One predator - two prey systems: Individual behaviour and population dynamics
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In this thesis, I study relatively simple food webs consisting of only two or three herbivores, a plant and a predator. Such systems are characterized by complex interactions, on the one hand, through the occurrence of apparent competition, apparent mutualism and intraguild predation, and on the other hand through the behaviour of predators and prey. Additionally, the temporal and spatial scales on which these interactions occur will intensify or weaken the effect of these interactions on population densities.

The aim of this thesis is to study interactions determining prey densities in a simple food web and how prey and predator behaviour influence the strength of these interactions.

Roos van Maanen
One predator - two prey systems:
Individual behaviour and population dynamics

PhD thesis, University of Amsterdam, The Netherlands

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Predator-prey interactions play an important role in shaping dynamics of populations of organisms. Whereas early theory has focused on interactions between populations of one species of prey and one species of predator (Volterra 1926, Lotka 1932, May 1999), there is increasing emphasis on multispecies food webs involving more complex interactions (Rosenheim et al. 1995, Janssen et al. 1998, Sih et al. 1998, Brodeur and Boivin 2006). In this thesis, I study the interactions between two or more prey species that share the same predator. Possible interactions in such systems are predation, competition for resources between the prey species and indirect interactions among the prey that are mediated by the predator. Predator-mediated interactions include apparent competition and apparent mutualism (Holt 1977, Morris et al. 2004). Theory predicts that adding a population of a new prey species to a system consisting of one predator and one prey results in a lower equilibrium density of the resident prey, even when the two prey species do not compete for resources (apparent competition; Holt 1977, 1984). This is because the equilibrium density of the shared predator increases with the increased equilibrium density of the added prey species. This interaction can even lead to exclusion of the resident prey species (Holt 1977, Bonsall and Hassell 1997). In the short term, before reaching an equilibrium, two prey species that share a predator may also affect each others’ densities positively because an increase in the numbers of one species may lead to predator satiation, resulting in decreased predation on the other species (apparent mutualism: Holt and Lawton 1994, Abrams and Matsuda 1996, Holt 1997). This predator satiation, however, occurs on a short time scale, because the higher availability of prey will result in a numerical response of the predator, which in turn leads to more predators that have to share the food (Holt and Kotler 1987). Consequently, there will be less prey per predator and predators will cease to become satiated. Like other predator-prey systems, two prey and a shared predator can also show cyclic dynamics. Models predict that such population cycles weaken apparent competition, or may even result in apparent mutualism. Such long-term apparent mutualism occurs when population densities of one prey show cycles, resulting in repeated satiation of the shared predators and repeated reduced predation on the other prey (Abrams et al. 1998, Brassil 2006). In short, theory predicts positive as well as negative effects of the indirect interaction between prey through a shared predator.
The theory discussed so far assumes that predators encounter and attack both prey simultaneously. In reality, this will often not be the case; predators may be capable of searching for only one of the prey species at a time. For example, when the two prey species occupy different patches or habitats, an individual predator obviously has to choose for which prey it will forage, and while hunting for one species, it cannot hunt for the other. Because predators tend to leave habitats in which their foraging yields are relatively low (Charnov 1976a), this provides the remaining prey individuals in this habitat with a refuge, thereby promoting coexistence of prey species sharing a predator (Holt 1984).

When predators have to switch from feeding on one prey to feeding on another, they are expected to devote more attention to the more abundant or more profitable prey until its density declines, after which they will switch to feeding on the other prey species (Murdoch 1969). In this case, the proportion eaten of a particular prey changes from less than expected to more than expected as the relative abundance of that prey increases (Murdoch 1969). Such switching may be the result of predators developing a ‘specific search image’ for abundant and profitable foods (Tinbergen 1960). Given that all species reach stable equilibrium densities, theory predicts the occurrence of positive indirect effects of one prey population on the equilibrium densities of others when predators exhibit switching (Abrams and Matsuda 1996, 2003).

Whereas with switching, the predator forages for either one prey or the other, but never for both, optimal foraging theory predicts that predators should forage exclusively for one prey species or for both (Charnov 1976b). This theory assumes that the predator encounters both prey in the same patch, and that predators forage in such a way as to maximize their net energy or food intake per unit time. Specifically, it predicts that generalist predators should switch from eating only prey of high quality to eating a mixture of prey when the density of the high-quality prey falls below a threshold (Charnov 1976b, Stephens and Krebs 1986). In systems with two prey species sharing a predator, such adaptive foraging can result in apparent competition or apparent mutualism (Holt 1983).

However, it is clear that the diet choice of a predator is not always density dependent, it can also be driven by the need of various nutrients. Herbivores are known to select an optimal diet through consuming various plant species (Waldbauer and Friedman 1991, Bernays and Bright 1993, Bernays et al. 1994, Singer and Stireman 2001). For example moose need to eat low-energy aquatic plants in order to obtain sufficient sodium (Belovsky 1978). Diet mixing in predators received relatively little attention (Greenstone 1979, Toft and Wise 1999), but predators are also known to prefer mixed diets for nutritional reasons (Tinbergen 1981), for example to obtain amino acids and fatty acids (Greenstone 1979, Uetz et al. 1992). Positive effects of mixing high- and low-quality prey, such as increased survival and fecundity, have

Such effects have, to my knowledge, not been incorporated into the theory of apparent competition. If mixed diets have a positive effect on predator survival or reproduction, it is easy to see from simple models of apparent competition that the equilibrium densities of the prey sharing a predator will decrease. Equilibrium prey densities are inversely related to the growth rate of the shared predator (Holt 1977), hence, if the predators’ growth rate increases because of a mixed diet, prey equilibria go down.

Besides the possible advantage of mixing diets in order to optimize nutrient uptake, there is another advantage of mixed diets to predators. To my knowledge, this mechanism has received very little attention so far and I will therefore explain it in some detail here. Predation rates of generalist predators are largely influenced by antipredator behaviour of prey (Sih and Christensen 2001), such as fleeing, hiding (Lima and Dill 1990) and counterattacking (Saito 1986, Janssen et al. 2002). To avoid wasting energy or time on antipredator behaviour, which would otherwise have been used for other essential activities, prey need to assess the risk posed by a predator and tune their behaviour accordingly. To this end, chemical cues associated with predators have shown to be helpful indicators (Chivers and Smith 1998). Many prey species can distinguish chemical cues of predators that fed on conspecific prey from those of predators that fed on heterospecific prey, and react stronger to the first (Wilson and Lefcort 1993, Chivers et al. 1996, Laurila et al. 1997, Venzon et al. 2000, Meng et al. 2006). If diet-related chemical cues enable prey to discriminate between harmless and dangerous predators, predators might be able to ‘chemically disguise’ themselves by eating two given prey species alternatingly. Therefore, I suggest that predators can increase their predation rate on a superior prey species by selecting a mixed diet. Although this mechanism has been suggested a few times (Venzon et al. 2000, Lima et al. 2003, Meng et al. 2006), it has never been investigated.

As explained above, diet mixing does not only occur in generalist predators, but also in herbivores (Belovsky 1978). Herbivores that mix plant food with animal food are referred to as omnivores, i.e. they consume resources from different trophic levels (Pimm and Lawton 1978). A special case of omnivory is called intraguild predation (IGP, Polis and Holt 1989). It occurs when one species is an omnivore, a second species is its prey and they share a third species as their food (Polis et al. 1989, Coll and Guershon 2002, Janssen et al. 2007, Lucas et al. 2009). Theory has demonstrated that intraguild predators and intraguild prey can only persist when the intraguild prey is the superior competitor for the shared food (Polis et al. 1989). This means that the most efficient competitor for the shared prey can persist with this prey at low productivity levels (Mylius et al. 2001, Kuijper et al. 2003), whereas at high productivity
levels the IG-predator will exclude the IG-prey and this will result in an increase of the equilibrium density of the prey (Holt and Polis 1997). The effect of IGP on the population densities of the prey will, like in the case of apparent competition, depend on prey preference of the IG-predator (Janssen et al. 1998) and antipredator behaviour of the IG-prey (Magalhães et al. 2005).

Concluding, relatively simple food webs consisting of only two or three herbivores, a plant and a predator are characterized by complex interactions, on the one hand, through the occurrence of apparent competition, apparent mutualism and intraguild predation, and on the other hand through the behaviour of predators and prey. Additionally, the temporal and spatial scales on which these interactions occur will intensify or weaken the effect of these interactions on population densities.

The aim of this thesis is to study interactions determining prey densities in a simple food web and how prey and predator behaviour influence the strength of these interactions. I concentrate on predator-mediated interactions and diet choice, and do not report on the interactions between the two herbivores mediated by the plant’s induced defences.

