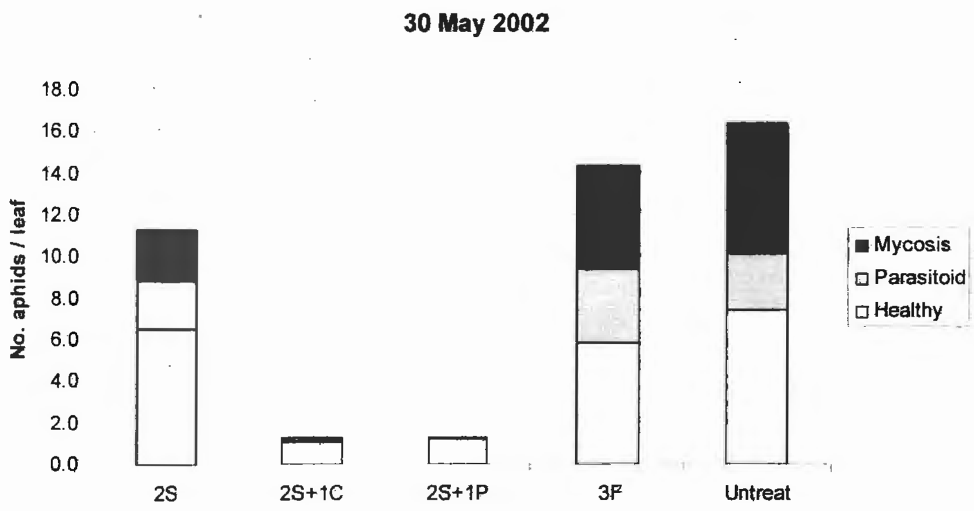

Figure 1. Mean numbers of healthy aphids and of mummies due to parasitoids or fungal mycosis per leaf on plots subject to different pesticide management in 2002. S = sulphur, C = chlorpyrifos, P = pirimicarb, F = fenpropathrin,
Untreat = untreated. Values are means of five replicates.

Blackcurrant aphid and currant sowthistle aphid were serious pests in two of the three years of the project. The blackcurrant aphid was especially numerous. Of the three varieties planted, Ben Gairn was noticeably more susceptible to aphids. The pre-blossom sprays of chlorpyrifos or pirimicarb gave well, though not 100%, control of the aphids. The 3x sprays of fenpropathrin gave little or no control of the aphids. Coccinellid adults and larvae were the most abundant predators. Numbers of coccinellids were greatly reduced by the 3x fenpropathrin treatment. Low numbers on the chlorpyrifos and pirimicarb treated plots may have been due to the smaller numbers of aphids present. A wide range of other predators occurred rather erratically and in small numbers. In 2002, parasitoids and the entomopathogenic fungus *Entomophthora planchoniana* parasitised a high proportion of the aphids and caused the high populations of aphids to crash rather rapidly, but not before the aphids had caused substantial plant damage. The wet humid but mild weather in May and June clearly favoured the *Entomophthora*.

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Potential of ground covers for manipulating pest, predator, and pollinator populations in highbush blueberry

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Abstract: Highbush blueberries are grown on approx. 18,000 hectares in Michigan, USA, making them the most significant of the soft fruit crops in this state. In many farms in this region, the ground between the rows of bushes is planted with perennial ryegrass or autumn-sown annual ryegrass to help maintain soil structure. Unfortunately, grasses are an ideal oviposition site for Japanese beetle, a primary insect pest of blueberries in this region, and grass roots are an optimal food source for their larvae. Recent research in our laboratory has shown that clean cultivation to remove grass from between rows reduces oviposition and survival of this pest within fields. However, this practice has negative environmental and horticultural implications, and ground covers may be an alternative approach to maintain soil structure and reduce colonization of fields by Japanese beetle, while providing resources to conserve beneficial insects. We have recently evaluated acid soil-tolerant ground covers in Michigan blueberries, and report our findings here. Reduced density of Japanese beetle larvae was observed under one potential ground cover, and ground beetle activity/density and pollinator abundance also responded to ground covers. The multifunctional role cover crops may play in integrated blueberry production is discussed.

Key words: Japanese beetle, Carabidae, pollination, blueberry, cover crop.

Introduction

Highbush blueberries, *Vaccinium corymbosum* L., are the most significant soft fruit crop grown in Michigan, USA, producing approx. 32 million kilograms of fruit annually on 18,000 hectares. Harvested fruit is primarily for the processing market, though about 25% of the fruit is sold fresh. In the past 10-20 years growers have increasingly monitored fields for insect pests and have applied insecticides only when needed, to protect the fruit against key insect pests such as the cranberry fruitworm, *Acrobasis vaccinii*, and the blueberry maggot, *Rhagoletis vaccinii*. In recent years, the Japanese beetle, *Popillia japonica* (Coleoptera: Scarabaeidae) has invaded the primary blueberry production regions of Michigan (Potter & Held, 2002), where suitable habitats for all life-stages of this pest occur, and where the level of biological control is extremely low (Cappaert & Smitley 2002). Japanese beetle has now become the key insect pest in Michigan blueberry because harvest of blueberries occurs in this region from July to September, the period during which adult beetles may be present on the bushes within infested fields. This situation causes a risk of contamination of the fruit, particularly in machine-harvested fields. The zero tolerance by fruit buyers for contamination of fruit with beetles has prompted many growers to adopt aggressive programs to control *P. japonica*, including applications of organophosphate and carbamate insecticides in the period just prior to harvest. There is a significant need to develop additional, non-chemical, approaches to management of Japanese beetle and other key pests, to help provide a more sustainable system of blueberry production.

We continue to evaluate reduced-risk chemical controls for adult and larval stages of *P. japonica* in highbush blueberry (e.g. Isaacs et al. in press), but long-term sustainable approaches to population suppression are seen as critical for the ability of growers of blueberry and other crops to reduce pressure from this pest. One such approach is the manipulation of the blueberry field and surrounding habitat to make it less suitable for pest development. Such cultural controls have been shown to reduce pest damage in other systems (Landis et al. 2000, Ostman et al. 2001).

Recent comparisons of blueberry fields in Michigan with different types of row middles (the area between rows of bushes) found that cultivated fields had nearly 80% lower density of *P. japonica* larvae in the row middles than those with permanent sod (Z. Szendrei, unpublished data). While this finding suggests that row-middle cultivation may be an effective control method for larval Japanese beetle in blueberry fields, there are horticultural considerations preventing widespread adoption of this practice. Many fields have relatively wet soils where bare ground could create conditions difficult to drive machinery through for spray applications and harvesting. Fields without ground cover may become dusty during mechanical harvesting and can be difficult for hand-harvesters to walk through.

The low soil pH (4.5-6) required for production of blueberry (Pritts & Hancock 1992) is a major limitation to the selection of potential ground covers for testing in this crop. Two acid-tolerant ground covers were tested in this study, to determine the potential of ground covers for reducing the suitability of blueberry fields for this insect. An alsike clover, *Trifolium hybridum* L., buckwheat, *Fagopyrum esculentum* Moench, and perennial ryegrass (*Lolium perennae*) were tested because of their suitability as ground covers in other fruit systems (Bugg & Waddington 1994) and because of their tolerance for acid soils (Bowman et al. 1998).

Integration of ground covers into blueberry fields for direct control of pest insects may have additional benefits that can increase their utility. These benefits may include enhancement of the habitat that is suitable for natural enemy development and movement within the blueberry field. Recently, we have demonstrated increased predation of fly pupariae in small plots in which the abundance of ground beetles (Carabidae) was enhanced (O'Neal et al. in prep.). Thus, blueberry field habitats can be made more suitable for ground beetles while also being suppressive for *P. japonica* oviposition. Additional benefits may include sustainable pest management of other key blueberry pests that overwinter on the soil, such as the blueberry maggot or the cranberry fruitworm.

Highbush blueberry is highly dependent on insect pollination for maximizing yields (McGregor 1976, Delaplane & Mayer 2000). Although growers typically rent hives of European honeybees to achieve pollination, there is a suite of native bees in eastern North America that are better adapted to pollination of *Vaccinium* flowers than *A. mellifera* (Batra 1997, McKenzie 1997, Javorek et al. 2002). Native bees may require nectar and pollen resources after blueberry bloom has finished, in the middle and end of the growing season. Ground covers that bloom after blueberry could enhance populations of native pollinators and thus increase the potential for a diverse suite of pollinators to provide sustainable yields of this crop.

In this study, the potential of ground covers to provide a multi-functional role for insect manipulation in blueberry fields was investigated. The response of *P. japonica*, ground beetles, and pollinators to perennial ryegrass, buckwheat, and alsike clover was investigated in a blueberry planting.

Material and methods

Experimental Design

This research was conducted in a planting of highbush blueberry, *Vaccinium corymbosum*, var. Bluecrop at the Trevor Nichols Research Complex, Fennville, Michigan, USA. The blueberry plants were four years old, on a 3.65 x 1.2 m planting spacing, with 12 bushes in each row over an area of 0.41 ha. Plots were cultivated in the spring of 2003 and the area between the rows on either side of a 12 bush length of row was planted with seeds to create blueberry bushes bordered by perennial ryegrass (*L. perennae*), white alsike clover (*T. hybridum*), buckwheat (*F. esculatum*), or bare ground (no seeds). Treatments were arranged within the planting in a randomized complete block design, with four replicates. Roundup (glyphosate) was sprayed with a small hand sprayer when needed to maintain these plots free of vegetation.

Japanese beetle

The number of adult Japanese beetles was counted on each plot weekly from July 15 to August 26 2003 by making two minute visual surveys in each plot. In October 2003, 6 soil cores were taken from each plot, using a 10 cm diameter golf course cup cutter (area = 95 cm^2) (Parmenter & Andre Inc., Grand Rapids, MI). Collected larvae were identified to species using the rastral pattern as described by Vittum et al. (1990). Data were not normally distributed and so the effect of ground covers on beetle numbers and the density of larvae was determined using the Kruskal-Wallis test.

Carabid beetles

Pitfall traps comprised of an inner and an outer plastic cup, each 11.8 cm in diameter and 14.6 cm deep. Holes were punched in the bottom of the outer cup to allow water drainage. In each plot, 2 pitfall traps were placed under two blueberry bushes in each plot on 12 August 2003. Traps were emptied 24 h after placement and then on two more consecutive days. Collected beetles were counted and identified to species when possible. The effect of ground cover on the total number of carabid beetles found in pitfall traps and the three most common species was determined by ANOVA. When a significant effect of ground cover occurred, mean comparisons (LSD) were conducted using the least-square means calculated from the total number of beetles recovered in the pitfall traps during the 3 days.

Pollinating insects

The number of pollinating insects was sampled in each plot on July 15, 2003. Three one-minute observations were conducted per plot between 2 and 4 pm, and the number of insects in each of the major pollinator groups (honeybees, bumblebees, andrenids, halictids, and syrphids) was recorded. Four replicate plots of each cover crop type were sampled for pollinating insects. The number of pollinators was compared among cover crop treatments using ANOVA followed by means comparison using the Protected Least Square Differences test.

Results

Japanese beetle

The abundance of Japanese beetle adults was significantly affected by the different ground covers (Table 1). Compared to the standard ground cover of ryegrass, clover had no effect on beetle abundance. However, the other two ground covers caused a significant change in adult beetle behavior compared to the ryegrass: beetles were rarely found on bare ground and they were almost eight times more common on buckwheat.

There was a different pattern in abundance of larval *P. japonica* compared to adults of this species. While bare ground remained the least colonized treatment, clover and ryegrass had the greatest density, while larvae were reduced by almost three quarters in the buckwheat plots compared to ryegrass (Table 1).

Table 1. Average abundance (± S.E.) of adults and larvae of Japanese beetle in highbush blueberry row middle plots planted with four different ground covers, 2003. Values within columns followed by the same letter are not significantly different, $P = 0.05$.

Ground cover	Adults/plot	Larvae/sample
Bare ground	0.1 ± 0.1 c	0.1 ± 0.1 c
Buckwheat	7.6 ± 2.9 a	0.7 ± 0.2 b
Clover	1.9 ± 0.3 b	2.1 ± 0.2 a
Ryegrass	1.0 ± 0.4 bc	2.7 ± 0.2 a

Carabids

A total of 312 carabids were collected, comprised of thirteen species (Table 2) during August 2003. Of these, *Harpalus pensylvanicus* (DeG.) represented over 50% of the species collected.

Table 2. List of carabid beetle species collected within plots planted with different cover crops during 12-15 August 2003, with the percentage of the total captures made up by each species.

Species	Percentage
Harpalus pensylvanicus (DeG.)	58.0
Chlaenius tricolor Dej.	19.6
Cratacanthus dubius (Beauv.)	10.3
Amara cupreolata Putz.	2.9
Harpalus indianus Csiki	1.6
Harpalus affinis (Schr.)	1.6
Agonum octopunctatum (Say)	0.6
Agonum cupripenne (F.)	0.6
Stenolophus lineola (F.)	0.6
Diplocheila obtusa (LeC.)	0.6
Colliuris pensylvanica (L.)	0.3
Harpalus puncticeps (Steph.)	0.3
Anisodactylus rusticus (Say)	0.3
Unidentified	2.6

Ground cover significantly affected the activity-density of all carabids in general ($F = 3.08$, df = 3, 47, $P = 0.04$) and *H. pensylvanicus* specifically ($F = 3.29$, df = 3, 47, $P = 0.03$). There was no significant effect of ground cover on the activity-density of *Chlaenius tricolor* or *Crataocanthus dubius*, the second and third most common carabid collected (Table 2; analysis not shown). When *H. pensylvanicus* was subtracted from the total of beetles collected within each pitfall trap, ground cover was not observed to affect the activity-density of the other species (Fig. 1). For all carabids and for *H. pensylvanicus*, the lowest activity-density was in pitfall traps under blueberry bushes surrounded by bareground (Fig. 1), with the highest level of activity-density observed in traps surrounded by clover or by ryegrass.

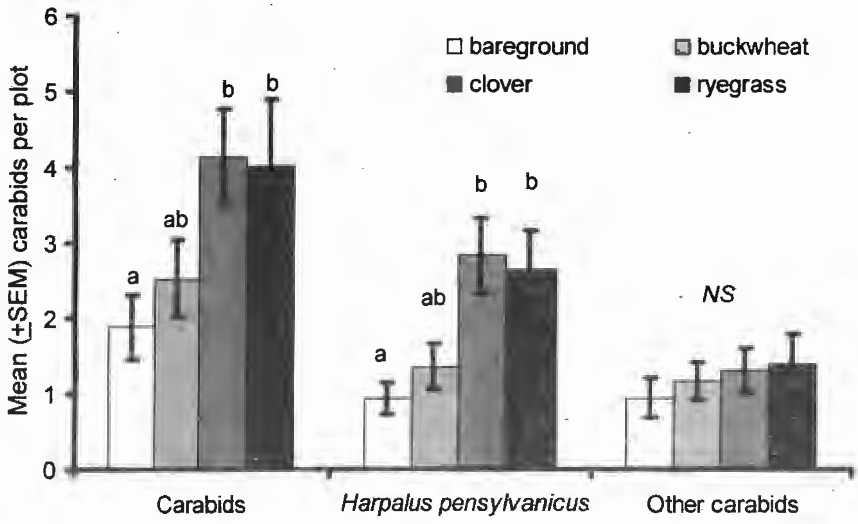

Figure 1. Activity-density of carabid beetles under blueberry bushes in response to type of ground cover planted within the row middles. Columns labeled with the same letter were not significantly different (LSD, $P = 0.05$). The first set of four columns represents all carabids collected, the second set is only *H. pensylvanicus*, and the final set is all other carabids.

Pollinators

Ryegrass plots were devoid of pollinators, except for a few syrphids that were flying near to flowering weeds (Table 3). Bumblebees were not found in any of the observed plots on the sampling date. Honeybee and andrenid bees were observed most often on clover, which was in partial flower at the time of the samples. In contrast, halictid and syrphid insects were most abundant on buckwheat.

Table 3. Average number of pollinators (± S.E.) observed on different ground covers planted between rows of highbush blueberry at the Trevor Nichols Research Complex, Michigan. Values in a column followed by the same letter are not significantly different, $P = 0.05$ (Fishers PLSD method).

Ground cover	Honeybee	Bumble bee	Andrenid	Halictid	Syrphid
Buckwheat	1.1 ± 0.5 b	0.0 ± 0.0	0.2 ± 0.1 b	0.5 ± 0.2 a	4.5 ± 0.6 a
Clover	4.0 ± 0.5 a	0.0 ± 0.0	1.5 ± 0.4 a	0.0 ± 0.0 b	2.8 ± 0.5 b
Ryegrass	0.0 ± 0.0 c	0.0 ± 0.0	0.0 ± 0.0 b	0.0 ± 0.0 b	0.3 ± 0.3 c

Discussion

This study demonstrates that manipulation of ground covers within highbush blueberry fields can affect the abundance of pests, beneficial insects, and pollinators. Japanese beetle adults were found over bare ground in only very low abundance, and this translated into very low density of grubs. This helps to explain the effectiveness of clean cultivation as a control strategy for Japanese beetle. Conversely, adult beetles were most abundant on buckwheat, and these plots had low abundance of grubs. This shows that cover crops can be an effective tool for disruption of egglaying by this key pest, although optimal deployment strategies remain to be determined. Buckwheat is promising for grub suppression in the acidic soils of blueberry but since attraction of adults to plots would be undesirable, deployment as a trap crop in field perimeters may be preferred to enable targeted chemical control of adults and suppression of grubs in this area. Recent sampling across blueberry fields has shown that the density of Japanese beetle grubs is greatest in field perimeters (Z. Szendrei, unpublished data), and so seeding cover crops in problem areas may be a more acceptable strategy for deployment.

The weed and invertebrate predatory feeding strategies of carabids coupled with augmentation of their populations through habitat manipulation make them attractive candidates for biological control in agroecosystems (Landis et al. 2000). At our study site, *Harpalus pensylvanicus* was the dominant species during August (Table 2) and had the greatest response to ground cover (Fig. 1), with increased activity/density in blueberry rows surrounded by ryegrass and clover. *Harpalus pensylvanicus* has been reported as a predator of insects (Riddick & Mills 1994) and weed seeds (White 1999), and is active as an adult from June to October (Kirk 1973). The augmentation of this species using cover crops may in turn reduce levels of blueberry pests that overwinter on the soil surface beneath blueberry bushes, such as blueberry maggot (*Rhagoletis mendax*) and cranberry fruitworm (*Acrobasis vaccinii*). Carabid larvae are also predaceous and in laboratory assays, *H. pensylvanicus* larvae were observed to feed on average of one Japanese beetle grub every two days (Hallock 1929). Further studies are needed to demonstrate the impact of both adult and larval carabid predation on pest abundance in highbush blueberry fields.

Bee abundance was affected by the presence of different ground covers (Table 3). A diverse complex of bees were detected in these samples, including honeybees (*Apis mellifera*), Halictidae spp., and Andrenidae spp. Native bees have been shown to have a high degree of pollination efficiency for blueberry flowers (Javorek et al. 2002). Bees were not captured on any of the plots planted with ryegrass, indicating that the typical ground covers adopted in blueberry have low suitability for native pollinators after blueberry bloom. Although they have been reported as highly efficient pollinators of blueberry, no *Bombus* species were detected in any of these samples. Further, more extensive sampling is required to determine the phenology and identification of the different pollinators found in blueberry, including bumblebees.

Deployment of flowering cover crops may require a specific spatial arrangement to maximize their benefit to pollinators and natural enemies while also reducing the risk to these insects. Flowering cover crops inside fields may increase exposure of pollinators to insecticides, and so if pollinators and natural enemies are sufficiently mobile, deploying cover crops at field perimeters could provide the required resources while minimizing exposure.

By changing the type of ground cover planted in blueberry fields, growers may be able to achieve multiple functions that will help them achieve pest management goals. Depending on the ground cover used, these functions may be suppressive to pests and beneficial for pollinators. However, it is clear from this study that further research is required to determine how to best integrate ground covers into blueberry production as an alternative to bare ground or grass. Future studies will need to take account of whether these ground covers are compatible with blueberry crop development, and if addition of a ground cover is best adopted as an annual planting that is cultivated into the soil, or as a perennial species.

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Vegetable biocidal pellets for biofumigation and fertilisation

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Abstract: The use of chemical compounds of vegetable origin in plant defence with a high biological activity shows several environmental (renewability, biodegradability, low toxicity) and agro-technical advantages of great interest both for conventional and organic agriculture. In particular, in these last years, some interesting perspectives have been arisen by the enzymatic hydrolysis glucosinolates (GLs) derived products, which many authors have pointed out as a typical defensive system of *Brassicaceae* and some other minor plant families. The high biocidal activity of these compounds and their volatility, in fact, could provide a soft and practical alternative to conventional chemical fumigants used at present for controlling some soil borne pathogen and pests, avoiding a so strong environmental impact. The presence in all Brassicaceae plant organs of a high GLs amount together with a sufficient myrosinase (MYR) activity for catalyzing the GLs hydrolysis, has suggested the practical possibility of amending soil with these biocidal compounds by their cultivation and green manure. The application of this technique, even at full field level, is easy and practical. In addition, it positively affects the following crop yield if it is compared with a not treated soil, but also with a conventional green manure. To overcome the typical green manure disadvantages, and to give a new option to the farmers based on the use of chemicals of vegetable origin in plant defence, recently we explored the possibility to produce some bioactive defatted meal mixtures. These formulations contain a high level of GLs-MYR system able to produce a series of bioactive chemicals in short time, as a consequence of the GLs catalyzed hydrolysis. In this way, formulations will be able to produce biocidal compounds only after water addition in soil. The production of these new organic formulations, their chemical composition (nitrogen, glucosinolate, myrosinase and oil content), their pelletisation and a preliminary evaluation of their effect on some widespread soil borne pathogen and pests *viz*. nematodes and wireworms will be reported and discussed. If these innovating compounds will confirm even at full field level their interesting biocidal activity, they could represent a real innovation not only for organic farming, but even in conventional agriculture. Thus these formulations appears to be a potential environmentally friend alternative to methyl bromide, opening interesting perspectives even on strawberry and other soft fruits.

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Application techniques of phytosanitary products adapted to small fruit crops

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Abstract: Production of high quality berries while respecting the environment requires targeted application of phytosanitary products. To do this, the product quantities and spray volumes must be adapted to the growing crop, and losses through drift and run-off must be limited.

Key words: Application techniques, pests, diseases, small fruit crops

Introduction

The production of berries with high quality implies, among other things, good management of disease and pest control. On the majority of farms in Switzerland, indirect methods to control pests and diseases (using resistant cultivars, healthy stocks, best choice of production systems) are not sufficient for reaching this goal, and the use of phytosanitary products is necessary (Mariéthoz et al., 2002; Ançay et al., 2003). In view of this, the role of phytosanitary product application becomes a determining factor in quality production.

Products must be applied to ensure optimum protection of the foliage and fruits while limiting losses caused by drift and run-off. Only perfectly adjusted sprayers which are adapted to the crop are capable of distributing the active ingredients evenly at the recommended doses. It is essential that application techniques of phytosanitary products remain under constant revision as new knowledge becomes available. At the same time, guiding rules for use of the products must be rigorously respected.

In collaboration with various cantonal stations, the Swiss Federal Stations of Wädenswil (Agroscope FAW Wädenswil) and Changins (Agroscope RAC Changins, Centre les Fougères) have developed a concept of spraying adapted to small fruit crops. This concept forms the basis for targeted, rational and economical use of phytosanitary products.

Product dose and quantity of water adapted to the volume of berry crops

In berry crops, the leaf area to be protected increases greatly between the time of the start of plant growth and harvesting. A simultaneous increase in plant volume of the crop can be noted (Figure 1).

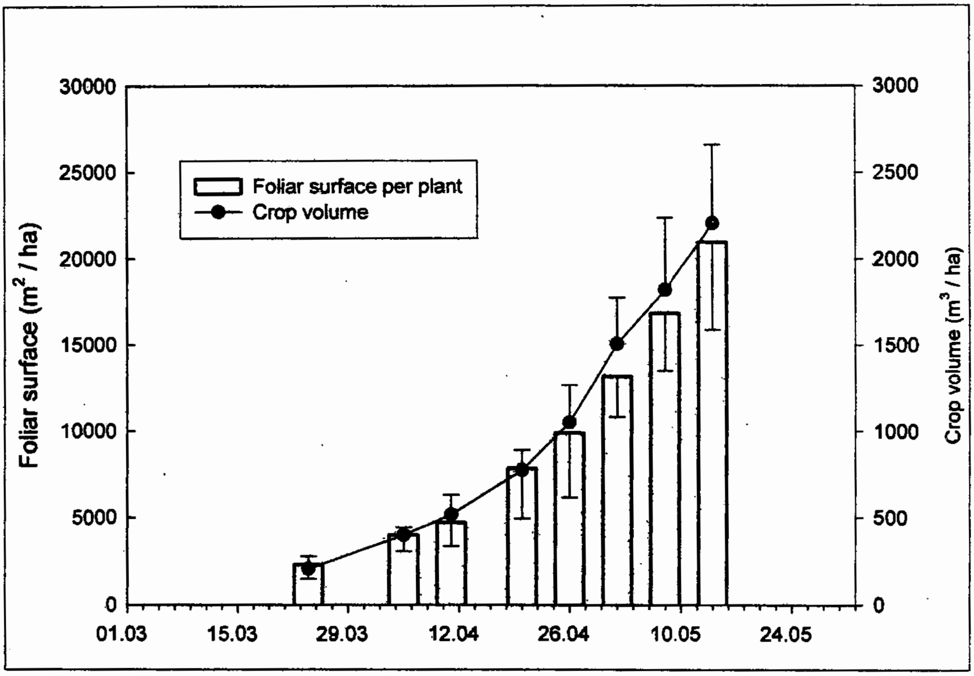

Figure 1. Development of leaf area and plant volume for a crop of strawberries.

Using traditional spraying techniques, the product quantity and spray volume used would remain constant throughout the growing season. However, the new spraying concept which has been elaborated for use on small fruits is based on the Tree Row Volume (TRV) concept (Viret et al., 1999). The TRV concept was developed in order to optimise product and water quantities in relation to crop development of fruit trees and has already been tested in arboriculture. This concept allows the dosage of phytosanitary products and the quantity of water applied per hectare to be adapted as berry crop volumes evolve, with lower water volume and amount of pesticide applied early in the growth cycle, rising during the season as the stage of plant development of the crop progresses.

This concept has been developed and tested over several years on berry crops using air propelled sprayers (turbo-diffusers), as well as traditional sprayers without air assistance in strawberry crops. The result of this study has been the establishment of reference tables for the different species of small fruits which permit a rapid determination of water volume and amounts of product to be applied per hectare according to plant stage and crop density. As an example, the volume of an adapted application of products on strawberry crops is shown (Figure 2).

Producers who have applied this new concept over the last few years reported that the experience had been positive giving very good results. In many cases, the amount of product used was reduced by up to 50%.

	First new leaves	Apparition of inflorescences	Beginning of flowering	Full flowering to beginning of fruit ripening
Phenological stages				

Plant height	5 - 15cm	16 - 29cm	30 - 39cm	40 - 50cm
Planting density	Spray volume (litres/ha) (with constant concentration of pesticide)			
3 plants or less per m²	250 ± 20 %	400 ± 20 %	600 ± 15%	900 ± 10%
4 plants per m²	250 ± 20 %	450 ± 20 %	700 ± 15%	1000 ± 10%
5 plants or more per m²	300 ± 20 %	500 ± 20 %	800 ± 15%	1100 ± 15%

Increase or decrease spray volume according to plant vigour

Figure 2. Reference table for strawberries for a rapid determination of spray volume and amount of product to be applied per hectare according to plant stage and crop density.

Steps to be followed by farmers

The spray volume to be applied per hectare according to the main stages of plant development of strawberries are given in Figure 2. The volume increases as the crop develops but the concentration of product in the spray volume remains identical. In this way, less pesticide is required early in the season, and the increase in spray volume guarantees that the quantity of product being used is sufficient to provide good coverage of crops while minimising losses to the environment. For summer raspberries and blackberries, autumn raspberries, redcurrants, gooseberries and blueberries, such data also exist and are available from the authors on request.

Using the concept described in the present study, the quantity of product to be applied per hectare is reduced early in the season, and increases as the growing season advances. Product rates are always given as a single concentration. Nevertheless, in case of need, for example, when using a turbodiffuser, experiments showed that concentrations of most homologated products can be doubled. In such cases, it is advised not to mix products. Moreover, the sensitivity of berry crops to products differs depending on the system of farming (under protective cover or in open fields), on weather conditions or the time of day. Therefore, before working with concentrated products, it is advisable to seek the manufacturer's approval.

Conclusions

- The concept of spraying phytosanitary products with adaptation of spraying volumes and product quantities as plant growth develops is suitable to all species of small fruits. It is flexible and applicable to different farming systems.
- An appropriate adjustment of the vaporizer is vital to the realisation of this concept.
- Compared with traditional systems, adapting the spray volume (with a fixed product concentration) to plant development allows considerable economies in product usage to be made. At the same time, residues are reduced as well on the harvested fruits as on the non-harvested plant parts and the environment.

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Cultural control methods against *Phytophthora* root rot of red raspberry

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Abstract: The combination of several cultural control methods to control red raspberry root rot caused by *Phytophthora fragariae* var. *rubi* was tested in a field experiment. The treatments were the use of hill planting, compost amendment and plastic mulch in different combinations studied in a naturally infected soil. In all three years after planting, cane survival was significantly increased for both cultivars tested, Zeva 2 and Tulameen, by planting on hills of a soil-compost mixture. Significantly higher yield than the control treatment were obtained when planted on hills in combination with compost and/or plastic mulch. In contrast, planting on hill alone did not increase yield significantly.

Key words: compost, hill planting, plastic mulch

Introduction

Root rot caused by *Phytophthora fragariae* var. *rubi* is a major soil borne disease of red raspberries (*Rubus idaeus*) (Ellis et al., 1997). In cases of high disease pressure and susceptible cultivars, complete loss of the crop can occur within a few years. Even at a lesser degree of disease economic losses are important and may cause the abandonment of this high-value, but also very labour-intensive crop.

Among the methods of control of *Phytophthora* root rot are the use of resistant cultivars and treatment with fungicides before or after planting. However, the utilisation of cultivars can be limited by other traits such as quality, yield or resistances to pests or other diseases than root rot. Limitations to the use of fungicides are high costs, limited efficacy in the soil and resistance of the pathogen. Therefore, the application of cultural control methods are gaining interest, especially in the context of the elaboraton of an integrated control strategy.

Cultural control methods can act upon two factors in the *Phytophthora* root rot – red raspberry complex. Hill planting to improve the drainage of the soil and decrease the risk of infection by zoospores which is favored by water logging (Heiberg, 1995; Wilcox et al., 1999). Reduction of water logging is also the goal of covering the soil with impermeable polyethylene plastic mulch. Amendement with compost not only improves soil drainage but can also enrich the soil with a number of fungal and bacterial antagonists (Hoitink et al., 1993). These ones can reduce the population of a number of soil borne pathogens and probably also of soil populations of *P. fragariae* var. *rubi*.

The goal of the study presented was to evaluate the effect over several years of several cultural control methods, on their own or in combination, on the cane survival and yield of two red raspberry cultivars.

Material and methods

Experimental design

In spring 2000, red raspberries were planted in a field experiment at the Domaine de Bruson, an experimental site belonging to Agroscope RAC Changins – Centre des Fougères, situated at 1080 m.a.s.l. near the Grand St-Bernard (Valais, Switzerland). The experimental field was naturally infected with *P. fragariae* var. *rubi*. Each experimental plot consisted of two rows of 5 m length and with 2 m between the rows. One row was planted with the highly susceptible cultivar Zeva 2 and one with the moderately susceptible cultivar Tulameen (Viret et al., 2002). All experimental plots were equipped with a drip irrigation system.

Cultural control methods

Three cultural control methods, singly or in combination, were tested next to the control treatment. The latter consisted of planting raspberries in low beds without compost and plastic mulch. All five treatments included planting on hills of 40 cm high and 60 cm wide (at the base). Amendment of composted vegetatif debris from households and home gardens (municipal compost) was tested alone or mixed with soil. The amount of compost added was 80 l per linear meter which corresponds to 350 m^3 per ha. Finally, hills of soil and soil mixed with compost were covered with black polyethylene plastic mulch as two additional treatments.

Results and discussion

Cane survival

In the three years following planting, the number of living, fruit-bearing canes of both cultivars was significantly higher compared to the control when planted on hills of soil mixed with compost (figure 1). Covering hills of soil with plastic mulch was the second treatment that increased significantly cane survival, especially of the more susceptible cultivar Zeva 2.

Improving soil drainage conditions only by building hills failed to enhance control of root rot. In contrast, when drainage conditions were further improved by mixing the soil with compost, cane survival significantly increased in all three years and for both cultivars. Covering hills with plastic mulch, another way to reduce water logging conditions in the soil, did significantly increase cane survival of the more susceptible cultivar Zeva 2 but, with the exception of the first year, not of Tulameen. This might be explaned by the reluctance of Tulameen to form primocanes. Eventually the plastic mulch, even when it was partially removed during primocane formation, might have hindered the formation of new canes. Consequently, even with a reduced mortality due to *P. fragariae* var. *rubi* the number of living fruit-bearing canes were not significantly higher than in the control.

The effect of compost on its own was less than the mixture of compost with soil. One reason could be the spatial separation of the compost at the upper part and the soil at the lower part of the hill. Therefore, red raspberries roots benefit from the improved drainage conditions and antagonstic effect of compost only as long as they grow in the top of the hill. Once they reach the lower part of the hill they are again exposed to the root rot infective conditions of the soil.

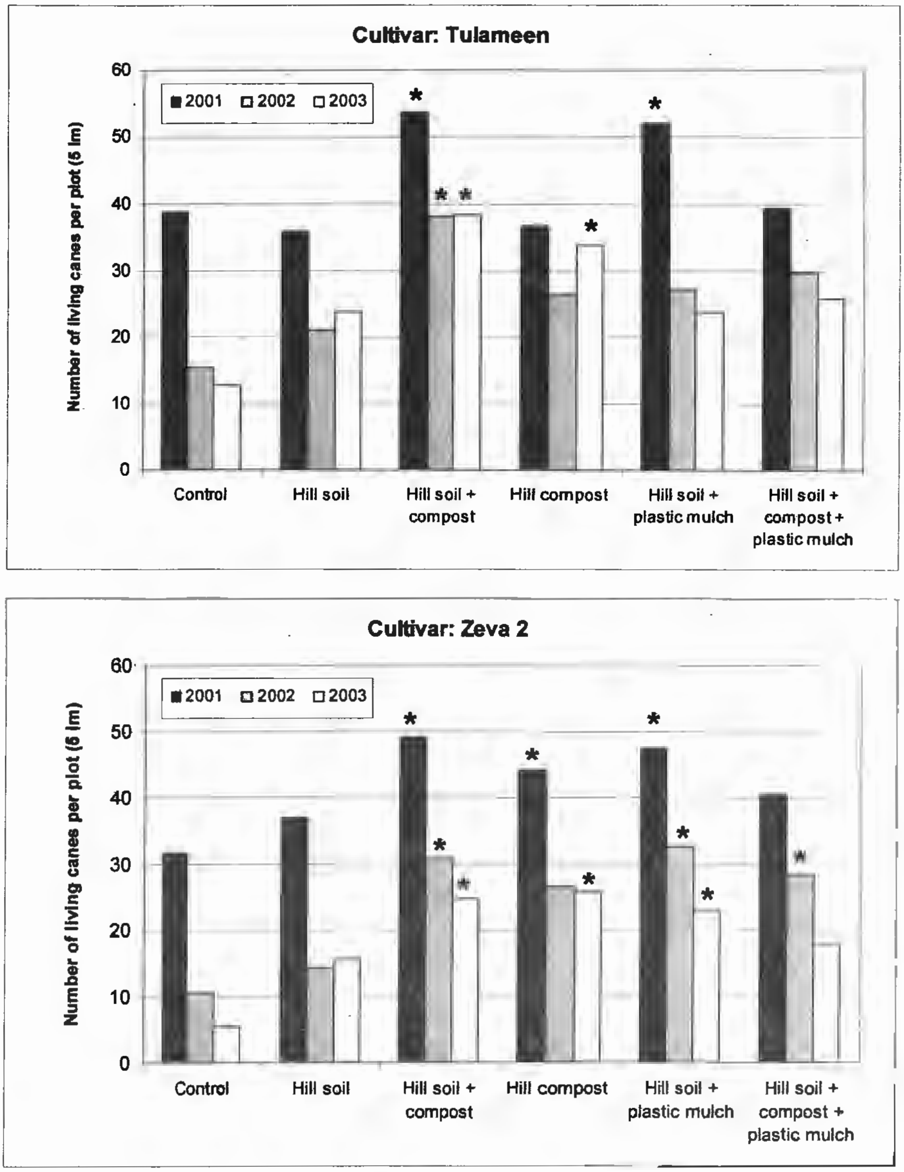

Figure 1. Effect of cultural control methods on the cane survival of cultivar Tulameen and cultivar Zeva 2, expressed as living, fruit bearing canes per experimental plot. Treatments were compared for each year separately. Treatments with an * were significantly ($P \leq 5\%$) different from control (Dunnet test).

Yield

Yield was measured in 2002 and 2003 on the cultivar Tulameen and in 2002 on the cultivar Zeva 2. In 2003, the latter cultivar could not be irrigated sufficiantly because of shortage of irrigation water. Therefore, yield was very irregular and data were omitted for analysis.

In both years and for both cultivars, yield was not significantly higher compared to the control when planted on hills (figure 2). In contrast, the use of compost, alone or mixed with soil, and of plastic mulch increased yield significantly in 2002. However, only Tulameen planted on hills of soil mixed with compost and of soil covered with plastic mulch had a significantly higher yield in 2003.

In similar pattern to the to cane survival, planting on hills alone had no effect and did not improve yield significantly. Higher yield was not always related to higher cane survival, e.g. in six year x treatment combinations with a significant higher yield of the cultivar Tulameen, four did not havesignificantly higher cane survival. Compost was involved in five of these six cases which indicates that a part of the yield effect was probably caused by the increase of nutirents and/or the improvement of the soil structure after adding compost.

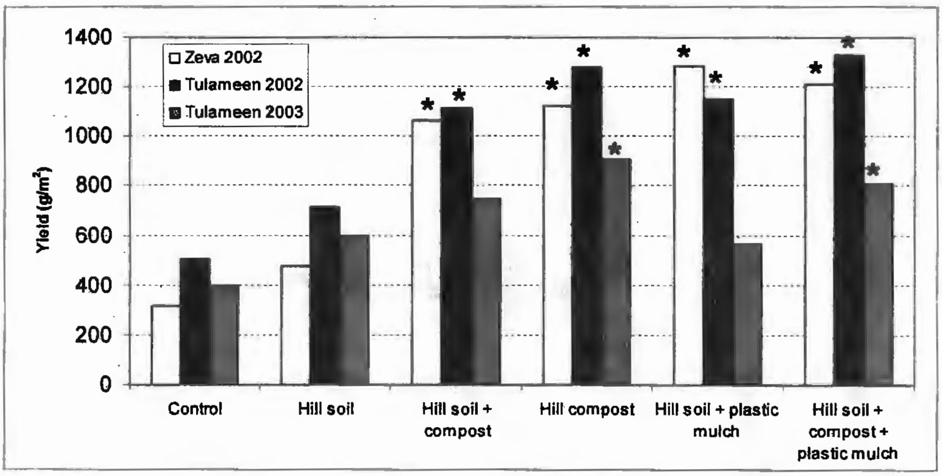

Figure 2. Effect of cultural control methods on the yield of cultivar Tulameen (2002 and 2003) and cultivar Zeva 2 (2002). Treatments were compared for each year and cultivar separately. Treatments with an * were significantly ($P \leq 5\%$) different from control (Dunnet test).