Biological control systems, especially in greenhouses, offer an ideal opportunity to assemble communities, in which relatively simple food webs can be studied experimentally. In this thesis, I study such a simple food web to contribute to the knowledge of the interplay between indirect predator-mediated interactions and behaviour and improve biological control. I use a system consisting of a biological control agent (the generalist predatory mite *Amblyseius swirskii*), three pest species (greenhouse whitefly, two-spotted spider mite and the Western flower thrips) and cucumber plants (see box ‘Experimental system’ for details and photos A-E). This offers the opportunity to experimentally test the occurrence of apparent competition and apparent mutualism, as well as to investigate behaviour of predators and prey that may affect the population dynamics of the species involved.

In contrast with the time scale studied in most theoretical models on predator – prey interactions, which often investigate the effects of interactions on equilibrium densities, the period of cropping in most agricultural systems is so short that equilibria may not be reached (Briggs and Borer 2005). Predictions of interactions from studies that consider transient dynamics can be quite different from those that only consider equilibrium dynamics (Janssen et al. 1998, Briggs and Borer 2005). For example, using one predator species to control two pests at the same time seems to favour biological control at first sight because it results in lower pest densities (apparent competition), but theory on short-term dynamics of systems with multiple prey that share a predator predicts that two prey species may also affect each other’s densities positively (apparent mutualism: Holt and Kotler 1987, Holt and Lawton 1994, Abrams and Matsuda 1996, Abrams et al. 1998), which is not in favour of bio-
logical control. Thus, the experiments reported in this thesis also serve to show whether shared predation results in better biological control.

Thesis outline
In Chapter 2, I address the question whether the dynamics of two prey species sharing a predator is characterized by apparent competition or apparent mutualism. As outlined above, most theory predicts that apparent mutualism occurs at a short time scale, and long-term dynamics are characterized by apparent competition. It is not clear however, when this transition from short-term to long-term dynamics occurs. The dynamics of Western flower thrips and greenhouse whiteflies were studied, separately as well as together. Experiments were carried out in the presence of a population of predatory mites on cucumber plants in small greenhouse compartments. In this system, the time scale was set by the length of the growing season of the greenhouse crop, which is about 10 weeks. In this period, there will be several generations of pests and predators, but the system will not reach equilibrium, hence, it is unclear whether the addition of a second pest will increase or decrease control of the target species.

In Chapter 3, the question is whether pest species can escape from control by a predator, and how this is affected by the presence of the second prey species. Again, I study the effects of the two pest species, but now assessments were done more frequently and earlier in the experiment, to investigate whether short-term positive indirect effects of the two prey on each other occur. In contrast to the experiment described in Chapter 2, pests and predators were released concentrated on one plant. This was done to compare spatial dynamics of all three species and to assess whether a temporal escape of one of the two prey occurs. Because the two pests can disperse rapidly relative to the non-flying predator, prey can possibly escape from predation in space and time.

In Chapter 4, the question is whether prey species diversity reduces populations of non-prey species because of the increased numerical response of generalist predators on mixed prey populations. Because predators feeding on a mixture of food sources often show increased survival and fecundity (Uetz et al. 1992, Toft 1995, Evans et al. 1999, Toft and Wise 1999, Harwood et al. 2009), they can reach higher densities. These high predator densities may significantly affect prey populations on which the predators have small per capita effects. I perform a similar experiment as in Chapter 2 and 3, but now I add a third pest species, the two-spotted spider mite *Tetranychus urticae* Koch to investigate whether there might be effects of apparent competition on prey that are only marginally suitable for the predator. This spider mite species is an important greenhouse pest and often co-occurs with thrips and whiteflies. Spider mites are not good prey for the predator, and the predator cannot con-
Experimental system

The experimental system consisted of the generalist predatory mite *Amblyseius swirskii* Athias-Henriot (Acari: Phytoseiidae), two of its herbivorous prey species; the Western flower thrips *Frankliniella occidentalis* (Per- gande) and the greenhouse whitefly *Trialeurodes vaporariorum* (Westwood) (Nomikou et al. 2001, see also Chapter 2), one non-prey herbivore (*Tetranychus urticae* Koch) and cucumber plants, *Cucumis sativa* L. (cv. Aviance RZ). Western flower thrips and greenhouse whitefly are major pest species of various crops, such as cucumber, sweet pepper and eggplant in Northern Europe and North America (Byrne 1990, Lewis 1997).

The Western flower thrips is native to North America but has spread to other continents, including Europe, Australia, and South America. Thrips lay their eggs in the tissues of the plant and feed on the flowers and foliage by piercing a hole in the leaf with their mandibles and subsequently ingesting cell contents via their maxillary stylets (Chisholm and Lewis 1984). When thrips feed on developing tissues, affected cells are unable to expand, resulting in distortion of mature leaves, fruits and petals. When thrips feed on expanded tissue, affected cells become filled with air, which imparts a silvery appearance. Thrips feed mainly on leaf parenchyma and plant pollen, but also on eggs of its natural enemies, the predatory mites *Iphiseius degenerans* (Berl.) and *Amblyseius cucumeris* (Oudemans), the eggs of another predatory mite *Phytoseiulus persimilis* Athias-Henriot (Faraji et al. 2002, Janssen et al. 2002), which is not an important enemy of thrips, and on the eggs of another herbivore competing for the same host plant, the two-spotted spider mite *T. urticae* (Trichilo and Leigh 1986, Pallini 1998, Agrawal and Klein 2000).
The greenhouse whitefly is common throughout the tropics and subtropics and in greenhouses in temperate zones. Whiteflies lay their eggs on the undersides of leaves. Eggs are pale yellow in colour, but turn dark prior to hatching. Newly hatched larvae, known as crawlers, are the only mobile immature life-stage. During the first and second larval instars, they are translucent and flat. During the fourth and final immature life-stage, referred to as pupa, compound eyes and other body tissues become visible as the larvae grow and rise from the leaf-surface (Byrne and Bellows 1991). The greenhouse whitefly mainly feeds on crops such as cucumber, eggplant, pepper, tomato, sweet pepper and zucchini. The whitefly crawlers and adults cause direct crop damage by inserting their stylet into leaf veins and extracting phloem sap and indirect damage by decreased photosynthesis as a result of sooty mould (Vet et al 1980, Lei et al. 1996).

The predatory mite *A. swirskii* occurs in the wild in Mediterranean countries, and is well adapted to warm and humid climate conditions. The development from egg to the adult phase takes only five to six days at 26 °C and includes the following stages of development: egg, larva, protonymph, deutonymph, and adult. If sufficient food is available, *A. swirskii* lays on average two eggs per female per day. This predatory mite has proven to be an effective control agent for thrips (Messelink et al. 2006), whiteflies (Nomikou et al. 2001, 2002, 2010) and broad mites *Polyphagotarsonemus latus* (Banks) (Acari: Tarsonemidae) (van Maanen et al. 2010). It is also shown that *A. swirskii* attacks chilli thrips (*Scirtothrips dorsalis* Hood) (Arthurs et al. 2009) and possibly also tomato russet mite (*Aculops lycopersici*) (Park et al. 2010). In Chapter 2 and 4, I additionally use the predatory mite *Euseius ovalis* (Evans), an effective control agent for thrips (Messelink et al. 2006) and whiteflies (pers. obs.), and the two-spotted spider mite *T. urticae* an important greenhouse pest often co-occurring with thrips and whiteflies.

control this prey. Here, I investigate whether the build-up of large populations of predators on thrips and whiteflies has an effect on spider mite dynamics. In Chapter 5, the question is addressed whether not only predators, but also herbivores profit from consuming mixed food sources. Thrips are known as omnivores and intraguild predators; they feed on spider mites, with which they also share the host plant (see box "Experimental system"). I investigated whether thrips feed on whitefly stages, such as eggs and nymphs and whether a mixed diet of whiteflies and plant tissue has a positive effect on life history parameters of thrips. This is important, because prey species diversity may not only result in higher densities of generalist predators, but could also favour herbivores that are intraguild predators of other herbivores.

As outlined above, predation rates are not only affected by predator behaviour, but also by antipredator behaviour of the prey (Sih and Christensen 2001). It has been suggested that predators can ‘manage’ antipredator behaviour by feeding on a mixture of prey (Lima 2003). This is because many prey species show stronger antipredator behaviour in response to chemical cues from predators that fed on conspecific prey than to cues of predators fed on prey of another species. Lima et al. (2003) therefore assumed that antipredator behaviour in prey increases with the number of conspecific prey that are attacked by a predator. In Chapter 6, I test whether predators that were marked with cues of whitefly indeed have higher predation rates on thrips than predators marked with cues of thrips.

In the final chapter, I summarize the results and discuss differences between these results and theory on apparent competition, apparent mutualism and diet choice, and formulate the consequences of my results for biological control.

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Biological control of thrips and whiteflies by a shared predator: Two pests are better than one

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We studied the capacity of one species of predator to control two major pests of greenhouse crops, Western flower thrips (Frankliniella occidentalis (Pergande)) and the greenhouse whitefly (Trialeurodes vaporariorum (Westwood)). In such a one-predator-two-prey system, indirect interactions can occur between the two pest species, such as apparent competition and apparent mutualism. Whereas apparent competition is desired because it brings pest levels down, apparent mutualism is not, because it does the opposite. Because apparent competition and apparent mutualism occur at different time scales, it is important to investigate the effects of a shared natural enemy on biological control on a time scale relevant for crop growth. We evaluated the control efficacy of the predatory mites Amblyseius swirskii (Athias-Henriot) and Euseius ovalis (Evans) in cucumber crops in greenhouse compartments with only thrips, only whiteflies or both herbivorous insects together. Each of the two predators controlled thrips, but A. swirskii reduced thrips densities the most. There was no effect of the presence of whiteflies on thrips densities. Whitefly control by each of the two predators in absence of thrips was not sufficient, yet better with E. ovalis. However, whitefly densities in presence of thrips were reduced dramatically, especially by A. swirskii. The densities of predators were up to 15 times higher in presence of both pests than in the single-pest treatments. Laboratory experiments with A. swirskii suggest that this is due to a higher juvenile survival and developmental rate on a mixed diet. Hence, better control may be achieved not only because of apparent competition, but also through a positive effect of mixed diets on predator population growth. This latter phenomenon deserves more attention in experimental and theoretical work on biological control and apparent competition.

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The use of different natural enemies for the biological control of different pest species results in the creation of complex artificial food webs in agricultural crops. This implies that pest densities are not only determined by the natural enemies of that pest, but also by direct and indirect interactions with other pests and enemies, and such interactions can affect biological control (Rosenheim et al. 1995, Janssen et al. 1998). The use of one natural enemy to control several pests will result in food webs simpler than those in which different enemies are introduced against...
each pest species. However, such a natural enemy may mediate indirect interactions between these pest species, which in turn may be beneficial or detrimental to biological control. One such interaction between two pest species through a shared natural enemy is apparent competition, which Holt (1977, 1984) defined as the indirect interaction between prey through shared predation. When two prey species share a natural enemy, the equilibrium density of one of the prey species decreases with increasing equilibrium density of the other species. This is because the density of the shared natural enemy increases with the increased equilibrium density of either prey species (Holt 1977, Müller and Godfray 1997, Janssen et al. 1998, van Rijn et al. 2002, Morris et al. 2004). This interaction can even lead to exclusion of one of the two prey species (Bonsall and Hassell 1997).

The addition of alternative food to better suppress a pest species through apparent competition has been used in biological control, often with the desired result (Karban et al. 1994, Hanna et al. 1997, van Rijn et al. 2002, Liu et al. 2006). However, the alternative food often consists of non-prey, such as pollen (van Rijn et al. 2002), or the alternative prey is not a pest, but serves primarily as alternative food to build up predator populations (Karban et al. 1994, Hanna et al. 1997, Liu et al. 2006). Here, we investigate the effects of apparent competition between two prey species that are both serious pests of various crops worldwide.

Two prey species that share a predator may also affect each other’s densities positively because an increase in the numbers of one pest species may lead to predator satiation, resulting in decreased predation on the other pest species. In addition, predators may switch to the most abundant prey species, thus releasing the other species from predation. Such positive indirect effects are referred to as apparent mutualism (Holt and Lawton 1994, Abrams and Matsuda 1996). Apparent mutualism occurs mostly at a shorter time scale than apparent competition (Harmon and Andow 2004, van Veen et al. 2006). Hence, depending on time-scale and prey preference, a natural enemy that feeds on two pest species can mediate mutualistic or antagonistic interactions between the two pests. The use of one species of natural enemy for biological control of two pests may thus result in reduced control in the short term, but increased control in the long term (van Rijn et al. 2002). It is therefore relevant to assess the time scale at which indirect interactions occur. In our system, this scale is set by the length of the growing season of the greenhouse crop.

In the literature on apparent competition, little attention has been given to the effect of mixed diets on the performance of predators. Mixed diets are known to have positive effects on reproduction in some predator species (Wallin et al. 1992, Toft 1995, Evans et al. 1999), and the effect of adding a new prey species would then surpass that of simply adding more prey items with the same nutritive value. Therefore, we also investigated the effects of a mixed diet on predator survival and reproduction.
The experimental system

Western flower thrips (*Frankliniella occidentalis* (Pergande)) and greenhouse whitefly (*Trialeurodes vaporariorum* (Westwood)) are two major pest species in various crops in Northern Europe and North America (Byrne et al. 1990, Lewis 1997). The phytoseiid *Amblyseius swirskii* (Athias-Henriot) (Chant and McMurtry 2004) has recently been shown capable of suppressing populations of the tobacco whitefly (*Bemisia tabaci* (Gennadius)) (Nomikou et al. 2001, Nomikou et al. 2002). *Euseius ovalis* (Evans), another species of predatory mite, also feeds and reproduces on a diet of *B. tabaci* (Borah and Rai 1989). An evaluation of phytoseiids for control of Western flower thrips in greenhouse cucumber showed that *A. swirskii* and *E. ovalis*, amongst others, are much more effective thrips predators than *Neoseiulus cucumeris* (Oudemans), a phytoseiid that is often used to control thrips, (Messelink et al. 2005, 2006). Thus, each of the two mites *A. swirskii* and *E. ovalis* can potentially control whiteflies and thrips.

We studied the dynamics of Western flower thrips and greenhouse whiteflies separately as well as together on cucumber plants (cv. Aviance RZ) with one of the two predatory mite species in small greenhouse compartments. Cucumber plants have a short cropping season, and short-term effects of shared predation, such as apparent mutualism, may determine the dynamics of the pests and predators. For biological control, it is therefore important to assess whether the effects of shared predation on pest levels are positive or negative. In order to detect an effect of mixed diet on the population dynamics of the predator, we also compared the effect of diets consisting of pest species separately or of a mix of both species on several life-history parameters of *A. swirskii* that are important for population dynamics (oviposition, juvenile survival, development).

Materials and Methods

Cultures

For the greenhouse experiments, the predatory mites were reared on flowering *Ricinus communis* L. plants in small greenhouses. The predators fed on the pollen, amply produced by these plants. Western flower thrips were reared on flowering chrysanthemum plants cv. Miramar. The greenhouse whitefly was reared on tobacco plants (*Nicotiana tabacum* L.). Cucumber plants cv. Aviance RZ were grown in rock wool blocks in a greenhouse compartment without any application of pesticides. The greenhouse experiments were carried out at Wageningen UR Greenhouse Horticulture, Naaldwijk, The Netherlands.

For the laboratory experiments, *A. swirskii* mites were reared on plastic arenas (8 x 15 cm), placed on a wet sponge in a plastic tray containing water (Nomikou et al. 2003a). They were fed cattail pollen, *Typha latifolia* L. twice per week. Western flower thrips were reared in climate boxes and greenhouse whiteflies in a walk-in climate
Greenhouse experiments
The effects of the predators A. swirskii and E. ovalis on Western flower thrips, greenhouse whitefly and a combination of these two pests was examined in a greenhouse experiment in twelve separate compartments (each 18 m²) that each contained two tables (1 x 3 m) on which cucumber plants were grown. The experiment was set-up as a split-plot experiment with four replicates. Each block contained one compartment with thrips, one compartment with whiteflies and one compartment with thrips and whiteflies. In each compartment, A. swirskii was released on plants on one table and E. ovalis on the other table. Note that we did not include control treatments in which the pest species had no predators. Hence, we have no data on the effects of the pest species on each other through the shared host plant. The plants in the treatments with both pest species had such low damage levels that exploitative competition between the two pests was improbable, certainly when we consider the large leaf size of a cucumber crop (in this treatment 250-450 cm²). However, the two pests possibly affected each other through induced plant responses (Karban and Carey 1984). This will be the subject of forthcoming research. Secondly, it should be realized that the predator treatments are strictly not independent, because thrips and whiteflies were able to migrate between the two tables in a compartment. This migration might result in an underestimate of the control capacity of the best performing predator, and an overestimate of the capacity of the other predator. However, for analyzing results we assumed the predator treatments to be statistically independent.