However, the relative yield increase of Tulameen in the treatment containing compost (compared to the control treatment) is lower in 2003 than in 2002. To understand if this is just the year specific effect or a general trend we decided to continue this field trial for at least one more year.

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Problems for strawberry culture in Latvia

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Abstract: Strawberries are one of the most important soft fruits in Latvia. The total area of strawberries is growing, but productivity is decreasing. They are grown mostly under the open-field conditions as a perennial crop. For establishing a new strawberry planting farmers often use planting material from production fields. This leads to an uncontrolled and wide spread of diseases. For a long time pathogens of strawberry were not studied in Latvia. The investigations on occurrence of strawberry diseases were started in 1998 in the Pure Horticultural Research Station. During the investigations the presence of viruses was not detected in plant samples, however most of the plants showed symptoms caused by fungi. The most common strawberry diseases observed were grey mold (*Botryotinia fuckeliana*), leaf scorch (*Diplocarpon earlianum*) and leaf spot (*Mycosphaerella fragariae*). Root and crown rot diseases were present in strawberry plantations. Affected plants had various types of symptoms and fungi belonging to different plant pathogenic genera such as *Fusarium*, *Cylindrocarpon*, *Verticillium*, and *Rhizoctonia* were recovered from diseased tissues. Widely grown cultivars showed different resistance level to diseases.

Key words: strawberry, root diseases, *Botryotinia fuckeliana*, *Diplocarpon earlianum*, *Mycosphaerella fragariae*, viruses

Introduction

Strawberry is the most popular commercial berry crop in Latvia today. It gives a rather high income in comparison to other agricultural crops (Bite et al., 1997). In 1996 there were about 500 ha of strawberries in Latvia and today it is significantly higher, however the yields have a tendency to decrease (Table 1).

Table 1. Strawberry production in Latvia.

Year	Area, ha	Average yield, t ha^{-1}	Total yield
1999	569	3.83	2179
2000	826	5.53	4566
2001	1230	3.15	3875
2002	1135	2.58	2928

Strawberries are mainly grown on flat fields in rows with a density of 30000 - 40000 plants ha^{-1} and usually organic materials are used for mulching. Fields usually last 3 - 4 years. The efficiency of mechanisation and plant protection is low in many farms. For establishing new strawberry plantations, farmers often use runners from production fields. The Regulations for Certification and Control of Planting Material were established only in 2002 and certified planting material is not available yet in the market. All these factors have led to an uncontrolled and wide spread of diseases and insect pests.

The aim of these studies was to determine the occurrence of strawberry diseases in Latvia, and evaluate the susceptibility of widely grown strawberry cultivars to the main diseases.

Materials and methods

The investigations were started in 1998 in different production fields at the Pūre Horticultural Research Station and in 12 farms from different strawberry growing areas in Latvia.

The occurrence and severity of diseases on different cultivars were noted. 20 widely grown cultivars were evaluated. The occurrence of diseases was noted as a percent of damaged plants from total plants evaluated. The severity of gray mold was evaluated as a percentage of damaged fruits from total yield. Severity of leaf diseases was evaluated visually as a percent of damaged leaf surface. To note root and crown diseases, plants showing wilt symptoms were evaluated visually; a 0 - 5 point scale was used, where 0 - plant without any symptoms of wilting and 5 – plant fully wilted. Assessments for diseases were done two times per season: in spring-summer and summer-autumn.

To identify the causal agents of wilting, surface sterilized tissues from strawberry roots and crowns were plated on several agar media for laboratory investigations.

To detect the presence of virus diseases, plants with symptoms similar to those shown by virus diseases were collected from different farms and tested in the Virus Disease Laboratory of the Research Institute of Pomology and Floriculture in Skierniewice, Poland. Virus testing was done using an ELISA test and grafting to indicator plants.

Results and discussion

Occurence of diseases

Observation of the strawberry fields showed that the most common strawberry diseases were grey mold (*Botryotinia fuckeliana*), leaf scorch (*Diplocarpon earlianum*) and leaf spot (*Mycosphaerella fragariae*). Many plants showing wilt and stunt symptoms were also present in strawberry plantations.

Botrytis rot is one of the most important diseases of strawberry, reducing yield and quality pre- and post-harvest in all strawberry production areas (Berrie et al., 2000; Legard et al., 2002). In these studies the amount of rotted berries greatly varied between cultivars and years. In conditions favorable for gray mold development, in some susceptible cultivars it caused yield reductions of almost 40%, while more resistant cultivars had about 10% rotted berries.

Leaf spot diseases were widespread in all plantings, but they did not cause severe damage. White spot was more widespread than leaf scorch.

There have been no investigations of the occurrence of strawberry root and crown diseases in Latvia for many years. The occurrence of wilting and stunted plants in our screening varied from 4 to 100% depending on farm, and 60 % of farms had more than 50% of damaged plants. Diseased plants had different types of symptoms. They were generally stunted in various stages; older leaves were wilted and curled up. Roots of such plants were poorly developed and black. In the 1970's the main causal agent of wilt was noted *Verticillium albo-atrum* (Dūks, 1976). In our studies many isolates of *Verticillium spp.* were also obtained. The majority of fungi recovered from diseased strawberry roots and crowns were *Cylindrocarpon spp.*, *Fusarium spp.*, *Verticillium spp.* and *Rhizoctonia spp. Fusarium,* *Cylindrocarpon*, *Rhizoctonia* and *Phoma* are the most common root pathogens on strawberries in other northern Europe countries like Finland (Parikka & Kukkonen; 2002). However, although the occurrence of wilting and stunted plants was high, the severity was low (0.1 - 1.5 points depending on farm). This could be explained if there were some antagonistic microorganisms present in the soil that restricted development of disease. In spite of low severity, root damage affected the growth, the yield and the over wintering of strawberry plants. Root diseases are difficult to control and there are no chemical fungicides registered for them in Latvia. The spread of these diseases can be reduced effectively in the long term only by using healthy planting material and resistant cultivars, and by establishing strict regulations for nurseries.

During observations of strawberry farms, 35 plant samples with symptoms similar to virus diseases were collected and tested. Results showed that none of plant samples contained antibodies of ArMV, SLRV, RRV and SMYEV viruses. Two graftings on indicator plants showed symptoms similar to Strawberry Crinkle virus, but due to the lack of specific antiserum it was not confirmed by an ELISA test.

Resistance of cultivars

As stated by Cravedi & Jörg (1996) the availability of resistant or less susceptible cultivars would solve many problems with fungal diseases in soft fruit production. Strawberry cultivars which are widely grown in Latvia, according to our investigation results, showed very different resistance levels to common diseases (Table 2).

According to Daugaard (1999), there are marked differences among cultivars in susceptibility to gray mold but no strawberry cultivar is completely resistant to this disease. It was confirmed in these studies too. `Kokinskaya Rannaya`, `Elsanta` and `Bogota` showed the highest resistance to gray mold among the tested cultivars. Rather low susceptibility was shown also by cultivars `Honeoye`, `Dukat`, `Darunok Vchiteliu` and `Tenira`. `Kokinskaya Pozdnaya`, `Pandora`, `Senga Sengana` and `Venta` were the most susceptible.

Higher levels of leaf spot diseases infection were observed in the oldest plantings, and severity was significantly influenced by cultivar and time of observation. Cultivars `Kokinskaya Rannaya` and `Darunok Vchiteliu` showed the highest resistance to white spot disease, but `Bogota`, `Eldorado`, `Induka`, `Jonsok`, `Kokinskaya Pozdnaya`, `Korona`, `Siurpris Olimpiade` and `Senga Sengana` were very susceptible. Cultivars `Bogota`, `Siurpris Olimpiade`, `Senga Sengana` that were very susceptible to white spot, showed high resistance to leaf scorch. In contrast, cultivars `Kokinskaya Rannaya` and `Darunok Vchiteliu` that were resistant to white spot showed high susceptibility to leaf scorch. Low infection by leaf scorch was observed also on cultivars `Dukat`, `Elsanta and `Pandora`.

The occurrence of wilting and stunted plants was significantly dependent on cultivar, growing year and time of observation. The highest amounts of damaged plants were observed on cultivars with the most early ripening time - `Darunok Vchiteliu` and `Kokinskaya Rannaya`, where more than 50% of plants were damaged. High susceptibility to these diseases was noticed also for cultivars `Zefyr` and `Tenira`. The cultivars `Senga Sengana`, `Eldorado` and `Dukat` had the lowest amount of damaged plants .

None of the tested cultivars showed good resistance to a complex of common diseases. Of those tested, higher levels of resistance were shown by `Honeoye`, of the early ripening cultivars, by `Dukat`, `Eldorado` and `Surpris Olimpiade` from cultivars with medium ripening time and by `Bogota` from late ripening time cultivars.

Table 2. Susceptibility of widely grown strawberry cultivars to diseases

Cultivar	Production time	Susceptibility to diseases			
		leaf scorch	white spot	wilt and stunt	grey mold
Darunok Vchiteliu	early	+++	+	+++	+/++
Honeoye	early	+++	++	+/++	+/++
Jonsok	early	+++	+++	++	++
Kokinskaya Rannaya	early	+++	+	+++	+
Zefyr	early	++	++/+++	+++	+/++
Dukat	medium	+	+/++	+	+
Eldorado	medium	++	+++	+	++
Elsanta	medium	+	++	+++	+
Festivalnaya Romashka	medium	+++	+/++	++/+++	++
Induka	medium	++	+++	++	++
Korona	medium	++	+++	++	++
Senga Sengana	medium	+	+++	+	+++
Siurpris Olimpiade	medium	+	+++	++	++
Tenira	medium	++	++	+++	+
Venta	medium	++	+/++	++	+++
Bogota	late	+	+++	++	+
Bounty	late	++	++	++	++/+++
Kokinskaya Pozdnaya	late	++	+++	++	+++
Pandora	late	+	++/+++	+/++	+++
Pegasus	late	++/+++	++/+++	++/+++	++/+++

+++= high; ++= medium; +=low

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Possibilities of integrated grey mould control on strawberry plantations in Poland

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Abstract: Effectiveness of Trichodex 40 WP (*Trichoderma harzianum*) and naturally originated compounds like Messenger (harpin), Biochicol 020 PC (chitosan) and Biosept 33 SL (grapefruit extract) was investigated in control of grey mould, the most destructive strawberry disease in Poland. Also, a new promising fungicide Switch 62.5 WG (cyprodinil+fludioxonil), with some curative activity, was included in postinfection programme. None of the bioproducts used alone gave satisfactory control of the disease. However, Switch 62.5 WG applied only twice during blossom period after occurrence of infection (based on prediction analysis) was very effective. The use of Switch® reduced the numbers of treatment applications by half compared to the preventative standard.Beside the chemical treatments in integrated grey mould control in Poland it is recommended to utilize the agricultural practices and tolerance of some cultivars to disease.

Key words: strawberry, grey mould, integrated plant protection, biological control, chemical control

Introduction

Grey mould (*Botrytis cinerea* Pers.) is the most important strawberry disease in Poland. Its severity is closely correlated to susceptibility of cultivars (Muller, 1965) and weather conditions (Jarvis, 1964). In some years, on many plantations grey mould destroyed more than 50% of Senga Sengana fruits. Until now, disease control based mainly on protective fungicides like procymidone, iprodione, tolylfluanid, pyrimethanil and recently fenhexamid. However, in the recent years some groups of farmers have started to grow strawberries under IPM (Integrated Pest Management) rules. Generally, the first step in developing an IPM program was to look for alternative methods (more resistant cultivars, proper agriculture). Also some researchers work on possibilities to move from preventive, calendar–based fungicide applications to use of pesticides only when there is a risk for significant crop damage (Cooley et al., 1996). Now, integrated grey mould control combines a few alternative methods such as: cultivar selection, agricultural practices and proper use of fungicides.

Cultivar selection

At present, none of registered Polish strawberry cultivars is highly resistant to grey mould. Some commercial cultivars such: Dukat, Honeoye, Karel and Polka are less susceptible than others: Senga Sengana, Elkat, Vega, Marmolada or Pegasus. On plantations of less susceptible cultivars it is possible to reduce fungicide use.

Agricultural practices

Reduction of primary source of infection is achieved by removing old, dead leaves and mummified fruits. Control of weeds improves air circulation and sunlight penetration. It decreases relative humidity and duration of leaf or fruit wetness. Proper nitrogen fertilization does not stimulate excessive foliar growth, which favors the development of grey mould and makes good application of fungicides difficult.

Proper use of fungicides
Rotation of dicarboximides with other groups of chemicals prevents the development of resistant forms of *B. cinerea*. Good technique of fungicide application (appropriate sprayers for each type of strawberry plantations) enhances the quality of grey mould control.

The aim of this presented work was to investigate the efficacy of Trichodex 40 WP (*Trichoderma harzianum*) and naturally originated compounds such Messenger (harpin), Biochikol 020 PC (chitosan) and Biosept 33 SL (grapefruit extract) in control of grey mould and the possibility to use a new fungicide Switch 62.5 WG (cyprodinil+fludioxonil) in the forecasting program for the control of this disease.

Material and methods

The effectiveness of different biological agents and Switch 62.5 WG used in postinfection programme in control of grey mould was tested in field conditions on several strawberry cultivars (Senga Sengana, Elkat, Polka and Elsanta) in Institute of Pomology at Skierniewice. The tested products were applied from beginning of blossom at 5-7 days intervals by a motor knapsack sprayer 'Solo' using 600 l of water per hectare. Switch 62.5 WG treatments were conducted according to the disease warning system, which is based on weather conditions (Bulger et al. 1987).

The evaluation of grey mould occurrence was made twice during the harvest. The affected fruits were counted in the field and in the laboratory after 48 hrs storage at the room temperature. From each plot a sample of 400 ripen fruits was collected.

Results and discussion

The severity of grey mould on strawberry fruits strongly depended on weather conditions, mainly temperature and humidity. In 1999 and 2001 seasons with a lot of rain during the blossom and harvest time the severity of disease was extremely high. In these years, the percent of affected fruits of Senga Sengana was 50 and 70%, respectively. Unfortunately, it was found that none of the biological agents gave satisfactory control of the disease. The efficacy of chitosan, harpin and grapefruit extract was about 20-30% (Table 1 and 2). However, Mazur & Waksmundzka (2001) showed that chitosan water solution had protective effect in control of grey mould and the decay of strawberry fruits was diminished to 15%. Our results suggest that these biological agents should possibly be used earlier in the season, to stimulate plant resistance. Antoniacci et al. (2000) showed that among the natural and microbiological products, only *T. harzianum* provided a certain efficacy against *B. cinerea*. Monchiero et al., 1996, confirmed that *Trichoderma* can be used in the integrated control of grey mould. In our experiments Trichodex 40 WP (*T. harzianum*) gave good control only in mixed programme with dichlofluanide, which was probably the only one responsible for control. Trichodex 40 WP used alone was ineffective and the percentage of affected fruits was the same as on untreated plots (Table 3). Similar results were obtained by Stensvand (1998). In his experiments *Trichoderma* was also ineffective as a biological control agent and he suggested that it may be more effective at higher temperatures.

Switch 62.5 WG applied twice after occurrence of infection was very effective. Effectiveness of postinfection programme was similar to a preventive one, when Switch 62.5 WG was used four times during blossom period (Table 4).

Table 1. Effectiveness of Biosept 33 SL in the control of grey mould on strawberry Cv. Senga Sengana, Miedniewice 2001.

Treatment and dose per ha	Active ingredient	Percent of infected fruits		
		26 June	2 July	Average number of infected fruits
Untreated	-	83.9 b*	87.9 c	85.4 c
Biosept 33 SL 2.0 l	extract of grapefruit	83.3 b	62.3 b	60.3 b
Sumilex 500 SC 1.5 l	procymidone	2.5 a	9.7 a	8.0 a

Means in columns followed by the same letter do not differ at 5% level of significance (Duncan's multiple range t-test)

Table 2. Effectiveness of Biochikol 020 PC and Messenger in the control of grey mould Cv. Senga Sengana, Dąbrowice 2002.

Treatment and dose per ha	Active ingredient	Percent of infected fruits		
		5 June	12 July	Average number of infected fruits
Untreated	-	69.9 b*	43.9 b	56.9 b
Biochikol 020 PC 0.5%	chitosan	54.7 b	41.3 b	48.0 b
Messenger 0.03%	harpin	48.7 b	42.1 b	45.4 b
Switch 62.5WG 1.2 kg	cyprodinil+ fludioxonil	0.7 a	8.1 a	4.8 a

Means in columns followed by the same letter do not differ at 5% level of significance (Duncan's multiple range t-test)

Table 3. Efficacy of Trichodex 25 WP in the control of grey mould on strawberry.

Fungicide and dose/ha	Active ingredient	Percent of infected fruits - 1999	
		Senga Sengana	Elkat
Untreated	-	73.5 c*	88.9 b
Euparen M 50 WP (E) 5 kg -4x	dichlofluanid	5.8 a	20.9 a
Trichodex 25 WP (T) 4 kg -4x	*Trichoderma harzianum*	56.3 b	85.8 b
T+E+T+E	*Trichoderma harzianum* + dichlofluanid	12.6 a	-

Means in columns followed by the same letter do not differ at 5% level of significance (Duncan's multiple range t-test)

Table 4. Postinfection efficacy of Switch 62.5 WG in control of grey mould

Fungicide and dose/ha	Percent of infected fruits		
	2002	2003	
	Senga Sengana	Polka	Elsanta
Untreated	56.9 b*	5.6 c	2.4 b
Switch 62.5 WG –1.2 kg preventively	4.8 a	0.1 a	0.0 a
Switch 62.5 WG - 1.2 kg according to infection	8.6 a	1.3 b	0.4 a
Sumilex 500 SC - 1.5 l preventively	-	1.6 b	0.03 a

Means in columns followed by the same letter do not differ at 5% level of significance (Duncan's multiple range t-test)

Conclusions

It is concluded that biological control of strawberry grey mould in the field is feasible, but effectiveness of different plants extract and various microorganisms has yet to be determined. Switch 62.5 WG can be applied in postinfection programme and the introduction of this fungicide to IPM creates a big chance to obtain effective control of grey mould with a low level of fungicide usage.

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Fungal flora in strawberry plants and relative importance of *Botrytis cinerea*

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Abstract: Fungal flora in strawberry plants (*Fragaria* X *ananassa* Duch.) imported from Italy and cultivated in Switzerland was analyzed. Among the 40 different fungal species isolated from symptomless plants, about half were common fungal strawberry pathogens. The gray mold caused by *Botrytis cinerea* Pers.: Fr. is one of the most destructive pathogen of strawberries worldwide. For this reason molecular markers were developed in order to detect specifically latent *B. cinerea* in apparently healthy tissues. A 0.76 kb DNA fragment specific to *B. cinerea* was identified by RAPD assays and selected to design specific primers for PCR amplifications. Specificity and sensitivity of PCR using the new primers on DNA of different fungal species were evaluated. Detection and quantification of the latent pathogen in infected tissues of strawberry, raspberry and grape was also performed by isolations on selective agar media and by quantitative-competitive PCR (QC-PCR) using the identified molecular marker. The results obtained confirm the specificity and sensitivity of the PCR, as well as the importance of latent *B. cinerea* for the expression of the disease on ripe fruits. The DNA fragment constitue a powerful molecular marker for the detection of latent *B. cinerea* in plant tissues.

Key words: *Botrytis cinerea*, strawberry plants, latency, PCR, QC-PCR.

Introduction

Strawberry (*Fragaria* X *ananassa* Duch.) is the major berry cultivated in Switzerland. Healthy, certified planting material is imported from Italian, French and Hungarian nurseries (Rigotti et al., 2003). Phytosanitary inspections, however, reveal often the presence of latent infections of bacterial or fungal origin, representing potential pathogens. If environmental conditions are suitable for their development, they may cause important diseases and severe economical losses.Beyond powdery mildew caused by *Sphaerotheca aphanis* (Wallr.) U. Braun, gray mold caused by *Botrytis cinerea* Pers.: Fr. (teleomorph *Botryotinia fuckeliana* [Wallr.] U. Braun) is one of the most destructive pathogen of strawberry worldwide. Flowers are the main pathway by which *B. cinerea* infects the fruits (Jarvis & Borecka, 1968), where it remains latent until fruit senescence. By high relative humidity, disease appears at or near harvest time, even during storage. Several cultural practices may reduce the incidence of *Botrytis* fruit rot. Presently, fungicide applications are the main control measure against gray mold.

The aims of this research were to better understand the biology of *B. cinerea* (i.e. ecology, epidemiology and genetics), in order to apply specific fungicides parsimoniously and accurately timed and to avoid the rapid apparition of resistant strains (Leroux et al., 2002).

Material and methods

Fungal flora in apparently healthy strawberry plants
Forty symptomless strawberry plants imported from Italy and grown in Switzerland were analysed for parasitic and saprophytic fungi. The plants were examined by conventional isoltation methods on nutrient media according to Rigotti et al. (2003).

Early detection of* B. cinerea *by PCR
In order to detect latent *B. cinerea* in plant tissues, the rapid and sensitive PCR amplification technique was applied to identify a DNA fragment specific to *B. cinerea*. The polymorphism of the genomic DNA of many fungal isolates was examined by RAPD according to Rigotti et al. (2002).

Quantification of latent* B. cinerea *by isolation and QC-PCR
The evolution and incidence of *B. cinerea* were evaluated on three main host plants: strawberry, raspberry and grape.

The first approach was by conventional isolations methods on specific nutrient media, prepared according to Kerssies (1990) and Bammé & Penaud (2000).

The second approach was the use of quantitative-competitive PCR (QC-PCR), allowing to quantify the DNA of latent *B. cinerea*, by using the identified DNA fragment (Rigotti et al., 2002). In competitive PCR, a known amount of internal standard (also called competitor) is introduced to the PCR reaction together with the target template. This internal standard is a DNA fragment which contain sequences for the same primers used to amplify the target. When the target DNA and the competitor are amplified together, both will compete for the same set of primers. Since the initial amount of the competitor is known, the initial amount of the target DNA can be estimated based on the ratio of the amounts of the two amplified products (McCulloch et al., 1995; Zimmermann & Mannhalter, 1996). The internal standard was prepared according to Rigotti (2003), by amplifying genomic DNA of *Escherichia coli* strains BL21 (DE3) (Amersham Pharmacia Biotech) with the primer sets specific to *B. cinerea* at low stringency (annealing temperature of 37°C). Products of interest were amplified at high stringency (60°C anneal) in the presence of both primers and singly to verify the presence of both primer sites. Fragments were purified from gel agarose according to Qiagen (QIAquick Gel Extraction Kit).

Results and discussion

Fungal flora in apparently healthy strawberry plants
Forty fungal species were isolated from symptomless strawberry plants (Rigotti et al., 2003). In spite of healthy certification, 17 were common strawberry pathogens (Fig. 1), the others being considered as saprophytes. The typical structures of each isolated fungal species were drawn after microscopical observation (Fig. 2 and 3, only pathogens are shown). Among the analyzed plants, 24 were imported frigo plantlets sampled during vernalisation, the others being field-grown plants. More than 20 fungal species were isolated from the plantlets, from which half were potential pathogens of strawberries, such as *Botrytis cinerea, Fusarim* spp., *Pythium* spp., *Hainesia lythri, Rhizoctonia fragariae* and *Zythia fragariae* (teleomorph: *Gnomonia comari*).

The great number of isolated fungi from apparently healthy strawberry plants reveal the importance of latency for fungal diseases transmission. In spite of phytosanitary inspections and healthy certification, many potential pathogens are present in the strawberry plants analyzed, which reveal the hazard of transmission of quarantine organisms (i.e. *Xanthomonas fragariae*, *Colletotrichum acutatum*, *Phytophthora fragariae* var. *fragariae*; EPPO/CABI, 1997).

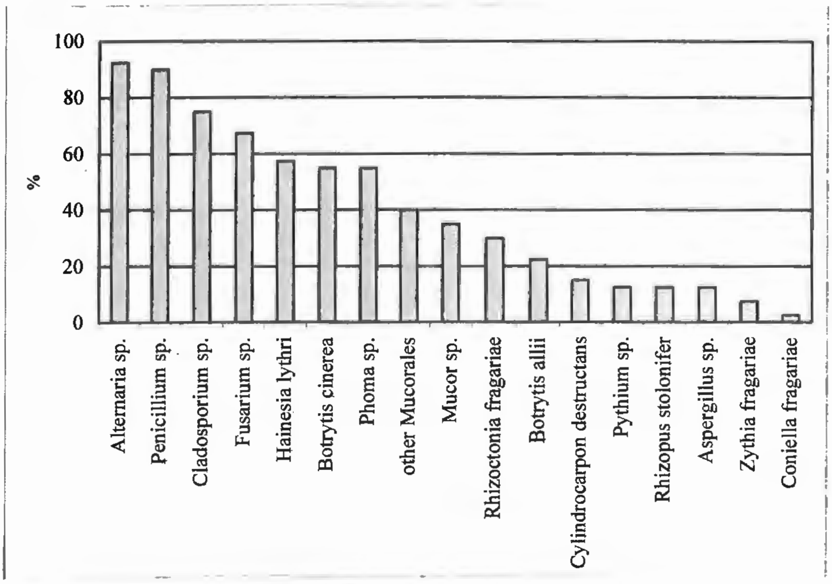


Figure 1. Frequency of common pathogens isolated from symptomless strawberries.

Early detection of* B. cinerea *by PCR

After RAPD assays, a 0.76 kb DNA fragment specific to *B. cinerea*, corresponding to a single copy sequence within the fungal genome, was selected to design specific primers (Rigotti et al., 2002). Specificity and sensitivity of the new primers were confirmed after PCR analyses on genomic DNA of several fungal species. This genetic marker allowed to distinguish two subgroups of *B. cinerea* isolates, as well as the species *B. cinerea* and *B. fabae* (Rigotti, 2003).

Quantification of latent B. cinerea by isolation and QC-PCR

The evolution and incidence of *B. cinerea* were evaluated on strawberry, raspberry and grape by isolation and QC-PCR.

In competitive PCR, standard curve was constructed after co-amplification of known amount of *B. cinerea* genomic DNA and internal standard (Fig. 4a and 4b). This approach enable accurate analysis of multiple samples by comparison with the standard curve (Zachar et al., 1993), in order to quantify the target DNA of latent *B. cinerea*.

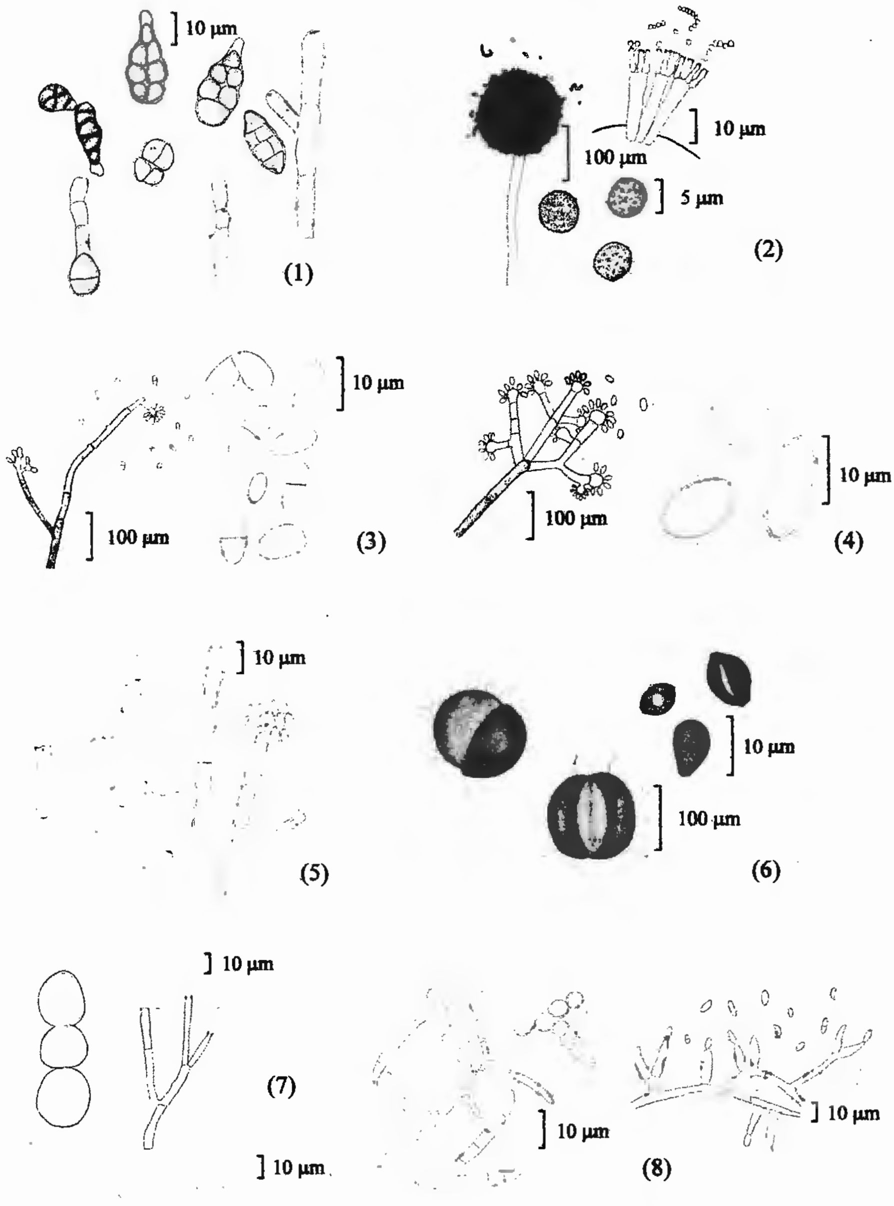


Figure 2. Typical identification structures of fungal pathogens isolated from strawberry plants (drawings S. Rigotti). (1) *Alternaria* sp., conidiophores and conidia; (2) *Aspergillus* sp., conidiophores, phialides and conidia; (3) *Botrytis allii*, conidiophores and conidia; (4) *Botrytis cinerea*, conidiophores and conidia; (5) *Cladosporium* sp., conidiophores and conidia (with scars); (6) *Coniella fragariae*, pycnidia and conidia; (7) *Cylindrocarpon destructans*, conidiophores, phialides, macroconidia, microconidia and chlamydospores (8) *Fusarium oxysporum*, phialides, macroconidia, microconidia and chlamydospores.

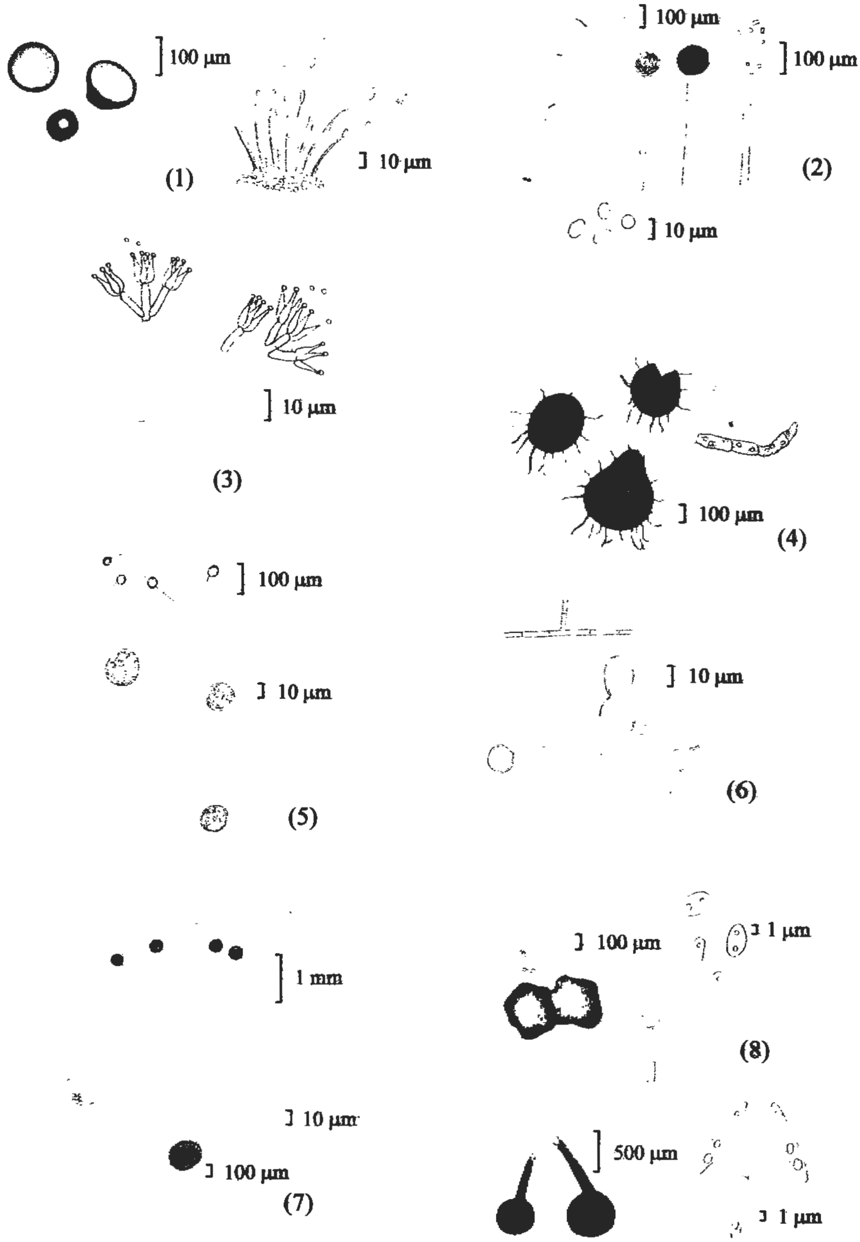
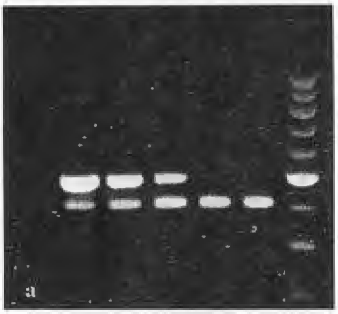


Figure 3. Typical identification structures of fungal pathogens isolated from strawberry plants (drawings S. Rigotti). (1) *Hainesia lythri*, acervuli, phialides and conidia; (2) *Mucor* sp., sporangiophores, sporangia, columella and spores; (3) *Penicillium* sp., conidiophores, phialides and conidia; (4) *Phoma* sp., pycnidia and conidia; (5) *Phytium* sp., antheridium and oogonia; (6) *Rhizoctonia fragariae*, sterile hyphae; (7) *Rhizopus stolonifer*, sporangiophores, sporangia, columella and spores; (8) *Zythia fragariae*, pycnidia and conidia (teleomorph *Gnomonia comari*, perithecia and ascospores).



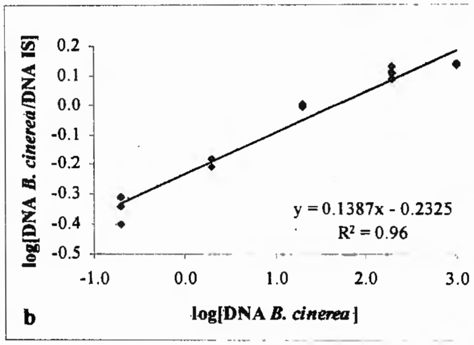


Figure 4.(a) Co-amplification of known amount of *B. cinerea* genomic DNA and constant amount of internal standard; (b) Standard curve constructed from the data obtained by quantitative analyses (ROI's analyses data) of the mean band intensity produced in (a), using 1D Image Analysis Software from Kodak Digital Science (Eastman Kodak Company). The coefficient of determination R^2 and the regression equation were calculated.

The two approaches allowed to confirm the increased susceptibility of the plant at the open-flower stage. Latent *B. cinerea* was mainly present in the receptacle area, and increased during fruit ripening. Moreover, the number of infected fruits increased during ripening, ripe fruits being more susceptible to secondary infections (Rigotti, 2003).

This research allowed to point out the importance of early detection of latent infection caused by *B. cinerea*. Two quantitative approaches were tested, the isolation method being time consuming and tedious. Thanks to the DNA fragment identified, the molecular approach permits to detect and quantify specifically the pathogen in one work day in many tissues samples. The development of the fungus my also rapidly be predicted, to improve control strategies.

Acknowledgements

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Effect of cultural methods on leaf spot (*Mycosphaerella fragariae* Tul.) incidence in strawberries

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Abstract: Leaf spot (*Mycosphaerella fragariae* Tul.) is one of the most common diseases of strawberry. In this study the efficiency of cultural methods like planting density (single or double row system) and removal of dead and leaf spot infested leaves (leaf sanitation) were tested to control leaf spot. In the single row system *M. fragariae* incidence (treatments without leaf sanitation) was lower and yield was higher than in the double row system. In both systems leaf sanitation decreased *M. fragariae* significantly and increased yield.

Key words: *Fragaria x ananassa* Duch. (*Rosaceae*), leaf spot, *Mycosphaerella fragariae* Tul., cultural methods, leaf sanitation, fruit weight, yield.

Introduction

Leaf spot caused by *Mycosphaerella fragariae* Tul. is one of the most common diseases of strawberry. The control of leaf spot mostly relies on the application of protective fungicides (Maas, 1998). In organic agriculture the use of copper products is effective (Vukovits, 1980) but this has a negative influence on soil fertility (Bergmann, 1993). Therefore its use in strawberry crops should also be reduced.

Material and Methods

The field study evaluated the effects of cultural methods on leaf spot incidence and yield. Influence of planting density and removal of dead and leaf spot infested leaves in March before sprouting were the main investigation criteria. The potted green plants cv. Senga Sengana were planted in August 2000 on raised beds. The planting systems are described in Table 1 below.

Table 1. Data of planting systems.

	Width / Height of raised beds	Distance of rows	Distance between each plant	Plants per m^2
Double row system	40 cm / 20 cm	Between the middle of the raised beds: 150 cm. Between the rows on the raised beds: 25 cm	33 cm	4
Single row system	25 cm / 20 cm	75 cm	33 cm	4

Results and Discussion

In the single row system *M. fragariae* incidence (treatments without leaf sanitation) was lower and yield was higher than in the double row system. In both systems leaf sanitation decreased *M. fragariae* significantly (Table 2) and yield increased.

Table 2. Effect of leaf sanitation on *M. fragariae* and yield in the double row system and the single row system at Frick 2001. cv. Senga Sengana.

	Double row system		Single row system	
	Without leaf sanitation	With leaf sanitation	Without leaf sanitation	With leaf sanitation
Leaf spot incidence (% infested leaf surface on June 21st, 2001)	71.8 a	5.6 b	34.2 a	4.1 b
Yield/plant (g)	199 a	245 b	238 a	325 b

Means within each field followed by different letter are significantly different (P= 0.05, Tukey Test).