Each greenhouse compartment had a small closed entrance corridor without windows and was equipped with an air pressure system in order to minimize contamination with organisms from outside. Plants were at the fifth-leaf stage when the experiments started, and roots were preventively treated with Propamocarb against Pythium spp. Four plants were placed on two pieces of a rock wool substrate slab on each table. The experiment started in week number 12. The rock wool slabs were continuously immersed in a nutrient solution that was automatically supplied once per day. Plants were cultivated vertically up to a 1.5 m high crop supporting wire. Side-shoots were removed until the top of the plant reached the crop supporting wire. Later on, plant shoots and side shoots grew down over the crop supporting wire. Contamination of treatments was avoided by applying insect glue to the wires supporting the crop and by keeping the plants isolated in the water layer on the tables.

Pests were introduced one day after the cucumber plants were planted. Forty adult female thrips, collected from the culture using an aspirator, were released on each
A total of 120 adult whiteflies were released per table. The population of whiteflies contained on average 42% females. The same numbers were released in the treatment with both pests. Predatory mites were released seven days after introducing the pests. Female predatory mites were sampled with a fine paintbrush in the laboratory and placed on leaf discs of sweet pepper (Capsicum annuum L.) (diameter 2 cm) containing cattail pollen. One leaf disc with 15 mites was introduced on the upper leaves of each cucumber plant.

The experiment lasted 11 weeks. During this period, the crop was inspected twice a week, and cucumbers were harvested as soon as they reached the standard fruit size. Different treatments were handled by different persons to avoid cross-contamination. The treatments with different pests were indeed not invaded by other pests, except for one compartment with whiteflies, which was invaded by spider mites in week 8. This spot was treated by releasing 100 adults of the predatory mite Phytoseiulus persimilis Athias-Henriot, resulting in successful control. Phytoseiulus persimilis does not consume thrips or whiteflies, and they were not observed after the spider mites had been eradicated, within a few days. Powdery mildew occurred occasionally during the experiment, but infections remained limited to small spots because the cucumber variety used is partially resistant to mildew.

The numbers of predatory mites and pests were assessed four, six, eight and ten weeks after introducing the predatory mites. Plants were not sampled during the first four weeks in order not to cause disturbance. Assessing the populations was done by collecting six leaves from each treatment by randomly choosing three shoots of which the sixth and eighth leaf from the tip was collected. These leaves were each put in a separate plastic bag and transported to the laboratory where they were cut into strips of 5 cm. The number of mites, thrips and whiteflies were counted on both sides of the leaves using a binocular microscope (40×). All mites were slide-mounted for identification to species, gender and life-stage under a microscope (400×). Only the larval stages of thrips were counted, and eggs, larvae and pupae of whiteflies were counted separately. When densities of whiteflies exceeded 500 individuals per leaf, densities were assessed on part of each leaf only, and extrapolated to the whole leaf. The leaf area of each collected leaf was measured with an optical area meter (LI-COR LI-3100) after mite and pest densities were quantified. The average temperature and relative humidity were comparable for each block treatment (Table 2.1).

<table>
<thead>
<tr>
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<th>1</th>
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<td>Mean temperature (°C)</td>
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<td>22.8</td>
<td>22.5</td>
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<tr>
<td>Mean relative humidity (%)</td>
<td>74</td>
<td>75.8</td>
<td>76.7</td>
<td>80.5</td>
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The results were analysed with linear mixed effects models (lme in R), with time as random factor nested in blocks to correct for pseudoreplication due to repeated measures (Crawley 2002). The numbers of thrips and whiteflies were log(x+1) transformed before the analysis, in order to stabilise variance. Treatments were compared through model simplification by combining treatments (Crawley 2002).

Oviposition, juvenile survival and development, and predation

The oviposition rate and predation rate of A. swirskii was measured during three days on three different diets: whitefly eggs, young first instars thrips and a mixture of whitefly eggs and young first instar thrips. These stages are most vulnerable to these predators (Nomikou et al. 2004; R van Maanen, pers. obs.). Adult female predators (8-11 days old since egg stage) were tested individually on a leaf disc (diam. 24 mm) with one of the three different diets. The numbers of whitefly eggs ranged from 21 to 59 eggs/leaf disc and the numbers of young first instars thrips were 8 or 15/leaf disc. Previous predation tests showed that these densities are high enough to ensure maximum prey consumption (Nomikou et al. 2002). Cohorts of whitefly eggs were prepared in advance (Nomikou et al. 2003b) and first instar thrips larvae were reared on cucumber leaves with cattail pollen placed on wet cotton wool in Petri dishes. The numbers of both thrips and whiteflies in the mixed diet were equal to the single prey diets, hence, supplied additively. Predation was recorded as the number of whitefly eggs consumed (as judged by the transparent cuticle) and first-instar thrips consumed (as judged by the remains) after 24 and 48 h. Because oviposition rates are affected by the previous food source of the adult predatory mites (Sabelis 1990), we discarded data from the second day of the experiment only (Sabelis 1986). Oviposition was compared between days and diets using a generalized linear mixed effects model with Poisson errors and a random factor within replicates to correct for pseudoreplication, using R (lmer, R Development Core Team 2005). Predation was analysed for thrips larvae and whitefly eggs separately with generalized linear models with Poisson error distributions and diet as factor.

For the effects of diet on juvenile survival and development, we placed between 80 and 100 female mites from the culture on a plastic arena with cattail pollen. After less than 24 h, we transferred their eggs to clean cucumber leaf discs, each egg on a separate disc. We transferred the larva, directly after emerging, to a leaf disc with whitefly eggs, young first-instar thrips or a mixture of whitefly eggs and young first-instar thrips. Every other day, mites were transferred to a new leaf disc with whitefly eggs, young first-instar larvae or the two prey together. Survival and stage of the predator were recorded daily until the mites reached adulthood. Kaplan-Meier survival curves on different diets were fitted and compared using the log-rank test (Hosmer and Lemeshow 1999, library survival, R Development Core Team 2005). Juvenile development was
analysed using a time-to-event analysis with log-rank tests. Three replicates were performed with 45, 45 and 37 individuals, the first two with 15 individuals per diet, the last with eight individuals on a mixed diet, 15 on a thrips diet and 14 on a diet of whiteflies.

Results

Greenhouse experiments

Thrips

There was a clear effect of treatment on thrips densities (Figure 2.1, lme: $F_{3,377} = 124.8$, $P<0.0001$). *Amblyseius swirskii* reduced thrips to very low densities (Figure 2.1A). The highest thrips densities were observed at the first assessment after four weeks, after which they went down to less than one larva per leaf in the following weeks (Figure 2.1A). *Euseius ovalis* was less successful in controlling thrips; the ultimate density was between 80 and 20 larvae per leaf (Figure 2.1B, difference between *A. swirskii* and *E. ovalis*: model simplification after linear mixed effects model (lme), thrips only: Log likelihood ratio (LR) = 167.1, df = 8,7, $P<0.0001$, thrips plus whitefly: LR = 140.1, df = 8,7, $P<0.0001$). This result is qualitatively similar to earlier experiments in which ultimate thrips densities were four times higher in the presence of *E. ovalis* than in the presence of *A. swirskii* (Messelink et al. 2006). In the treatments with the two pest species present, densities of thrips were not affected by the presence of whiteflies, irrespective of whether *A. swirskii* or *E. ovalis* was the shared predator (Figure 2.1A,B, *A. swirskii*: LR = 0.019, df = 8,7, $P = 0.89$, *E. ovalis*: LR = 2.17, df = 6,7, $P = 0.14$).

Whiteflies

There was a significant effect of treatment on densities of whiteflies (Figure 2.2, $F_{3,377} = 69.2$, $P<0.0001$), but neither of the two predators could prevent an increase in the populations of whiteflies, resulting in thousands of immature whiteflies per leaf (Figure 2.2A,B). However, the predators delayed the population increase of whiteflies considerably compared to that expected under exponential growth and to that observed in earlier experiments without predatory mites (GJ Messelink, pers. obs.). *Euseius ovalis* was more successful in controlling whiteflies than *A. swirskii* when thrips were absent (Figure 2.2A,B, LR = 64.1, df = 8,7, $P<0.0001$).

In the treatments with both thrips and whiteflies, the two predators reduced densities of whiteflies clearly more than in the treatments without thrips (Figure 2.2A,B, *A. swirskii*: LR = 113.1, df = 8,7, $P<0.0001$; *E. ovalis*: LR = 24.2, df = 8,7, $P<0.0001$). *Amblyseius swirskii* reduced whitefly densities more than *E. ovalis* (Figure 2.2A,B, LR = 146.0, df = 8,7, $P<0.0001$). In the case of *A. swirskii*, whiteflies went practically extinct (Figure 2.2A), and with *E. ovalis*, densities of whiteflies went down from a peak of approximately 1000 immatures per leaf in the sixth week, to approximately 500
immatures per leaf in the tenth week (Figure 2.1B). This suggests an indirect interaction between thrips and whiteflies mediated by the shared predator (apparent competition) or via the host plant.