Conclusions

It can therefore be suggested for Central European conditions to use a single row system combined with leaf sanitation in early spring. Leaf sanitation should be done carefully, healthy green leaves should remain intact, otherwise fruit weight could decrease significantly (data not shown). The best period to conduct leaf sanitation is during a frost period in early spring before plants begin to sprout. Removal of dead leaves can be done by besom (broom), but removal of by *M. fragariae* infested green leaves should be done by hand. The time required for leaf sanitation is about 40–100 hours per ha^{-1} which corresponds to 2 -5 % of the amount of working time in strawberry. It is conceivable to do this work in future with a crop adapted machine with brushes, similar to the one used for cleaning streets.

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Occurrence of diseases in Austrian elderberry orchards

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Abstract: Between 1999 and 2002 a survey investigated the occurrence of diseases in 14 Austrian elderberry orchards. The very humid summers in 1999 and 2002 led to high crop losses caused by a wilt disease of the umbels, where the peduncles of the umbel suddenly became blighted. *Fusarium sambucinum* and *Phoma sambuci-nigrae* both frequently isolated out of diseased shoots were able to cause wilt symptoms in artificial inoculation experiments. In 2002, a *Colletotrichum* species attacked the ripening fruits. In contrast to the wilt, which occurred only in orchards situated at unfavourable sites (valleys), *Colletotrichum* attacked favoured sites, such as upper slopes. Other fungi isolated occasionally from rotting fruits were *Botrytis cinerea, Ascochyta* sp., *Cylindrocarpon* sp. and *Penicillium* sp. In contrast to the wilt fruit rots where characterised by shrivelled berries remaining attached to the healthy peduncles. In the dry summers of 2000 and 2001 wilt and fruit rots were observed occasionally. Several fungi, such as *Phoma sambuci-nigrae, Alternaria* sp., *Ascochyta* sp., *Cercospora* sp., *Ramularia* sp. and *Stagonospora* sp. were observed to cause leaf spots.

Key words: *Sambucus*, wilt, *Fusarium sambucinum*, *Phoma sambuci-nigrae*, *Colletotrichum*

Introduction

Since the 1970s the elderberry production area in Austria steadily increased and has currently reached a level of 1200 ha on approximately 1000 farms. The main production is in Styria, in the south-east of Austria. The main variety is Haschberg. The berries are used in food processing and pharma-industries, while flowers are used for juice production. The aim of this 4-year survey was to investigate the cause of umbel wilt in Austrian elderberry orchards. On that occasion the fungi colonising berries, leaves, shoots and perennial wood were recorded. This paper gives a summary of the occurrence of diseases in Austrian elderberry orchards.

Material and Methods

Between 1999 and 2002, 14 elderberry orchards in different parts of Styria were surveyed and the occurrence of diseases on fruits, leafs and wood was recorded. Umbels were taken at different growth stages to investigate the influence of pathogens associated with umbel wilt on the occurrence of wilting symptoms. The samples (basal internode and peduncle) were washed, surface sterilised (Schulz et al., 1993) and transferred to a isolation medium (Bills, 1996). Leaves showing symptoms of leaf spots were washed and incubated in a moist chamber for the development of sporulating structures. Wood samples were taken from branches and trunks of all 14 sites with a 5 mm increment borer (Wright, 1933) and the cores were flamed, and after incubation, examined for endophytes.

Results and discussion

Wilt of the umbels

First symptoms of wilt occur during summer after periods of rain, when lateral peduncles of the umbel suddenly became blighted. The attacked parts of the umbel falls to the floor at the slightest movement. Several factors favour the wilt: a) the site: the disease only occurs in orchards with long periods of leaf wetness (valleys, bases of slopes); b) the climate during the growing season: cool summers with high precipitation favour an outbreak; c) the age: the older the orchard, the more severe are the symptoms.

Results of the survey showed that the occurrence of wilt symptoms correlates with the fungi infection of the umbels. An increasing rate of infection of the peduncles was found 2-3 weeks before first symptoms appear. In infection experiments that were conducted in greenhouse and field both *Fusarium sambucinum* and *Phoma sambuci-nigrae* induced wilting symptoms and were reisolated (Steffek, 2002). Both fungi are frequently detected in one year old shoots (mainly in the bark around buds) of diseased trees. *Fusarium sambucinum* attacked the phloem and cambium of infected shoots. Controlled infections with a set of other fungi (*Alternaria* sp., *Fusarium* sp., *Cladosporium* sp., *Colletotrichum* sp. and *Ascochyta* sp.) which were isolated from shoots did not cause any symptoms.

Fusarium sambucinum is described to cause cankers and diebacks on many trees and shrubs, *Sambucus* sp. is a main host (Wollenweber, 1932; Booth, 1971; Domsch et al., 1993). *Phoma sambuci-nigrae* (Sacc.) Montel et al., 1991 (former described as *P. exigua* var. *sambuci*) is associated with leaf spots and diebacks of shoots (Boerema & Höweler, 1967).
Analysis of nutrients between flowering and harvest revealed no differences in the contents of K, Mg and Ca between different sites. The nitrogen level in the peduncles was consistently higher in trees where the disease occurred, especially at early growth stages.

Fungal Diseases of the Fruit

In humid summers *Colletotrichum* causes high losses in many elderberry orchards. First symptoms appear shortly before harvest, the rot spreads rapidly, revealing salmon-coloured acervuli with masses of spores on the surface of the berries. As a consequence the berries shrivel but remain attached to the peduncles until late in the autumn. At unfavourable sites mixed infections of both wilt- and fruit rot causing fungi might lead to a confusion of symptoms.

Other fungi isolated from rotting fruits were *Botrytis cinerea* (which might occur epidemic too, but can easily be controlled), *Ascochyta* sp., *Cylindrocarpon* sp. and *Penicillium* sp. While *Collettotrichum* occurred frequently the others were only found occasionally.

Fungal diseases of the leaves

Ascochyta-, *Alternaria-*, *Cercospora-*, *Phoma-*, *Ramularia-* and *Stagonospora* sp. were isolated from leaf spots and scorch. Leaf spots of *Phoma sambuci-nigrae* might play a role in the disease cycle of umbel wilt. They emerge in June, some weeks after the release of spores from pycnidia on 1 year old twigs and herald an forthcoming outbreak of the wilt. In some orchards *Cercospora* sp. caused defoliation of the attacked trees in summer. The other fungi where not associated with other symptoms than leaf spots.

Fungi in wood

Different genera of Basidiomycetes were isolated especially from the wood of older orchards. In many of them also *Graphium* sp. and *Fusarium* sp. were found. Their occurence in single orchards was quite uneven and not linked to the occurence of wilt symptoms. For that reason a detailed determination was not conducted.

A bacterial blight, reported from plant protection services in Germany and Hungary to cause severe damages in orchards, did not yet occur in Austria.

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Strawberry diseases imported into Switzerland with young plants

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Abstract: Successful integrated production depends on young plant material of high quality. Surveillance of the health status of random samples of young strawberry plants imported into Switzerland showed that too often plants were in a bad condition, lacking in vigour and had weakly developed roots. These plants failed to develop in the field, and, depending on different factors like climate and soil conditions, symptoms of latent systemic infections became visible. Comparing the results of import monitoring and of the investigations of our extension service, it must be concluded that at least three diseases have been inadvertently introduced into Swiss strawberry growing areas since 1993.

Key words: strawberry, imported plants, monitoring, health status, disease

Introduction

Small fruit production is still increasing in Switzerland (Bosshard et al., 1998) but producers have difficulty in purchasing young plants of high quality at a reasonable price. This is true not only for strawberries, but also for all kinds of small fruits.

Alarmed by the poor quality of some lots of imported young strawberry plants, the Swiss Plant Protection Service ordered the monitoring of the health status of random samples starting in 1995 (Bosshard et al., 1997). Plants were analysed for quarantine organisms and pathogens affecting plant quality. The EC lists *Phytophthora fragariae* (red core), *Colletotrichum acutatum* (black spot, anthracnose) and *Xanthomonas fragariae* (angular leaf spot) as well as *Aphelenchoides fragariae* and several viruses as quarantine-organisms.

Systemic diseases caused by *Gnomonia, Hainesia* or *Verticillium* might influence the development of the plants and reduce the yield of high quality fruits.

Materials and methods

Different kinds of strawberry plants are imported, depending on the time of the year. Nurseries buy cold-store plants during winter, and garden centres and fruit producers import fresh dug plants or lifted green plants from June til August.

In the diagnostic lab of the Federal Research Station at Wädenswil, combinations of different conventional, serological and molecular biological methods were used. Test series were developed which allowed rapid and reliable identification of quarantine and quality-affecting microorganisms.

Table 1 shows the quarantine organisms which were sought and the detection methods used. Under certain circumstances the samples were also investigated for nematodes such as *Ditylenchus dipsaci* and *Aphelenchoides fragariae* and for strawberry mites (*Phytonemus pallidus*). Viral diseases were not taken into account.

C. acutatum (black spot) was detected in petioles by the paraquat test developed by Cook (1993) and identified with monoclonal antibodies following the method described by Hughes et al. (1997). Roots affected by red core disease *(P. fragariae)* were found by visual inspection or with nested PCR (Bonants et al., 1997). *X. fragariae* causing angular leaf spot was detected by DAS-ELISA in thin slices of the crowns (Bosshard et al., 1997).

By placing little pieces of surface-sterilized crown tissue on PDA medium containing antibiotics, pathogens affecting plant quality such as *Gnomina coumari* (stem-end rot), *Hainesia lythrie* (tan brown rot) and *Verticillium sp.* (wilt disease) were detected.

Table 1. Strawberry pathogens which may not be imported into EC member countries.

Organism	Disease	Symptoms	Detection Method
Xanthomonas fragariae	Angular leaf spot	Leaf spots, reduced growth, brown calyxes	ELISA DAS PCR
Phytophtora fragariae	Red stele root rot (red core)	Red central cylinders	Visual, ELISA Duncantest nested PCR
Colletotrichum acutatum	Anthracnose (black spot)	Leave spots, reduced growth, brown calyxes, fruit rot	isolation on PDA paraquat-test ELISA mab, PCR

Results and discussion

The health status of cold store plants is difficult to judge by visual inspection as no leaves are present. However, the texture and colour of the crown are indicators of diseases such as crown rot (*Phytophthora cactorum*), other systemic diseases or frost damage. Strawberry roots affected by red core show dark red coloured steles, but only at certain times of the year. A reddish discolouration of steles can also be an effect of cold storage.

Green plants may show the typical symptoms of angular leaf spot, but more often the latent disease can be detected only by DAS-ELISA in crown tissue. Leaf symptoms of black spot (anthracnose) and tan brown rot look very similar; a clear diagnosis is only possible after acervuli and the typical conidia have been developed. Latent infections with *C. acutatum* can detected by the paraquat test; however, the identification of the pathogen by ELISA with monoclonal antibodies was possible only after the isolation of the fungus. Using the method described by Hughes et al. (1997) it was found that the washing water of the paraquat-treated plant parts caused a significant quenching of the ELISA reaction. The conventional method used to isolate pathogens from crown tissue proved to be very reliable for the detection of *C. acutatum, G. coumari, H. lythrie* and *Verticillium sp.*.

The percentage of latently diseased plants was found to be very high (fig. 1), depending on the exporting nursery. The kind of pathogens detected varied from year to year, although plants from some nurseries showed the same contaminants each year. It is well known that nurseries have great difficulties to get rid of pathogens once they are established.

After the monitoring of over 400 samples it can be stated that false negatives were much more frequent than false positives. In some cases, tests failed to show positive results even in plants with clear symptoms. It must be assumed that young plants are thoroughly treated with fungicides which suppress the pathogens but don't eradicate the disease. Plant parts or DNAextracts used in our tests might contain fungicide residues, which inhibit the test reactions and retard the growth of the pathogens on the isolation medium.

Regarding the monitoring results it is not surprising that at least three strawberry diseases have been inadvertently introduced by imports into Switzerland since 1992: angular leaf spot (Grimm et al., 1993), black spot (anthracnose) and tan-brown rot.

The introduction and spread of the tan-brown rot has been observed since 1996 (tab.2). *H. lythri* was more or less frequently isolated from samples of imported young plants and occasionally from diseased plants sent in by growers and extension services. As the fungus is described as a weak wound parasite (Compendium on strawberry diseases) and as it is not mentioned in the European publications on small fruit diseases no attention was paid to this pathogen at the beginning. At the end of July 1998, samples of severely diseased Pegasus plantlets were sent by two different nurseries to the extension service for inspection. Plants showed brown leaf spots and reduced root growth. *H. lythrie* was isolated from both samples. If young strawberry plants were inoculated with these isolates the symptoms described developed. *H. lythrie* could be reisolated from the leaves as well as from all other parts of the inoculated plants. In the following years *H. lythrie* seemed to be spread all over Europe which is not surprising as strawberry plants are widely traded from country to country.

From monitoring results and from the incidence of the above mentioned diseases in Swiss strawberry fields it can be concluded that latently infected plants will not necessarily develop symptoms in the field. This observation can be explained by the fact that the growth of pathogens existing in the plant system is influenced by the variety, the climate, the planting system and the soil conditions. Wet autumns followed by mild, humid winters, and wet cold springs followed by sudden high summer temperatures at the beginning of June might have a disastrous effect on plant growth and fruit quality. In the often heavy soils of Swiss small fruit growing areas, unfavourable weather conditions can induce damage which is not known in other European countries. The analysis of Swiss weather data shows that in years with significant deviations from the long-term average temperature and rain fall, the incidence of diseases increases.

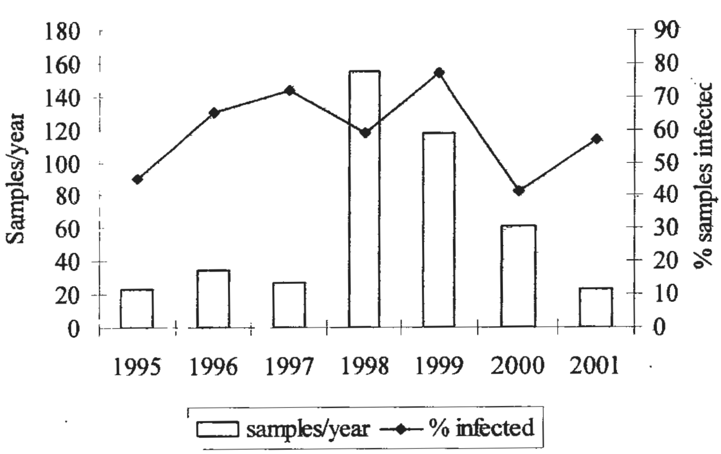

Figure 1. Samples monitored 1995-2001.

Table 2. Increasing contamination of imported strawberry plants with *Hainesia*.

Year	Number of samples monitored	Number of samples infected by *Hainesia* in different countries							Total	% with *Hainesia*
		D	GB	F	I	NL	HU	USA		
1995	23									
1996	34			1		2	1		4	11.8
1997	27	2		1				1	4	14.8
1998	155	3		1		3			7	4.5
1999	118		3	8			46		57	48.3
2000	61	6		9		5	1		21	34.3
2001	23	2		2	5	1	4		14	60.9

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Fungi colonising elderberries (*Sambucus nigra*) and their impact on the occurrence of wilt symptoms

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Abstract: With an area of 1.200 ha European Elder (*Sambucus nigra*) is the most important soft fruit crop in Austria. Because of their high content of anthocyanes the berries are used in food industries. At suitable sites elder can be produced without high effort in plant protection. Wilt symptoms of the corymbs occur only at unfavourable sites especially in years with high precipitation. On such sites corymb wilt is a disease of high economic importance, it can cause crop losses of up to 100 %.
From 1999-2002 the influence of fungal pathogens associated with corymb wilt on the occurrence of wilting symptoms were investigated. In addition to that controlled infection experiments in greenhouse and in the open field were carried out with fungi which had been frequently isolated from diseased corymbs.

Results showed that in years with high infestation the occurrence of corymb wilt correlates with the fungi infections of the corymbs. Increasing fungal colonisation in the corymbs was detected 2-3 weeks before first symptoms are visible. The controlled infection experiments showed that both *Fusarium sambucinum* and *Phoma sambuci-nigrae* can induce wilting symptoms in corymbs. Both fungi are frequently detected in the one year old shoots of diseased trees. Controlled infections with a set of other fungi which were frequently isolated from shoots did not cause any symptoms.

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Chitinase as a control agent of *Didymella applanata* causing the raspberry spur blight

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Abstract: *In vitro* and *in vivo* studies were done to assess the efficacy of the microbial chitinase (Chi) (from *Streptomyces sp.*) on *Didymella applanata*, the fungus which causes spur blight of raspberry (*Rubus idaeus*). *D. applanata* was isolated from canes of diseased raspberries in a plantation in Novosibirsk, Russia. *In vitro*, the effective concentration of Chi that reduced the growth of *D. applanata* was 400 mU/ml. In inoculation experiments on raspberry canes, chitinase at the rate 500 mU/ml reduced fungal development. In plantation where canes were inoculated after spraying with chitinase, fruiting bodies of fungus failed to form in all enzyme treatments, whereas a significant number of these fungal fruiting bodies (12.8 per cm^2) developed in control treatments lacking chitinase spraying. The chitinase reduced the size of lesions and limited the infection of internal tissues of canes.

Key words: chitinase, ecologically safe control, *Didymella applanata*, raspberry, spur blight

Introduction

Spur blight caused by *Didymella applanata* (Niessl.) Sacc. (anamorph *Phoma argillacea*) is one of the most harmful cane diseases of red raspberry (*Rubus idaeus* L.) (Williamson & Hargreaves, 1981; Jenning, 1988). In Russia, the first report of this fungus as causal agent of raspberry spur blight was published by Lebezhinskaya (1959). Later, this disease was registered in different regions of Russia. To prevent serious damage, chemical control of this fungus is necessary in many production areas. However, recent public concern over the potential harmful effects of chemical fungicides on the environment and human health has given more emphasis to the search for alternative ecologically benign control methods. Ecologically safe control of raspberry cane diseases, including the use of biological controls, is poorly developed.

Recently, *in vitro* fungicidal and fungistatic effects of biological preparations involving bacteria of genera *Pseudomonas* and *Bacillus* and fungi of the genus *Chaetomium* on *D. applanata* were evaluated. The preparations containing bacteria of the genus *Pseudomonas* and fungi of the genus *Chaetomium* were most efficient in suppressing *D. applanata* (Shpatova et al., 2003). It is also valuable to consider some microbial metabolites (enzymes, toxins) as potential agents of ecologically benign methods for the spur blight suppression.

Due to the important role of chitinolytic enzymes in fungal growth and development, these enzymes have been examined as possible agents for plant disease suppression (Cohen-Kupiec & Chet, 1998; Herrera-Estrella & Chet, 1999; Ordentlich et al., 1998). For example, chitinases produced by the bacterium *Serratia marcescens* or S. *liquefaciens* were used for control of *Sclerotium rolfsii* (Ordentlich et al., 1998) and *Botrytis* cinerea (Whiteman & Stewart, 1998). The chitinase gene has also been used to create transgenic plants for control of fungi (Moffat, 1992; Shi, 2000). In field testing of a chitinase produced by *Streptomyces sp.*, we have shown the effect of this enzyme on raspberry spur blight (Shternshis et al., 2002a). A more detailed study of the effect of chitinase enzyme on *D. applanata* and spur blight was undertaken.

This paper describes the effect on *D. applanata* by chitinase (Chi) produced by *Streptomyces sp.*, *in vitro* and in field experiments in which raspberry canes were wound-inoculated and given prophylactic treatments with chitinase.

Materials and Methods

Source of chitinase

The chitinase, Chi is a crude commercial chitinase preparation derived from *Streptomyces sp.* and supplied by "Biostart" company (Novosibirsk, Russia). The Chi was additionally purified with ammonium sulfate precipitation at 80% saturation to remove possible impurities of low molecular weight metabolites. Specific activity of this preparation was 11 U/mg of protein.

Preparation of inoculum

Didymella applanata, isolate Da-99 was obtained from diseased raspberry canes in Novosibirsk. The isolate was maintained on Czapek's medium at 4°C. To obtain inoculum, pseudothecia were placed on the medium and incubated at 25°C for 2 weeks up to picnidia production. Single spore isolations were made by transferring pycnidia to sterile distilled water to release pycnidiospores. Concentrations of pycnidiospores were determined with the aid of haemicytometer.

Enzyme assays

Chitinase activity was assayed by the rate of hydrolysis of a water-insoluble chromogenic chitin substrate in 50 mM sodium acetate buffer (pH 5.5) (Duzhak et al., 2002). Since we intended to use chitinase in the investigation of plant disease control, an important feature of the enzyme preparations was the relative absence of other hydrolytic activities. To establish the presence of impure enzymes, Chi preparation was assayed for β-1,3-glucanase, α-mannanase, lipase, and protease activities. β-1,3-glucanase and α-mannanase activities were determined by measuring the release of reducing groups from a corresponding substrate (0.1% laminarin and 0,1% yeast α-mannan, respectively) in 50 mM sodium acetate (pH 5.5). The concentration of reducing sugars was estimated by the reaction with 3,5-dinitrosalicylic acid (Miller, 1959).

One unit of chitinase, β-1,3-glucanase or α-mannanase activity is defined as an amount of enzyme that liberates 1mM of N- acetylglucosamine, glucose or mannose, respectively, per min at 37°C. Lipase and protease activities were measured at pH 5.5 as described previously (Shternshis et al., 2002b). Protein concentrations were determined by the method of Coomassie Blue binding assay (Spector, 1978), using bovine serum albumin as a standard.

Bioassays of chitinase in vitro

1.5 ml of chitinase solution, sterilized by filtration through 25 mm Millex disc Durapore filters (0.22 μm), added to 1.5 ml of autoclaved and cooled to 42°C medium, containing 40g/l maltose, 2 g/l $(NH_4)_2SO_4$, 10 g/l yeast extract, 0.3 g/l $MgSO_4 \times 7H_2O$, 6 g/l KH_2PO_4, and 30 g/l agar, and immediately poured into 35 mm Petri dishes. Chi remained stable at 42°C at least 30 min. After the agar had solidified, single spore suspensions of *D. applanata* Da-99 were streaked on plates at the rate of 15-20 spores per plate and grown at 28°C in the dark. After 3 days exposure to chitinase, the diameter of the fungal colonies was measured under a dissecting microscope (x8), and the percentage of inhibition was calculated. The control experiment was performed in the same way without chitinases omitted from the medium. All experiments were done in 6 replicates.

Inoculation of raspberry canes

These experiments were carried out on 1-year-old vegetative canes (primocanes) in the collection of the raspberry plantations of the Novosibirsk Fruit Experimental Station. Cv. Kirzhach, selected by Dr. V. Kichina at the All-Russian Institute for Breeding and Technology in Horticulture, Moscow. This selection is highly susceptible to spur blight and was used for inoculation with *D. applanata*. Agar plugs 10 mm diameter were cut out of plates of 15–day-old *D. applanata*. The inoculum consisted of mycelium and native pycnidia from the fungus grown.

For inoculations, primocanes of a height of 70 cm were used. A zone 30 cm above soil level and 15 cm wide was pruned to remove leaves (petioles of 3 cm were left after cutting). This zone was damaged with the aid of glass (?)powder. The size of each wound was 2 cm^2. Before application of fungal inoculum, the canes were sprayed once with chitinase solution (500 mU/ml) applied at a volume application rate of 10 ml/cane using a hand held Orion- 6 sprayer (Quazar Corp. Warsow, Poland). The agar plugs were then positioned so that the fungus was in direct contact with the damaged epidermis and wrapped with moist cotton wool. Each inoculation was then individually covered with plastic film (15x15 cm) to provide adequate humidity and temperature conditions for fungal growth. Preliminary experiments discovered that the temperature inside did not exceed 30^o C. The plants were incubated for 7 days. After this incubation period, the cotton with film was removed. Five replicates were used for each enzyme application treatment. The control canes were treated with fungal inoculum, but not with chitinases. A randomized design was used in the experiment. Pathogen was inoculated on 21 June.

Data collection and analysis

To estimate the effect of the enzyme on spur blight, the lesion squares were measured after 7 and 30 days, and at the end of vegetation period. The area of tissue necrosis of the xylem and pith on cane cross-section, the extent of internal necroses along the cane and the area with fungal fruiting bodies and their number were measured at the end of vegetation only. The area of lesions was quantified using Cellophane film overlays of cane damage. The extent of internal necroses expressed as the lesion length along the cane was determined by measuring length and depth of lesions. The area of the necrotic zone in pith was also measured. The area of necrosis patch on cross-section was determined with aid of Cellophane film to copy the contour of the lesion. To quantify the number of fruiting bodies of *D. applanata* per 1 cm^2 of patch, a light microscope was used with a calculated eyepiece reticule. Data were analyzed by paired t- test. The alpha level used was 0.05.

Results and discussion

Effect of chitinases on* D. applanata *growth

The protease, lipase and α-mannanase could not be detected in chitinase preparation. β-1,3-glucanase was present at 0.01 U/mg of protein in the Chi. The results of laboratory experiments are presented in Table 1.

Chi significantly reduced the growth of *D. applanata* at concentration 400 and 750 mU/ml (up to 1.3 and 2 times respectively) with significant differences between these two concentrations (t-test 3.1, $p<0.05$). Although there were significant differences between the control and Chi at 400 and 750 mU/ml (t-test 2.4 and 6.9, $p<0.05$), the differences between control and Chi at 80 and 150 mU/ml were not significant.

Table 1. Effect of chitinases on *D. applanata* growth

Treatments	Concentration of chitinase, mU/ml	Diameter of colony, mm (mean ±SE)
Chitinase	80	6.8 ± 1.0
	150	7.4 ± 1.6
	400	5.8 ± 2.0
	750	3.8 ± 1.2
Control	0	7.6 ± 1.5

Inoculation of canes with D. applanata

Data on the influence of chitinase on spur blight lesion development under the treatment with Chi are presented in Table 2. Area of lesions was measured at 7 and 30 days after inoculation.

Table 2. Influence of chitinase on cane lesion due to spur blight development

Treatments	Area of lesion in cm^2 (mean ± SE), days after treatment	
	7	30
Chitinase	0.2 ± 0.1	0.2 ± 0.1
*Control	0.2 ± 0.1	10.5 ± 3.8
Untreated canes	0	1.6 ± 1.6

*Control includes inoculation by the fungus lacking sprayed chitinase

There were no significant differences between the control and Chi 7 days after treatment, but after 30 days the treatments with Chi led to complete inhibition of *D. applanata* development (t-test 2.7, $p<0.05$), whereas the control had large lesions (10.5 ± 3.8 cm^2).

Data on the influence of chitinase on spur blight development in inoculations are shown in Table 3. All parameters were measured 80 days after inoculation.

The data show the efficacy of chitinase at rate of 500 mU/ml in experiments on inoculation of canes by *D. applanata* (480 pycnidia/cm^2). The size of lesion caused by spur blight was reduced significantly under the influence of Chi (t-test 3.04, $p<0.05$). In addition, preliminary spraying of canes with enzymes restricted infection of internal tissues. The treatment with Chi reduced the development of pith necrosis 2 times (t-test 3.3, $p<0.05$). The necroses of xylem were reduced by the application of Chi by 4.5 times (t-test 3.1, $p<0.05$). The extent of pith necrosis (distally and proximally from point of inoculation) was reduced 6.5 times (t-test 3.3, $p<0.05$) by Chi. Fruiting bodies failed to form in spray treatments, but a significant number of these bodies (12.8 per cm^2) was observed in control the treatment lacking sprayed chitinase.

Table 3. The effect of chitinase on spur blight development in inoculation experiment with *D. applanata* (mean ±SE)

Treatments	Area of lesion, cm^2	Relative area of xylem necrosis on cross-section, %	Relative area of pith necrosis on cross-section, %	Pith necrosis (along the cane), cm	Area with fungal fruiting bodies, cm^2	Number of fruiting bodies/ cm^2
Chitinase	4.3 ± 2.8	1.0 ± 0.3	10.0 ± 3.3	0.3 ± 0.1	0	0
*Control	29.2 ± 4.7	4.5 ± 1.1	20.5 ± 1.9	2.6 ± 0.7	1.1 ± 0.4	12.8 ± 4.3
Untreated canes	1.6 ± 0.5	0	0	0	0.9 ± 0.3	7.4 ± 1.2

*Control includes inoculation by the fungus lacking sprayed chitinase

Thus, we have made the first attempt to estimate the direct effect of microbial chitinase on *D. applanata,* the causal agent of raspberry spur blight. This fungus belongs to the Ascomycotina, and the main components of cell walls of these fungi are glucans and chitin (Bartnicki-Garcia, 1968). The results obtained here provide some evidence that it may be possible to reduce spur blight by application of microbial chitinase damaging chitin of fungal cell walls.

Therefore, chitinase would be considered as effective agent of ecologically safe disease control on raspberry.

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Resurgence of virus problems in *Rubus* in the United Kingdom: possible effects on crop production and certification

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Abstract: Recent surveys of commercial *Rubus* crops in the UK have shown a large increase in the incidence of the pollen-borne *Raspberry bushy dwarf virus* (RBDV). Much of this is attributable to the widespread displacement in commerce of RBDV-immune cultivars by those with improved agronomic qualities but that are infectible with common isolates of this virus. In England and Wales there is also evidence for the spread of resistance-breaking isolates of RBDV. In addition, the widespread development of biotypes of the main virus vector aphid, *Amphorophora idaei,* able to overcome gene A_1 (that confers resistance to this aphid), has resulted in large increases in the incidence of the four viruses transmitted by this vector aphid. It is also of concern that biotypes of this aphid able to overcome gene A_{10} (that confers strong resistance to the four known biotypes of this aphid) are also appearing in some crops. *Raspberry vein chlorosis virus* transmitted by the small raspberry aphid, *Aphis idaei,* is increasing in prevalence in crops. The response of several raspberry cultivars to infection with these viruses in a field experiment is reported, together with possible strategies to minimise their levels of infection in crops.

RBDV and aphid-borne viruses infect many cultivars symptomlessly but the results of trials have established that such symptomless infections by individual viruses can decrease the production of quality cane, fruit size and the time of fruit ripening. Such effects are likely to be greatly increased by multiple virus infections, the situation that normally prevails under commercial conditions. In addition to such losses, symptomless infection poses problems for the certification of healthy planting material.

Key words: aphids, pollen transmission, virus resistance, virus tolerance, virus vector resistance, fruit quality

Introduction

About 15 virus or virus-like diseases are reported in *Rubus* crops in Europe (Jones, 1986) and several of these cause diseases that damage severely crop production and/or fruit quality. Information on the impact of symptomless infection is sparse. The most important viruses and virus complexes in crops and the vector species associated with their transmission are given in an accompanying paper (Jones, 2004; this volume).

The viruses found most commonly infecting *Rubus* in Europe are transmitted by the large raspberry aphid, *Amphorophora idaei,* that transmits four of the five most important aphid-borne viruses. No immunity to these viruses has been detected in *Rubus* germplasm (Jones & Jennings, 1980), but several sources of strong resistance to the aphid vector are known (Knight et al., 1959; Keep & Knight, 1967). This resistance has been introduced into most commercial raspberry cultivars with a dramatic effect on decreasing the incidence of the four viruses transmitted by this aphid (Jones, 1976; 1979a; 1988). However, the widespread and prolonged cultivation of these resistant cultivars in the UK has created a strong selection pressure on the aphid to overcome such resistance. Several biotypes of *A. idaei* are now recognised in the UK and one or more of these biotypes are capable of colonising plants containing the most widely deployed *A. idaei*-resistance gene, A_1 (Birch & Jones, 1988; Jones et al., 2000). Such biotypes are now prevalent in crops in the UK and this has resulted in large increases in the incidence of the four viruses this aphid transmits. It is also of concern that other biotypes able to overcome gene A_{10} (that confers strong resistance to four known biotypes of this aphid) are also appearing in some crops. However, there is some evidence that the effectiveness of the same *A. idaei*-resistance gene can differ between cultivars (Jones et al., 2000).

The small raspberry aphid, *Aphis idaei,* that transmits only *Raspberry vein chlorosis virus* (RVCV), has increased in prevalence in crops in northern Europe in recent years. No sources of resistance to this aphid have been identified in *Rubus* germplasm but resistance to the virus has been identified in some *R. strigosus var. idaeus* material. Crosses between RVCV-infectible and -resistant material produced a significant number of progeny with high resistance/immunity to RVCV (Jennings & Jones, 1986). Its effects on raspberry production in Europe are not known.

Raspberry bushy dwarf virus (RBDV) is transmitted in association with infected pollen and is therefore very difficult to control. The virus occurs worldwide wherever *Rubus* is grown (Jones et al., 1996). Recent surveys of commercial *Rubus* crops in the UK have shown a large increase in the incidence of this virus (Knight & Barbara, 1999; Chard et al., 2001). Much of this is attributable to the widespread displacement in commerce of RBDV-immune cultivars by new cultivars with improved agronomic qualities but that are infectible with common isolates of this virus. Immunity to common isolates of RBDV is present in many raspberry cultivars and this character is determined by the single dominant gene, *Bu* (Jones et al., 1982; 1998) that has provided effective virus control for more than 40 years, even under strong inoculum pressure. However, in England and Wales and parts of northern Europe and Russia, RBDV isolates able to overcome gene *Bu* (termed resistance-breaking; RB) occur and are spreading in crops (Knight & Barbara, 1999). Such isolates are capable of infecting all UK commercial raspberry cultivars making control of such isolates very difficult (Jones et al., 1996).

This paper reports on a recent field experiment to assess the incidence of these different aphid-borne viruses and RBDV in raspberry cultivars with different resistance genes. As many viruses induce no obvious leaf symptoms when they infect most cultivars, it is often assumed that they have little impact on fruit production. Therefore, this paper also summarises some earlier unpublished field data on the effects of symptomless infection with aphid-borne viruses in raspberry.

Materials and Methods

A field experiment, using six red raspberry cultivars differing in the resistance genes they contained (Table 1), was planted in 1996 and arranged as four randomised blocks; each block was a replicate of eight rows of 20 plants of each cultivar and the cultivars were randomised within each block. No insecticide was applied to the crop for the duration of the experiment. In early summer, 10% of all plants in two blocks were assayed for RBDV by ELISA and for *Rubus yellow net virus* (RYNV) by PCR (Jones et al., 2002). All plants in these blocks were also assessed visually for symptoms of Raspberry leaf spot mosaic disease (RLSMD), caused in sensitive cultivars by infection with either Raspberry leaf mottle (RLMV) or Raspberry leaf spot (RLSV) viruses (Jones & Jennings, 1980; Jones, 1987), and RVCV (Jones et al., 1987).

In a field experiment planted in 1975, the effects on growth and yield of infection with Black raspberry necrosis BRNV; (Jones & Roberts, 1977), RVCV (Jones et al., 1987), RLMV and RLSV (Jones, 1987) was studied in the *Am. idaei*-resistant cultivars, Glen Prosen and Malling Orion (containing gene A_1) and cv. Leo (containing gene A_{10}). These cultivars were chosen because their resistance to *Am. idaei* prevented healthy control plants from becoming infected, and because they show no obvious symptoms when infected with the *Am. idaei*-transmitted viruses and only very faint, or no, symptoms when infected with RVCV (Jones & McGavin, 1998). The precise experimental details have been given elsewhere (Jones, 1979b). Cultivation and management of the experiment followed normal commercial practice in Scotland. Hand-picked fruit was collected every 4-9 days during the fruiting months. Fruit was weighed in the field directly after picking and sub-samples of the total pick on each picking date were weighed, fruit numbers determined and the mean berry weight calculated. Data were analysed by ANOVA and, for simplicity and space, representative data for only 1978 is presented, based on mean values per plant for the combined cultivars.

Results and Discussion

Virus incidence in different cultivars

Incidence of RYNV and RLSMD.

Table 1 shows that after 6 years, cultivars with the strongest Am. idaei-resistance gene, A_{10} (Glen Rosa, Leo) were amongst those with the lowest incidence of RYNV, whereas the two gene A_1-containing cultivars (Glen Prosen, Malling Landmark) and cv. Glen Clova were those with the highest incidence. However, cv. Glen Ample that contains gene A_1 had the second lowest incidence of RYNV. The incidence of RYNV was therefore 9-53% within cultivars containing the same resistance gene, A_1. All red raspberry cultivars are infectible with RLMV and RLSV (Jones, 1987; Jones & McGavin, 1998) but only four of the cultivars studied here show RLSMD on infection. Table 1 shows that two such cultivars (Malling Landmark, Glen Rosa) showed one of the highest and lowest incidences of RYNV infection respectively, and one of the highest and the lowest incidences respectively of RLSMD. There was little difference between the incidence of RLSMD in cvs Glen Clova and Glen Ample. These data confirm the increase in incidence of Am. idaei biotypes on cultivars containing gene A_1, the differing effectiveness of gene A_1 in different genetic backgrounds (Birch & Jones, 1988; Jones et al., 2000), and the relative stability of minor gene resistance to this aphid (Jones, 1979a; 1988).

Incidence of RVCV

None of the 6 cultivars are resistant to RVCV or its vector, Ap. idaei (Jennings & Jones, 1986; Jones & McGavin, 1998; A.T. Jones, unpublished data), yet the incidence of infection (based on symptoms) was 0.3-100% (Table 1), indicating strong field resistance to infection in cvs Glen Rosa and Leo both of which contain gene A_{10}. However, the low incidence of RVCV in Malling Landmark is probably a gross underestimate due to the very high incidence and the severity of RLSMD symptoms masking those of RVCV.

Incidence of RBDV

Table 1 shows that the gene *Bu*-containing cv. Glen Clova was not infected with RBDV after 6 years exposure in the field, whereas all the other cultivars were infected. However, the incidence of RBDV infection varied greatly (0.3-78%) indicating very strong resistance to natural infection in some cultivars. Some of this apparent resistance to pollen-borne infection might be due to the different flowering times of the cultivars, but other studies suggest that intrinsic resistance to natural infection is a feature of some RBDV-infectible cultivars, though the reason for this resistance is unclear (Barbara et al., 1984; 2001). The data show the stability of gene *Bu* resistance to common isolates of RBDV and supports earlier evidence that RB isolates of RBDV are probably not present in crops in Scotland (Chard et al., 2001).

Table 1. Percent incidence of RBDV, RYNV and symptoms of RLSMD and RVCV in 6 raspberry cultivars containing different resistance genes to *Amphorophora idaei* and RBDV, 6 years after planting.

Cultivar	**Resistance gene to:** *Am. idaei*	**RBDV**	**RBDV**	**RYNV**	**RLSMD**	**RVCV**
Glen Clova	Minor genes	Yes	0	21.9	32.4	12.8
Glen Ample	A_1	No	0.3	9.4	29.5	41.2
Glen Prosen	A_1	No	78.1	53.1	symptomless	100
Malling Landmark	A_1	No	21.9	18.8	80	1.2
Glen Rosa	A_{10}	No	1.5	0	0.6	2.8
Leo	A_{10}	No	21.9	9.4	symptomless	0.3

Effects of Latent Virus Infection

Table 2 shows the overall effects on fruit and cane characters of latent virus infection in 1978 and represent the major effects observed over several years in all three cultivars (data not shown). A consistent feature in all years was a significant decrease in fruit size and advance in fruit ripening, although there was generally little statistically significant effects on total fruit yield. Similarly, overall total cane production was usually unaffected statistically by virus infection but analysis of the different components of the cane produced show that cane length and quality was decreased significantly.