**Predators**

There was a significant effect of the pest species on the densities of predators (lme: \(A. \text{ swirskii} \): \(F_{2,282} = 151.0, P<0.0001; \ E. \text{ ovalis} \): \(F_{2,282} = 131.1, P<0.0001\). Compared to the treatments involving single pest species, the presence of thrips and whiteflies together resulted in higher densities of \(A. \text{ swirskii} \) (Figure 2.3A, thrips vs. mix: LR =

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**Figure 2.1** The dynamics of Western flower thrips during a 10-week greenhouse experiment in presence of the predatory mites \(A. \text{ swirskii} \) (A) and \(E. \text{ ovalis} \) (B). Shown are average densities (± SE) of thrips larvae in the presence (triangles) or absence (circles) of greenhouse whiteflies.
128.2, df = 7.6, P<0.0001; whitefly vs. mix: LR = 189.5, df = 7.6, P<0.0001), but the difference in densities between treatments with single pest species was also significant (Figure 2.3A, thrips vs whitefly: LR = 9.3, df = 7.6, P = 0.0023). The same was found for *E. ovalis*, (Figure 2.3B, thrips vs. mix: LR = 126.3, df = 7.6, P<0.0001; whitefly vs. mix: LR = 168.4, df = 7.6, P<0.0001, thrips vs whitefly: LR = 6.4, df = 7.6, P = 0.011). Together with the lower densities of whiteflies in the treatments with both pests, these findings are suggestive of apparent competition between thrips and whiteflies on whitefly densities.

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**Figure 2.2** The dynamics of greenhouse whitefly on cucumber plants during a 10-week greenhouse experiment in presence of the predatory mites *A. swirskii* (A) and *E. ovalis* (B). Shown are average densities (± SE) of immature whiteflies in the presence (triangles) or absence (squares) of thrips.
Oviposition, juvenile survival and development and predation

The average oviposition rates of *A. swirskii* on a diet of whitefly eggs, first instar thrips or on a mixed diet were not significantly different (Figure 2.4, mixed effects model). Predation, however, differed significantly with diet (Figure 2.5, thrips: $F_{1,69} = 23.5$, $P<0.0001$, whitefly: $F_{1,66} = 61.6$, $P<0.0001$). When offered the mixed diet, *A. swirskii* consumed roughly half the numbers of each prey, as compared to experiments with single-species diets. All individuals under test consumed the two prey species. Hence, the population of predators tested did not consist of a mixture of individuals that specialized on one prey.

![Figure 2.3](image_url)

*Figure 2.3* The dynamics of predatory mites on cucumber plants during a 10-week greenhouse experiment. Shown are average densities (± SE) of (A) *A. swirskii* and (B) *E. ovalis* with thrips (circles), whitefly (squares) or a combination of thrips and whiteflies (triangles).
Juvenile survival was significantly affected by diet ($\chi^2 = 10.7$, df = 2, $P = 0.0047$). There was no mortality on a diet consisting of thrips or on the mixed diet, whereas some 34% of the juvenile predators that were feeding on whitefly eggs died. The rate of juvenile development (egg-to-adult) was strongly affected by diet (Figure 2.6, log-

**Figure 2.4** Average oviposition rates of young adult females of A. swirskii on a diet of thrips larvae, whitefly eggs, or a combination of the two (mixed diet). Shown are average numbers of eggs (+ SE) per female per day measured after 48 and 72 h since the predators were allowed to feed on these prey.

**Figure 2.5** Average predation rates by young adult females of A. swirskii on thrips larvae and whitefly eggs when offered either of these two prey species separately or in combination (mixed diet). Data are from the same experiment as shown in Figure 2.4. Shown are average number of prey consumed (+ SE) per female per day measured after 48 h since the predators were allowed to feed on these prey.
rank test: $\chi^2 = 28.9$, df = 2, $P<0.001$). The difference in development was significant among all three diets (Figure 2.6, all $P$'s<0.035).

**Discussion**

We investigated whether the use of a single species of natural enemy for biological control of two pests resulted in better control of the two pest species through an increase in predator densities (apparent competition). Alternatively, it could result in temporal escape of one of the two pest species (apparent mutualism). Our results support the first hypothesis and not the second; we found significantly higher abundance of the shared predators and lower levels of whiteflies, one of the two herbivore species. Thus, control of whiteflies was improved by the presence of thrips, but the control of thrips was not affected by the presence of whiteflies. Such asymmetric effects of prey species on each other through a shared predator have also been referred to as indirect amensalism rather than apparent competition (Chaneton and Bonsall 2000). Perhaps the lack of an effect of the presence of whiteflies on the population densities of thrips was caused by the low initial densities of thrips. Possibly, experiments with higher initial densities of thrips would reveal a positive effect of the presence of whiteflies on the control of thrips.

We found no evidence that either of the two pest species had a positive effect on the other species in the greenhouse. In the laboratory, however, predation rates on each prey was halved in the presence of the other species. This was probably caused by
satiation of the predators owing to the higher total density (whitefly plus thrips) of prey. Because the total density of prey in the greenhouse experiments was also higher in the treatments with mixed prey, there might have been similar short-term positive effects of the two prey on each other through satiation of the predators, but they were probably not detected because they were masked by an increase in predator populations during the first four weeks. Experiments should therefore be done in which the populations of prey and predators are sampled more frequently early in the experiment.

Densities of predator species reached much higher levels in presence of both pest species; predator densities were up to 15 times higher compared to the situation with only thrips or whiteflies. This may have been partly caused by differences in the amount of food present: in the treatment with \textit{A. swirskii} and thrips, densities of thrips were low (<1 larva/leaf). Hence, the low numbers of predators may have been a direct consequence of low numbers of prey. The density of thrips larvae in combination with \textit{E. ovalis} was always high (>17 larvae/leaf). Thus, the low numbers of this predator cannot have been caused by low prey densities. Moreover, in the treatments with whiteflies and predators, densities of whitefly immatures were always high (>67 immatures/leaf with \textit{A. swirskii} and >138 immatures/leaf with \textit{E. ovalis}), suggesting that food was also not limiting in these treatments. Thus, the high numbers of predators in the treatments with both pests must have another cause besides prey densities.

To assess whether the higher predator densities were caused by differences between a mixed diet and the two single-species diets, we measured oviposition and survival in the laboratory. Oviposition on a mixed diet was not higher than on a diet of each of the two pest species alone. However, pest densities in the laboratory were never limiting for oviposition – the predators consumed at most half of the prey present – whereas densities in the greenhouse might have been. Hence, it cannot be ruled out that higher predator densities in the greenhouse were caused by increased availability of prey. In the laboratory, juvenile survival was affected by diet, and was significantly higher on a mixed diet than on either prey species separately. Likewise, developmental rate was also highest on a mixed diet. These differences in survival and developmental rate were not caused by increased availability of prey in the treatments with the mixed diet, because densities were always sufficiently high to avoid prey shortage. Hence, the higher juvenile survival and developmental rate were a result of the mixture of prey. This better juvenile performance on a mixed diet may also explain the differences in predator density observed in the greenhouse. Assuming exponential population growth of the predatory mites, the small changes in growth rate due to an increase in juvenile survival and developmental rate as observed here, would have enormous effects on numbers of predators: after 8
weeks, the predator population on the mixed diet would be 7 times as high as on a single thrips diet and 27 times as high as on a single whitefly diet. We suspect that the high densities of *E. ovalis* in the mixed species treatment has a similar cause. However, it must be noted that both thrips and whiteflies were present in sufficiently high numbers in the laboratory experiment, whereas thrips levels in the greenhouse were relatively low compared to whitefly densities. Possibly, a few thrips larvae in a diet of whiteflies already have a strong impact on population growth of the predatory mites. However, it is also possible that most first–instar thrips larvae were consumed by the predators, present in high numbers, shortly after emerging from the eggs, resulting in an underestimate of the density of thrips larvae in the greenhouse.

When comparing *A. swirskii* with *E. ovalis*, it can be concluded that *A. swirskii* is a much better predator of thrips, confirming earlier experiments on cucumber (Messelink et al. 2006), and a better predator of whiteflies when thrips is also present as prey (this experiment). These differences might even be higher in reality because of the free migration of pests between the two predator treatments.