Table 2. Overall effects of latent infection with four different aphid-borne viruses on raspberry fruiting and cane production in 1978.

Character	**Healthy**	**RVCV**	**BRNV**	**RLSV**	**RLMV**
Total fruit yield (kg)	15.2	17	16.9	15.6	17.6*
Mean berry weight (g)	3.7	3.5*	3.5*	3.3*	3.5*
Days to 50% pick	7.5	6.5**	7.5	6.6*	6.4*
Tied total cane length (m)	13.2	11.5**	12.6	11.5**	13.4*
Tied cane number	6.8	6.4	7.1*	6.5	7.4*
Tied cane mean length (m)	1.93	1.77**	1.75**	1.72**	1.81**
Thinned cane mean length (m)	1.62	1.25**	1.41	1.33*	1.49

*, **, *** = statistically different from healthy plants at 5, 0.5 and 0.1% probability respectively.

These data indicate that symptomless infection with aphid-borne viruses can have significant effects on the quality both of cane and fruit production. This is in keeping with data from field experiments in North America (Converse, 1963; Freeman & Stace-Smith, 1970).

Furthermore, the effects observed in this experiment were from single virus infection whereas multiple virus infection is the norm under field conditions. Other work has shown that such multiple virus infection has a much greater effect than the combined effects observed in single infections (Converse, 1963; Freeman & Stace-Smith, 1970; Jones, 1979c). This feature is now reflected in the UK where some gene A_1-containing cultivars have to be replaced in half their expected life-span because of their decreased productivity.

Because these aphid-borne viruses infect most cultivars symptomlessly, they will go undetected in visual inspection for certification of propagation cane beds. Until more rapid and sensitive diagnostics for these viruses become available, the presence of such viruses in certified planting stock is therefore a serious possibility. One possible way to give some indication of their likely presence in such propagation stock is to include in the production beds raspberry cultivars sensitive to infection with these viruses so as to provide a visual indication of infection. However, this may not meet with acceptance by growers because of the risk of providing a source of inoculum within the crop and of the downgrading of their stock.

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Sampling of the two-spotted spider mite *Tetranychus urticae* Koch and its predators *Amblyseius andersoni* (Chant) and *Phytoseiulus persimilis* Athias-Henriot in protected Swiss raspberry

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Abstract: The relationships between the percentage of terminal leaflets occupied by one or more mobile stage of *Tetranychus urticae* and by its predators *Amblyseius andersoni* and *Phytoseiulus persimilis* and their respective densities were calculated by means of the Nachmann function. Results obtained allow development of a practical sampling method based on the percentage of occupation of terminal leaflets by two new species of predatory mites. Some suggestions for use of this information for practical control are discussed. We expect this method to contribute to enhancement of biological control against spider mites in raspberry crops.

Key words: *Tetranychus urticae, Amblyseius andersoni, Phytoseiulus persimilis*, sampling method, raspberries

Introduction

Biological control of the two-spotted spider mite, *Tetranychus urticae* Koch, in raspberry crops is being developed in several countries and has so far met with varying degrees of success (Charles et al., 1985; Wood et al., 1994; Höhn et al., 1995; Baillod et al., 1996; Meesters et al., 1998; Tuovinen et al., 2000; Linder et al., 2003). This is due to the fact that crop systems often differ from one country to another or even from one region to another, and species of predatory mites being employed in the control of *T. urticae* vary from study to study. In zones climatically similar to Switzerland, studies carried out to date mainly involve the use of native predatory mites, such as *Typhlodromus pyri* Scheuten, *Amblyseius andersoni* (Chant), *Neoseiulus cucumeris* (Oudemans) and *Euseius finlandicus* (Oudemans). With the recent introduction of cultivation under plastic, species originating from more southern regions such as *Phytoseiulus persimilis* Athias-Henriot and *Neoseiulus californicus* (McGregor) may play a more significant role in mite biological control programmes.

Studies by Mariéthoz et al. (1994) and by Baillod et al. (1996) have demonstrated that it is possible to use the terminal leaflet (TL) in the practical control of *T. urticae* and its predator *T. pyri*. In order to ensure that biological control, which is increasingly being used in raspberry crops, is effective in production under plastic, it has become necessary to check the effectiveness of this control method for two new major predatory mites frequently present naturally or introduced artificially into crops of Switzerland: *A. andersoni* and *P. persimilis*. Additional studies were conducted on a farm employing biological control methods over a two year period in order to recalculate the relationship between the percentage of terminal leaflets occupied by a mobile stage of *T. urticae* and its density, based on a much greater number of samples than those studied by Mariéthoz et al. (1994).

Material and methods

Sampling took place in 2000 and 2001 on a farm located on the eastern edges of Lake Constance (at an altitude of 450m). Observations were made in 6 plastic tunnels measuring 45 x 6 m, planted with the variety Autumn Bliss (3 rows, each 40cm wide per tunnel). Estimates of *T. urticae* populations and of phytoseiid populations were made by examining 25 terminal leaflets per tunnel approximately once every two weeks from May to the end of November. Sampled leaves were taken from the interior and exterior of the vegetation near the middle of the plant at the beginning of the season. As the season progressed, leaves were sampled randomly from upper halves of the raspberry canes. The density of *T. urticae* populations was estimated by means of a ranking system (Guignard, 1968 unpub.), while the phytoseiids *A. andersoni* and *P. persimilis* were determined by counting the number of mites per leaf. Results were also recorded as a percentage of occupation by one or more mobile stage , i.e. a terminal leaflet was considered to be occupied as soon as one mobile mite was observed.

The relationship between the occupation percentage and the density of mites per terminal leaflet was established by means of the Nachmann function, an empirical binomial model (Nachmann, 1984).

Results and discussion

***Percentage of occupation and density* – T. urticae**

Figure 1 shows the relationship between percentage of occupation and the density per terminal leaflet, in which a high coefficient of correlation (0.968) was obtained. In Table 1, mean theoretical values up to 60% occupation are given as well as the prediction interval (PI) at 95%, compared with the theoretical values obtained by Mariéthoz et al. (1994). The 60% occupation level of terminal leaflets by one mobile stage of *T. urticae* or more corresponds to the average theoretical value of 2.79 individuals, but the prediction interval shows that with a new sample having an identical level of occupation, there would be a 95% chance of falling within the interval of 1 to 7.73 mites. Even though the relationships in the present study have been calculated on a far greater sampling base than Mariéthoz et al. (1994) (162 samples compared with 22), the similarity of the results is striking, at least up to the 60% occupation level. Above this limit, the latter authors' values still fit largely within the prediction interval of this study's relationships.

Table 1. Relationships between % of occupied terminal leaflets (TL) and mobile *T. urticae* (mf) per terminal leaflet using the Nachmann function equation with prediction interval (PI) at 95%.

% occupation ≥ 1 mf	Linder et al. (2004)		Mariéthoz et al. (1994)
	T. urticae / TL		
	mf	± PI 95%	mf
20	0,59	0,21 – 1,61	0,49
30	0,99	0,35 – 2,73	0,92
40	1,47	0,53 – 4,06	1,48
50	2,05	0,73 –5,67	2,20
60	2,79	1,00 – 7,73	3,20

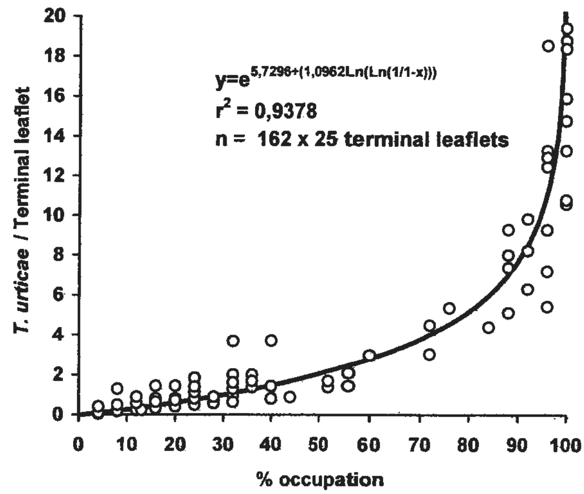

Figure 1. Relationships between % of occupation and mite densities for *T. urticae*, on raspberry grown under plastic, developed using the Nachmann function. The x-value in the formula has to be in decimal format.

***Percentage of occupation and density* – A. andersoni**

Figure 2 illustrates the relationship between the percentage of occupation and the number of mobile stages per terminal leaflet, with a high coefficient of correlation (0.959). In Table 2, the mean theoretical values together with the prediction interval (PI) values at 95% are given and compared with the theoretical values obtained by Baillod et al. (1996) for *T. pyri.*

Analogies between the two species are striking at low percentages of occupation but *A. andersoni* showed higher densities as soon as more than 50% of terminal leaflets were occupied. According to Sabelis (1985), *A. andersoni* possesses a superior rate of intrinsic natural growth (r_m) than *T. pyri*. In other words, this species produces a greater number of descendants of the feminine sex per female per day than *T. pyri* under equal conditions of nourishment, temperature and relative humidity. Although the observations were not made under the same climatic conditions in this study as in those of Baillod et al. (1996), they were made on the same kind of prey so the present results could be interpreted as a confirmation of these different reproductive strategies.

***Percentage of occupation and density* – P. persimilis**

Figure 3 illustrates the relationship between the percentage of occupation and the number of mobile stages per terminal leaflet, in which a high coefficient of correlation (0.986) was obtained. In Table 2, the mean theoretical values together with the prediction interval (PI) at 95% are given and compared with the theoretical values of other predatory species. Only in the season of 2000 are the monitoring results representative and, although the number of observations is smaller than that which was used to establish the A. andersoni curve (41 instead of 210), they do cover a sufficiently wide range of occupation percentages to enable validation of this study's results. Among the three species compared, P. persimilis is the one with the highest r_m (Sabelis, 1985). It would therefore appear logical that this species, at equal rates of occupation, would have higher population densities.

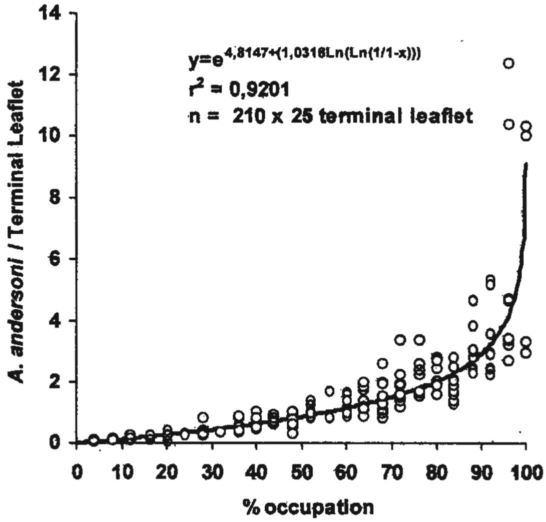

Figure 2. Relationships between % of occupation and mite densities for *A. andersoni*, on raspberry grown under plastic, developed using the Nachmann function. The x-value in the formula has to be in decimal format.

Table 2. Relationships between % of occupied terminal leaflets (TL) and mobile *Phytoseiidae* (mf) per terminal leaflet using the Nachmann function equation with prediction interval (PI) at 95%.

% occupation ≥ 1 mf	Linder et al. (2004)				Baillod et al. (1996)
	A.andersoni / TL		*P. persimilis* / TL		*T. pyri* / TL
	mf	± IP 95%	mf	± IP 95%	mf
20	0.26	0.12 – 0.52	0.29	0.15 – 0.52	0.25
30	0.42	0.20 – 0.86	0.49	0.26 – 0.89	0.40
40	0.61	0.29 – 1.25	0.73	0.38 – 1.34	0.54
50	0.84	0.40 – 1.73	1.02	0.54 – 1.88	0.68
60	1.12	0.54 – 2.32	1.39	0.73 – 2.58	0.83
70	1.49	0.71 –3.09	1.88	0.99 – 3.50	0.97
80	2.01	0.96 – 4.17	2.59	1.36 – 4.83	1.11

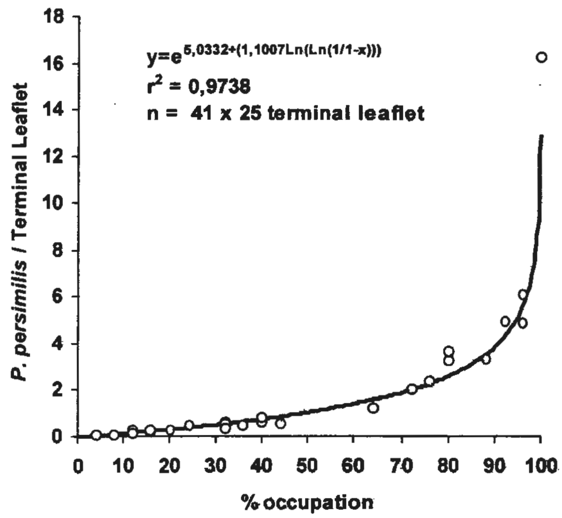

Figure 3. Relationships between % of occupation and mite densities for *P. persimilis* on raspberry grown under plastic, developed using the Nachmann function. The x-value in the formula has to be in decimal format.

Monitoring practices

This article is not intended to describe the practical details of monitoring for mites in raspberry already covered by various other authors (Baillod et al., 1996 ; Anonymous, 2002). It is, nevertheless, important to note that in the case of predator presence, control by acaricides becomes superfluous provided that the percentage occupation of predators is equal to or greater than that of pests. Experience has shown that, as in arboriculture, momentary excesses of 20% to 30% on average of the pest at the level of unit production may be tolerated.

Such situations, however, necessitate particular care by pest management practitioners. Thus, a population of *T. urticae* with a 60% terminal leaflet occupation rate will develop differently according to whether 1 or 7.73 mites per leaflet are present. In other words, although a simple comparison of pest-predator percentages will be sufficient in the majority of cases, in extreme cases the quality of observations becomes vital. In the above example, careful monitoring will be sufficient without too much extra investment of time in order to estimate the position of the pest population within the given range and consequently to take the required action.

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Interactions among predatory and phytophagous mites on strawberry

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Abstract: In order to maximise the effects of releases of biocontrol agents against pest mites we need to understand the pattern of feeding relationships in a system that includes two species of phytophagous (pest) mites, and several species of naturally occurring and released predatory mites. Leaf samples were taken to determine the spatial occurrence of the different species. Old, medium aged and recently expanded leaves, plus leaves that had not yet opened, and blossom/fruiting clusters, were taken from strawberry plants. *Tetranychus urticae* were most abundant on the older leaves, and *Phytonemus pallidus* on the unopened leaves and blossom/fruiting clusters. All adult phytoseiids found were mounted on slides and identified to species *Phytoseiulus persimilis* and *Neoseiulus californicus* were found more frequently on the older leaves, and *N. cucumeris* and *N. aurescens* on the unopened leaves and fruiting clusters. No *Typhlodromus pyri* were found in these samples.

Choice experiments were done on strawberry leaf disc arenas to determine if the phytoseiid species commonly found on strawberry showed a preference for prey species. Leaf discs were held lower surface up on wet cotton wool in Petri dishes. There was no apparent preference for either prey for *Typhlodromus pyri* and *Neoseiulus cucumeris*. Mean cumulative numbers of *T. urticae* and *P. pallidus* consumed after 48 hours were 5.3 and 3.0 for *T. pyri* and 9.8 and 11.0 for *N. cucumeris* respectively. *Neoseiulus californicus* showed a preference for *T. urticae* compared with *P. pallidus* with mean cumulative numbers consumed of 13.3 and 7.3 respectively. *Phytoseiulus persimilis* did not consume *P. pallidus*. The preferences of *N. aurescens* were not tested.

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Impact of two-spotted spider mite *Tetranychus urticae* Koch infestation in early season strawberry crops, and efficacy of different control strategies

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Abstract: The impact of the two-spotted spider mite *Tetranychus urticae* on the early season strawberry variety Madeleine planted under plastic tunnels in central Valais, Switzerland was measured in relation to various control strategies. Acaricide applications during autumn of the planting year or during the following spring controlled the pest well. Despite visible leaf damage and populations reaching 120 individuals per leaf and pressures of up to 3,800 mite-days per leaf, the yield, number and sugar content of fruits were not significantly affected. However, with increasing mite pressure, the size of first class fruits and photosynthesis showed a decreasing trend. These results suggest that strawberry plants are able to tolerate relatively high mite pressure and they should create renewed interest in biological control by means of predatory mites.

Key words: *Tetranychus urticae,* strawberries, chemical control, yield, quality, photosynthesis.

Introduction

The two-spotted spider mite, *Tetranychus urticae* Koch, is the main pest in early season strawberry crops cultivated under plastic tunnels in Switzerland (Antonin et al., 1997; Bosshard et al., 1998; Anonymous, 2002). Numerous studies on the damage caused by *T. urticae* in strawberry crops and the resulting economic impact have been published (Sances et al., 1979a,b; Oatman et al., 1981; Sances et al., 1981; Oatman et al., 1982; Raworth, 1986; Butcher et al., 1987), but no such research has been carried out under Swiss field conditions.

Mite thresholds currently used in Switzerland have been empirically defined and are based on a strategy aiming to obtain low pest populations at the end of the first year of growth and to restrict acaricide treatments to a single application at the start of flowering in the second year (Antonin et al., 1997). Although practical application of this strategy is generally satisfactory, cases of failure have been recorded. These poor results may be a result of inadequate application techniques or acaricide resistance, but may also be due to inappropriate thresholds. In order to better define the reasons for poor results, growers requested Agroscope RAC Changins, the Swiss Federal Agricultural Resarch Station re-examine these empirical estimations. Thus, a study was carried out in central Valais, Switzerland in 2000 and 2002 with the aim of measuring the influence of the two-spotted spider mite on yield, leaf photosynthesis and fruit quality of strawberries grown under plastic tunnels.

Material and methods

All the trials were made at the Centre for Arboriculture and Horticulture « Les Fougères » in Conthey (Valais, Switzerland). Various control strategies together with their impact on fruit yield and quality were compared in randomized replicated 19 x 2 m plots. In 2000, an acaricide treatment called "autumn" or post-plantation was tested with or without a complementary springtime treatment, whereas in 2002, the acaricides were applied only in the spring following plantation. A summary of the principal cultivation data is given in Table 1.

The acaricide Zenar (80 g/ha; 20% tebufenpyrad) was used in all applications for treating *T. urticae.* Pest populations were recorded as a percentage of occupation by one or more mobile stages. This is the type of control frequently used by Swiss strawberry growers. Density was estimated under a binocular microscope using a system of ranking (Guignard, unpub.) and was then used to calculate the load expressed in mite days/leaf. Yield was determined for all of the replicate plots. Sugar content measurement was considered to be the criterion of quality (Carlen et al., 2001), and so this was analysed several times during the harvest period (replicate sampling of 500g of first class fruits). °Brix values were determined using an Atago PR-1 refractometer (Kunzmann, Switzerland). Leaf photosynthetic activity was measured only in the 2002 trial. Gas exchange were measured instantaneously using a LiCor 6200 closed portable system linked to an infra-red gas analyser. In 2000, mite monitoring was compared between treatments using a *t* test and in 2002 by one-way analysis of variation (on log (x+1) transformed data)and the Tukey test ($p < 0.05$). The relationship between mite-days/leaf and parameters of yield and quality were determined by linear regression.

Table 1. Main cultivation data of *T. urticae* trials (2000 and 2002)

Year	Variety (type of plants)	Planting date	Densities plants/m^2	Treatments *	Replicates	Leaves / replicate	Harvest date
2000	Madeleine (cold-stored)	08.07.99	4.4	1. A 2. A + S	6	30	05.05 - 24.05
2002	Madeleine (Motted)	06.08.01	4	1. U 2. 1 x S 3. 2 x S	4	20	29.04 - 27.05

* A: Autumn; S: Spring; U; untreated

Results and discussion

Autumn and spring treatments

Figure 1 A illustrates the dynamics of mobile stages of *T. urticae*. No significant differences were observed between treatments before 03.05, i.e. two days before harvesting. After that time, populations in the autumn and spring treated plots were significantly lower than those treated only in the year of planting. It should be noted that the threshold level fixed at 10 to 20% of occupied leaves at the start of flowering was reached in both treatments, despite the autumn treatment being applied over the whole of the plot when 80% of leaves had been colonised. The additional acaricide spring treatment provided excellent control of *T. urticae.*

In the plots where a single treatment in the year of planting was applied, an overall intensity of attack 12 times greater was measured compared to the plots which received an extra treatment in spring, this was still a low level of mite pressure (811.6 mite-days/leaf).

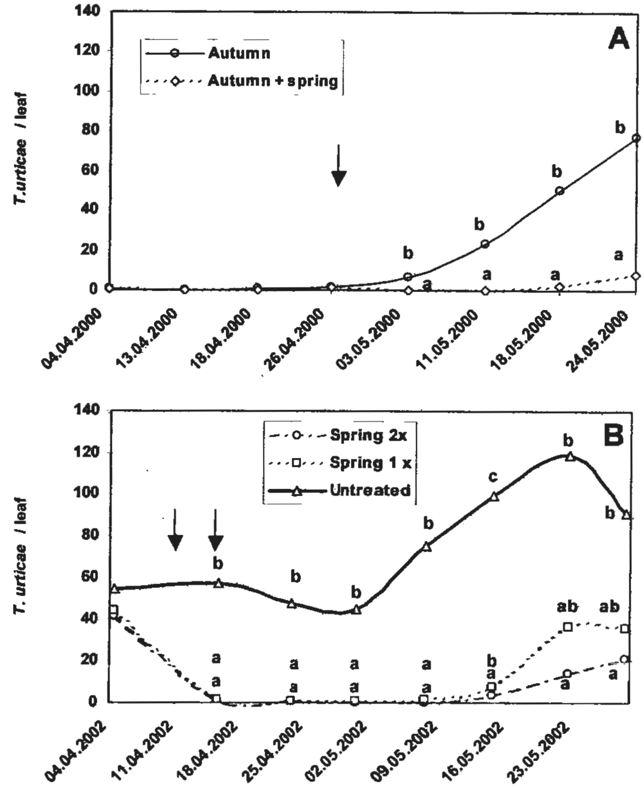

Figure 1. Population dynamics of *T. urticae*. A: 2000 trial. B: 2002 trial. Values for treatments on the same date with the same letter are not significantly different. Arrows represent the timing of acaricide applications.

Spring treatments

The first acaricide treatment applied to populations which were well above recommended limits (100% occupation in the three treatments at the beginning of April) resulted in a considerable reduction of density. Mite populations on plants which received two applications in spring in order to heavily reduce populations, differed from the plots receiving a single application only once, during the period of harvesting. The mite population flourished in the control treatment, , reaching 5 to 9 times greater density (3800 mite-days/leaf) over the entire season than in the treatments receiving one or two miticide applications. The plots that were treated twice suffered a significantly lower mite load than the two other treatments (Figure 1). A single acaricide intervention before flowering was sufficient to bring *T. urticae* well under control even in populations which had already greatly surpassed the currently recommended threshold limits.

Yield, quality and photosynthesis

Values of yield and fruit quality are provided in Table 2. As far as the 2000 season was concerned, the global yield of first class fruit did not differ statistically between treatments.

The proportion of non-commercially viable fruit rose to 20% of the total yield for the autumn treated plants, compared with 17.1% for those treated twice. Under the trial conditions, the weight of first class fruit did not differ between the treatments (Table 2). More surprisingly, the overall number of fruits per plant showed a significant difference in favour of the plants which had received two treatments. However, in view of the fact that yield and average fruit weight showed no statistical differences, this result must be interpreted carefully. In 2002, no differences among treatments were identified. The proportion of non-commercially viable fruit harvested was 14.1% of the total yield for the untreated plants, 13.3% for those treated once and 14.8% for those treated twice. No significant differences in sugar levels were detected between the different treatments in either 2000 or 2002 (Table 2).

Table 2. Yield parameters and sugar content 2000 – 2002 (mean global values)

Year	Treatments*	Fruits / plant	Weight / fruit (g)	Weight / plant (g)	Sugar Content (°Brix)
2000	A	11.8 b	19.5	212.7	8.2
	A+S	15.2 a	19.3	239.7	8.1
2002	U	19.2	17.3	394.8	8.4
	1xS	21.0	19.5	434.7	8.2
	2xS	21.6	19.2	421.6	8.3

*A: Autumn; S: Spring; U; untreated

Different letters show a significant difference between the treatments.

In contrast to the yield findings, infestations by two-spotted spider mites had a significant effect on leaf photosynthesis in 2002 (Table 3). With populations reaching 100 mobile stages per leaf and an accumulated load of approximately 2500 mite-days, photosynthetic activity in leaves of the control plants at mid-harvest was 23% lower than that in the plants receiving two spring treatments (1.1 motiles /leaf). However, two days later and in spite of being below 15%, the photosynthetic capacity of leaves in the control experiment was no longer significantly different from that in the two treatments receiving acaricide application. These results are in agreement with those of Sances et al. (1981) who recorded a lowering of photosynthetic activity as soon as two-spotted spider mite populations exceeded 100 individuals per strawberry leaf. Within the framework of our study, and despite reduced photosynthesis, no difference in the levels of sugar content in fruits was observed. This can most probably be explained by compensation, the Madeleine variety having very strong plant growth. Moreover, although yield was statistically similar to that in the pots receiving acaricides, the lower yield in the control study at mid-harvest may also have played a secondary role. With reduced yield, lower carbon requirements of fruits may indeed lead to a decrease in the photosynthetic capacity of leaves (Henriot et al., 2002).

Table 3. Leaf photosynthesis at mid-harvest in relation to the densities of *T. urticae*/leaf. Mean values of 4 replicates. 2002 Trial.

Variants	*T. urticae* / leaf	Photosynthesis (μmol $CO_2/m_2/s$)	
	14.05.02	13.05.02	15.05.02
Untreated	99.8 a	11.5 b	13.8
Spring 1 x	7.4 b	13.6 b	14.5
Spring 2 x	1.1 c	14.9 a	16.1

Different letters show a significant difference between the treatments ($p<0.05$).

Relationships between mite load, yield and quality

Table 4 indicates the values of correlation coefficients between mite load expressed in mite days/leaf and the various parameters of yield and quality which were measured. Levels of mite pressure observed within this study had no influence on yield per plant. The average number of fruits/plant was influenced by the rise in the mite load in 2000. This phenomenon was not observed in 2002, however, when loads were greater and the harvesting period longer. In 2000, the mean weight of first class fruits did not suffer any negative influence due to *T. urticae*. On the other hand, in 2002, a significant tendency towards a decrease in average fruit weight was noted. Sances et al. (1981) pointed out a decrease in the weight and number of fruits in response to about 10000 mite-days during early or late attacks. Although differences between varieties (plant strength, leaf/fruit relations, sensitivity to *T. urticae*, length of harvesting period) should also be taken into account when establishing these limiting values, these critical thresholds were far from being reached in the present study. The above remarks, together with the findings that yield and number of fruits/plant was not influenced by the levels of mite attack observed, lead to a cautious interpretation of the observed tendency for reductions in fruit weight. Sugar content of fruits was not influenced by observed mite loads.

Table 4. Correlation coefficients (r^2) between mite load expressed in mite days/leaf, and yield and quality parameters (n=12).

Mite load (mite days/ leaf) and...	r^2 values	
	2000	2002
Yield/plant	0.24	0.057
Weight/fruit	0.075	0.78*
Number of fruits	0.41*	0.0042
Sugar content	0.0065	0.059

* significant ($p<0.05$)

Conclusions

Autumn or post-plantation treatments of miticides provides significant reduction in hibernating populations, and this prevents significant development of *T. urticae* the following spring. Better control can, however, be obtained with a springtime acaricide treatment and this is true even when populations largely exceed currently recommended threshold levels. This finding suggests that the recorded cases of acaricide failure probably did not result from inadequate thresholds. Applications techniques and/or resistance to miticides might play a major role in these failure. Fields that do not receive post-planting acaricide treatments may experience high spring *T. urticae* populations. In our study, *T. urticae* populations developed exponentially during the harvesting period. Recorded mite pressure (maximum mean of 120 *T. urticae*/leaf) lead to a reduction in fruit weight and photosynthetic activity, but plant yields and sugar content in fruits were not affected. It seems that early season strawberry crops cultivated under plastic tunnels are able to withstand relatively high levels of *T. urticae* attack without any significant loss of yield, even in the presence of visible leaf damage symptoms. These results cannot be extrapolated without risk to varieties with a lower leaf/fruit ratio than cv. Madeleine or a longer harvesting period (4 to 5 weeks). The perspectives developed from this research offers encouragement for renewing investigations into biological control of *T. urticae* using predatory mites, for use in plastic tunnels.

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Open forum discussion: 'What do growers expect from research?'

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Abstract: The question 'What do growers expect from research' was addressed in an open discussion amongst delegates at the at the 4th IOBC workshop on Integrated Soft Fruit Production. A wide range of opinions was expressed by the delegates, reflecting variation in soft fruit industries, research organisations and research funding arrangements. Crop protection, registration of plant protection products and variety testing were generally considered by growers to be of high priority. Growers in some countries considered ways of reducing labour costs and protected cultivation important. Plant breeding and genetic resources were considered to be of low priority in some countries, whereas in others private breeding programmes by individual co-operatives predominated. It was generally recognised that government research funding is increasingly being devoted to public good aspects of agriculture and horticulture, especially environment, food safety and sustainability. Researchers in different institutes are motivated by widely varying drivers. In many research institutes, there is a strong requirement for researchers to maintain a good scientific publication record, steering researchers away from the more practical requirements of soft fruit grower. It is probable that many growers believe that too much horticultural research is being focussed on highly scientific subjects from which they do not see direct benefits. A co-ordinated investigation across Europe of grower research priorities would be an interesting collaborative project for the IOBC soft fruit sub-group.

Key words: crop protection, research priorities, fruit quality, sustainability, cultivar testing,

An open discussion amongst delegates at the workshop of the soft fruit sub-group debated the question 'What do growers expect from research?' The discussion was lead by Christoph Carlen from Agroscope RAC, the Swiss Federal Agronomical Research Station, Centre Les Fougères Conthey where the meeting took place, and lasted approximately 1 hour.

A wide range of opinions was expressed by the delegates which revealed a lack of a consensus amongst the participants. Attitudes were strongly coloured by different situations in different countries, both differences in the fruit industries and fruit production and marketing but also by the way research was organised and funded in different countries.

Results of a questionnaire survey of approximately 50 growers done recently in Switzerland were reported by Christoph Carlen. This survey had been premised on the need for research to be focussed on a limited number of specific topics due to reducing research funding and resources. Growers were asked to prioritise the topics for research. The survey showed that crop protection and attendant plant protection product registration were given the highest research priority, closely followed by cultivar testing, then R&D on crop production techniques including fertilisers and irrigation. Plant Quality, conservation and fruit quality were middle ranking priorities and breeding and genetic resources were considered to be the lowest priority.

These grower's research priorities was not considered to be entirely consistent with the understanding of grower's research priorities by researchers from other countries who participated in the discussion. A number of important general points were made:

1. Grower's priorities should be driven by the priorities of the market place. Multiple retailers are the dominant influence. Important market place requirements are
    - improving fruit quality
    - reducing the incidence of pesticide residues
2. Growers take a short-term view of research priorities and there is a need for researchers to take a longer term view. Actual priorities should not be over-influenced by grower's priorities. To some extent grower's priorities are influenced by the research that can be delivered.
3. Several delegates considered the need to improve plant quality was of higher priority than indicated by the Swiss survey.
4. Growers research priorities are affected by the way researchers, advisory services and growers interact. In some countries there is strong competition between researchers and advisors. In some countries private advisors have a strong influence.
5. Most delegates concurred with the high priority given by growers to cultivar testing. The lower priority given by growers to the more strategic needs of plant breeding and genetic resources are understandable. However, in some countries there is a strong grower emphasis on soft fruit breeding and many soft fruit co-operatives have their own exclusive breeding programmes. The problem of de-listing of cultivars without notice by Multiple Retailers was highlighted.
6. In many countries, research to reduce labour costs, especially for harvesting, was regarded as of high priority. Research to develop new systems of growing which present fruit to pickers more easily to reduce harvesting costs is an important priority.
7. In many countries, there is an increasing trend for cultivation of soft fruits under semi-protection. Research to develop crop production and crop protection systems for protected cropping are a high priority in some countries
8. Government research funding is increasingly being devoted to public good aspects of agriculture and horticulture, especially environment, food safety and sustainability. Global climate change, water quality and biodiversity are important topics. This research is diverting a large proportion of funding from other areas of agriculture and horticulture. Growers are, however, generally pleased to participate in such research because they understand the wider benefits.
9. An increasing proportion of soft fruit grower funding for 'research' is being devoted to pesticide registration, especially to acquiring off-label approvals. These activities are administrative rather than research, but the diversion of funds is impoverishing research.
10. Most growers believe that too much horticultural research is being focussed on molecular biology, plant genomics and other highly scientific subjects which they do not understand and from which they see little benefit. The strong resistance of the public to GM in many EU countries makes this research appear of little relevance to growers.
11. Researchers in different institutes are motivated by widely varying priorities. In many research institutes, there is a strong requirement for researchers to maintain a good scientific publication record in high impact refereed scientific journals. Unfortunately, most journals that publish agricultural and horticultural research have a low impact factor. This steers researchers into more fundamental research areas and away from the more practical requirements of soft fruit growers. In other institutes, the pressure for scientific publication is less acute and grower articles and consultancy are recognised. This latter approach is clearly more appropriate for horticultural research.

The discussion thus indicated considerable variation in the understanding of researchers on what growers expect from research. The Swiss survey was an interesting approach and there would be value in undertaking similar surveys in other countries. A co-ordinated investigation across Europe would be interesting collaborative project for the IOBC soft fruit group. However, researchers have to take into account the views of many different interests including those of governments, the public and the market place. 'He who pays the piper plays the tune'.

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Molecular techniques to determine mite predator / prey interactions on strawberry

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Abstract: Several methods, such as isoenzyme analysis and monoclonal antibody techniques, can be used to identify species and to determine prey consumption of predators. However, molecular techniques have the potential to be much more sensitive and specific than isoenzyme analysis and much quicker to develop than MAbs. We have developed a molecular technique that enables us to identify not only the prey species consumed but also the species of phytoseiid mite present. The multi-copy hypervariable non-coding ITS1 and ITS2 regions were used to detect length variation between mite species using PCR. Primers were developed that differentiated between two pest species, *Phytonemus pallidus* and *Tetranychus urticae*. These primers did not amplify DNA from phytoseiids, and were detectable for up to 18 hours after they had been consumed by predatory mites.

Primers were also designed that would amplify DNA from the five phytoseiid species (*Typhlodromus pyri, Phytoseiulus persimilis, Neoseiulus californicus, Neoseiulus cucumeris, Neoseiulus aurescens*) identified in previous field collections in strawberry, but not from the prey mites *T. urticae* and *P. pallidus*. The primers were designed to amplify a product approximately 350bp long, which did not interfere on the gel with the much smaller PCR products designed to recognise prey species in the predator's gut. The phytoseiid primers were multiplexed with *T. urticae* and *P. pallidus* specific primers. Both sets of primers successfully amplified their products, enabling us to confirm the identity of the phytoseiid as well as prey remains in its gut. The technique has been validated using phytoseiids fed on prey on leaf arenas and on potted plants as well as on mites collected from strawberry plantations.

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Pest control in blackcurrant IFP in Poland using the new neonicotinoid – thiacloprid as Calypso 480 SC

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Abstract: Calypso 480 SC (thiacloprid) was tested at the Research Institute of Pomology and Floriculture in Skierniewice (Poland) for control of some pests in black currant plantations in 1999-2003. Calypso 480 SC at a rate of 0.1 and 0.15 l/ha showed very good control of aphids (mainly *Hyperomyzus lactucae* and *Aphis schneideri*). Calypso 480 SC at the rate 0.15 and 0.20 l/ha gave good control of the black currant stem midge (*Resseliella ribis*). The effectiveness of Calypso 480 SC (0.15 l/ha) for control of the currant clearwing moth (*Synanthedon tipuliformis*) in one year's experiments was not fully satisfactory. Results were similar to those obtained with acetamiprid (Mospilan 20 SP) and worse than the results obtained with the standard fenitrothion (Sumithion Super 1000 EC).

Key words: thiacloprid, black currant aphids, *Hyperomyzus lactucae, Aphis schneideri, Resseliella ribis, Synanthedon tipuliformis,* currant pests

Introduction

Blackcurrant is an important crop in Poland. There are about 42 thousand hectares of currants and blackcurrants plantations comprise 75 % of this area. Blackcurrant crops are attacked by several pests and they cause serious damages to stems and leaves (Łabanowska, 1996). In the last few years the main pests of blackcurrants were: big bud mite (*Cecidophyopsis ribis*), two-spotted spider mite (*Tetranychus urticae*), blackcurrant stem midge (*Resseliella ribis*), currant clearwing moth (*Synanthedon tipuliformis*) and aphids. There are several other less important pests. Chemical control of these pests is quite difficult, especially in plantations, where an Integrated Fruit Production (IFP) programme is used (Gajek et al., 1998, Łabanowska, 2001, Łabanowska et al., 2002). While fenitrothion and phosalone are recommended only for control of certain pests, there is a need to find new, selective insecticides and acaricides that are safe to beneficial organisms that can be used in blackcurrant IFP. Among the prommising chemicals is thiacloprid as Calypso, a novel and highly active chloronicotinyl insecticide with a broad spectrum of efficacy against insects (Elbert et al., 2002). In apple IFP the new group of neonicotinoid insecticides is recommended to control *Tortricidae*, codling moth and others (Olszak & Płuciennik, 2001; 2002). They also gave good control of aphids and pear leaf blister moth on apples (Maciesiak & Olszak, 2002; 2003). Some of them also gave good results in control of the strawberry blossom weevil on strawberry and the rose tortrix moth on black currant (Łabanowska, 2002; 2003). Until now only acetamiprid as Mospilan 20 SP is registered against the above pests on strawberries and on currants.

The aim of this work was to estimate the usefullness of thiacloprid as Calypso 480 SC for control of the following pests on black currant: aphids, black currant stem midge and currant clearwing moth. Some selected results are shown in the Tables 1 - 3 and Figures 1 - 2.

Material and Methods

The experiments were carried out in 1999-2003 at the Research Institute of Pomology and Floriculture in Skierniewice (Poland) on black currant plantations.

Aphids (Aphididae) control

Two experiments were conducted in the spring of 2002-2003 on 10-11 year-old blackcurrant plantations of the cultivar 'Titania' in Klementynowo, near Włocławek. Insecticides to control aphids were used once, just after blossom of currants. At this time the aphids were feeding on the youngest leaves at the tops of the shoots. A knapsack "Stihl" motor sprayer was used to apply the tested compounds in a volume of spray liquid of 750 l per ha. The plots were 100 m^2 each. Pirimicarb as Pirimor 500 WG was used as a standard chemical. The effectiveness of insecticides was evaluated at 3, 7 and 14 days after treatment. The shoot tops (4 replicates of 25 items) damaged by aphids were randomly picked and the number of alive insects was counted in laboratory, under a stereomicroscope. The results were analysed statistically on data transformed according to logaritmic formula $Y = \log(x+1)$, where x was the number of alive aphids per 25 shoots.