In conclusion, our results demonstrate that increased control of pests can be achieved through apparent competition between two pest species. Whereas earlier studies focused on the use of a non-pest species to increase predator densities (Karban et al. 1994, Hanna et al. 1997, Liu et al. 2006), this study shows that similar results can be reached with two pest species that share a predator species. A further novelty of this study is the indication that the consumption of a mixed diet increases the densities of the natural enemies, not just because of a higher availability of food, but because a mixed diet results in higher growth rates of the predator populations through an increase in juvenile survival, resulting in further reduction of pest densities. We therefore suggest that the effects of mixed diets should be addressed in experimental and theoretical studies of apparent competition.

Nowadays, *A. swirskii* is increasingly used for biological control of thrips and whiteflies in many crops. Biological control strategies in these crops might be improved by tolerating acceptable levels of both thrips and whiteflies in order to stimulate population growth of predatory mites. Further research is needed to clarify relationships between pest levels and yield to enable implementation of results like these.

Acknowledgements
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References


CHAPTER 2 | TWO PESTS ARE BETTER THAN ONE

vан Rijn PCJ, van Houten YM, Sabelis MW. 2002. How plants benefit from providing food to predators even when it is also edible to herbivores. Ecology 83:2664-2679.
Prey temporarily escape from predation in the presence of a second prey species

R. van Maanen, G.J. Messelink, R. van Holstein-Saj, M.W. Sabelis & A. Janssen

In systems consisting of two prey and a shared predator, indirect interactions mediated by the predator such as apparent competition and apparent mutualism can play an important role. Depending on the time scale, these indirect interactions may result in positive (apparent mutualism) or negative effects (apparent competition) of the prey on each other’s densities. The effectiveness of using one species of predator as biological control agent against more than one pest is therefore difficult to predict. Using an experimental system consisting of the two pest species Western flower thrips and greenhouse whitefly and the shared predatory mite Amblyseius swirskii, we found evidence for a short-term escape of thrips from control when whiteflies were present, thus confirming the occurrence of apparent mutualism. More thrips larvae escaped because the predators dispersed slower in compartments with two pests. After six weeks, this effect was overruled by a higher total number of predators in greenhouses with both prey species due to positive effects of a mixed diet on the predator population growth rate. This shows that, despite the occurrence of apparent mutualism, the population dynamics of thrips and predatory mites were characterized by apparent competition after a few generations. Moreover, the average cucumber yield did not differ between compartments with or without a second pest present. Successful biological control of thrips was achieved, despite their temporarily higher numbers.

Submitted
species (so-called ‘apparent mutualism’; Holt and Lawton 1994, Abrams and Matsuda 1996, Holt 1997). Predator satiation, however, only occurs on a short time scale, because the higher availability of prey will result in a higher numerical response over subsequent predator generations (Holt and Kotler 1987). Long-term apparent mutualism may occur when population densities of one prey show cycles, resulting in repeated satiation of the shared predators and repeated reduced predation on the other prey (Abrams et al. 1998, Brassil 2006). Whereas there is much evidence for apparent competition, proof for apparent mutualism is scarce (Chaneton and Bonsall 2000). We studied the occurrence of apparent mutualism in a system consisting of two pests and a shared predator. Biological control systems, especially in greenhouses, offer an ideal opportunity to assemble communities, in which relatively simple food webs can be studied experimentally. Biological control traditionally used a single natural enemy for each pest species, whereas more recently, growers use one species of natural enemy against several pest species (Rosenheim et al. 1995, Janssen et al. 1998, Sih et al. 1998, Brodeur and Boivin 2006, Messelink et al. 2008), making predictions from models on apparent competition and apparent mutualism relevant for improving biological control (Janssen et al. 1998, Harmon and Andow 2004, van Veen et al. 2006). Several studies have demonstrated that the control of a pest species can be improved by the positive effect of another pest species or alternative food on predator density (Karban et al. 1994, Bonsall and Hassell 1997, Hanna et al. 1997, van Rijn et al. 2002, Liu et al. 2006, Messelink et al. 2008, 2010), whereas short-term disruption of biological control through predator satiation has also been demonstrated (Koss and Snyder 2005, Symondson et al. 2006, Xu et al. 2006, Desneux and O’Neil 2008). In most of these latter studies apparent mutualism occurred within a single predator generation, through satiation or switching behaviour of the shared natural enemy (Murdoch 1969, Abrams and Matsuda 1996). In theory, both short- and longer-term effects of shared natural enemies can lead to apparent mutualism (Holt and Lawton 1994), but empirical evidence for this is limited (Tack et al. 2011).

We conducted an experiment to investigate whether pest species can escape from control due to satiation of a predator, and how this is affected by the presence of a second pest species. We used a system consisting of the generalist predatory mite Amblyseius swirskii Athias-Henriot (Acari: Phytoseiidae) and two pest species, the greenhouse whitefly (Trialeurodes vaporariorum (Westwood)) and the Western flower thrips (Frankliniella occidentalis (Pergande)). These two herbivores are important pests in greenhouses in Western Europe and frequently invade the same greenhouses (Messelink et al. 2010), where they cause damage to crops such as cucumber, tomato and sweet pepper. The whitefly crawlers and adults cause direct crop damage by inserting their stylet into leaf veins and extracting phloem sap and indirect damage by decreased photosynthesis as a result of sooty mould (Vet et al. 1980, Lei et al. 1996).
Thrips feeding on immature cucumber fruits causes silvery scarring, or even malformation of the fruit. As a result, fruits may be downgraded because of quality loss of their appearance, thereby reducing the market price significantly. A. swirskii occurs in the wild in Mediterranean countries, and is well adapted to warm and humid climate conditions. This predatory mite has proven to be an effective control agent for thrips (Messelink et al. 2006), whiteflies (Nomikou et al. 2001, 2002, 2010) and both thrips and whiteflies at the same time (Messelink et al. 2008, 2010).

In our experiments we introduced large numbers of pests and predators in order to detect a possible escape of a pest species in time due to predator satiation (apparent mutualism). The pest species were released all on one plant in order to investigate whether predatory mites and thrips dispersed differently in the presence of whiteflies. This would offer the prey with the highest dispersal rate the possibility to escape to plants further away, while the predator population would lag behind and interact with the pest that disperses less far.

Materials and Methods

Cucumber plants (cv. Aviance RZ) were grown in rock wool blocks in a greenhouse compartment without applying pesticides. The predatory mite was reared on pollen, amply produced by flowering Ricinis communis L. plants in small greenhouses (6 × 6 m). Western flower thrips were reared on flowering chrysanthemum plants (Dendranthema grandiflora Tzvelev, cv. Miramar) and greenhouse whiteflies were reared on tobacco plants (Nicotiana tabacum L.). The greenhouse experiment was carried out at Wageningen UR Greenhouse Horticulture, Naaldwijk, The Netherlands. The experiment started in week 37 in 2006 and lasted until week 48, corresponding to the length of a crop cycle of greenhouse cucumber plants. Plants were at the fifth-leaf stage when the experiments started. Before planting, roots were preventively treated with propamocarb (Previcur N®, Bayer Crop Science) against Pythium spp. Rock wool slabs in which the plants rooted were supplied with a nutrient solution through drip irrigation. Plants were cultivated vertically up to a 2 m high crop-supporting wire. Side-shoots were removed until the top of the plant reached the wire. Thereafter, side-shoots were allowed to grow down over the wire. The main stem of the plants was trained using a white polyethylene string that was suspended from the crop supporting wire (Messelink et al. 2008).

We used two compartments (9.6 × 7.9 m), each containing one replicate with twelve sub-replicates represented by 12 rows, each with ten cucumber plants (Figure 3.1). In both compartments, the two pests were released in the first week after planting and predatory mites were released three days later. In each compartment, 720 adult female thrips (collected from the culture using a aspirator) and 360 (male + female) whitefly adults were released on every second plant of row 1-6 and on the ninth plant of row 7-
12 (hereafter termed release-plants) (Figure 3.1). Three days later, 135 female predatory mites were released in both compartments on the same plants as where pests had been released before. Female predatory mites were sampled in the laboratory using a fine paintbrush and placed on leaf discs of sweet pepper (*Capsicum annuum* L.) (diameter 2 cm) containing cattail pollen (*Typha latifolia* L.). Nine leaf discs with 15 mites per leaf disc were introduced on each cucumber plant. As a control, we used three compartments of the same size and the same number of cucumber plants, in which we released only 720 adult female thrips and, three days later, 135 female predatory mites.