Currant clearwing moth (**Synanthedon tipuliformis**) *control*

Detection of currant clearwing moth and observation of the adult flight period was based on pheromone traps. Spray treatment was applied during the mass flight of adults (Fig. 1), in a part (about 500 m^2) of a larger currant plantation, about 10-12 years old, cv. Ben Lomond or Ojebyn. A tractor sprayer "Turbine" was used to apply the spray solution at the rate of 900 l/ha. The effectiveness of insecticides was evaluated in October. The number of one-year-old shoots damaged by larvae of currant clearwing moth was counted.

Blackcurrant stem midge (**Resseliella ribis**) *control*

The experiments were conducted in 1999, 2000 and 2002 on 10-12 year old currant plantations of the cultivars 'Titania' and 'Ojebyn' in Mokra Lewa, Trzcianna and Godzianów (central part of Poland). The egg laying period of black currant stem midge was determined by observing eggs laid into artificially made cuts in one-year-old shoots (Fig. 2). The spray treatments were applied at the peak of egg laying by the first generation of females and then against the second generation, just after harvest. The size of the plots and the sprayer were the same as used in the aphid control experiments. The effectiveness of insecticides was evaluated in the field at the end of September or in October. The number of damaged one-year-old shoots on bushes was estimated. The results are shown on Figures 1 - 2 and Tables 1 - 3.

Results and discussion

Aphids (Aphididae) control

Calypso 480 SC (thiacloprid) used at the rate 0.1 and 0.15 l/ha showed very good results in controlling aphids on blackcurrant (Table 1). At the end of experiment, 14 days after treatment, the mortality of aphids on Calypso 480 SC treated bushes was very high, 99.8-100%. The results obtained with Calypso 480 SC were similar to those obtained with the standards acetamiprid (Mospilan 20 SP) and pirimicarb (Pirimor 500 WG). On untreated bushes the population of aphids was very high, 190-340 per shoot, at the beginning of the experiment. Results gained with Calypso 480 SC confirmed earlier findings, where it was used to control apple aphids (Maciesiak & Olszak, 2002) and other aphids (Elbert et al., 2002).

***Currant clearwing moth* (Synanthedon tipuliformis)**

Calypso 480 SC at the rate 0.15 and 0.2 l/ha used 2-3 times gave quite good control of the currant clearwing moth (Table 2). The results were similar to those obtained with acetamiprid (Mospilan 20 SP), but a little bit poorer compared to fenitrothion (Sumithion Super 1000 EC) used as a standard. These results confirmed those obtained in controlling other moths (Olszak & Płuciennik, 2002; Łabanowska, 2003; Maciesiak & Olszak, 2003).

***Black currant stem midge* (Resseliella ribis)**

Calypso 480 SC at the rate 0.15 or 0.2 l/ha used 2-3 times, during the egg laying period of black currant stem midge on the youngest shoots showed good control of this pest (Table 3). The efficacy of Calypso 480 SC was similar to that obtained with the standard chemicals acetamiprid (Mospilan 20 SP), etofenprox (Trebon 10 SC) and chloropyrifos + cypermethrin (Nurelle D 550 EC). On the bushes treated with Calypso 480 SC the number of damaged shoots was about 83-90% lower compared to the untreated controls (check). On the untreated bushes the black currant stem midge damaged from 7 to about 37% of shoots, depending on the year and the field (Table 3).

Table 1. Effectiveness of Calypso 480 SC for the control of the aphids (*Aphididae*) on black currant (Klementynowo 2002/2003).

Insecticides (active ingredient)	Rate l/kg/ha	No. of aphids per shoot tip after 7-14 days					
		2002			2003		
		3	7	14	3	7	14
Calypso 480 SC (thiacloprid)	0.1	0.9 bc*	2.9 b	0.3 c	36.0 c	6.4 c	0.03 a
Calypso 480 SC	0.15	0.3 b	0.5 ab	0.1 b	5.2 b	3.6 bc	0.0 a
Mospilan 20 SP (acetamiprid)	0.125	2.3 c	1.1 b	0.7 d	1.7 ab	2.1 abc	0.1 a
Pirimor 50 WG (pirimicarb)	0.75	0.0 a	0.0 a	0.0 a	0.4 a	0.3 a	0.0 a
Control– untreated	-	189.5 d	283.3 c	169.1 e	339.5 d	193.3 d	14.0 b

Date of treatments: 3 of May, 2002 and 16 of May, 2003.

* Means followed by the same letter are not significantly different at the 5% level (Duncan's multiple range t-test)

Table 2. Effectiveness of Calypso 480 SC for the control of the currant clearwing moth *Synanthedon tipuliformis* .

Insecticides (active ingredient)	Rate l/kg/ha	Damaged shoots in % %		Efficacy %
		Muchnice	Końskowola	
Calypso 480 SC (thiacloprid)	0.15	27.0 b	20.0 a	57-74.4
Calypso 480 SC	0.2	-	19.0 a	74.4
Mospilan 20 SP (acetamiprid)	0.15	28.0 b	-	54
Sumithion Super 1000 EC (fenitrothion)	1.125	8.0 a	12.0 a	88-84
Control - untreated	-	60.0 c	72.0 b	-

Date of treatments: Muchnice - June 6 and July 1, 2002; Końskowola - June 3 and 16 and July 24, 2003.

* Explanations see under Table 1

Table 3. Effectiveness of Calypso 480 SC for the control of the black currant stem midge *Resseliella ribis.*

Insecticides (active ingredient)	Rate l/kg/ha	Damaged shoots in %%		
		Mokra Lewa	Trzcianna	Godzianów
Calypso 480 SC (thiacloprid)	0.15	-	-	3.7 a
Calypso 480 SC	0.2	0.9 a	0.9 a	-
Mospilan 20 SP (acetamiprid)	0.2	0.2 a	0.6 a	-
Mospilan 20 SP	0.125	-	-	6.2 b
Trebon 10 SC (etofenprox)	0.9	0.4 a	0.6 a	-
Nurelle D 550 EC (chloropiryfos+cypermethrin)	1.5	-	-	6.0 b
Control – untreated	-	6.9 b	11.1 b	36.7 c

Date of treatments: Mokra Lewa - May 26 and July 19, 1999; Trzcianna - May 20 and June 6, 2000; Godzianów - May 20, June 7 and July 8, 2002.

* Explanations see under Table 1

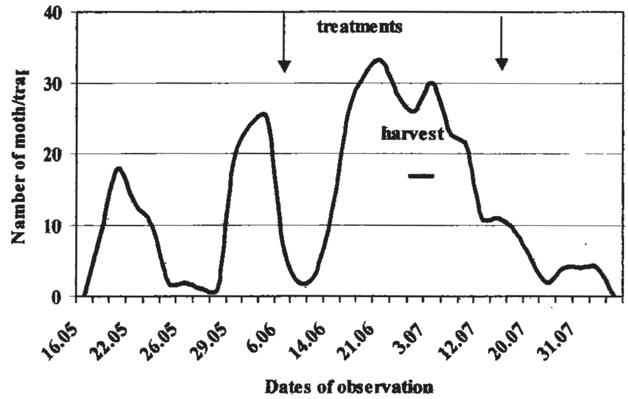

Figure 1. Currant clearwing moth - *Synanthedon tipuliformis* captured on pheromone traps (Muchnice, 2002).

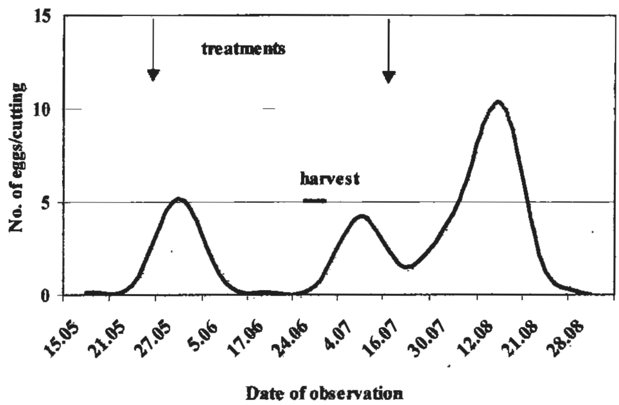

Figure 2. Numbers of eggs laid by blackcurrant stem midge *Resseliella ribis* (Godzianów 2002).

Conclusions

1. Calypso 480 SC (thiacloprid) at the rate 0.1-0.15 l/ha gave very good control of aphids.
2. Calypso 480 SC at the rate 0.15-0.2 l/ha gave good control of the blackcurrant stem midge and satisfactory control of the currant clearwing moth on blackcurrant.
3. Calypso 480 SC will be very useful in the IFP program on currant.

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Adopting Integrated Pest Management for the raspberry beetle, *Byturus unicolor* Say (Coleoptera: Byturidae), for Washington State red raspberries

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Abstract: The raspberry beetle, *Byturus unicolor*, is a major fruit contaminant of red raspberries in North America. Currently, over two-thirds of Washington State growers use calendar-based, non-IPM methods for controlling raspberry beetles. A two-year grant project has been embarked upon to assist growers' adoption of new pest monitoring and decision-making skills. Using a new raspberry beetle pest-monitoring tool, the Rebell® Bianco trap, developed at the Swiss Federal Research Station of Wädenswil, eight raspberry growers representing over 800 hectares of red raspberry in Washington State have participated in this project. Using trap data, all growers agreed that IPM is possible for managing raspberry beetles. Two growers chose not to apply chlorpyrifos as a preventative measure for reducing raspberry beetle contamination of fruit. Growers were able to relate trap data information, estimated fruit damage and management decisions in order to build a functional threshold for raspberry beetle. Additionally, growers gained new knowledge and understanding of raspberry beetle population and spatial dynamics, biology and crop border interactions.

Key words: *Byturus unicolor*, raspberry, integrated pest management

Introduction

The raspberry beetle, *Byturus unicolor*, is an insect contaminant of machine-harvested raspberries (*Rubus idaeus*). Tolerances for this contaminant range from zero tolerance to moderate depending on the berry grade and production method. In Whatcom County, Washington State, two-thirds of raspberry growers apply an insecticide at 5% bloom, just prior to introducing pollinators, for control of *B. unicolor* (Menzies, 1999). The insecticide application is one of the remaining few calendar-based treatments in the developing IPM program for raspberries. This insecticide application is preventative; little information about pest occurrence, abundance or distribution is available for growers to aid in the decision-making process of IPM.

Growers currently use diazinon as the standard insecticide for raspberry beetle prevention. The United States Environmental Protection Agency, under the Food Quality Protection Act of 1996, has sought to reduce risks associated with organophosphates, such as diazinon, through greater restrictions on use and cancellations of registrations. The red raspberry industry, along with many others, is looking towards a reduced reliance on diazinon in preparation of its cancellation and/or use reduction in red raspberry production.

This project engaged raspberry growers in developing IPM methods for raspberry beetles, using the successful model of the Nooksack IPM project (Menzies & MacConnell, 1998). Using a new raspberry beetle pest-monitoring tool, the Rebell® Bianco trap developed at the Swiss Federal Research Station of Wädenswil (Gordon & Woodford, 2000), eight raspberry growers representing over 800 hectares of red raspberry in Whatcom County, Washington State participated. Each growers' production methods, based on past decisions for managing raspberry beetles, were categorized. The production methods were: organic (no synthetic pesticides), conventional (with Bifenthren pre-harvest application) with a diazinon application specifically for raspberry beetles, and conventional (with Bifenthren pre-harvest application) without a raspberry beetle treatment.

Material and methods

Raspberry Beetle Flight

B. unicolor abundance and flight activity were monitored in commercial raspberry fields throughout the 2003 growing season. Raspberry beetle flight appears to originate outside conventionally produced raspberries. In past monitoring programs, beetles were encountered more at field margins. Based on these observations, traps were placed along field perimeters to identify trends of immigration and understand border interactions.

Traps were placed at various densities along the perimeter of each field, every 10 (30 meters), 20 (60 meters) or 40 (80 meters) rows, depending on the area of the field. The number of traps placed in the field ranged from 10-40 per field. [How many traps per field on average?] Traps were set on the top wire of the raspberry trellis (1.5 meters high) and one post-length (10 meters) inside the field. Newly trapped adult beetles were counted weekly and the information was provided to the grower. Traps were replaced every 3 weeks. Over 163 traps for all eleven fields were maintained from mid-April to mid-August.

Alternative Host Survey in Field Borders

Because immigrating populations may influence monitoring data and trap placement, six *Rubus* species were surveyed at seven locations adjacent to raspberry fields for *B. unicolor* populations. Up to one hundred berries from each host plant at each location were collected and evaluated for raspberry beetle presence or damage. Data were analyzed using an ANOVA and Dunn's pair-wise comparison for unequal sample sizes.

Fruit Husk Evaluation

During August and September, after harvest, an estimate of fruit infestation was made. At each trap site, 40 fruit laterals were collected, 20 from each side of the trap site. Lateral collections were made 4 rows (12 meters) away from the trap site to minimize trap influence on the husk infestation rate. Laterals were brought into the lab and evaluated for evidence of husk feeding by raspberry beetles. The number of damaged and undamaged husks was recorded.

Results and discussion

Representative flight patterns for each growing method are provided in Figure 1. The first detectable flight occurred in the first week of May. *B. unicolor* abundance was higher in organic fields compared to conventionally managed fields. Diazinon applications in late May reduced adult beetle activity for most growers that used this treatment. Beetles continued to be trapped in the field until the end of July, well into harvest.

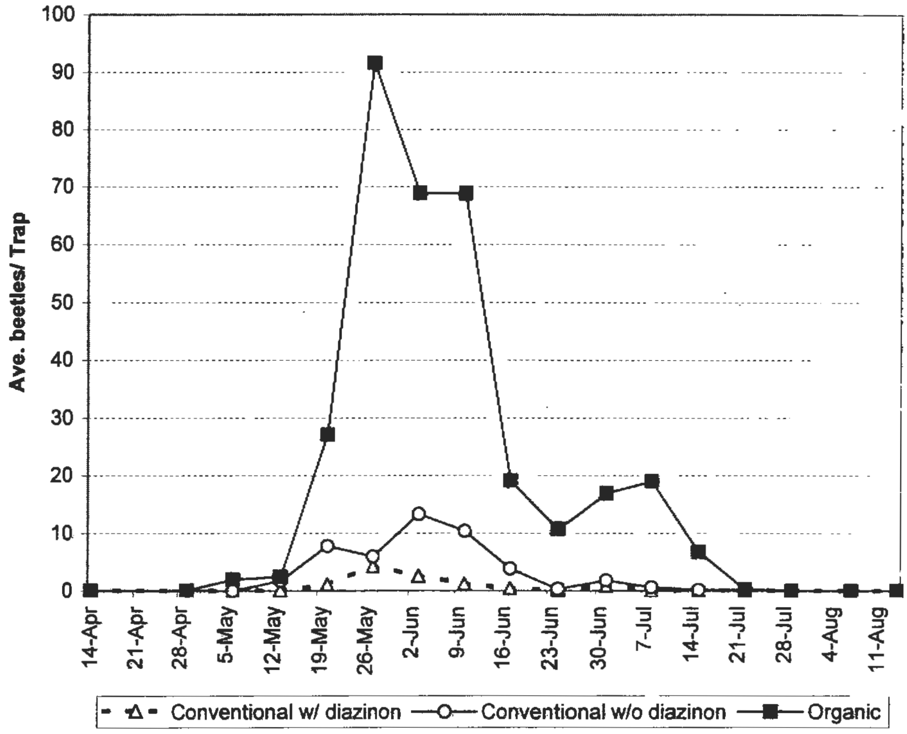

Figure 1. Representative flight activity (average number of beetles per trap per week) at three different fields with different production methods.

Figure 2 illustrates the average number of infested berries for each *Rubus* species in field borders. Beetle larvae and damage were found on all six species, with *Rubus parviflorus* (thimble berry) housing the highest reservoirs of *B. unicolor*. Consideration of the field border while placing traps is important for future monitoring of these fields. Based on these findings, trapping areas of raspberry fields that are bordered by alternative *Rubus* hosts will provide more accurate representation of beetle immigration into fields.

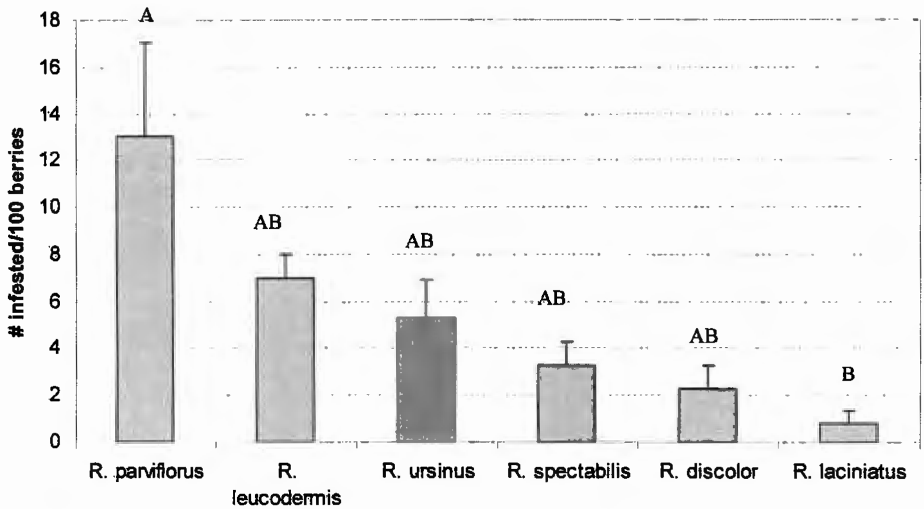

Figure 2. Number of infested fruit per 100 berries +/- S.E. Bars with the same letter are not significantly different (Dunn's pair-wise comparison of unequal samples P<0.003).

Table 1 contains the average number of beetles at 5% bloom (May 22), total number of beetles after the flight period and the percentage of fruit infested by the season's end. Organic growers had higher infestations than conventional growers. Growers that used a diazinon application had similar fruit damage as those growers that only use a pre-harvest bifenthrin treatment. Relating pre-bloom beetle numbers, management decisions and percent husk damage aided understanding of how to use monitoring information for making pest control decisions.

For the first time, Whatcom County raspberry growers were able to monitor *B. unicolor* abundance and flight activity. Growers also were able to measure the impact of management strategies and production methods on the population of raspberry beetles. Understanding the relationship between beetle numbers, management decision and percentage of fruit infestation is important for each grower's ability to develop a threshold for raspberry beetles (Höhn et al., 1995).

During a mid-season survey, growers expressed that having monitoring data aids their ability to make management decisions and accurately time treatments. Four of the conventional growers used timed diazinon applications. One grower used a selective diazinon application only for areas of the field with high beetle captures in the monitoring traps. One grower continued to monitor and felt that beetle numbers were under the threshold for treatment.

Continued development and adoption of IPM for raspberry beetles will empower growers to make decisions without relying on preventative treatments. Using monitoring tools and evaluating management practices will allow growers to develop thresholds that accommodate their specific standards. It is the authors' expectation that adopting this behaviour will reduce unnecessary diazinon applications without compromising fruit quality.

Table 1. Average beetles per trap, % of husks infestation and treatment decision.

	Average Total Beetles per trap until May 20-22	Average Total Beetles per trap for entire season	Average % of husks with infestation damage for entire field	Treatment Decision
Farm 1	25.00	306.50	7.050	Organic
Farm 2: Field 1	3.25	20.63	0.00019	Diazinon
Farm 2: Field 2	0.56	2.56	0.094	Diazinon
Farm 2: Field 3	2.17	7.42	0.103	Diazinon
Farm 3	0.50	17.08	0.000	Limited Diazinon Treatment
Farm 4	0.36	5.93	0.000	Diazinon
Farm 5	1.30	5.50	0.033	Diazinon
Farm 6	31.63	501.56	6.015	Organic
Farm 7: Field 1	0.75	7.58	0.088	No Diazinon
Farm 7: Field 2	9.54	46.45	0.039	No Diazinon
Farm 8	4.29	22.57	0.205	Diazinon

Acknowledgements

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Developing a "lure and kill" system for raspberry beetle, *Byturus tomentosus*, in *Rubus* production.

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Abstract: The raspberry beetle, *Byturus tomentosus,* is an important pest of raspberries in the UK and continental Europe. Until now, the main method of controlling this damaging pest was the use of insecticides. However, consumer pressure and perceived risk to the environment, have led to the EU reviewing the pesticides available to control agricultural pests, weeds and diseases. With the reduction in the number of insecticides available, growers are seeking alternative ways of controlling pests that do not rely as heavily on insecticide use. One such method being developed at SCRI is a "lure and kill" system for raspberry beetles in raspberry production. This paper shows the result of a set of experiments which tested the efficiency of combining an identified flower attractant with a standard white non-UV reflective trap on the number of beetles caught. There was a 30 fold increase in the number of beetles caught on the enhanced traps. A comparison of the levels of berry damage in areas surrounding the traps showed that there was no significant difference in the level of damage in the areas containing the standard trap and the areas containing the enhanced trap

The overall purpose of this research is to use these visually attractive traps, in combination with the identified key flower volatiles, to develop a method of reducing the amount of damage caused by the raspberry beetle below a level that requires the input of insecticides. This should enable conventional growers to reduce insecticide inputs to acceptable levels and offer new control strategies for organic growers.

Key words: *Byturus tomentosus, Rubus*, "lure and kill" system, floral attractants, traps

Introduction

The raspberry beetle, *Byturus tomentosus* Degeer, (Coleoptera: Byturidae) is the most important pest of commercial crops in the UK and in many parts of continental Europe. The adults emerge in the late spring and feed on the developing flower buds. Once the flowers are open, the adults mate and the females lay their eggs in the flower. The main damage is caused by the larvae tunnelling into the developing fruit (Taylor & Gordon, 1975). At present, control of this pest in commercial plantations involves applying an insecticide to the ripening fruits a few weeks before harvest which kills the newly emerged larvae (Gordon et al., 1997).

Raspberry beetles are known to use visual and olfactory cues to locate raspberry flowers (Woodford et al., 2003). After a range of coloured sticky traps were tested, it was found that white, non-UV reflective traps were the most effective. Höhn et al. (1995) suggested that the numbers of beetles caught on the sticky traps was related to the amount of beetle damage observed in the plantation and that in some instances the use of sticky traps could help growers avoid the need for routine applications of insecticides.

A recent EU funded project, 'Reduced Application of Chemicals in European Raspberry Production' (RACER) tested the use of these traps for monitoring raspberry beetles (Woodford et al., 2003). Adult raspberry beetle activity was monitored at twenty three sites in Scotland, Switzerland and Finland and there was found to be a great variation in the numbers of beetles caught between sites and years. The extent of damage was not closely related to the number of beetles caught although there was very little damage at sites with fewer than 5 beetles caught per trap before flowering.

This beetle monitoring and trapping system was taken further at SCRI by Birch et al. (1996) with the identification of two flower volatiles which are recognised by the beetles and involved in their attraction to flowers. This required the use of combined automated thermal desoportion-gas chromatography-mass spectrometry (GC-MS) with an electro-antennogram (EAG) to identify volatiles emitted from raspberry flower (Robertson et al., 1993; 1994). EAG assays combined with behavioural studies in olfactometers and wind tunnels identified two attractants (coded chemical A and chemical B) for testing under field conditions.

This paper describes a series of experiments which tested combinations of visual and olfactory cues, undertaken to observe the efficiency of the identified volatiles at attracting raspberry beetle in a commercial plantation. The amount of husk damage was assessed to indicate whether the amount of beetle damage could be related to the number of beetles caught and whether the lure system was sufficient to reduce damage by raspberry beetle to a level similar to that achieved with insecticides .

The current glass dispenser for the test volatile chemical is not sufficient if this system is to be released for use by commercial growers. At present, the chemical in the dispenser has to be replaced once or twice a week. This is not acceptable due to health and safety requirements in the use of the chemical and because of excessive handling time. A biological control company, AgriSense-BCS Ltd, has designed a new slow release dispenser which is sealed and made of slightly porous plastic. This allows slower release of the chemical over several weeks under field conditions. Our experiments also investigated how this new dispenser compared in evaporation rate and attractiveness to the original glass dispenser.

Material and Methods

Comparison of standard sticky traps and chemically enhanced traps

The site used for the experiment was located in Blairgowrie, Scotland (OS NO 135 435). The white non-UV reflective traps (AgriSense-BCS Ltd) were used either on their own or in combination with a 2 ml amber glass vial containing 1.7 ml of chemical B and sealed with a plastic stopper, modified by the addition of a 2mm diameter hole housing a cotton wick to aid in the evaporation of the volatile. Two traps with volatile chemical added (enhanced traps) and two traps without volatile (standard traps) were positioned along one row of the raspberry plantation. A distance of 40 m was left between the two traps types, to avoid the odour plume affecting the efficiency of the standard traps. A distance of 20 m was left between the traps of the same type. This trap placement design was repeated in four areas of the plantation, resulting in a total of eight enhanced traps and eight traps standard traps. The traps were put in position during early green bud stage (1st May 2003) and were changed once a week until after the start of flowering (19th June 2003). The vial with test chemical was also replaced once a week. The sticky traps were wrapped in cling film and stored at 4 °C until the number of beetles could be counted.

Comparison of beetle numbers and beetle damage

One area containing two standard traps and two enhanced traps was covered with a large polythene sheet to protect it from insecticide application against the raspberry beetle. One hundred husks were collected from a section of the row 5 m either side of each sticky trap. These husks were collected not more that 24 hours after fruit harvest, to ensure that they were still fresh. To compare the amount of damage observed in the area that had been protected from insecticide application with an area that had been subject to insecticide application, 100 husks were sampled from an area of the plantation that had not been protected. These husks were frozen until analyses could be completed. Replication was not possible because of restrictions on experimental use within the grower's field.

Trial of a new dispenser
A new dispenser made from porous plastic was developed by AgriSense-BCS Ltd. The evaporation rate from this new dispenser (containing 2.5 ml of each test volatile) was compared with the evaporation rate from the amber glass dispenser with wick (containing 1.7 ml of volatile), for both attractants individually (chemical A and chemical B) To obtain comparable results, the dispensers were tied to a support wire, 70 cm from the ground in the plantation, which is a similar location to where they would be found if they were being used in combination with the sticky traps. There were four replications of the four combinations of dispenser type and volatile chemical (A or B). These dispensers were weighed on several days, to obtain the evaporation rate during the 13 days of the experiment.

The relative attractiveness of the new dispensers was assessed by comparing the number of beetles caught using the four dispenser combinations to captures in standard sticky traps as the control.

Results and Discussion

Comparison of standard traps and chemically enhanced traps
During the first two weeks of the experiment no beetles were caught on either the enhanced traps or on the standard traps (Figure 1). An increase in the number of beetles caught in the third week suggests that this was the main period of beetle emergence.

The greatest number of beetles were caught on the enhanced traps in the third week (118 beetles on 8 traps) and the comparison with the number of beetles caught on the standard traps in the same week (6 beetles on 8 traps) suggests that the use of the volatile attractants increases the number of beetles caught by up to 30 fold. The number of beetles caught on the enhanced traps remained high in the fourth week. The sudden drop in week five coincided with the beginning of raspberry flowering. This drop in numbers suggests that during flowering the traps are not as attractive, because vegetation obscures the trap and volatiles released from the flowers compete with the test volatiles released from the vials.

Comparison of beetle numbers and beetle damage
Overall, there was very little beetle damage observed in the three areas sampled. In the area surrounding the standard traps there was no beetle damage recorded. This was the same in the area that had received an application of insecticide against the raspberry beetle. In the area surrounding the enhanced traps there was 1.5 % damage. Although the results suggests that the enhanced traps may in fact increase the amount of berry damage, the sample size was not large enough to give a true indication of the effect of the enhanced traps and the difference was not statistically significant in this experiment.

In plantations exposed this number of beetles, the use of the standard traps may be enough to control the number of beetles and therefore an application of insecticides would not be required.

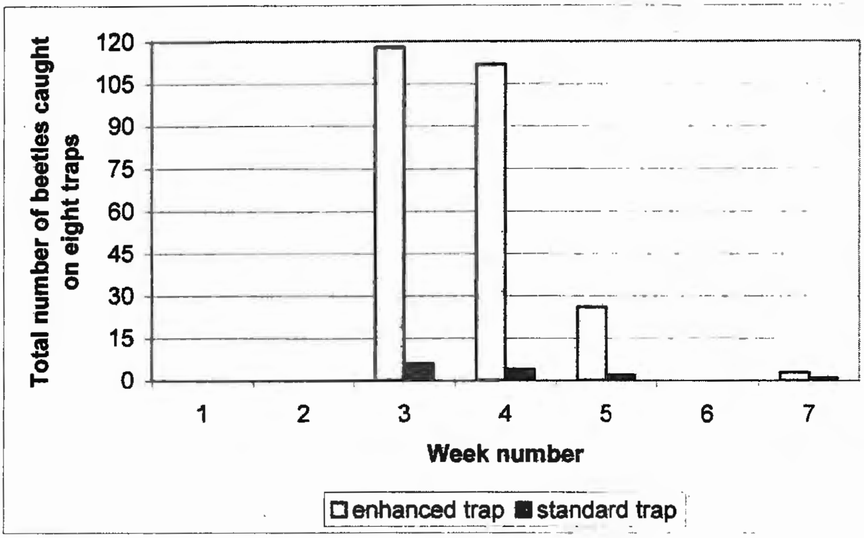

Figure 1. The total number of *B. tomentosus* beetles caught each week on the enhanced traps and the standard traps.

Trial of a new dispenser

The evaporation rate of both chemicals from the plastic dispensers was much slower than from the glass dispensers with wicks (Figure 2). After 12 days, there was 91% left of chemical A and 90% of chemical B. This is in contrast to the glass vials, where there was only 34% left of chemical A after 6 days and 0% of chemical B left after 6 days. These results suggest that at the present rate of evaporation, the chemicals in the plastic vials would last for approximately 17 weeks, which is much longer than the time span over which the lures would be required. Further work will indicate the level of release required to attract the beetles and trap them to a level below that which causes economic damage.

There were more beetles caught using chemical B (Figure 3), suggesting that this chemical is more attractive than chemical A under these field conditions. For both chemicals, the number of beetles caught on the traps used in combination with the glass dispensers was greater than the number found on the trap with the corresponding plastic dispenser, but these differences were not statistically significant. The slower evaporation rate from the plastic dispensers may be reducing the numbers of beetles attracted to the traps compared with the glass dispensers. The evaporation rate of the chemicals from the prototype plastic dispenser could be increased and still last the duration required to monitor *B. tomentosus* in Scottish *Rubus* production (6-8 weeks). This increase in evaporation rate may raise the number of beetles trapped.

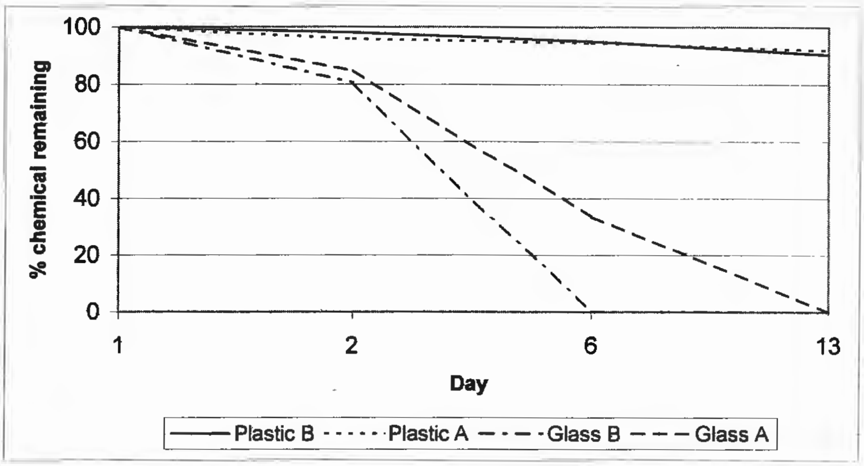

Figure 2. Evaporation rate of chemicals A and B from plastic or glass dispensers.

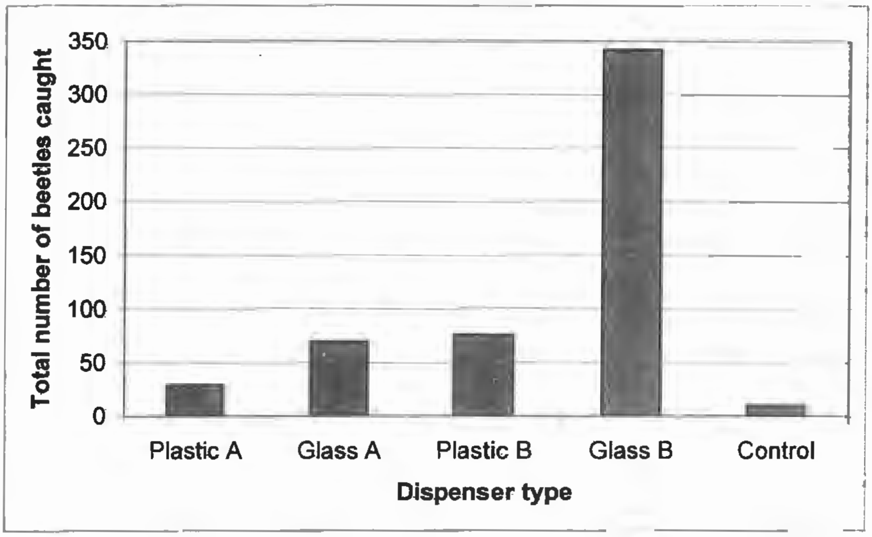

Figure 3. Total number of *B. tomentosus* beetles caught over two weeks using sticky traps in combination with chemicals A and B in the plastic dispenser (P) and the glass dispenser (G).

At present, these techniques could be developed into an enhanced monitoring system for the raspberry beetle, in order to target optimal timing of insecticide sprays when numbers per trap exceed a threshold. If this proves successful, with the appropriate registration, it could be modified further into a non-insecticidal management strategy for the raspberry beetle.

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Economic evaluation of two plant protection programmes, Standard and IPM, in blackcurrant production in Poland

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Abstract: The economics of standard and IPM blackcurrant protection programmes were studied. The investigation was conducted at a plantation located at the Research Station in Dabrowice (Central Poland). The plantation was divided in two equal blocks, standard and IPM. In one block, routine plant protection was provided whereas in the second block IPM rules were followed. During the whole production period all material and labour costs were noted. After the harvest, the yields and the incomes obtained form both blocks were compared. In 2000 and 2001 berries were picked manually whereas in 2002 and 2003 a Polish Arek currant harvester was used. During the four years of the study the IPM programme was more economical than the standard in only one season. This was mainly due to the yields from the IPM block generally being lower than from the standard block. However, the price for the fruit from both blocks was the same. In some countries, fruit produced using IPM programmes obtains a better price for the producers. Hopefully a similar situation will develop in the Polish fruit market, making plantations managed under IPM programmes more economically viable.

Key words: revenue, production costs, IPM, blackcurrants

Introduction

Poland is one of the world leaders in currant production, and is the biggest exporter of these soft fruits. The blackcurrant is much more important than the white and redcurrant in Poland. The most commonly grown varieties are Ben Lomond, Titania and Ojebyn, andvery promising Polish varieties are increasingly planted, such as Tiben and Tisel. These new varieties are even more productive and will probably soon replace commercial plantations of the old varieties.

IPM production is getting more and more important on world currant markets, both for processors and consumers. It will be increasingly difficult to sell fruits grown with the standard plant protection model. Although most currant growers in Poland use fewer chemicals that their colleagues in the Western Europe, residues of some pesticides can be occasionally be found. IPM is not popular yet among Polish currant growers, so this practice has to be introduced widely on Polish plantations, with education so growers know the difference in costs between standard and IPM production. This will be particularly important in the beginning, when implementing IPM production practices could yield very low benefits or even cause losses.

The main aim of this study was to compare the economics of Standard and IPM programmes in blackcurrants in relation to production profitability.

Material and Methods

The comparison of economical efficiency of two methods of blackcurrant production was carried out as a biological experiment established in Dabrowice on a 1.3 hectare planting. One half of the area (0.65 ha) was devoted to Standard Plant Protection Programme and the second half to IPM Plant Protection Programme. The conditions of blackcurrant production in the experiment were the same as commercial plantations. Only the scale of the production was smaller, and this caused higher indirect labour costs and lower labour productivity. To eliminate this problem the average values of labour requirements, obtained on commercial farms, were used in the evaluation of labour costs. The plantation for the experiment was established in the autumn of 1998. For each plot 3200 black currants of cultivar 'Ben Lomond' were planted with the spacing 4 m x 0.5m. The material and labour costs were counted separately for both programs to obtain the costs of the black currants production. As well as the yields during the period of harvesting. The fixed costs were counted as a percent of the real fixed costs for the Experimental Station.

Results

Yields and profits in 2000, the first period of the experiment after establishing of the plantation, were very low and the investments very high. Therefore the results from the years 2001, 2002 and 2003 are presented in this study. Each year is represented by one table where all expenses are reported. First, two columns consist of the real numbers from the experiment area 0.65 hectare, next two columns show these numbers converted to costs per hectare and the last two show expenses per hectare in Euros.

In the year 2001, low yields on young bushes caused high costs of production. The revenue per hectare (Table 1) was -462.55 Euros per hectare in the Standard Block comparing to as much as -677.46 Euros in IPM Block. The unit production cost per kilo was 0.65 Euro and 0.77 Euro, respectively.

Due to higher yield, losses in the year 2002 were much less than the year before in both Standard and IPM Blocks (Table 2). In the Standard programme, a rate of -110.19 Euros per hectare was calculated and -260.98 Euros per hectare was calculated in the IPM programme. The unit costs were much lower, as well as the differences between them, than the previous year. This finding indicates that the IPM programme costed less than in the previous year.

During 2003, production on both blocks was profitable (Table 3), and the difference between the IPM and Standard programmes was much lower than in the previous two years.

Conclusions

- Overall, blackcurrant production was more expensive using IPM methods than using a standard production programme, but the difference in the total costs per hectare was very small, because of the low proportion of the total costs used for chemicals.
- Probably the Integrated Pest Management programme had a big influence on the yields of blackcurrants in Dabrowice, and caused lower profits. Especially big differences were noted in years 2001 and 2002, but as the bushes got older, the differences between programmes were smaller.
- The future results from the experiment will show whether differences in yields between the programs will be eliminated or will appear again.
- There is no price difference for fruits grown under IPM or Standard production rules in Poland. Further development of fruit markets and as a result better prices for blackcurrants grown with IPM Standard will be necessary to help with the adoption of IPM by Polish blackcurrant growers.

Table 1. Production costs and revenues for black currants grown under Standard and IPM production programmes in 2001.

Item	Polish zloty/0.65 Ha		Polish zloty/ Ha		Euro/ Ha	
Block	Standard	IPM	Standard	IPM	Standard	IPM
Yield in kilograms	2465.80	1841.00	3797.33	2835.14	3797.33	2835.14
Labour	2489.22	1926.90	3833.40	2967.43	1008.79	780.90
Labour (machine)	585.00	585.00	900.90	900.90	237.08	237.08
Total material costs	1267.73	1110.76	1952.30	1710.57	513.76	450.15
Chemicals	1041.73	884.76	1604.26	1362.53	422.17	358.56
- Pesticides	732.16	575.19	1127.53	885.79	296.72	233.10
- Herbicides	309.57	309.57	476.74	476.74	125.46	125.46
Fertilizers	226.00	226.00	348.04	348.04	91.59	91.59
Operating costs	4341.95	3622.66	6686.60	5578.89	1759.63	1468.13
Fixed costs	1731.00	1731.00	2665.74	2665.74	701.51	701.51
Total costs	6072.95	5353.66	9352.34	8244.63	2461.14	2169.64
Gross net product	4931.60	3682.00	7594.66	6104.56	1998.60	1492.18
Revenue	-1141.35	-1671.66	-1757.68	-2140.07	-462.55	-677.46
Costs per kilogram	2.46	2.91	2.46	2.91	0.65	0.77

Table 2. Production costs and revenues for black currants grown under Standard and IPM production programmes in 2002.

Item	Polish zloty/ 0.65 Ha		Polish zloty/Ha		Euro/Ha	
Block	Standard	IPM	Standard	IPM	Standard	IPM
Yield in kilograms	2238.00	1982.00	3446.52	3052.28	3446.52	3052.28
Labour	672.84	626.76	1036.17	965.21	252.73	235.42
Labour (machine)	982.50	982.50	1513.05	1513.05	369.04	369.04
Total material costs	1130.90	1066.42	1741.58	1642.28	424.78	400.56
Chemicals	891.34	826.86	1372.66	1273.36	334.80	310.58
- Pesticides	560.11	495.63	862.57	763.27	210.38	186.16
- Herbicides	331.23	331.23	510.09	510.09	124.41	124.41
Fertilizers	239.56	239.56	368.92	368.92	89.98	89.98
Operating costs	2786.24	2675.68	4290.81	4120.54	1046.54	1005.01
Fixed costs	1983.13	1983.13	3054.02	3054.02	744.88	744.88
Total costs	4769.37	4658.81	7344.83	7174.56	1791.42	1749.89
Gross net product	4476.00	3964.00	6893.04	6104.56	1681.23	1488.92
Revenue	-293.37	-694.81	-451.79	-1070.00	-110.19	-260.98
Costs per kilogram	2.13	2.35	2.13	2.35	0.52	0.57

Table 3. Production costs and revenues for black currants grown under Standard and IPM production programmes in 2003.

Item	Polish zloty/ 0.65 Ha		Polish zloty/Ha		Euros/Ha	
Block	Standard	IPM	Standard	IPM	Standard	IPM
Yield in kilograms	6814.00	6122.00	10493.56	9427.88	10493.56	9427.88
Labour	951.40	882.20	1465.16	1358.59	325.59	301.91
Labour (machine)	1382.50	1382.50	2129.05	2129.05	473.12	473.12
Total material costs	1187.79	1123.01	1829.20	1729.43	406.49	384.32
Chemicals	936.93	872.15	1442.87	1343.11	320.64	298.47
- Pesticides	575.92	511.14	886.92	787.16	197.09	174.92
- Herbicides	361.01	361.01	555.95	555.95	123.55	123.55
Fertilizers	250.86	250.86	386.32	386.32	85.85	85.85
Operating costs	3521.69	3387.71	5423.40	5217.07	1205.20	1159.35
Fixed costs	2302.92	2302.92	3546.50	3546.50	788.11	788.11
Total costs	5824.61	5690.63	8969.90	8763.57	1993.31	1947.46
Gross net product	7836.10	7040.30	12067.59	10842.06	2681.69	2409.35
Revenue	2011.49	1349.67	3097.70	2078.49	688.38	461.89
Costs per kilogram	0.85	0.93	0.85	0.93	0.19	0.21

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Exploiting the sex-aggregation pheromone of strawberry blossom weevil (*Anthonomus rubi*)

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Abstract: A sticky stake design of pheromone trap was developed and found to be effective for monitoring populations of the strawberry blossom weevil adults in strawberry crops in the UK. It comprised a pointed wooden stake (2 x 2 x 50 cm) inserted vertically into the ground to a depth of 5-10 cm and thickly coated round its circumference with a c. 10 cm band of polybutene sticker ('Oecotac') to within 2 cm of the top of the stake. A 7 x 7 cm corrugated plastic (Correx) board fixed horizontally on the top of the stake and secured with a tack provided protection of the sticky surface from rain. A polythene sachet pheromone dispenser containing 100 µl of a blend of Grandlures I, II and (±)-lavandulol in the ratio 1:4:1 was hung from a corner of the board by a bent paper clip as a lure. Field experiments in commercial strawberry crops using a similar prototype design indicated that the pheromone trap could be used as a predictor of flower bud severing damage caused by the weevil. In most cases the first catch of weevils in the traps preceded the onset of damage by a week. The average number of flower buds severed per plant was generally in the range 0.5-2 buds severed per adult weevil caught.

Adult weevil catches started at a low level in April or early May with similar numbers of males and females. The catches showed a marked increase in mid June coinciding with the emergence of new adults (in reproductive diapause) from the current seasons damaged flower buds. Catches beyond this date were predominantly of males by a factor of approximately 2:1.

No evidence that traps were affecting weevil damage or distribution or evidence of interference between traps was found in two large scale replicated field experiments. Attempts to exploit the pheromone trap for control of strawberry blossom weevil damage in 0.5 ha plots in commercial strawberry crops by surrounding the crop with a perimeter of lures, by the same method but with a boarder spray of the pyrethroid insecticide bifenthrin or by mass trapping with sticky stake pheromone traps at a density of 1000 per ha were all unsuccessful and did not result in a reduction in crop damage compared to untreated plots. Possible reasons for this lack of success are discussed.

Keywords: semiochemical, pest monitoring, pheromone trap, mass trapping, lure and kill, attract and kill, crop damage assessment

Introduction

The strawberry blossom weevil, *Anthonomus rubi* Herbst, is a common pest of strawberry in western and central Europe. Adults overwinter outside the strawberry field in hedge bottoms etc. They emerge in spring and invade strawberry crops. The females insert eggs, generally singly, into unopened flower buds. After oviposition, the female nips the stalk just below the flower bud of strawberry with her rostrum, partially severing it. The flower bud withers and the larvae develops inside. Adults emerge about six weeks later but are in reproductive diapause. They feed for a few weeks on strawberry but do not reproduce or cause flower bud severing damage. They migrate to overwintering sites in August. Severing damage to strawberry can cause yield loss depending on the number of flower buds severed in relation to the yield compensation capacity of the plant (Cross & Burgess, 1998).

Innocenzi et al. (2001) characterised the male sex aggregation pheromone of the weevil, which attracts both males and females, as Grandlure I, Grandlure II and lavandulol. The components occurred naturally in the ratio 1:4:1 respectively. Traces of Grandlures III and IV were also detected. The lavandulol was shown to be a single enantiomer but the absolute configuration was not determined. Germacrene-D, a known volatile from strawberry plants, was also collected in increased amounts in the presence of pheromone-producing weevils. In subsequent work, we developed low cost, robust and reliable polythene sachet dispensers containing 100µl of the pheromone blend which were shown to have a constant release rate (of 0.64 mg/day at 20 °C) and a life of over 6 weeks in the field.

In this paper we outline three years of field experiments which had 4 principle objectives as follows:

1. To develop a more effective and more practical trap design than the prototype used by Innocenzi et al. (2001).
2. To investigate use of the pheromone trap for monitoring populations of *A. rubi* adults and crop damage to determine whether it could be used for predicting and estimating the intensity of severing damage.
3. To investigate the range of attraction of the pheromone and possible interference between traps.
4. To determine whether the pheromone could be exploited for controlling the pest by lure and kill or mass trapping approaches.

Results and discussion

Trap design

In 2000 and 2001, a series of replicated field experiments was done in *A. rubi* infested commercial strawberry crops in southern and eastern England to optimise the design of pheromone traps for catching *A. rubi* adults. Most of the work aimed at improvement to the first prototype design used by Innocenzi et al. (2001) but other traps designs were also evaluated. The first prototype consisted of a horizontal 20 x 20 cm, double-sided, white sticky plastic board, affixed to the top of a 50 cm long, 2 x 2 cm diameter vertical wooden stake with the lure held on top in a small cage made from a hair curler. Standard 2.5 x 2.5 cm polythene sachet lures containing 100 µl of the standard pheromone blend (1 GLI: 4 GLII: 1 ±lavandulol) were used to bait the traps and traps without lures were included as control treatments.

In a series of steps, the first prototype was improved leading to the development of a sticky stake trap as the most effective design. The sticky stake trap consisted of a 50 cm long 2 x 2 cm diameter vertical wooden stake with a 10 cm band of a viscous polybutene sticker ('Oecotac') round its circumference and with a small Correx board on top to act as a rain shield, from the corner of which the lure was suspended on a wire hook. Weevils enter the trap mainly by walking up the stake. Other trap designs tested were less effective. The sticky stake trap design is a suitable trap design for use for pest monitoring.

Relationship between pheromone trap catches and crop damage

In early April 2001 and 2002, a row of five *A. rubi* pheromone traps spaced 5 m apart, each with a horizontal 20 x 20 cm white correx board with sticky underneath and standard lure suspended by a wire hook from a corner, were set out in a row at the edge in each of 5 commercial strawberry crops in Kent and Hampshire (different sites were used in the two years). A further row of five traps were set out in a similar row in the centre of each of the crops. The traps were examined and refreshed at 1-2 week intervals up to fruiting. Records were taken of the catch of each sex of *A. rubi* in each trap and of other incidental weevils. The numbers of severed blossoms on 10 plants in each of 4 sampling locations (at the edge of the field adjacent to the traps, at the edge of the field 30 m distant from the traps, in the centre of the field adjacent to the traps in the centre of the field 30 m distant from the traps) were recorded on each occasion. The average numbers of blossoms per plant were also determined so that data on severing could be converted to percentages.

Good correlations were obtained at each sampling location between the cumulative numbers of weevils (of either sex or both sexes combined) and the amount of severing damage on the crop. The lowest correlation coefficient was 0.67, but in most instances the coefficients were considerably greater (typically > 0.8).

For simplicity, the data from the different sampling locations (edge and middle, near and 30 m distant from the traps) at each site were combined and graphs of the cumulative trap catch (both sexes combined) and the average number of blossoms severed per plant were plotted on the same axes against sampling date. The graphs of the cumulative trap catches and severing damage over time showed that trap catches increased as severing damage increased (see example in Figure 1). In 9 out of 10 data sets, the numbers of flowers severed per plant were approximately 0.5-2.0 times the cumulative trap catch of weevils during the period when the damage was occurring. The worst case was at one site (Sopley) in 2001, when the amount of damage was 6-8 times the total cumulative catch. First catches of weevils, though small, did precede the first occurrence of damage by one week or more in most instances.

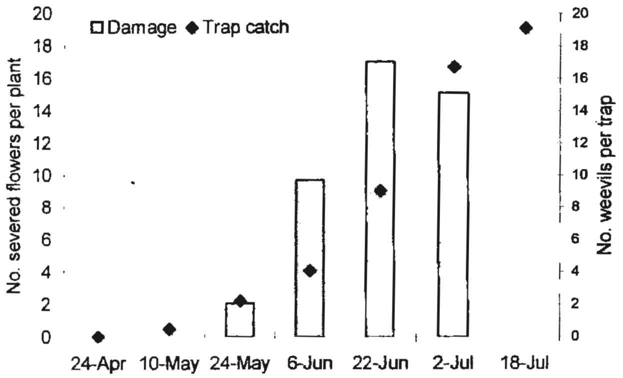

Figure 1. Mean cumulative number of weevils captured per pheromone trap and mean number of flower buds severed per plant in one of the five commercial crops (at Marden, Kent) monitored in 2001.

The results indicate that the pheromone traps would be useful for pest monitoring in commercial crops. The traps would be of low cost in comparison to the value of the crop and it would be easier to examine the traps for weevils than search the crop for damage. The first occurrence of catches of weevils would indicate when damage was likely to start. This would indicate the timing of preventive treatment with a broad-spectrum insecticide against adults. The results indicate that the expected number of flowers severed would usually be in the range of 0.5-2.0 times the cumulative number of weevils captured.

Seasonal dynamics of pheromone trap catches

The results obtained from monitoring pairs of traps in 14 commercial crops in 2000 together with the data above were used to explore the seasonal dynamics of catches of males and females in pheromone traps including whether weevils that are in reproductive diapause are caught by the traps. Records of catches were continued well beyond the end of flowering into August. Histograms of the numbers of each sex caught over time, averaged for all sites, were plotted for each year. Weevils were sexed by the presence of a prominent thorn on the mesothoracic coxae, which occurs on males only (Innocenzi et al, 2002)

In each of the three years, weevil catches started at a low level in April or early May with similar numbers of males and females (see Figure 2 for 2000 results). However, in all three years the catches showed a marked increase in mid-June (about week 25). The increase coincided with the emergence of new adults from the current seasons damaged flower buds. These adults are in reproductive diapause. Catches beyond this date were predominantly of males by a factor of approximately 2:1. This data provides circumstantial evidence that newly emerged diapausing adults are caught. If they were not caught, catches would be expected to decline as overwintered adults die out as the season progresses.

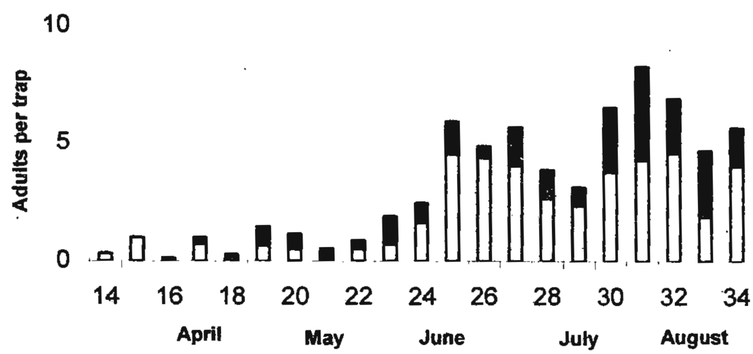

Figure 2. Mean catches of male (white bar) and female (black bar) strawberry blossom weevil per trap in 2000.

Range of attraction and trap interference

Two large-scale replicated (4 reps) experiments were conducted in a large strawberry field in Norfolk in 2000 and 2001 respectively to investigate the range of attraction of traps, to determine whether there was any interference between traps and whether groups of traps could be used to depress weevil populations.

In 2000, four different sizes of lattices of 5 traps (arranged in a cross like the five spots of the five of a dice), were compared to an untreated control as follows:

1. 1.7 m x 1.7 m lattice of 5 traps
2. 5.1 m x 5.1 m lattice of 5 traps
3. 10.2 m x 10.2 m lattice of 5 traps
4. 20.4 m x 20.4 m lattice of 5 traps
5. Untreated control with no traps

In 2001, there were three treatments as follows:

1. 5 x 5 square lattice of 25 traps spaced 3.4 m apart
2. single trap
3. untreated control (no trap)

Catches of weevils in the traps and the amount of flower bud severing damage on the surrounding crop were recorded.

In the 2001 experiment, small numbers of weevils were caught but, there were no significant differences between the trap spacings. Thus there was no evidence of interference between traps and no information was gained about the range of attractancy of the traps from this experiment. A small number of flower buds suffered severing damage by blossom weevil. However, there was no evidence that the amount of severing damage was suppressed by the presence of traps or that the amount of severing was influenced by the spacing of traps at any of the distances from the centre traps. In the second experiment in 2002, the number of weevils caught in the pheromone traps in the centre of the lattices was small but as in the first experiment, there was no evidence for interference between traps. The outer rings of traps were not greatly reducing the numbers caught in the centre. Thus there was no statistically sound evidence that the traps were affecting weevil populations or damage. If anything, the trend was to greater damage and higher numbers of weevils in the lattice of 25 traps, suggesting that the traps might be attracting weevils into the vicinity without actually catching them. Thus, neither of the two trap spacing / lattice field experiments showed any evidence that the lures were affecting weevil damage or distribution. Indeed, no evidence of interference between traps was evident in either experiment. There are several explanations for these results. One possible explanation is that the traps were of low attractancy to the weevils and only attracted a small proportion of the population, either because the blend and/or the release rate were incorrect or because the traps were competing with the pheromone produced by the natural population. Another possible explanation is that the weevils are only attracted weakly, and at certain times in their life cycle, by the pheromone. A third alternative explanation is that weevils were attracted into the vicinity of traps but were not actually caught, possibly because the first prototype trap design was inefficient. This latter explanation is not strongly supported by the data because severing damage was not markedly increased in the vicinity of traps.

Exploiting the pheromone for control by perimeter trapping, lure and kill and mass trapping

In 2002, a series of 6 experiments was conducted to examine 3 different strategies of exploiting the strawberry blossom weevil pheromone for control of flower bud severing damage by strawberry blossom weevil in the field. The three strategies evaluated were:

1. A ring of lures, spaced 3 m apart, round the perimeter of the field starting from before spring emergence of blossom weevil and maintained throughout the flowering period.

2. As for 1, but with one or more sprays of the insecticide bifenthrin sprayed in a 3 m wide band round the headland of the crop, including the strawberry plants at the edge of the field before flowering. This strategy was named 'LASH' (Lure And Spray Headland)
3. Mass trapping ('MASS') with 1000/pheromone lured sticky stake traps /ha.

Two field experiments were done to evaluate each of the three strategies (a total of 6 experiments). For each experiment, two approximately 70 m x 70 m (0.5 ha), similar areas of a 1-2 year old commercial strawberry crop of the June bearer variety Elsanta grown on raised polythene mulched buds with a history of strawberry blossom weevil were selected. In each experiment, one area was treated with one of the three control strategies the other was left untreated.

In all 6 experiments, the mean numbers of flowers severed on the treated and the untreated plots did not differ markedly. The distributions of damage along the two transects across the field were also similar. Thus none of the three control stratgies affect the levels or distribution of severing damage significantly. There was no evidence that any of the strategies was providing effective control of the pest and preventing damage These results are disappointing. It appears that the strawberry blossom weevil sex aggregation pheromone is inherently less attractive in comparison with the sex pheromones of moths. Another possible explanation for comparatively low attractancy is that there is more competition with pheromone naturally emitted by males in the field.

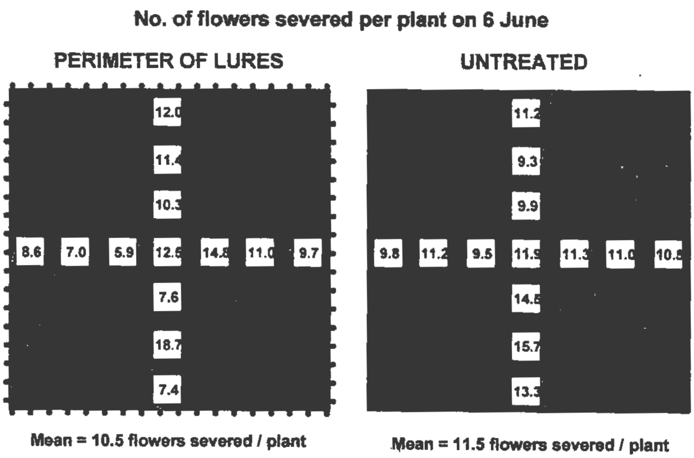

Figure 3. Perimeter of lures field evaluation, Grange Farm, Tunstead, Norfolk, 2002.

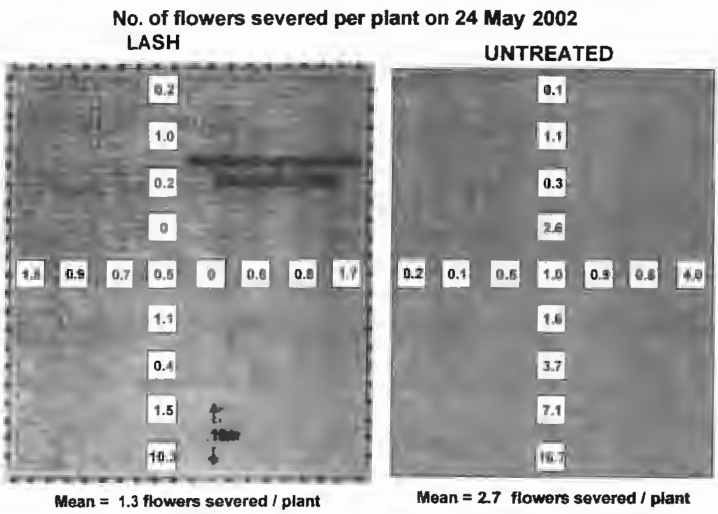

Figure 4. LASH field evaluation, Boughton Monchelsea, Kent, 2002.

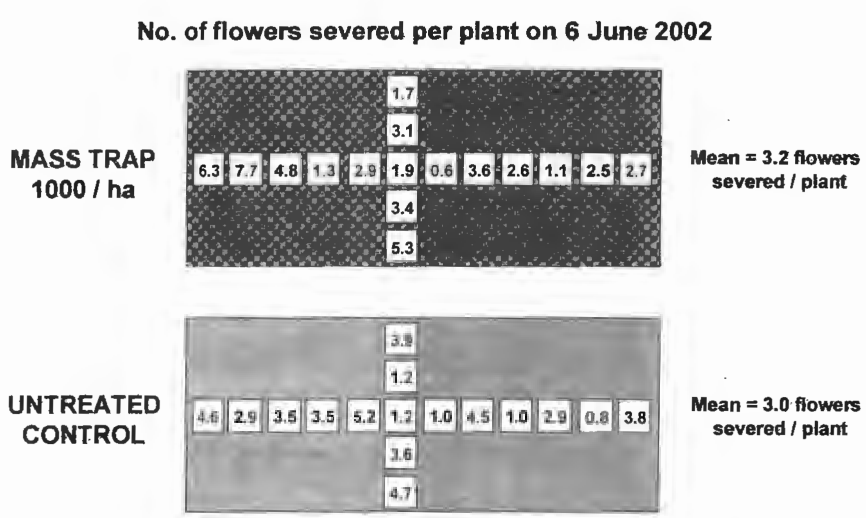

Figure 5. Mass trapping field evaluation, Norfolk, 2002. Mean number of flower buds severed per plant are shown in the boxes.

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The first field experiences with sex-aggregation pheromones of the strawberry blossom weevil, *Anthonomus rubi*, in Austria

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Abstract: The first field experiments were carried out in Austria in 2002 and 2003 to investigate the control effect of specific sex-aggregation pheromones of *Anthonomus rubi* (source HRI East Malling, UK) in strawberry fields within a mass trapping strategy. No reduction of strawberry blossom weevil infestation was achieved. At present the use of sex-aggregation pheromones for the control of *Anthonomus rubi* cannot be classified as effective to reduce the pest below the economic treshold level.

Key words: *Anthonomus rubi*, strawberry blossom weevil, strawberries, aggregation pheromone, control

Introduction

The strawberry blossom weevil, *Anthonomus rubi* Herbst (Coleoptera; Curculionidae), causes severe damage on strawberries throughout Europe (e.g. Alford, 1987) and is responsible for harvest losses of 60% or more in Austria. Broad-spectrum insecticides are currently used for the control of *A. rubi*, but resistance problems, adverse ecotoxicological effects and the prohibition of use of such pesticides in organic farming (biological production) create a demand for alternative, especially biological or biotechnical, control methods.

Innocenzi et al. (2001) succeeded in the identification and production of the sex-aggregation-pheromone of *Anthonomus rubi*, which attracts both males and females. Several experiments have been carried out to investigate these pheromones in the field for practical use (pest monitoring and control). In Austria a mass trapping strategy (sticky stake trap design) for the control of *A. rubi* was tested within an international project in 2002, and studies were continued with a modified trap design (sticky plate trap design) in 2003.

Material and methods

The experiments were carried out in the first year (2002) in Lower Austria (Raasdorf), about 15 km south from Vienna, and in the following year (2003) in Burgenland (Wiesen), about 70 km south-east from Vienna. Both sites are situated in typical Austrian strawberry production regions. The investigated field in Raasdorf was located in an open landscape with windy conditions; the experimental area was only a part of a large strawberry production site (2800 m^2) with Honeoye as the predominant cultivar. The level of infestation with the strawberry blossom weevil was low during the trial season. In contrast the strawberry field in Wiesen was small (720 m^2) and wind protected by a partially surrounding forest. Several strawberry varieties of different ages were present and were highly infested at the start of the season.

In 2002 studies were carried out within an international ring-test-project under the leadership of Horticulture Research International, East Malling (UK). A sticky stake trap experiment (see Fig. 2), including 2 plots each with 5 sticky stake traps and 2 plots without traps at either the edge or in the centre of the field, was used to test the effectiveness of the pheromones. The sticky stake trap (Fig. 1) consisted of a wooden stake (2 x 2 x 50 cm) in the ground with a 10 cm wide band of tangle-foot-glue round its circumference to 1 – 2 cm below the top to catch the weevils. The pheromone lure (polythene sachet dispenser) was fixed on a small plate on the top of the stake. The traps (+ pheromones) were set up on April 9th 2002.

Figure 1. Sticky stake trap (left) and sticky plate trap (right).

The number and sex-ratio of caught weevils was assessed regularly during the investigation period. In addition, 10 strawberry plants were checked for damage in all 4 plots in order to determine the relationship between trap catches and crop damage.

In 2003 a modified trap design (Fig. 3) using sticky plate traps was investigated in one test-field (720 m^2) and one control-field (852 m^2) . Two plots (rows), each with 5 pheromone traps (sticky plate traps), were evenly set up from one field edge to the opposite edge in order to cover up the whole field with the pheromone odour; the pheromones should attract over a distance of about 10 m (Cross, pers. comm.). The sticky plate traps (Fig. 2) looked like the sticky stake traps, but a white plastic plate (20 x 20 cm) was fixed on top of the stake. Sticky glue was distributed on the upper- and the lower side of each plate and later renewed only on the lower side, in order to catch the weevils. The traps were installed on April 9^{th}, 2003. The pheromone catches were recorded during the whole season. The damage to the strawberry plants was additionally assessed during the period of pre-blooming and blooming (7 rows in the test-field, 4 rows in the control-field, every row with 10 plants).

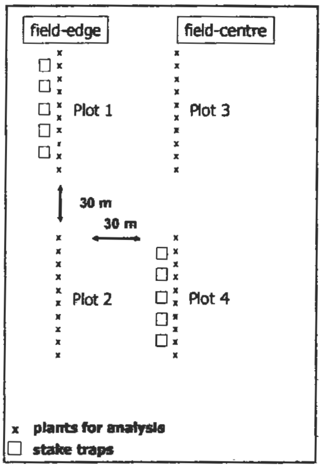

Figure 2. Investigation site and experimental design in Raasdorf, Austria, 2002.

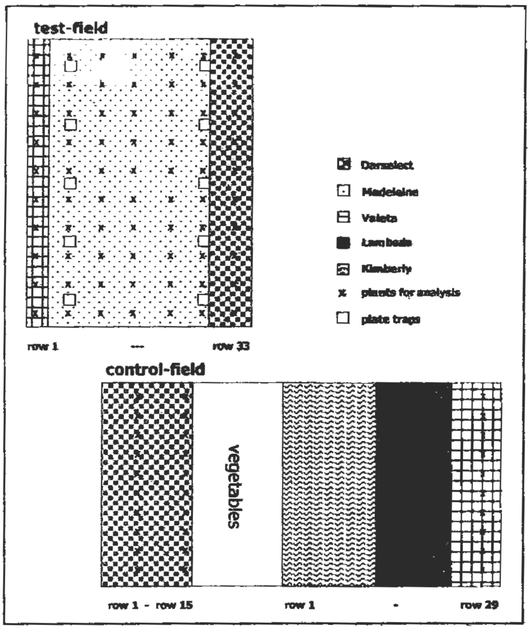

Figure 3. Investigation area and experimental design in Wiesen, Austria, in 2003.

In both experiments the pheromone traps were assessed every 7 or 14 days during the investigation period to record the number and sex-ratio of attracted strawberry blossom weevils and their distribution in the field. Due to the very low number of weevils caught no statistical evaluation was done. The infestation of the strawberry plants (results of 2002) was statistically evaluated with the Kruskal-Wallis-Test. The results of 2003 were not statistically analysed.

Results

Pheromone trap catches (Raasdorf)

The low infestation with *Anthonomus rubi* in Raasdorf (2002) was reflected in the low abundance of weevils caught in the traps. Only a small part of the weevil population was attracted and caught in the traps during the investigation period from the end of May to the end of June (Fig. 4). Furthermore, there was no difference in the total number of all *A. rubi* individuals and in the sex-ratio between catches at the edge of the field and at the centre of the field.

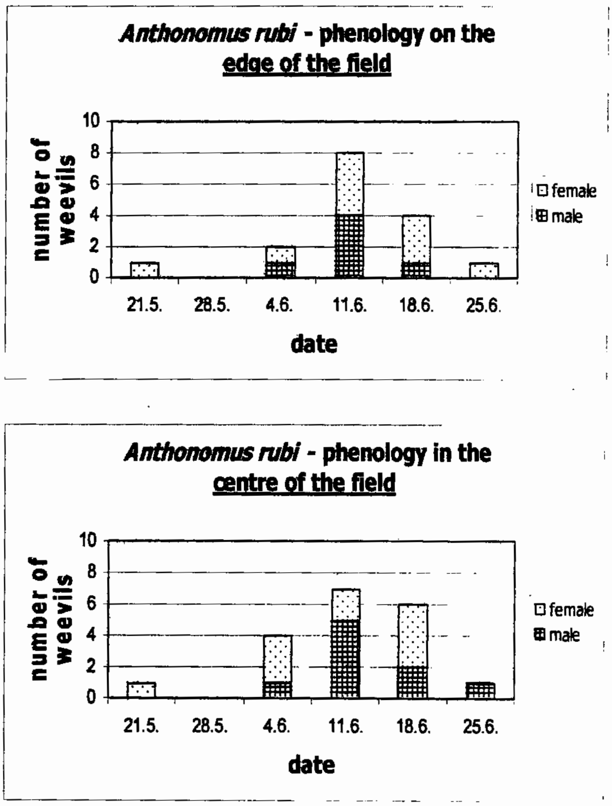

Figure 4. Total number of strawberry blossom weevils (sum of 5 pheromone traps) in Raasdorf caught by sticky stake traps during the period of May to the end of June 2002.

Infestation analysis (Raasdorf)

The infestation of the strawberry plants was assessed on May 7th and May 14th, 2002. The assessed damage did not exceed 30% of severed flowers per plant at the first evaluation date and increased only slightly one week later (Fig. 5). Data analysis showed a significant difference in the mean infestation rate of both pheromone-plots (field-edge and field-centre) and between both centre-plots of the different treatments (with/without pheromones) on May 7th, 2002. In contrast, no significant difference between both plots at the edge of the field of the different treatments (with/without pheromones) were observed. For the second evaluation date (May 14th, 2002) no significant difference in the mean infestation rate of the 4 tested plots was found.

No reduction of the infestation with *A. rubi* was achieved with this mass trapping strategy.

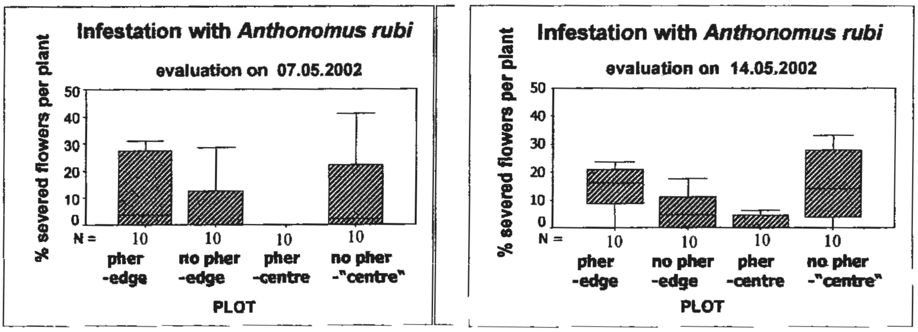

Figure 5. Boxplots of the infestation rate with *Anthonomus rubi* on strawberries for 4 different plots in the field (edge/centre of the field, with/without pheromones) in Raasdorf (Austria) in 2002.

Pheromone trap catches (Wiesen)

Despite the known and visually observed high infestation in Wiesen (2003) only a low number of *A. rubi* individuals was caught with the pheromone traps. Males and females were trapped almost in the same ratio during the whole season showing no tendency for an increased occurrence of males or females in spring- or summer-generation (Fig. 6).

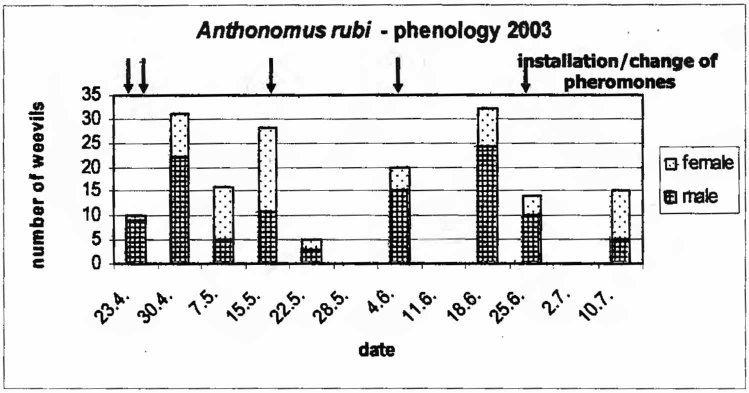

Figure 6. Number of strawberry blossom weevils in Wiesen (Austria), caught with sticky plate traps from the end of April to the middle of July 2003. The number of weevils per date is always the total of all 10 pheromone plates.

Infestation analysis (Wiesen)

Some parts of the test-field (with pheromone traps) and the whole control-field (without pheromone traps) were protected with fleece. For comparison of treatments the infestation of the strawberry plants is only presented for the rows with fleece-protection, in both fields (Fig. 7). Not only was the development of the fleece protected plants faster than without fleece, but also the infestation started much earlier in these parts. Eventually higher infestation was observed on the plants without fleece protection, and strong damage, up to 73% severed flowers per plant on average occurred. Infestation of the protected plants did not give such high percentages of damage (maximum up to 42%), but there was higher infestation in the test-field than in the control. Furthermore, there was no difference in the infestation level between the tested strawberry cultivars. Thus, this mass trapping strategy with the sex-aggregation pheromones did not lead to a successful reduction of the damage by the strawberry blossom weevil.

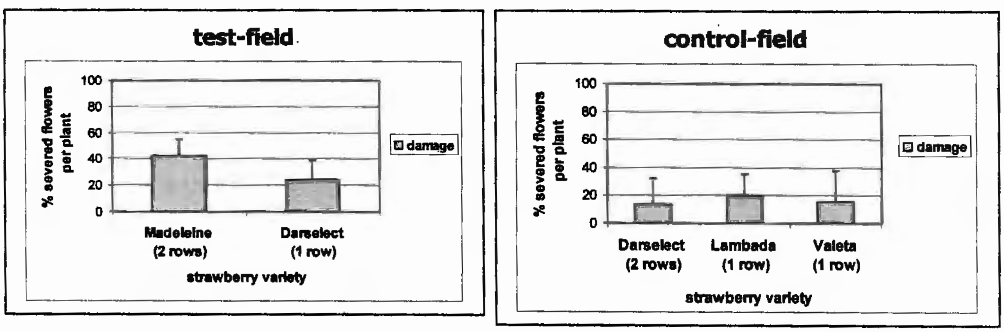

Figure 7. Evaluation of the infestation with *Anthonomus rubi* on different strawberry varieties for the test-field (with pheromones) and the control-field (without pheromones) on May 7th, 2003 in Wiesen (Austria).

Discussion

With regard to the obtained results the use of sex-aggregation pheromones for the control of *Anthonomus rubi* cannot be classified as effective in reducing damage to an economically feasible level.

There are some possible explanations and reasons for these results. One of the most important preconditions for the use and effectiveness of pheromone controlling methods is a low density of the pest population in the treated area (e.g. Krieg & Franz 1989; Howse et al. 1998). Thus, perennial strawberry fields have more initial infestation and therefore worse preconditions than "new" fields – which was obvious for the experiments in Wiesen. Other important points influencing the effect of pheromones are the environmental conditions of the treated field, such as the surrounding area and climatic conditions (wind, temperature). In contrast to Raasdorf, one side of the strawberry field in Wiesen was adjacent to a forest, which on the one hand favours the immigration of *A. rubi* to the production site, as the strawberry blossom weevils prefer to overwinter in forests, but on the other hand the forest reduces the influence of air movement (wind), which supports the prolongation of the presence of the pheromone-odour plume in the field.

Another important factor affecting the successful use of pheromones is the pheromone itself concerning its quality and attractiveness and the method of its use (for example the trap design). It is interesting that in fact many weevils seemed to be attracted into the field due to the pheromone odour, but not also into the traps. This was confirmed by visual observations of weevils remaining on strawberry plants next to an artificial pheromone source without being attracted and caught in the trap. Both an insufficient attraction of the pheromone and the weevil's behaviour could be responsible for this phenomenon. One presumption is that *A. rubi* seems to show different susceptibility to the pheromones during its life (Cross, pers. comm.). Finally, it has to be taken into consideration that the pheromone used is only a sex-aggregation-pheromone and not a specific sex-pheromone as usually applied for pheromone controlling methods.

In conclusion, further research is needed to investigate the possible potential of the *Anthonomus rubi*-aggregation-pheromone for use as an effective control method in future.

Acknowledgements

We are very grateful to Manfred Radl and Anna + Werner Schreiner who made their strawberry fields available to us for carrying out the experiments and to Brigitte Walestin for her help in the field.

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Effectiveness of conventional and integrated control of blackcurrant pests

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Abstract: Conventional and integrated methods of arthropod pest control were used separately in two blocks of blackcurrant crop planted in 1999 at the Experimental Station in Dąbrowice (Central Poland). During the first five years after planting the most important problems in the integrated block were blackcurrant gall mite (*Cecidophyopsis ribis* Westw.), two-spotted spider mite (*Tetranychus urticae* Koch. and aphids. It was observed that removal of infested buds as the only method of gall mite control was not sufficient to effectively protect the integrated block against this pest. Use of this method resulted also in an increase in the density of aphids. The number of aphid colonies was usually higher in the integrated block compared to the conventional block, in which endosulfan was applied. Three standard treatments with this pesticide each year against blackcurrant gall mite also reduced aphid populations effectively. In contrast, additional sprays against aphids were necessary in the integrated block. Promising results for the biological control of two-spotted spider mite with phytoseiids after release of *Typhlodromus pyri* were also obtained.

Key words: IPM, blackcurrant, Cecidophyopsis ribis, Aphididae, Tetranychus urticae, Synanthedon tipuliformis, Resseliella ribis, Typhlodromus pyri

Introduction

Poland is one of the biggest producers of blackcurrant fruits. At present, the total production is about 175 thousand tons, which constitutes nearly 25% of the world production of this fruit. Due to free market requirements the implementation of IP rules in blackcurrant plantations has become necessary in Poland. A first step in this direction was to establish the Polish guidelines for integrated production of currants. These guidelines were elaborated and published by the Research Institute of Pomology and Floriculture at Skierniewice in 2002 (Proekologiczne Technologie Produkcji Owoców Porzeczki, Skierniewice, 2002). The principles included in this document closely comply with IOBC Euro-guidelines published in 2000 (Guidelines for integrated production of soft fruits, IOBC wprs Bulletin Vol. 23 (5) 2000).

As integrated production of blackcurrants is not widely practiced it was necessary to establish pilot experimental plots in this crop, in which IP rules of arthropod pest control could be studied.