The experiment was divided into two periods, in which we used different methods to assess presence of insects and mites. Two days after releasing all insects and mites, three flowers and three top leaves of each plant were checked for the presence of adult thrips and whiteflies, and once per week, we examined six randomly chosen leaves per plant for female adult predatory mites with a magnifying glass. We started inspecting the release plant and subsequently every following plant until we failed to find any mites on two consecutive plants. In the second period, from week 4-10, two leaves of previously designated plants were collected and the number of predators and pests on these plants were assessed every two weeks. Designated plants were marked ‘close’, ‘middle, and ‘far’ (Figure 3.1). Plants marked as ‘close’ were next to the so-called release plant. Plants marked as ‘middle’ were four plants away from the release plant and the plants marked ‘far’ were seven plants away (Figure 3.1).

![Figure 3.1 Map of the greenhouse compartment with 12 rows of 10 cucumber plants. Every square represents one plant. Black squares represent the plants where predator and pests were released. Gray squares represent the plants that were sampled. Plants marked as ‘close’ were next to the so-called release plant. Plants marked as ‘middle’ were four plants away from the release plant and the plants marked ‘far’ were seven plants away.](image-url)
3.1. The sixth and eighth leaf from the tip of a randomly chosen shoot were collected. The leaves were each put in a separate bag and transported to the laboratory, where they were cut into strips of 5 cm (Messelink et al. 2008). The number of mites and thrips were counted on both sides of the strips using a binocular microscope (40x). The average temperature and relative humidity were comparable among compartments. After six weeks, cucumbers were harvested three times a week and categorized in three different groups; (I) no damage, (II) little thrips damage (silver damage) or (III) severe thrips damage (malformed).

Differences in distances that predatory mites dispersed during the first three weeks were subject to a Kruskal-Wallis ANOVA because data were not normally distributed, also not after transformations. Differences in numbers of thrips larvae and adult female mites between treatments were analysed with a linear mixed effect model on log(y+1) transformed data. For thrips larvae, data from week 8 onwards were excluded from the analysis because of the absence of thrips in many replicates. In order to find differences in spatial distribution of mites and thrips between treatments, we analyzed proportions of thrips larvae and female mites on plants close by, in the middle and far from the release plant. Although data were repeated measures in time, we could not analyse them with an appropriate repeated measures model (generalized linear mixed effects model) because the residuals were not adequately distributed. We therefore performed a generalized linear model with quasi-binomially distributed errors for each time step separately. Differences in yield (weight of cucumbers with no damage) per treatment were analysed with generalized linear mixed effects model.

Results

Two days after releasing thrips and whitefly adults, they were found on 100 and 87% of all plants, respectively. There was no difference in maximum distance reached by the predatory mites during the first two weeks of the experiment, but after three weeks, predatory mites dispersed over significantly longer distances when one instead of two prey species was present (Figure 3.2). Through time, numbers of thrips larvae were significantly higher when whiteflies were present instead of when there were no whiteflies present (Figure 3.3). Although most thrips larvae were found ‘far’ from release plants (Figure 3.3), there was no significant difference in the proportion of thrips larvae at plants close by, in the middle or far from the release plants (Figure 3.3). Through time, numbers of predatory mites were significantly higher when two instead of one pest species was present (Figure 3.4). Only after eight weeks, the fraction of mites ‘close’ to the release-plants differed significantly between treatments (Figure 3.4). This suggests that the spatial distribution of mites was not affected much by the presence of whiteflies. The average production of undamaged fruits, suitable for selling, did not differ between treatments ($F_{1,3} = 4.37, P = 0.13$).
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**Figure 3.2** Dispersal of predators through the plant rows during the first 3 weeks after release. Shown are the plants most distant from the release plants on which *Amblyseius swirskii* was present after 1, 2 and 3 weeks following predator release, averaged (+ SEM) per row (Figure 3.1). Thrips (*F. occidentalis*) were released earlier on the same plants, either with (two pests) or without greenhouse whiteflies (*T. vaporariorum*) (one pest). Asterisks indicate a significant difference (P<0.005) in maximum distance reached in the presence or absence of whiteflies (Kruskal-Wallis ANOVA week 1: $\chi^2 = 1.53$, df = 1, P = 0.22; week 2: $\chi^2 = 0.597$, df = 1, P = 0.44; week 3: $\chi^2 = 7.28$, df = 1, P = 0.007).

**Figure 3.3** Numbers of thrips larvae per compartment either with one prey (thrips, three replicates, shaded bars) or with two prey species (thrips plus whitefly, two replicates, hatched bars) through time since predator release. Bars give average numbers of thrips larvae per row of plants, error bars are SEM. Each bar is subdivided into the numbers found on plants close by the release plant (lower section), in the middle (middle section) and at the end of the row (top section). Significantly more thrips larvae were found in the presence of both pests than with one pest ($F_{1,16} = 8.78$, P = 0.008). Although most thrips were found ‘far’ from release plants ($F_{1,16} = 5.31$, P = 0.033), there was no significant difference in the proportion of thrips larvae at plants close by, in the middle or far from the release plants (all $F_{1,35}<1.15$ and all P>0.29).
Discussion

Significantly higher numbers of thrips were found when whiteflies were present than when no whiteflies were present, confirming the occurrence of apparent mutualism. These results support the theory that two prey species that share a predator may affect each others’ densities positively. The mechanism that causes this indirect positive effect in population dynamical models is predator satiation through an increase in the numbers of one of the prey species (Holt and Lawton 1994, Abrams and Matsuda 1996, Holt 1997). We did not measure predator satiation levels, hence we cannot confirm the importance of this mechanism, but think it is a likely explanation. Although densities of thrips were initially higher in the presence of whiteflies, (temporary escape), after three weeks the number of predators was equally high in compartments with only thrips and with thrips and whiteflies together. After ca. six weeks, the density of the shared predator reached much higher levels in the presence of two instead of one pest species (Figure 3.4). Higher density levels in presence of two pest species was shown earlier by Messelink and co-authors (2008). They showed that a mixed diet of whiteflies and thrips results in higher growth rates of the predator populations through an increase in juvenile survival and developmental rate of A. swirskii (Messelink et al. 2008).
Although we found differences in numbers of prey and predators between treatments, there was limited (in the case of predators) or no (in the case of thrips) evidence for different spatial distributions between treatments after four weeks. However, we did find differences in the maximum distance dispersed by predators after three weeks. We therefore suggest that thrips larvae had a small window of opportunity to escape, because the predators dispersed slower in the presence of two pests than with one pest. Predators might have had longer residence times in the presence of two prey species because there was a higher overall density of prey. Several studies have shown that the dispersal rate of phytoseiid mites decreases with an increase in prey densities (Croft 1995, Zemek and Nachman 1998). Adult thrips and whiteflies disperse relatively fast compared to predatory mites; two days after releasing pests and predators, we found thrips in flowers of all 120 cucumber plants per compartment and whitefly adults in approximately 104 plants, whereas mites were more or less evenly distributed over all plants per compartment after eight weeks. The results confirm our earlier studies in that the presence of more than one pest species can enhance pest control by the numerical response of the predatory mites (Messelink et al. 2008, 2010). Towards the end of the experiment, low numbers of thrips were found in all compartments and there was no difference in the number of fruits with thrips damage and the average cucumber yield did not differ between compartments with or without a second pest present. Hence, successful biological control was achieved, despite the early temporary escape of thrips leading to higher thrips numbers.

The predatory mite used here has a short generation time: ca. seven days from egg to adult on a mixed diet of thrips and whiteflies at 25 °C (Messelink et al. 2008). Hence, the predators could probably produce 7-8 generations during the experiments. In our study system, both the predators and the prey species went through several generations before the initial effects of apparent mutualism were overruled by those of apparent competition. Thus, the theoretical prediction that long-term dynamics will be characterized by apparent competition and not by apparent mutualism (Holt and Kotler 1987) was confirmed.

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Pest species diversity enhances control of spider mites and whiteflies by a generalist phytoseiid predator

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To test the hypothesis that pest species diversity enhances biological pest control with generalist predators, we studied the dynamics of three major pest species on greenhouse cucumber: Western flower thrips, Frankliniella occidentalis (Pergande), greenhouse whitefly, Trialeurodes vaporariorum (Westwood), and two-spotted spider mites, Tetranychus urticae Koch in combination with the predator species Amblyseius swirskii Athias-Henriot. When plants were infested with spider mites prior to predator release, predatory mites were not capable of controlling spider mite populations in the absence of other pest species. A laboratory experiment showed that predators were hindered by the webbing of spider mites. In a greenhouse experiment, spider mite leaf damage was lower in the presence of thrips and predators than in the presence of whiteflies and predators, but damage was lowest in the presence of thrips, whiteflies and predators. Whitefly control was also improved in the presence of thrips. The lower levels of spider mite leaf damage probably resulted from (1) a strong numerical response of the predator (up to 50 times higher densities) when a second and third pest species were present in addition to spider mites, and (2) from A. swirskii attacking mobile spider mite stages outside or near the edges of the spider mite webbing. Interactions of spider mites with thrips and whiteflies might also result in suppression of spider mites. However, when predators were released prior to spider-mite infestations in the absence of other pest species, but with pollen as food for the predators, we found increased suppression of spider mites with increased numbers of predators released, confirming the role of predators in spider mite control. Thus, our study provides evidence that diversity of pest species can enhance biological control through increased predator densities.