Material and methods

The investigation was conducted in blackcurrant plots planted in 1999 at the Experimental Station in Dąbrowice (Central Poland). The whole experimental area of this crop constituting 1.3 ha was divided into two blocks. The conventional methods of arthropod pest control were applied to one block whereas integrated control methods were used in the second. Both blocks were treated according to the same programme of disease control. During the first years of the investigation special attention was paid to the following pests: blackcurrant shoot borer (*Lampronia capitella*), blackcurrant gall mite (*Cecidophyopsis ribis*), aphids (*Aphididae*), two-spotted spider mite (*Tetranychus urticae*), currant clearwing moth (*Synanthedon tipuliformis*) and the gall stem midge *Resseliella ribis*.

The density of these pests was regularly monitored every year. The number of buds damaged by blackcurrant shoot borer was sampled during bud burst stage. During the dormant period the whole experimental area was thoroughly inspected for the buds infested with blackcurrant gall mite. The number of aphid colonies as well as the population density of two-spotted spider mite were determined several times during each season. The numbers of shoots damaged by clearwing moth and gall stem midge were estimated every autumn. Additionally, pheromone traps were used for monitoring clearwing moth.

Results and discussion

Blackurrant gall mite

Blackcurrant gall mite was and still is the most important pest of blackcurrant. In the conventional block three treatments of endosulfan were applied each season. As a result, no symptoms of gall mite feeding were observed in the first four years of the investigation. At the beginning of the planting the only method of control of this pest in the integrated block was the removal of infested buds each winter. Unfortunately this method did not protect the blackcurrant plants effectively and an increase in gall mite population was observed every year (Fig. 1).

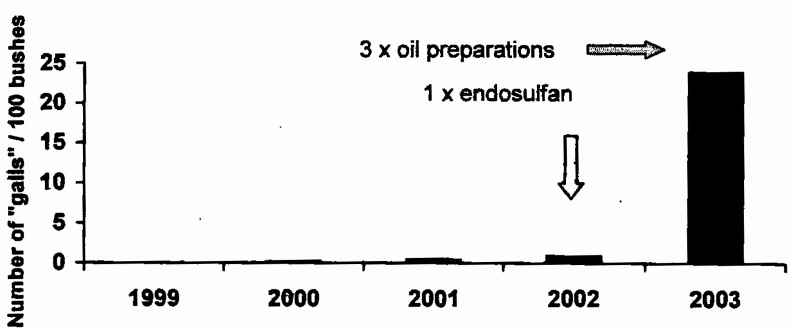

Figure 1. Population density of blackcurrant gall mite *(C.ribis)* in integrated block; Dąbrowice 1999-2003.

In consideration of this fact, in 2002 the removal of infested buds was supplemented with one treatment of endosulfan. This programme of control of the pest proved to be ineffective and the level of infestation significantly increased afterwards. Therefore, in 2003 mineral oil preparations were sprayed in addition to removal of infested buds.

The mentioned above mineral oil preparations were previously found to be effective weapons against blackcurrant gall mite (Gajek, 2001). It seems that these products will soon be extensively used in practice instead of endosulfan, which may not be applied any more because of new regulations of pesticide residue levels applied recently in Poland.

Aphids

Among the aphids occurring in experimental blocks, the most common was the permanent currant aphid (*Aphis schneideri* (Börner)). It was observed that the density of aphid colonies was usually higher in the integrated block than in the conventional block. The reduced density of aphids under conventional control was as a result of endosulfan applications used to control the gall mite. As this pesticide was not used in the integrated block, it was necessary to apply treatments with other insecticides there.

Examples of this situation are shown in figure 2. In 2001, acetamiprid and in 2002 pirimicarb had to be additionally applied in the integrated block for control of these pests.

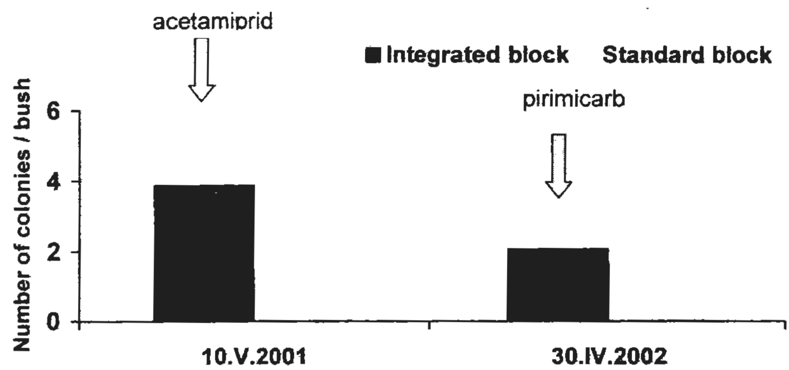

Figure 2. Density of aphid colonies in experimental blocks; Dąbrowice 2001 and 2002.

Two spotted spider mite

Two-spotted spider mite was another important pest infesting the experimental blocks. In the integrated control block the aim was to use phytoseiid mites. *Typhlodromus pyri* was successfully introduced into this block in the spring of 2001. Unfortunately, this release did not give noticeable results in the same season. Hot weather during the summer was a factor triggering dynamic development of spider mite population and the selective acaricides, hexythiazox and propargite, had to be applied in this block.

A similar situation occurred in 2002 and treatments with selective acaricides were again required, though, the presence of predatory mites was more easily noticed and their density at the end of the season reached the level of 0.1 mite per leaf.

The weather conditions in 2003 were again excellent for development of spider mites. It was necessary to control the pest using acaricides just at the beginning of the season. Therefore hexythiazox was applied in the integrated block and mixture of hexythiazox with propargite was applied in the standard block (Figure 3). Later in the season, especially in the integrated block, the density of phytoseiids significantly increased. Thanks to that, the population of two-spotted spider mite was maintained at a low level, below the damage threshold.

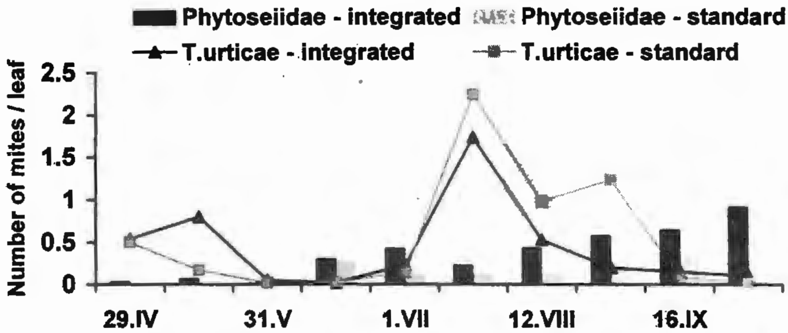

Figure 3. Population densities of two-spotted spider mite (*T.urticae*) and phytoseiids (*Phytoseiidae*) – 2003.

Other pest species systematically monitored in the experimental blocks included clearwing moth (*Synanthedon tipuliformis*), blackcurrant shoot borer (*Lampronia capitella*) and black currant stem midge (*Resseliella ribis*). However, all of them occurred at densities below economic thresholds in both blocks, therefore, no treatments have been applied against them so far.

Conclusions

Considering the research conducted hitherto, the most important conclusions are as follow:

1. The investigation showed that during the first five years of the plantations life the most serious problems in blackcurrant protection were blackcurrant gall mite, two-spotted spider mite and aphids.
2. The removal of buds infested with gall mite in the integrated block did not protect the blackcurrant plants effectively against this pest. The use of endosulfan has not been permitted in Poland since this year, and thus new, effective chemicals against this pest will have to be found.
3. Endosulfan, applied against blackcurrant gall mite, also reduced aphid populations effectively. As a result of withdrawal of this product, aphid control in blackcurrant crops may require an additional treatment.
4. The investigation showed promising results for the biological control of two-spotted spider mite with phytoseiids.

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Strawberry blossom weevil – recent research in Norway

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Abstract: Chemical control of the strawberry blossom weevil (*Anthonomus rubi*) prevents the use of predatory mites to control phytophageous mites in open field strawberries. In addition, growers report that available pesticides work poorly. This has led to investigations on the biology and alternative control methods of the weevil. To monitor the weevil, tapping plants above a bowl is faster and provides more information about future damage than counting cut buds. Without any direct control measures the bud number cut by *A. rubi* increases year by year, but as the plants also grow, the % bud loss does not necessarily increase. Studies of the relation between bud loss and yield suggest a lower damage threshold in Norway than in milder climates (e.g. the UK). In a pesticide trial in 2003 normal concentrations of esfenvalerate and deltamethrin did not have any effect on the number of weevils or buds cut, indicating a tolerance of pyrethroids in the local weevil population. We did preliminary trials with two alternative control methods: 1) A mechanical barrier (a 2 m fence made of fine-meshed net); and 2) An entomopathogenic nematode (*Heterorhabditis megidis* /Nemasys H). Both methods show promise as control methods against *A. rubi*.

Key words: *Anthonomus rubi*, *Heterorhabditis megidis*, exclusion fence, pesticides

Introduction

The strawberry blossom weevil (*Anthonomus rubi* Herbst) is a pest because it uses the flower buds of strawberry and raspberry as a larval habitat (e.g. Jary, 1932). The damage occurs when females oviposit in closed flower buds in the spring. After ovipositing the female severs the bud stalk, and the progeny is protected by the withering flower bud throughout its development. The new adults emerge from the buds in late summer, and seem to be in a reproductive diapause, at least in the northern parts of Europe. *A. rubi* thus has one generation per year, overwintering as an adult. Adult weevils also feed off the strawberry plant (leaves, pollen), but this damage is of minor importance.

The present control of *A. rubi* in Norway consists of spraying the plants with a pesticide (pyrethroid or organophosphate, the latter not available after 2005) when cut buds begin to appear in the spring. However, growers in parts of S-E Norway have started reporting that this chemical control is no longer effective. An additional problem is that the pesticides used against *A. rubi* destroy the potential for phytoseiid biocontrol of phytophageous mites. This has led to investigations on biology, damage threshold, and alternative control methods of the weevil. All the trials reported here took place in open fields in S-E Norway.

Material and methods

Comparing methods for monitoring A. rubi in the spring (2000)
Tapping whole plants by hand above a white washing bowl (Ø=30-40 cm, with a sector of 120° removed) has been suggested as a monitoring method of adult *A. rubi* in Finland (Tuovinen & Parikka, 1997). The method exploits the weevil's behaviour of slipping off the plant when disturbed. A more common monitoring method is to count the number of cut buds per plant. We used the two methods on portions of neighbouring rows (one method per row) about every 7 days in 11 'Korona' fields from mid May to late June. Weevils found in the bowl were put back on the plant they came from. Cut buds, either still on the plants or on the ground, were removed after counting.

Relation between cut buds and yield loss (2002+2003)
A simulation trial was carried out in three weevil-free fields at 59°40'N: 1) 4-yr old 'Zephyr' in 2002; 2) 2-yr old 'Honeoye' in 2002; 3) 2-yr old 'Korona' in 2003. Weevil activity was simulated by cutting the largest buds still tightly closed. The bud removal was done in one day (11 May in 2002; 7 June in 2003), at the stage when the first primary buds started opening. Three levels of bud removal in single plants (7 replicates) were used in each field, ranging from 12 to about 50% of the buds plus flowers present. During harvest the weight and number of healthy berries >25 mm were recorded from each plant. The trials were part of a collaboration within COST 836 to obtain data on *A. rubi* damage thresholds in different climates, cultivars and growing systems (Faby et al., in press).

Pesticide trial (2003)
Two new pesticides (indoxacarb and thiacloprid; not registered in Norway) were compared with the pyrethroids esfenvalerate and deltametrin in a commercial field just at the start of flowering (6 June). The grower had reported a poor effect of esfenvalerate. Indoxacarb, which is best known as a lepidoptericide, was included because it can control *Anthonomus* weevils in top fruit according to information from DuPont Agricultural Products. The experimental unit was 6 m of double row (4 replicates). The spray volume was 20 l per 100 m of double row, corresponding to about 1100 l per ha. The recommended dosages were used, with the exception of thiacloprid, which was tried in a reduced dosage (63% of the one recommended by Bayer Cropscience against *A. rubi*). Weevil activity was recorded before spraying (by counting cut buds and using the bowl method) and after spraying (bowl method 4 and 24 Days After Treatment; bud count 20 DAT).

Damage and population size in organic fields (2000-2003)
The weevil activity was monitored at regular intervals throughout the oviposition period in three organic fields. Field 1 was 0.5 ha of 'Korona' planted 1999 at Toten (60°40'N; 420 m.a.s.l.). Plants in 5 plots along a diagonal transect were monitored for 4 years (counting buds of 14 plants per plot + bowl method on 30 plants pet plot). Field 2 and 3 were small experimental fields, 250 m^2 each, at Kise (60°46'N; 140 m.a.s.l). In field 2 (planted 2000), all cut buds in 4 plots of 20 'Korona' plants were counted. In field 3, a cultivar test field (planted 2001), the bowl method was used on all plants (8 varieties x 4 replicates x 20 plants).

Exclusion fence (2003)
A mesh fence can keep cabbage root flies (*Delia*) from entering brassica fields (Vernon & MacKenzie 1998). In 2003, a small preliminary trial with an exclusion fence in strawberry was carried out to study its potential to prevent *A. rubi* from colonizing new fields. The fence

was erected on 16 May 2003 as a 5.5 x 43 m rectangle in a 1.2 ha new planting (at 60°45'N, planted July/Aug 2002). This new planting bordered an older planting with a large *A. rubi* population (⅓ of normal yield in 2003). The 2 m high fence was made of insect-net (mesh 0.49x0.77 mm, type REDE/PE 1410). At the top a 0.3 m overhang made by folding the mesh acted as a trap for insects climbing the fence from outside. At the base, the net was secured by heavy sacks at regular intervals. Insecticides were not applied inside the fence, while the area outside was sprayed 3 times. The weevil population was sampled (bowl method on 30 plants) outside and inside the fence at 5 times.

Bioassay with the entomopathogenic nematode* Heterorhabditis megidis *(July 2003)
Two filter paper circles (Ø=90 mm) were placed in the lids of 16 Petri dishes (also 90 mm) and moistened with a little tap water. Half of the dishes were then treated with 1 ml tap water (control) and the remaining half with a 1 ml suspension containing 1000 infective juveniles (IJ) of *H. megidis*. Ten buds severed by the strawberry blossom weevil (collected in organic field 1, see above) were put in each dish. The presence of a larva in each bud was not checked before the experiment. The Petri dish bottoms were used as lids, and each dish wrapped in aluminium foil to keep it dark. The buds were examined for live or infected *A. rubi* larvae after two days in room temperature (approx. 22°C). The short time between application and examination was due to previous experimentation showing that such small larvae rapidly disintegrate after nematode infection.

Results and discussion

Comparing methods for monitoring* A. rubi *in the spring
The number of cut buds correlated well with the number of weevils tapped in the neighbouring row ($y=16x+3.7$, $r^2=0.7$, $p<0.01$, where y is the total severed buds per plant and x the sum of weevils per plant in 7-8 tappings). Monitoring the adults provides information about the incidence of future damage (i.e., cut buds; Figure 1) and thus is better than counting the cut buds present. Tapping is also much faster to perform than the bud counting, but does require fine weather and dry plants. The bowl method should be considered a very useful monitoring method, but we need more studies on how the relationship between weevils tapped and buds cut is affected by plant age, cultivar and growing system, and on how many plants to sample, when to sample, etc.

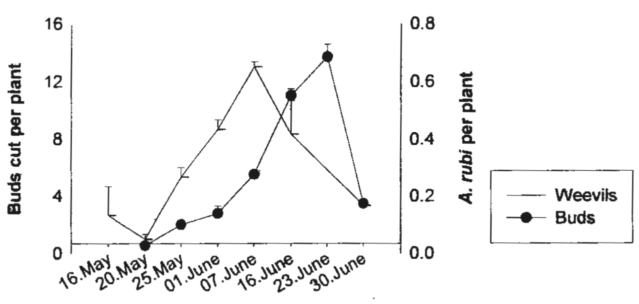

Figure 1. Weevil tapping and bud counts through the season. Data from unsprayed plants in the field (4 years old) with most weevils in the survey of 11 fields, 2000.

Relation between cut buds and yield loss

Removing buds did affect the yield (Figure 2), but due to large variation between plants the difference between control and the lowest level of damage was not statistically significant in any of the trials. Looking at the total picture, however, it is likely that the damage threshold in Norway is <12% bud loss in most cases. The damage threshold used so far is 10 cut buds per 40 m of row, corresponding to <1% bud loss even in very small plants. This seems very low. Further studies should concentrate on damage levels between 1 and 15%. The trials showed some compensation (i.e. % decrease in yield < % bud loss), but far from a full one. In the UK, Cross & Burgess (1998) found that 'Elsanta' planted the previous summer could sustain a bud loss up to 50% without a significant decrease in yield, while those planted in the autumn did not compensate. The Norwegian plants were planted in May-June. The compensation ability of strawberry plants may be larger in milder climates where the yield per plant in general is higher (Faby et al in press).

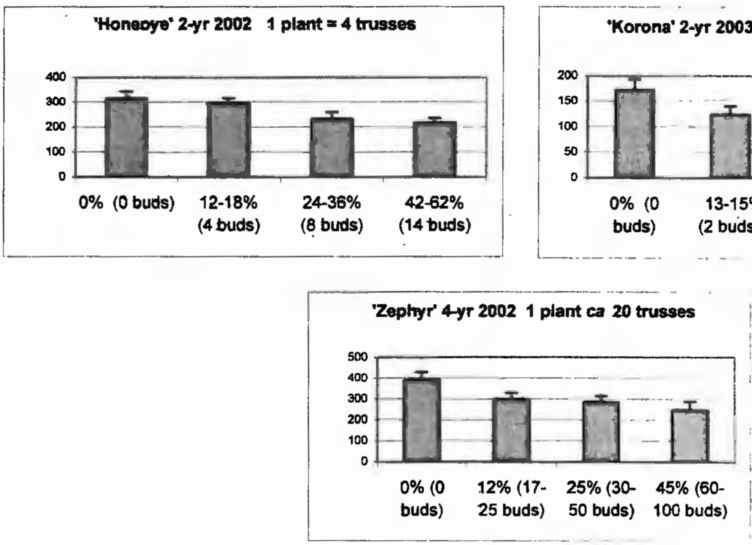

Figure 2. Saleable yield (>25mm) in g per plant* at different levels of bud removal. SE=Vertical bars (n=7). The effect of treatment was significant in 'Honeoye' and 'Korona' ($p<0.03$, one-way ANOVA after checking for block effect) and nearly so in 'Zephyr' ($p=0.057$). *In the 2 yr old plants, crowns were scattered and plant size was defined beforehand (tagging a fixed number of trusses).

Pesticide trial (2003)

The pesticide application did not have any effect on the large number of weevils or cut buds (Figure 3). This result agreed with the grower's experience with pyrethroids, indicating a possible tolerance for pyrethroids in the local weevil population. The area of the trial only took up a small proportion of the field, and theoretically the failure to detect any effect also could be due to weevils rapidly recolonizing the sprayed plants. As there was not even a tendency of less weevils in the treatment with thiacloprid (which is systemic), the most likely explanation is that the absence of effect was real, also in the case of thiacloprid and indoxacarb. It is important to keep in mind that thiacloprid was applied in a low dosage. A full dosage should be tested, perhaps also an earlier spraying time. Indoxacarb, which was used in full dosage (57 g.a.i/ha), does not seem to be a very promising pesticide against *A. rubi*.

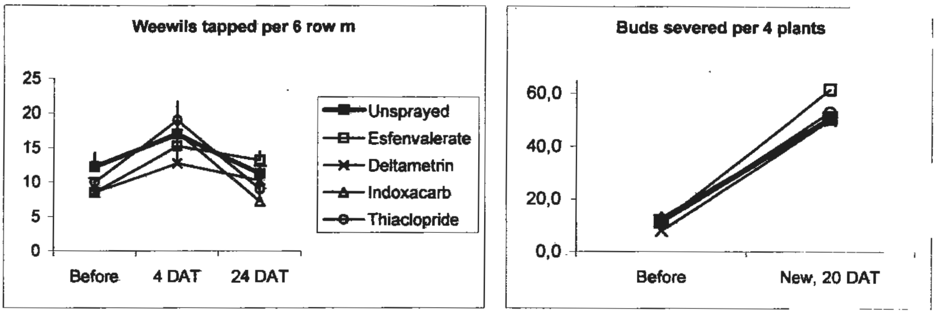

Figure 3. *A. rubi* registrations before and after spraying (DAT= Days After Treatment) with four pesticides in strawberry *cv* Korona at the start of flowering. Temperature at spraying was 22°C. The dosages per hl were: 50 ml SumiAlpha (esfenvalerate), 50 ml Decis EW15 (deltametrin), 17 g Steward 30WG (indoxacarb), and 25 ml Calypso 480SC (thiacloprid). The spraying volume corresponded to 1100 l/ha. Vertical lines are SE (n=4).

Damage and population size in organic fields

The number of cut buds increased in the first three years of harvest (Figure 4). However, this does not necessarily mean that % damage increased. In the first year of harvest there were 4.7 cut buds per flower truss (field 1); in the third year (with bigger plants), it had decreased to 3.7. This corresponds to a bud loss of 35-65% (depending on the number of buds per truss), which is far above most damage thresholds. In field 2, which was smaller and more isolated from other strawberry fields, the level of damage was lower. In the fourth year (field 1) the weevil population decreased, both in absolute and relative terms (2.6 cut buds per flower truss). The most likely reasons for this decline are reduced plant quality (the number of flower trusses per plant decreased from 19.4 in 2002 to 11.9 in 2003), increased mortality due to natural enemies, and/or less favourable climatic conditions for *A. rubi*.

The weevil phenology differed among the cultivars in field 3 (Figure 5). The weevil number in the late flowering cultivars (Symphony, Tyee) peaked after the earlier cultivars. If the bowl method reflects the true number of weevils present in the plants, this means that weevils prefer different plants at different times within a field. Trials comparing the weevil attractiveness or resistance of strawberry cultivars must therefore include the whole oviposition period to avoid biases created by phenological differences between cultivars.

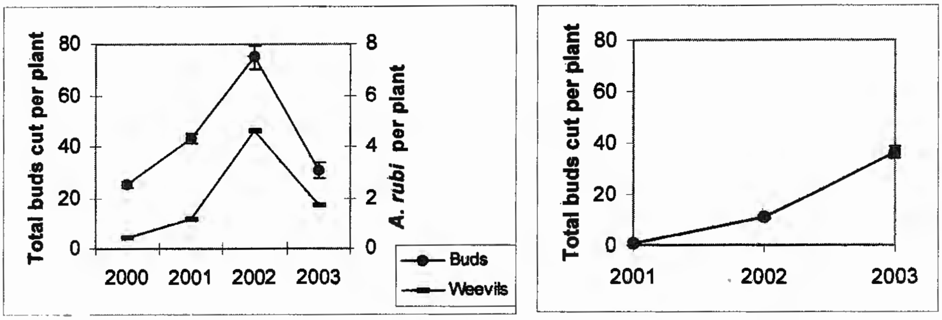

Figure 4. *A. rubi* development in two organic 'Korona' fields. Field 1 to the left (n= 5 plots; weevil number is the sum of weevils tapped in 4 annual visits). Field 2 to the right (n=4 plots).

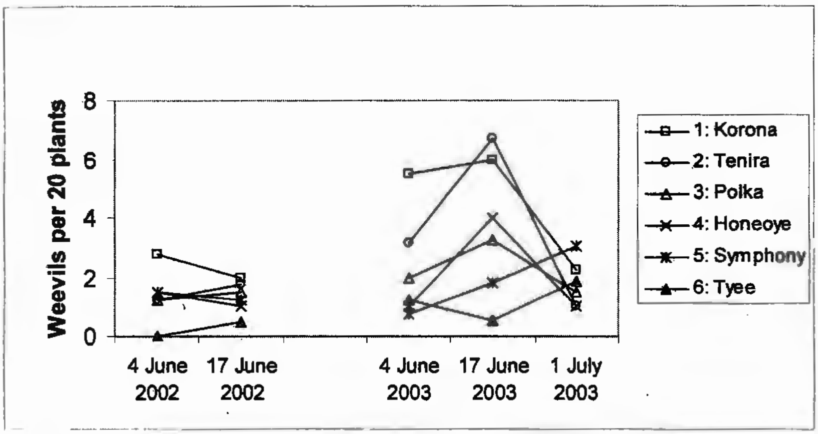

Figure 5. *A. rubi* registered with the bowl method in 6 organically grown cultivars during two seasons (mean of 4 replicates). The 2002 season was exceptionally early.

Exclusion fence

The number of adult weevils found outside the fence was 20 and inside it 13 (sum of the 5 registrations). This and other observations in the field indicated that the fence may have an effect on the weevil immigration pattern, but trials with replications and closer monitoring are needed to prove this. The presence of adult strawberry blossom weevils inside the fence could be due to: 1) Immigration of weevils looking for overwintering sites the previous autumn (immigration in spring not likely, the weather before the fence was put up did not favour weevil activity); 2) weevils flying above the fence (possible); or 3) weevils crawling beneath the fence (the most likely explanation - the fence was not as well secured at the base as it should have been).

***Bioassay with* Heterorhabditis megidis**

In many of the buds (57%) there was no visible *A. rubi* larva at the time of examination. Of the larvae present in the treated dishes, 68% were infected with *H. megidis* (0% in the control). This preliminary bioassay indicates that in two days IJs of *H. megidis* are able to penetrate strawberry buds on moist paper and infect the *A. rubi* larva developing within the bud. The rate of nematode infection was fairly high considering that the nematodes were required to find their way into the bud to reach the larva. The bioassay was conducted in moist, ideal conditions. In the field, conditions are probably somewhat drier and precarious. Another nematode species, *Steinernema carpocapsae,* is more resistant to desiccation and could be worth trying in more realistic experiments. Further investigations are necessary to investigate whether entomopathogenic nematodes are able to substantially reduce *A. rubi* populations in the field.

The large proportion of buds without a larva may indicate a high natural mortality rate of eggs that it would be interesting to study further. Also our attempts to rear parasitoids from cut buds have suggested a poor survival rate for immature weevils (as well as a low parasitisation rate). However, the large *A. rubi* damage in the organic field where the buds came from demonstrates that no natural factors were present to control the weevil at an agronomically satisfactory level.

Conclusions

In areas with a long history of intensive strawberry growing we need control strategies for *A. rubi* that are not based on pesticide application only. Both conventional and organic growers would benefit from this, as none of them have access to really efficient control methods at present. A better strategy will most likely integrate several methods. Less susceptible cultivars, microbiological control, mechanical exclusion, and manipulating weevil behaviour with semiochemicals (pheromones, plant volatiles) are possibilities presently being researched in Europe. Preliminary trials in Norway indicate that both mechanical exclusion with a mesh fence and application of entomopathogenic nematodes are worth investigating further. We also need research on the reasons for the poor performance of pesticides (application techniques, pesticide resistance/tolerance), and on the damage threshold (measured as weevil number) in different growing conditions. The bowl method is an efficient way to monitor the strawberry blossom weevil.

Acknowledgements

We thank the growers who let us do trials in their fields, and Inger Nordengen and Olav Sørbu for help in the field. We received financial support from our institutions, plus the Norwegian Agricultural Inspection Service (pesticide trial) and VIPS - The Norwegian warning service of plant pests and diseases (exclusion fence trial).

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Efficacy of chemical and biological control of the strawberry root weevil (*Otiorhynchus ovatus* L.) and the vine weevil (*Otiorhynchus sulcatus* F.) in strawberry plantations in Poland

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Abstract: Strawberry root weevil (*Otiorhynchus ovatus*) and sometimes also black vine weevil (*Otiorhynchus sulcatus*) destroy many plants in strawberry plantations in different regions of Poland. There have been only two compounds registered to control those pests on strawberry for last few years. One of them is diazinon as Basudin 10 GR or Diazinon 10 GR, which are registered to control the larvae in the spring before blossom and also after harvest of strawberry. The other is chlorpyrifos as Dursban 480 EC or Pyrinex 480 EC – used to control the adults and young larvae just after harvest. As the soil pests are still very important, we looked for new ways of controlling them. The effectiveness of some new compounds were studied in the field for the last two years. The treatment (to plants and the soil under them) was made in July, just after fruit harvest, when the adults were feeding on the leaves and laying the eggs in the soil and when some young larvae were present in the soil. Of the neonicotinoid insecticides evaluated, the best results were obtained with Actara 25 WG (thiametoxam) at a rate of 0.6 and 0.8 kg/ha (effectiveness was 81-99%, depending on the year and experiment). Mospilan 20 SP (acetamiprid) at rates of 0.3 and 0.6 kg/ha gave variable results, (efficacy 83-92% in 2001/2002, but 46-92% during 2002/2003, depending on plantation). Calypso 480 SC (thiacloprid) at a rate of 0.6 l/ha in experiments in 2001/2002 showed 73-93% of efficacy, but in the next year Calypso 480 SC used at a rate of 0.8 l/ha reduced the pest infestation by only 58-60%. The standard insecticide Dursban 480 EC (chlorpyrifos) in some experiments gave better results or similar effectiveness to the neonicotinoid insecticides (2001/2002), but in later experiments all the neonicotinoids gave better results than chlorpyrifos. The hot and dry weather was probably the reason for this result.

Biological control of root weevils with Larvanem (*Heterorhabditis megidis* entomopathogenic nematodes) gave excellent results (effectiveness 98.8%) in experiments in 2001/2002, but poorer results in the following year when the weather was dry and hot. Biological control with the fungus *Beauveria bassiana* in 2001/2002 gave good results.

Key words: strawberry root weevil, vine weevil, *Otiorhynchus ovatus, O. sulcatus*, chemical and biological control, strawberry, neonicotinoid insecticides

Introduction

Root weevils are very important pests in many strawberry plantations in Poland. The main species is strawberry root weevil *Otiorhychus ovatus*, but the black vine weevil *O. sulcatus* can also be damaging.

The main damage is caused by the larvae of strawberry root weevils, which can destroy many plants, mostly in older plantations. The most serious damage is caused by the more mature larvae in May or early June, when large numbers of larvae feed on roots and crowns. Strawberry plants can even be totally destroyed (Penman & Scott, 1976; Łabanowska, 1994;

Łabanowska et al., 2003). Circular patches of weak or dead plants can be observed in heavily infested plantations, especially in those that are 2-3 years old or more. Currently, only two compounds are registered for control of root weevils on strawberries. They are: 1. granular Basudin 10 GR and Diazinon 10 GR (diazinon) for control of larvae in the spring before the blossom of strawberry, after harvest; or before strawberry planting. 2. chlorpyrifos as Dursban 480 EC and Pyrinex 480 EC for control of the pest before planting or to control adults and young larvae just after harvest. The above pests are not easy to control. Several other researchers have looked for new compounds and methods to control the root weevils (Bogatko & Łabanowski, 1993; Cross & Burgess, 1993; Malinowski et al. 2001; Willmott et al, 2002; Łabanowska & Olszak, 2003).

Entomopathogenic fungi are the most common pathogens isolated from larvae and adults of *Otiorhynchus* sp. (Zimmermann, 1996). The most important species causing heavy infections of larvae in field populations are: *Metarhizium anisopliae*, *Beauveria bassiana* and *Paecilomyces fumosoroseus* (Marchal, 1977; Miętkiewski et al. 1992; Zimmermann,1996).

We are still looking for some modern way to control soil pests, suitable especially in strawberry IFP, but also for traditional methods of fruit production. The effectiveness of some new, mainly neonicotinoid compounds (insecticides) and some biological agents were studied in the field during last years.

Material and methods

The experiments were carried out in 2001-2003 by the Research Institute of Pomology and Floriculture in Skierniewice. The field tests on strawberry root weevil control were performed in several plantations located in the central part of Poland. Randomised complete blocks experimental designs were used, each one 70-1000 m^2 in area and containing 4 replicate plots. For chemical control the emulsified, mainly neonicotinoid insecticides were tested. They were as follows: acetamiprid as Mospilan 20 SP, imidacloprid as Confidor 200 SL, thiacloprid as Calypso 480 SC and thiametoxam as Actara 25 WG. Chlorpyrifos as Dursban 480 EC was used as a standard. These insecticides were applied as plant and soil sprays before bloom or after harvest. A knapsack motor sprayer "Stihl" was used for the small blocks and a tractor sprayer with "Fragaria" ending on big blocks was used to apply the tested compounds in a volume of spray liquid of 750 l per ha.

The fungus *Beauveria bassiana* on wheat seeds and *Paecilomyces fumosoroseus* (spore concentration of 10^6 ml^{-1}) isolated from *Otiorhynchus ovatus* collected from a strawberry field (Miętkiewski et al., 1992), as well as entomopathogenic nematode *Heterorhabditis megidis* as "Larvanem" in water suspension were applied to the soil in the spring or after harvest.

The efficacy of the treatments was evaluated in June or early July by counting larvae, pupae and adults. Six plants per plot, i.e. 24 in each treatment were removed, their roots and sieved soil were searched for the pests. The results were elaborated with analysis of variance on data transformed according to the formula y=log (x+1), were x is the number of specimens. Differences between means were checked with Duncan's multiple range "t"-test at 0.05% significance level. The results are presented in Figures 1 - 4.

Results

Chemical control

Field summer experiments 2001/2002 and 2002/2003

Neonicotinoid insecticides: Actara 25 WG (thiametoxam), Confidor 200 SL (imidacloprid), Calypso 480 SC (thiacloprid) and Mospilan 20 SP (acetamiprid) used after fruit harvest gave quite good results in controlling the strawberry root weevils (Figs.1 - 2).

Field spring experiments 2002

Neonicotinoid insecticides: Actara 25 WG, Mospilan 20 SP and Calypso 480 SC used in the spring, about 2 weeks before bloom, gave good control of the strawberry root weevils (Fig. 3).

Biological control

The fungi *Beauveria bassiana* (exp. 2) and *Paecilomyces fumosoroseus* (exp. 1) applied just after harvest, gave good control of the strawberry root weevils. Also the entomopathogenic nematode *Heterorhabditis megidis* as "Larvanem" (exp. 3) showed very high reduction of this pest (Fig. 4). However, when the biological compounds were applied during the hot and dry summer of 2002, the results were poor.

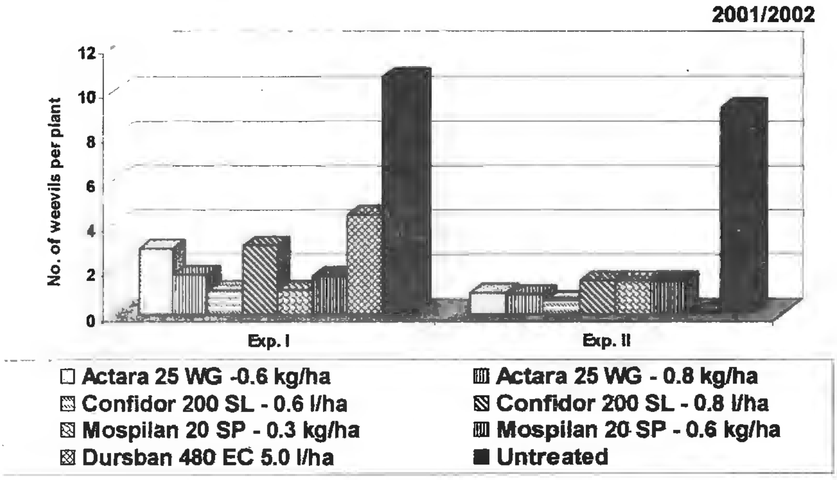

Figure 1. Effectiveness of neonicotinoid insecticides for control of the strawberry root weevils on strawberry after harvest (Treatment July 2001, checked July 2002).

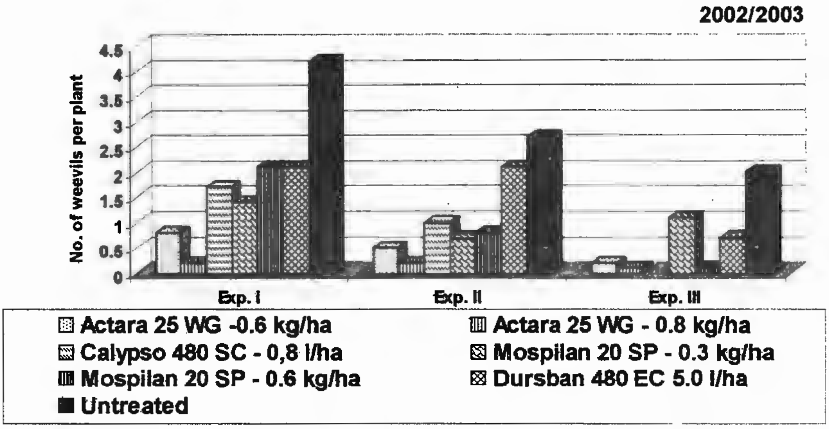

Figure 2. Effectiveness of insecticides for control of the strawberry root weevils on strawberry after harvest (Treatment July 2002, checked June 2003).

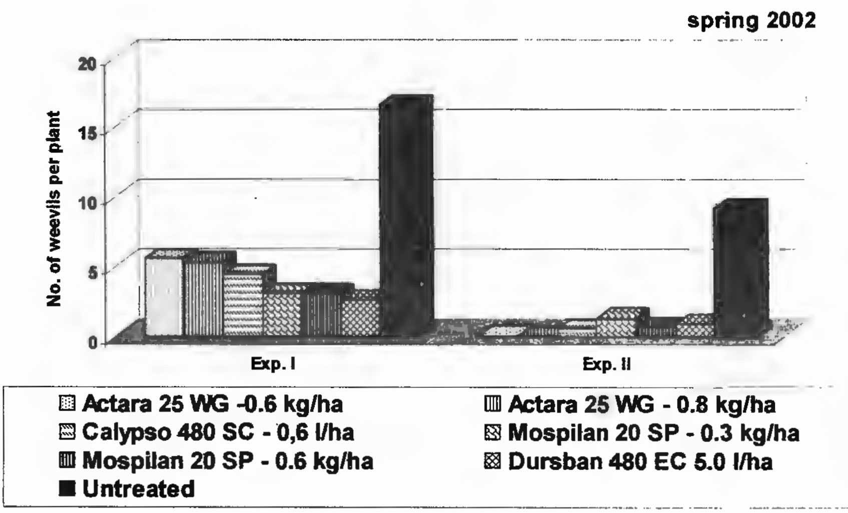

Figure 3. Effectiveness of neonicotinoid insecticides used in the spring before bloom of strawberry for control of the strawberry root weevils.

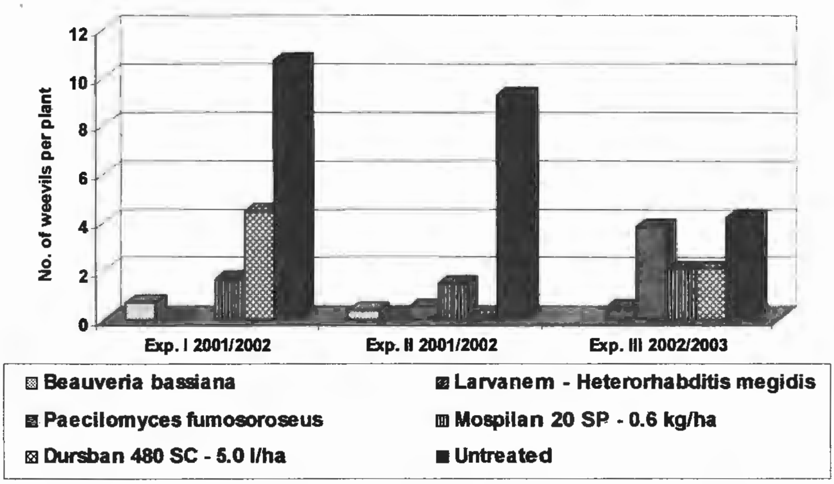

Figure 4. Effectiveness of biological control of the strawberry root weevils on strawberry.

Discussion

Chemical control

Actara 25 WG at the rate of 0.6 and 0.8 kg/ha used after fruit harvest of strawberry showed quite good reduction of the strawberry root weevils. The efficacy ranged from 82-96.7%, depending on the dose and field location. Results obtained with Actara 25 WG were similar or better than those obtained with the standard insecticide - Dursban 480 EC (5.0 l/ha). Mospilan 20 SP at the rate of 0.3 and 0.6 kg/ha, applied after harvest, reduced the number of root weevils from 51 to 96.7%, depending on the rate and the field. The results were similar to those obtained with standard insecticide – chlorpyrifos as Dursban 480 EC. Calypso 480 SC at the rate of 0.8 l/ha applied after harvest reduced numbers of strawberry root weevils by 59-61%. The results were better or similar to those obtained with the standard – Dursban 480 EC. Usually the results obtained with Dursban 480 EC were very good, good or satisfactory. However, the efficacy was worse when it was applied during hot and dry summer weather (2002). Results obtained with imidacloprid and thiametoxam confirmed the first experiments with these insecticides (Łabanowska & Olszak, 2003).

Biological control

The best results were obtained with "Larvanem" – containing the entomopathogenic nematode *Heterorhabditis megidis*. The efficacy was 89-99%, but when used into dry soil efficacy declined to 74%. Efficacy of the fungus *B. bassiana* was 93-95%, but it was difficult to apply under plants. In earlier experiments it gave poorer results (Łabanowska & Olszak, 2003)

The fungus *Paecilomyces fumosoroseus* gave good control. However, the results were poor, when it was applied during dry weather. There are only a few data on microbial control of *O. sulcatus* and *O. ovatus* in outdoor experiments. Isolates of *B. bassiana. B. brongniartii*, *P. fumosoroseus* and *M. anisopliae* failed to reduce larvae populations significantly on strawberry plants in the field trial (Soares et al., 1983; Vainio & Hokkanen 1993;

Zimmermann,1996) and efficacy depending on fungus species achieved from 30% up to 97.2% (Zimmermann, 1996).

Nematodes and fungi gave best results, in high moisture conditions and when they were applied onto warm soil. It confirmed results obtained by Kakouli-Duarte et al. (1997), that the nematode *Heterorhabditis megidis* can be effective in controlling larvae of *O. sulcatus* in moist soil.

Conclusions

- Neonicotinoid insecticides: Actara 25 WG (thiametoxam) and Mospilan 20 SP (acetamiprid) gave satisfactory results in control of the strawberry root weevils. They will be useful to control these pests on strawberry after being registered.
- "Larvanem" - a formulation of the entomopathogenic nematode *Heterorhabditis megidis* - gave good control of the strawberry root weevils and will be useful in warm and wet weather conditions.
- Calypso 480 SC (thiacloprid) gave satisfactory results in first experiments, but further experiments are needed.
- The fungi *B. bassiana* and *Paecilomyces fumosoroseus* gave good control, but further experiments are needed.

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Importance of simulated damage to flower buds by strawberry blossom weevil on raspberries

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Abstract: The strawberry blossom weevil (*Anthonomus rubi* Herbst) is an important pest of raspberry crops in Switzerland. To get more information for managing this pest, the ability of raspberries to compensate for flower bud removal was analysed. A simulation of the damage by blossom weevil was conducted by removing manually an increasing percentage of flower buds (0, 10, 20, 30 % for cv. Zeva 2 and 0, 5, 10, 15 % for cv. Glen Prosen). The results showed that berry yield was strongly related to the residual number of flowers per cane. However, mean berry weight was not influenced by the number of flowers per cane. It is concluded that raspberries are not able to increase berry weight with a decreasing number of flowers and that the damage caused by blossom weevil reduces yield linearly. The consequences of these results for defining the damage threshold value for flower bud severing by blossom weevil in raspberries are discussed.

Key words: *Anthonomus rubi*, berry weight, flowers per cane, *Rubus idaeus*, yield

Introduction

The strawberry blossom weevil (*Anthonomus rubi* Herbst) is an important pest of cultivated raspberry in Switzerland. The major damage is caused by the female weevil incising the stalk of the flower bud after oviposition. The damaged flower bud ceases to develop and does not set fruit. Locally the blossom weevil can cause important damage severing up to 33 % of buds (Mariéthoz, 1993).

The effects of severing damage caused by *A. rubi* on yield and size of fruit are unclear for raspberries. In contrast, Höhn & Neuweiler (1993) showed, that for strawberries the importance of the damage depended on the cultivar and the intensity of flowering. Cultivars with a high flower number per plant, such as Polka and Elvira, had no yield reduction even when 20-30 % of flower buds were severed. Other studies on strawberries with simulated removal of flower buds showed that moderate severing damage did not significantly affect yield (Terrettaz et al., 1995; Cross & Burgess, 1998, English-Loeb et al., 1999). The compensation for bud removal seems to be achieved by increasing mean berry weight and increasing the number of higher order buds that mature. However, a recent study showed that the removal of the largest strawberry flowers at the beginning of the flowering period can reduce yield (Faby et al., 2003). The fruit size increased only slightly and could not compensate for this loss. Nearly no experiments have conducted on this topic on raspberries.

To get more information to aid management of *A. rubi* in raspberry crops, the effects of the incidence of different intensities of flower bud removal on yield and berry weight were evaluated. These results should help definition of a damage threshold value for flower bud severing by blossom weevil in raspberries.

Material and methods

Two experiments were done at the research station in Bruson (1100 m a. s. l.) of the Centre for Arboriculture and Horticulture (RAC, Switzerland). The raspberry crops were planted in 1993 on a light soil (10 % clay) with 3.5 % organic matter and with a pH of 6.5. Between the rows (distance of 2.20 m) the soil was covered with a frequently cut grass mixture. In the row, herbicides were used to control weeds. The raspberry plants were irrigated by overhead sprinklers and fertilisation was made according the norms for an estimated yield of 20 t of raspberries per ha (60 kg N, 40 kg P_2O_5, 85 kg K_2O, 15 kg Mg per ha). Pyrethroid was sprayed at the beginning of flowering to avoid an interference with real damage of the *A. rubi.*

In the first experiment with the cultivar Zeva 2, the treatments consisted of 4 intensities of artificial flower bud severing: 0, 10, 20, 30 % of the flowers were manually removed on 13 June 1995, after counting all the buds/flowers per cane. A similar number of first, second and third order flower buds were clipped. Plots consisted of 4 canes. Thus there were 4 treatments replicated 3 times in a randomised complete block. The harvest period lasted from 24 July to 14 August 1995 (9 picks).

In the second experiment with the cultivar Glen Prosen, 0, 5, 10, 15 % of the flower buds were clipped on 21 June 1996 on 4 canes per replication. There were also 4 treatments replicated 5 times in a randomised complete block. The harvest period lasted from 16 July to 19 August 1996 (14 picks).

The impacts of decreasing flower bud numbers per cane on yield and on average fruit weight were analysed. Berry weight was measured from a sample of 100 g for each harvest.

Results and discussion

Effects of removal of flower buds on yield and fruit weight

In the first experiment, examining the effects of manual removal of flower buds of raspberries, the yield of the cultivar Zeva 2 decreased significantly with increasing severing intensity from 0 to 30 % of cut flower buds (Table 1). However, there was no significant difference in the mean berry weight between treatments. The second experiment with the removal of 5, 10, 15 % of the flower buds per cane on the cultivar Glen Prosen showed similar results as the first one (Table 2).

Table 1. Effects of manual removal of flower buds on yield per cane and mean fruit weight of the cultivar Zeva 2. Values are the mean of 3 replicates.

Flower buds removed	Flower number per cane before removal	Flower number per cane after removal	Yield per cane (g)	Fruit weight (g)
0 %	156	155	421	3.7
10 %	150	135	378	4.1
20 %	155	124	360	3.9
30 %	152	106	307	4.0
LSD[1)]	*n.s.*	*15.8*	*16.7*	*n.s.*

[1)] 'LSD' means the least significant difference according to the Fisher test with $P<0.05$

Table 2. Effects of manual removal of flower buds on yield per cane and mean fruit weight of the cultivar Glen Prosen. Values are the mean of 5 replicates.

Flower buds removed	Flower number per cane before removal	Flower number per cane after removal	Yield per cane (g)	Fruit weight (g)
0 %	269	269	820	3.7
5 %	257	244	710	4.0
10 %	243	219	683	3.9
15 %	265	225	630	3.8
LSD[1)]	*n.s.*	*36.4*	*123*	*n.s.*

[1)] 'LSD' means the least significant difference according to the Fisher test with $P<0.05$

For both cultivars the relation between the yield per cane and the residual number of flower buds per cane was very strong (Figure 1 and 2). However, there was no relation between the mean fruit weight and the residual number of flower buds per cane (Figure 1 and 2). These results showed that there was no yield compensatory effect. The decreasing flower number per cane did not influence the mean fruit weight as observed in some trials with strawberries (Höhn & Neuweiler, 1993; Terrettaz et al., 1995; Cross & Burgess, 1998; English-Loeb et al., 1999). For strawberries, the compensation for severing damage is apparently related not only to the increase in berry weight but also to the increase in the number of higher order buds matured. However for raspberries the latter mechanism was not available, because nearly all the fruits are harvested.

In conclusion, raspberries are not able to increase berry weight with decreasing number of flowers. Severing damages by *A. rubi* reduces the yield linearly.

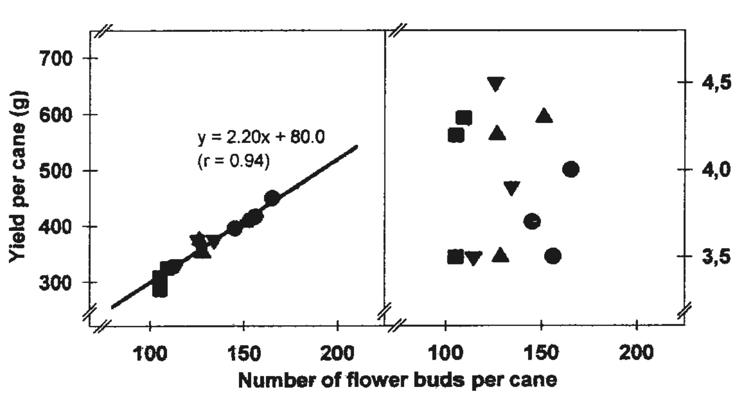

Figure 1. The relation between the number of flowers per cane and the yield per cane, together with the relation between the number of flowers per cane and the mean berry weight for the raspberry cultivar Zeva 2 after manual removal of an increasing percentage of flower buds: 0 % (●), 10 % (▲), 20 % (▼), 30 % (■).

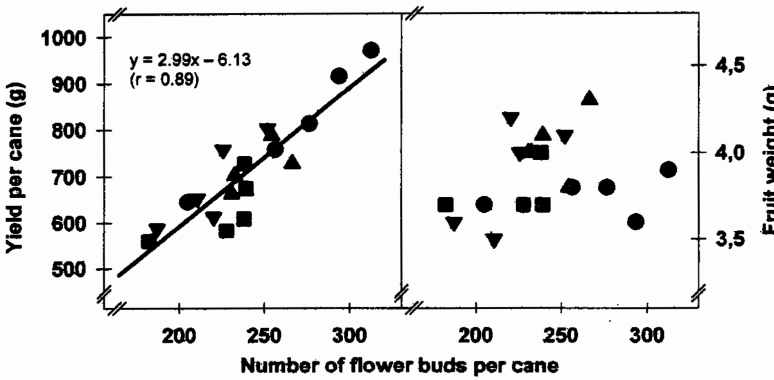

Figure 2. The relation between the number of flowers per cane and the yield per cane, together with the relation between the number of flowers per cane and the mean berry weight for the raspberry cultivar Glen Prosen after manual removal of an increasing percentage of flower buds: 0 % (●), 5 % (▲), 10 % (▼), 15 % (■).

Damage threshold value

The economic threshold value for flower bud severing by *A. rubi* in raspberries is lower than 1 % loss in yield (Mittaz et al., 2002). Assuming that losses of flower buds by *A. rubi* linearly decreases yield (Figure 1 and 2), that a raspberry cane develops about 200 flower buds (Table 1 and 2) and that a female severs 20 – 30 flower buds (Bovey *et al.*, 1972), it can be concluded that one female of *A. rubi* per 10 to 15 canes is sufficient to reach the economic damage threshold value. A damage threshold value of one female of *A. rubi* per 1 linear row metre (corresponding to 10 to 12 canes) is proposed.

The tapping method is recommended for monitoring of *A. rubi* (Mariéthoz, 1993). The advantage of the tapping method is that *A. rubi* can be detected before the occurrence of damage, in contrast to visual methods based on the density of clipped flower buds. By tapping lightly on the middle of the canes and putting a collection tray against the raspberry hedge at 30 cm above the soil, the presence of the pest can be detected. Assuming that only half of the total adults fall from the canes into the tray (the other half on the other side of the hedge falls on the soil) and that the sex ratio is 1:1 as described for *A. signatus* (Mailloux & Bostanian, 1993), a provisional damage threshold value can be defined:

Adults of A. rubi collected $\geq$ *lenght of the recipient (m)* x *number of samples.*

For example with a 0.4 m length recipient and 10 samples repeated on different sites of the field (control on 4 m or 40 to 48 canes) the damage threshold value is reached when 4 adults of *A. rubi* are captured. Farmers should begin to check their fields early in the season, as soon as the flower buds are visible. The monitoring should be repeated at least once a week. The captured adults of *A. rubi* with this monitoring method have to be cumulated. In the example mentioned above, if in addition 4 adults of *A. rubi* were captured, the economic threshold value is achieved and a pesticide treatment seems to be justified.

Further experiments have to confirm the sex-ratio of *A. rubi*, the potential of severing damage of one female (number of severed buds per female) and the validity of this monitoring method.

Conclusions

The manual removal of 5 to 30 % of the flower buds decreased the yield of raspberries linearly and mean fruit weight was not influenced by these treatments. No compensatory ability could be identified and therefore raspberry yield was very strongly related to the number of flowers per cane. Consequently, severing damage by *A. rubi* reduces yield linearly. In relation to these results, a provisional damage threshold value of 1 female of *A. rubi* per 1 linear row metre (corresponds to 10 to 12 canes) is proposed.

Acknowledgements

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Jaapiella vacciniorum Kieffer (Diptera: Cecidomyiidae); a new pest of highbush blueberry (*Vaccinium corymbosum*) in Trentino, Italy

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Abstract: In the early 1990s, blueberry producers in Trentino (alpine region of north Italy) reported severe attacks by a new gall midge *Jaapiella vacciniorum* (Kieffer, 1913) syn. *Dichelomyia vaccinii* (Rübsaamen, 1895). The larvae fed on vegetative buds thus preventing their development. In 1997 and 1998 investigations on the biology, behaviour, monitoring and control were done in a plantation near Pergine Valsugana where the effectiveness of two insecticides, vamidothion and fenitrothion were also assessed. The adult flight periods were monitored using white and yellow sticky traps (Rebell ® bianco and amarillo). Visual inspections of tips were used to study egg laying and larval population development on bushes, and to compare the effectiveness of the insecticides. Results indicate that the midge probably spends the winter as a mature larvae or pupae in the soil under infested plants. First adults emerge in late March/early April, when blueberry buds are just opening. During the summer, the midges complete a generation (from egg to adult) in about 24-28 days. Six to seven generations may to be possible in Trentino. The peak of larval infestation normally occurs during August. White sticky traps and visual inspections of tips were shown to be equally effective for detecting the start of adult activity in the field, but they are difficult for the growers to use. Since the larvae live protected inside the tips and the pest completes many generations in a year, its chemical control seems to be very difficult. In our trials, fenitrothion gave the best results, keeping infestation below 5% of infested tips in 1998, but 10 applications were required to get this result. Further trials are needed to obtain more information about biology, monitoring tools, effectiveness of new chemical compounds and control strategies of this pest.

Key words: blueberry, *Jaapiella vacciniorum*, biology, monitoring, chemical control, fenitrothion, vamidothion

Introduction

Highbush blueberry (*Vaccinium corymbosum*) has been cultivated in Trentino since the early 1980s and by 2002, approximately 50 ha were grown, producing some 200 t of fresh fruit, mainly for the Italian market. In addition, some 30-40000 nursery plants are produced annually.

As a recently introduced crop, there are few indigenous pests of highbush blueberries in Trentino. The pests that do occur include the otiorhynchid weevils, some geometrid and noctuid moths (e.g. *Opheroptera brumata*, *Conistra vaccinii*, *Eupsilia transversa*) and some unidentified species of scale insects and aphids. In early 1990, larvae of the gall midge *Jaapiella vacciniorum* Kieffer were first detected by the growers and identified by Dr. Marcela Skuhrava. *J. vacciniorum* is a European species known to develop in galls in the shoot tips of the native blueberry (*Vaccinium myrtillus* L.). It occurs in mountainous areas in Europe, especially in the Alps. The native blueberry is widespread in the main areas of highbush blueberry production in Trentino, and it is likely that this is the source of infestation. As there is very little information known about this insect we studied its biology and behaviour in 1997 and 1998. In addition, monitoring and control strategies were investigated. This paper reports the results of these trials.

Material and methods

Population development and biology of the pest

During both years, investigations were carried out in the same plantation, near Pergine Valsugana, Trentino at an altitude of about 600 metres above the sea level. Observations on the biology and population development were carried out on fruiting plants (grown in field soil and in pots) from the middle/end of March (buds breaking) until October, and on nursery-potted plants, exposed in the open field at the end of April. These potted plants were renewed each year and were used to test the effectiveness of insecticides against the pest.

The flight periods of the adults were monitored in 1997 by means of white and yellow sticky traps (Rebell ® bianco and amarillo; single panels). Ten traps (five per colour) were placed in treated blocks of the nursery plantation (1 trap in the middle of each replication). Information about the adult flight activity was obtained by means of 10 additional traps (5 per colour) placed on fruiting plants prior to placement of the nursery potted plants.

The sticky traps were changed every 7-15 days and examined in the laboratory under a dissecting microscope. Adults in the traps were identified by comparing them with individuals (males and females) that emerged from mature larvae collected from infested bushes and reared in vials with sand for a short time.

Visual inspections (under dissecting microscope) of tips randomly collected (1 tip/plant) at 7-15 days of interval were used to study egg laying and larval development on bushes. After the potted plants were placed in the plantation sampling was done exclusively on these plants. No samples from fruiting plants were taken in 1998. Air temperature data were collected from a meteorological station close to the trial site.

Effectiveness of insecticides

In 1997, a trial was carried out with the aim of evaluating the effectiveness of two insecticides, fenitrothion and vamidothion, against *J. vacciniorum.* About 17000 nursery potted plants (90% cv. Brigitta; 10% cvs. Spartan and Berkeley) were exposed in the field at the end of April. They were arranged in 11 square plots (each of about 1500 pots). Two rows, each with 5 plots/row and separated by a path 1.5 m wide were treated with vamidothion and fenitrothion in alternate plots. One untreated plot placed by the side of the row was used as a control block. Application was done by a hand held lance mounted on a sprayer machine and towed by a tractor. The control block was treated with insecticide at the end of the summer to try to limit the economical damage for the grower. In 1998, a trial was repeated using only fenitrothion, the product that gave the best results in 1997. Around 17000 pots were arranged in two blocks; 15000 plants were treated with fenitrothion every 7-10 days since the beginning of eggs deposition. Because of an error, this block was treated with vamidothion on the 6th of August. The remainder of the plants were untreated and used as control block, about 20 metres away from treated block. The sequence of the treatments is indicated in Table 1.

The effectiveness of the treatments was assessed in both years by visual inspections (under dissecting microscope) of randomly collected plant shoot tips every 7-15 days from the middle of each replicate. The sample consisted of 25 tips/replicate (1 tip/plant) in 1997; 200 tips from the treated block and 50 from the control block in 1998. The percentage of occupied apices and the number of viable eggs and larvae were recorded at each inspection.

Table 1. Treatments applied on nursery plants in 1997 and 1998.

Year	Insecticides Product	Active ingredient	Rate (g/hl)	Application volume (l/ha)	Application dates	Replications or blocks
1997	Fenitrocap	fenitrothion	250	850–1200	23,29 May 6,17,30 Jun 9,22 Jul 20 Aug	5 repl. On 15 Sep, treated also the control block
	Kilval	vamidothion	125			5 repl. On 20 Aug and 30 Aug, treated also the control block
1998	Fenitrocap	fenitrothion	250		4,13,22 May 3,9,20,30 Jun 10,24 Jul 22 Aug	1 block. On 6 Aug, treated with vamidothion

Results and discussion

Population development and biology of the pest

We describe the probable life cycle of the midge in Trentino. *Jaapiella vacciniorum* probably over-winters as mature larvae or pupae in the soil under infested plants. The first adults emerge from the soil very early the next season, at the end of March/beginning of April, when the buds of blueberry are just opening. We recorded the first eggs on tips the 3 April in 1997 (see Fig.1).

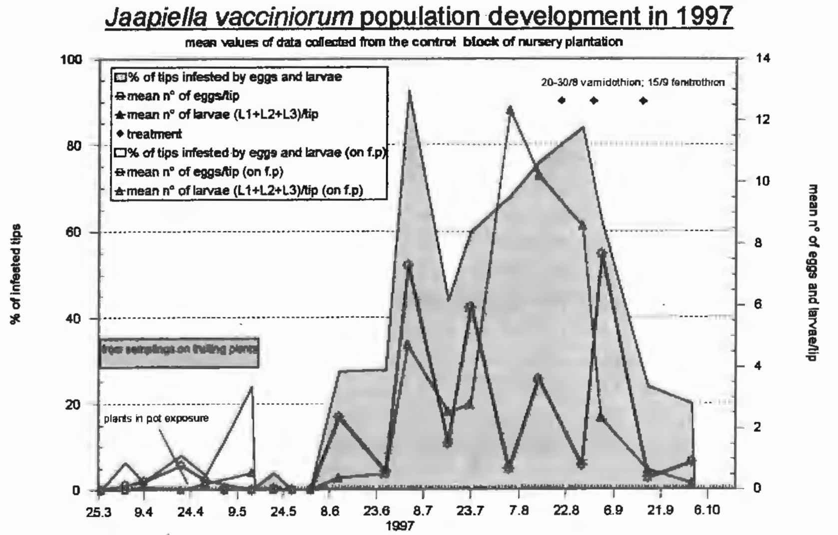

Figure 1. *Jaapiella vacciniorum* population development in 1997.

In the period between the 26 March and the 3 April 1997, we also caught the first adults on sticky traps (see Fig.2). It is possible that adults of *Jaapiella*, as those of many other cecidomyiid midge species, have a very short life-span and the females caught on the sticky traps may have been mated shortly after emergence. These females have long telescopic ovipositors and use them to lay the eggs between the layers of the densely folded leaves in the shoot tips of *Vaccinium corymbosum.*

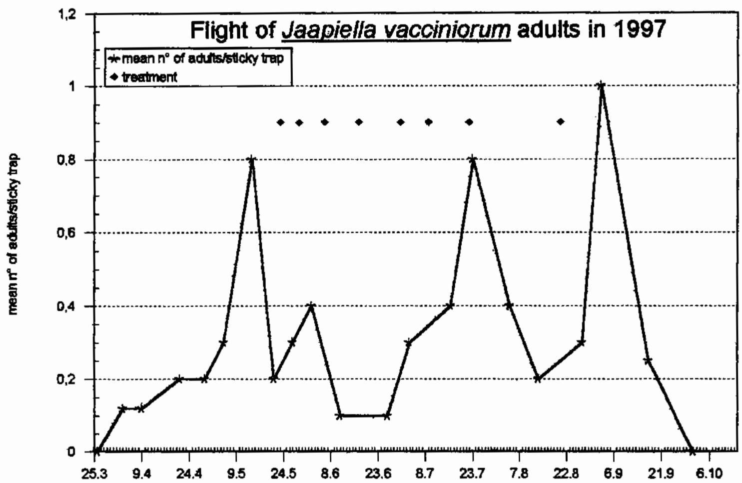

Figure 2. Flight of *Jaapiella vacciniorum* in 1997. After the 5th of May, the flight was recorded with traps placed only in nursery plants treated plots.

The eggs hatch in a few days. In 1997, the first young larvae (first generation larvae) were found in tips the 9 April (see Fig.1), 6 days after first eggs discovery. The young larvae migrate towards the inside of the shoot tip to find protection and to start to feed.

These first steps of the infestation seem to be the most difficult and crucial for the pest. Figurel shows that the development of the larvae on fruiting plants in April 1997 was erratic despite regular and continuous egg deposition. Normal development was not observed until the end of April and in later samples. Data from the meteorological station indicated that during April 1997 the minimum air temperature was particularly low, much lower than that recorded in April 1998. The monthly average of the minimum temperature was 0.85 °C in April 1997, while in April 1998 it was 4.4 °C. The minimum air temperature was below 0°C on 12 occasions and during the period from 15 - 19 April the average of the minimum temperature was –2.9 °C, with a low of -4.6 °C. This may indicate that, in some seasons the eggs laid by the first emerged females and the very young larvae are exposed to a high risk of mortality, probably due to very cold temperatures.

Therefore, in 1997, few of the eggs laid before the 20 April hatched and most of the larvae (first generation) that were recorded on the 29 April and the 14 May (see Fig.1) probably developed from eggs laid after 20 April. The cold temperatures during mid April probably delayed emergence of over-wintering adults from the soil. Most adults were caught on sticky traps in the period between the beginning and middle of May (Fig. 2) and this coincided with peak colonisation of the shoot tips (Fig. 1).

The development from egg to adult of the first generation was completed in about 30-34 days. When fully mature, the whitish larvae dropped out of the shoot tips and pupated in the soil. By the end of May an increase in flying adults (first generation) was observed and this partially overlapped with the flight of the over-wintering adults and peaked at the beginning of June (Fig. 2). This was confirmed by a peak in egg laying around the 10 June (Fig.1).

Subsequent generations followed at 24-28 day intervals. Looking at the number of the peaks of egg deposition in Figure 1, we suppose that a total of 6-7 generations possibly occurred in 1997.

Monitoring of the pest

Sticky traps were unable to differentiate between the summer generations. However, both the two main summer peaks visible in Figure 2 developed shortly after the two peaks of the larval population (Fig.1). The number of adults caught during the maximum summer flight activity was extremely low and comparable to the number of adults caught during the spring (May). These observations indicate poor effectiveness of attraction of the traps during the summer.

White traps were slightly more attractive than the yellow traps. White sticky traps and visual inspections of shoot tips were shown to be equally effective in detecting the start of adult activity in the field. Identification of the adults of *Jaapiella* from other flying midges on the sticky traps is difficult. Also the inspection of the tips is difficult, because of the very small size of eggs and larvae.

***Description of the damage on* Vaccinium corymbosum**

Feeding by larvae of *J. vacciniorum* within the tightly folded shoot tips prevented normal development. At first, infested tips appear to be swollen and turgid and then rapidly twist before turning brown and breaking off the stem. New shoots develop from healthy buds below damaged tips but they may become infested by later generations.

The damaged shoots tend to produce vegetative buds rather than flower buds affecting next seasons fruiting potential. The midge does not readily infest fruiting buds. Therefore, damage is important on nursery plants and on young fruiting plants.

Effectiveness of insecticides

Although insecticide treatments were applied at 10 day intervals in 1997, there was only a moderate reduction in midge infestation in the treated plots compared with the control block (Fig.3) and fenitrothion was more effective than vamidothion.

By the end of the growing season moderate economical damage was not avoided on treated plants.

The trials were repeated in 1998 with the aim of evaluating the effectiveness of fenitrothion. The whole nursery plantation (except for the control block) was treated with this chemical, resulting in damage of less than 5% of infested shoot tips. Our results suggest that chemical control of this pest is very difficult and the high frequency of applications of single chemicals may lead to the development of resistant strains of the midge. Beneficial species within the plantations may also be adversely affected.

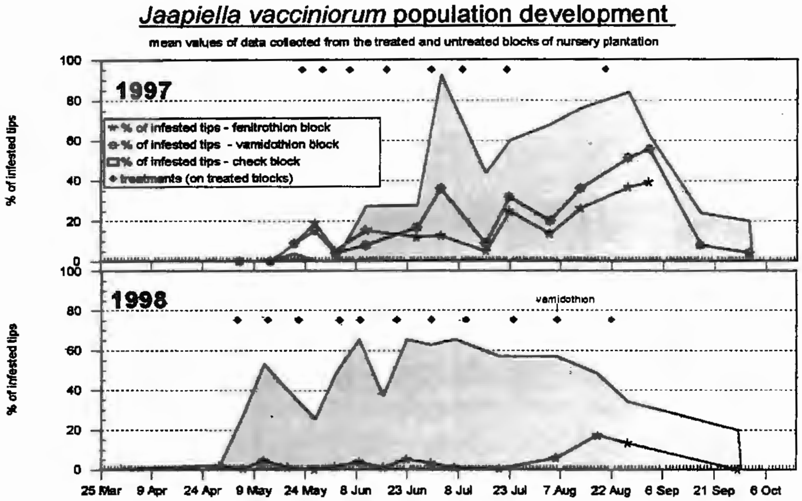

Figure 3. Development of the infestation in sprayed and unsprayed plots in 1997 and 1998

Conclusions

The understanding of the biology of this pest on blueberry has been improved. Additional studies are necessary to get more information on the biology and population development of this insect, especially on the native blueberry. Observations should be carried out in order to verify if other similar cecidomyiid midge species are present in Trentino (e.g. *Dasineura oxycoccana*, which has been already found in Piemonte, Italy) (Bosio et al., 1998).

Native blueberry may support indigenous parasites and predators that may be used to manage *J. vacciniorum*. Additional monitoring tools such as a forecasting emergence model based on day degree or pheromone trap catches should be considered and more effective insecticides should be sought.

Acknowledgements

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Annex

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The Potential for Biocontrol of Rosy Apple Aphid (*Dysaphis plantaginea*) using Entomopathogenic Fungi

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Abstract: Rosy apple aphid (*Dysaphis plantaginea*) is a serious pest of apples in the UK. Whilst there are several entomopathogenic fungi developed for biological control of aphids, their efficacy against the rosy apple aphid is unknown. A laboratory bioassay method was developed which allows rapid evaluation of the pathogenicity of fungi to apterae of rosy apple aphids on the autumn host, plantain (*Plantago* spp.). The efficacy of the mycoinsecticide products Bontanigard ES™, Vertalec® and Mycotal® against aphids was tested using this method. Mortality of aphids treated with mycoinsecticides was large (greater than 87% for all treatments after 7 days) and mortality of control aphids was small (less than 3% after 7 days). Future use of this bioassay method for evaluating the potential for using entomopathogenic fungi in the biocontrol of rosy apple aphids is discussed.

Key words: *Dysaphis plantaginea*, entomopathogenic fungi, biological control, bioassay

Introduction

The rosy apple aphid (*Dysaphis plantaginea* (Passerini)) is one of the most serious pests of apple (Massee, 1954). The aphid has a holocyclic life cycle and overwinters as eggs on the primary host, apple (*Malus* spp.). During the summer alate aphids migrate to the secondary host, plantain (*Plantago* spp.) although some colonies of aphids may persist on apple until late summer. The aphids remain on plantain until early autumn when aphids migrate to apple. The aphid causes characteristic damage by distorting and curling outer rosette leaves (Forrest & Dixon, 1975), making it difficult to control using conventional pesticides. Additionally, strains of rosy apple aphid in central and southern Europe have developed resistance to some aphicides. This has led to investigation of other methods of pest control such as the use of entomopathogenic (insect pathogenic) fungi as biological control agents.

This project is focussed on determining the potential for using entomopathogenic fungi as components of an integrated approach to managing the rosy apple aphid in the UK. Although several species of Hyphomycete fungi have been developed as mycoinsecticides for controlling aphids both in glasshouses and field crops (Copping, 1998), there are no reports of their efficacy against the rosy apple aphid. As part of an integrated approach to aphid management, we report on initial studies to evaluate the efficacy of some currently available mycoinsecticides against the rosy apple aphid on the autumn host, *P. lanceolata*.

Materials and methods

Insect rearing

Apterae of rosy apple aphid have been continually reared since summer 2001 on *P. laneolata* under controlled environmental conditions (16:8 light:dark photoperiod; temperatures 20°C in the light period and 18°C in the dark period). Aphids of known age were reared under the same environmental conditions for the bioassay. Approximately 20 adult apterae were transferred to each of 6 small *P. lanceolata* plants and allowed to produce nymphs. The adult aphids were removed after 2-3 days. The nymphs developed to apterae within a further 7-9 days.

Bioassay method

Excised leaves of *P. lanceolata* were surface sterilised in 10% hypochlorite and rinsed in sterile distilled water. The surface sterilised leaves were embedded in 2% (w/v) sterile water agar in 9cm Petri dishes. Aphids of known age were transferred to the embedded leaves and allowed to settle for 24 hours.

The following day, aphids were sprayed *in situ* using a Burkhard computer controlled precision sprayer (Burkhard Manufacturing Co. Ltd., UK). The commercial formulations of the mycoinsecticide products Bontanigard ES™ (based on *Beauveria bassiana*), Vertalec® and Mycotal® (both based on *Verticillium lecanii*) were applied at the maximum commercial rate recommended for aphid control for each product respectively. Following spraying, aphid mortality was monitored daily. Dead aphids were removed and incubated on 1% water agar (w/v) to confirm fungal sporulation. Live aphids were transferred every 48 hours post-inoculation to fresh, sterilised *P. lanceolata* leaves embedded in 2% (w/v) sterile water agar. Data were analysed using actuarial life tables analysis (Lee, 1992) and the LT_{50} value with associated standard error was calculated for each mycoinsecticide tested.

Results and discussion

Mortality of aphids in all mycoinsecticide treatments was greater than 87% (Table 1) whilst mortality of control aphids was very small (less than 3% after 7 days). Although it has been suggested that excised leaves provide poor quality food for aphids in such assays (Chandler, 1997), the small control mortality in this bioassay indicates that aphids are able to survive well on excised leaves using this particular combination of aphid and host plant. This bioassay method is therefore very simple and provides a quick and efficient method for evaluating the efficacy of a number of fungal pathogens to the rosy apple aphid.

Table 1. LT_{50} values (days) for three commercial mycoinsecticides assayed against apterous rosy apple aphids.

Mycoinsecticide treatment	LT_{50} (days)	± se LT_{50} (days)	% infection in aphids*
Botanigard ES™	5.08	0.14	87.50 (80)
Mycotal®	3.31	0.15	100.00 (82)
Vertalec®	4.24	0.19	91.46 (82)

*Values in parentheses = n, excluding missing individuals

The LT_{50} values which were obtained, indicate that 50% mortality of aphids was achieved in less than 6 days for the three mycoinsecticides tested. However, these promising early results were obtained under ideal abiotic conditions (high relative humidity and warm, constant temperatures); further investigation of the efficacy of fungi under more stringent conditions is needed to determine the efficacy of these mycoinsecticides under less favourable abiotic conditions, both in the laboratory and semi-field experiments.

Aphids started to succumb to infection with fungus after 3 days in the Botanigard™ and Vertalec® treatments (Figure 1). However, it is interesting to note that some aphids died after only 24 hours in the Mycotal® treatment and the cadavers subsequently sporulated with fungus. The speed of kill noted in this treatment may be related to the production of toxins by the fungus or because of unknown components such as wetters or spreaders in the formulated product. Additionally, very large quantities of conidia were deposited on leaves (data not shown) and hence, it is likely that at these rates aphids were exposed to very large numbers of conidia. Death in these cases may be due to shock and mechanical damage associated with numerous penetrations of conidia or suffocation because of occlusion of the respiratory apparatus. Further investigation of this result is required.

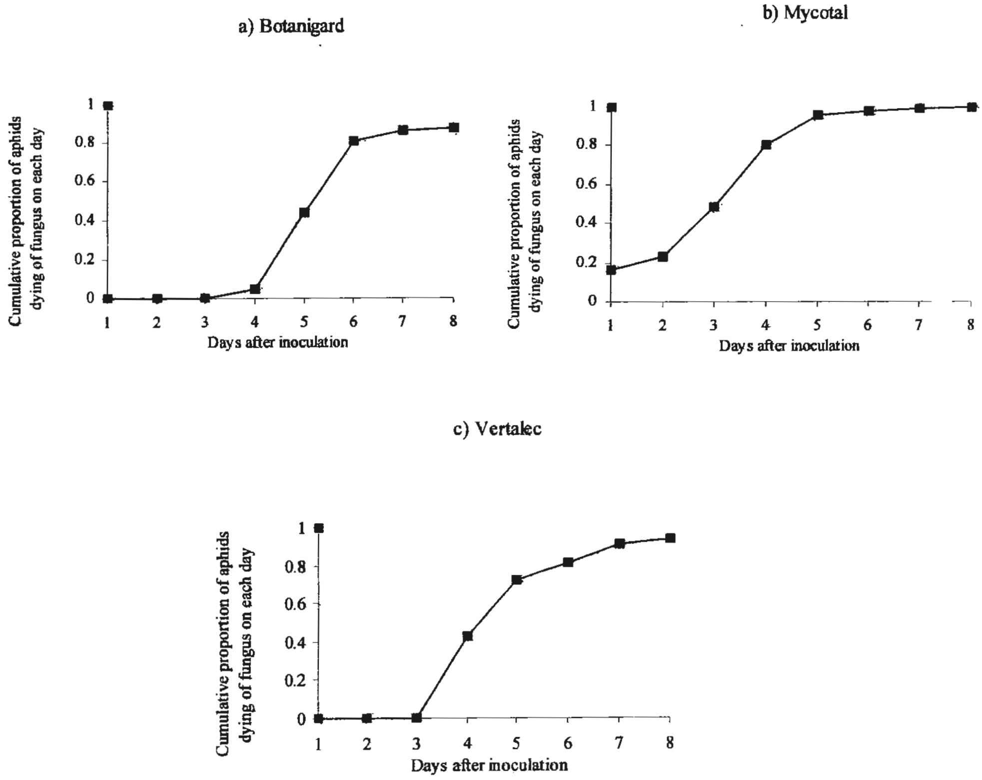

Figure 1: Cumulative proportion of *D. plantaginea* dying due to fungus on each day following spraying at maximum recommended rates with the mycoinsecticides a) Botanigard ES™, b) Mycotal® and c) Vertalec®

This was single run of the bioassay method and further experiments are required to evaluate the repeatability of the method. However, this initial experiment indicates that this will be a very useful method of evaluating the efficacy of mycoinsecticides again rosy apple aphids. Further assays will be done using aphids reared on apples to determine whether the different host plants or aphid morphs affect the efficacy of the mycoinsecticides tested.

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