Much research has been devoted as to whether the presence of multiple natural enemies leads to more efficient pest suppression than the presence of single enemy species (Rosenheim et al. 1995, Denoth et al. 2002, Cardinale et al. 2003, Casula et al. 2006). Empirical studies show that increasing diversity of natural enemies can result in a full spectrum of outcomes, including additive, antagonistic, synergistic, or no effects on biological control (Casula et al. 2006, Janssen et al. 2006, 2007). Less attention has been paid to the impact of pest species diversity on biological control. Most pest management programs in modern greenhouse cropping...
systems are focused on excluding and eliminating pest species as much as possible, resulting in low pest species diversity. However, indirect interactions occurring among various pest species may enhance biological control (Janssen et al. 1998, Harmon and Andow 2004, Prasad and Snyder 2006, van Veen et al. 2006). Such an indirect interaction occurs when the density of one prey species affects the density of a polyphagous natural enemy, which consequently affects the density of a second prey species. Holt (1977) was the first to develop theory on this mode of indirect interaction. He showed that the equilibrium density of a population of one prey species decreases when that of another, non-competing prey species is increased. Holt coined the term ‘apparent competition’ because it appears as if the two species compete for a shared resource, whereas in fact the two prey populations interact via the shared predator. Subsequently, theory was developed for the case of short-term dynamics of systems involving multiple prey that share the same natural enemy (Holt and Kotler 1987, Abrams and Matsuda 1996, Abrams et al. 1998). Such short-term, non-equilibrium dynamics are a more realistic scenario in agricultural systems with a short production cycle than the equilibrium dynamics studied by Holt (1977) and Holt and Lawton (1994). The theory on short-term dynamics shows that predators can not only mediate apparent competition between two of their prey species, but also apparent mutualism. In the latter case, predator satiation results in a short-term positive indirect interaction between its prey species. With respect to biological control, some studies have indeed demonstrated that the control of a pest species can be improved by the presence of another pest species (Collyer 1964, Karban et al. 1994, Liu et al. 2006, Messelink et al. 2008), whereas disruption of biological control through predator satiation in the short-term has also been demonstrated (Koss and Snyder 2005, Symondson et al. 2006).

So far, both theory and experiments on the effects of a shared predator have ignored the effects of a mixed diet on predator populations. Different prey can have complementary nutritional values (Wallin et al. 1992, Toft 1995, Evans et al. 1999), and this can amplify the effects of predator-mediated apparent competition. Hence, the presence of several prey species can increase predator populations through the increased availability of food as well as through the higher quality of a mixed diet. Based on these mechanisms, pest species diversity in combination with predators attacking various prey species can enhance biological control (Messelink et al. 2008). This study was designed to further evaluate the hypothesis that increasing pest species diversity can enhance biological control with generalist predators. We studied the dynamics of three major pest species in greenhouse crops, i.e. Western flower thrips, Frankliniella occidentalis (Pergande), greenhouse whiteflies, Trialeurodes vaporariorum (Westwood), two-spotted spider mites, Tetranychus urticae Koch and the predator Amblyseius swirskii Athias-Henriot (Zannou et al. 2007). The predatory mite
A. swirskii has proven to be an effective control agent for thrips (Messelink et al. 2006) and whiteflies (Nomikou et al. 2001, 2002), whereas its effect on spider mites is still unclear. Moreover, control of whiteflies is improved when thrips are present in low densities (Messelink et al. 2008). Although A. swirskii does feed on spider mites (Momen and El-Saway 1993), greenhouse observations suggest that the webbing produced by spider mites impedes effective control because A. swirskii is not able to enter it (GJ Messelink, personal observations). It has been suggested that one of the functions of spider mite webbing is defence against predators, and the way in which predatory mites cope with this webbing is suggested to depend on the dorsal chaetotaxy of the predators (Sabelis and Bakker 1992). The predator A. swirskii has short dorsal setae, and is therefore expected to be hindered by spider mite webbing. We first verified this by measuring predation rates of spider mite eggs by A. swirskii in the presence and absence of spider mite webbing. Subsequently, we verified that A. swirskii is not capable of controlling spider mite populations in the absence of other pest species. Finally, we investigated whether spider mite control by A. swirskii can be enhanced by the presence of the other pest species, i.e. thrips and whiteflies. A further experiment was done to shed some light on the possible mechanisms responsible for increased spider mite control in the presence of other pests. It is not our aim here to completely disentangle how multiple pest species interact, i.e. directly, indirectly via the plant, indirectly via the shared predator or via any combination of these mechanisms. Our primary goal is to establish the extent to which multiple pest species affect control efficacy and discuss the interactions mentioned.

Materials and Methods

Cultures

The predatory mite A. swirskii was reared on a diet of cattail pollen (Typha latifolia L.) in climate rooms, under 16 h of artificial illumination per day, at 22 °C and 70% RH on plastic arenas of a type described by Overmeer (1985). For the experiment with predator densities, A. swirskii was obtained from Koppert Biological Systems (Berkel en Rodenrijs, The Netherlands). These mites were reared on bran containing the sugar mite Carpoglyphus lactis L. All pest species were reared on plants in greenhouse compartments. Two-spotted spider mites were reared on bean plants, Phaseolus vulgaris L., Western flower thrips were reared on flowering chrysanthemum plants (Dendranthema grandiflora Tzvelev, cv. Miramar) and the greenhouse whitefly was reared on tobacco plants (Nicotiana tabacum L.). For assessing the effects of spider mite webbing on predatory mites in the laboratory, we reared the predatory mite species Phytoseiulus persimilis Athias-Henriot and Neoseiulus californicus (McGregor) on spider mite-infested cucumber plants in greenhouses.
Effect of webbing on predation
A laboratory experiment was set up to assess the extent to which spider mite webbing hinders predation by *Amblyseius swirskii*. Although spider mite eggs are less suitable for development of *A. swirskii* than the mobile stages of this prey (R van Maanen, personal observations), we used eggs because they do not move from the web where they are deposited. Predation by *A. swirskii* was compared with that of two other species of predatory mite, *P. persimilis* and *N. californicus*, commonly used for control of spider mites. This comparative test served to assess the impact of webbing on predation of spider mite eggs. We placed cucumber leaf discs (3 cm diameter, cut from the inter-vein area of 4-week-old plants) upside down on water-saturated cotton wool in plastic boxes (14 \times 20 cm), six leaf discs per box. Three female spider mites were placed on each leaf disc for two days, resulting in a colony with 40 eggs on average. Spider mites were prodded with a small paintbrush to make them move out of the webbing without harming the web structure, and the eggs were counted. The webbing was removed from half of the leaf discs with a fine needle. Single, young adult female predators (1-6 days old since their last moult, starved for one day), were placed on the leaf discs and the boxes with discs were incubated in a climate room (25 °C and 16:8 light:dark). The surviving eggs were counted after 24 h. Treatments were replicated 12 times with new predator individuals. Differences between treatments involving web or web removal and treatments involving different predator species were analysed using an ANOVA on the log-transformed numbers of eggs, followed by Fisher’s LSD (Least Significant Difference) test at the 5% confidence level.

Spider mite control by *Amblyseius swirskii*

We studied the population dynamics of spider mites and *A. swirskii* in the absence of other pest species on cucumber plants (cv. Aviance, powdery mildew resistant) in two separate greenhouse compartments (18 m²). So there was only one treatment, in which the spider mites were released prior to the predatory mites. Each compartment contained two tables (1 \times 3 m) on which plants were grown up to a 1.5 m high wire to support the crop. Plants were grown in rock wool blocks. Side-shoots were removed until the top of the plant reached the crop supporting wire, and all further plant shoots were suspended from the wire. Each greenhouse compartment had a small entrance corridor, and was ventilated with an air pressure system in order to minimize contamination by organisms from outside. The plants had six leaves when the experiment started in March 2006. Roots were preventively treated against *Pythium* spp by soaking the rock wool blocks in a 0.1% solution of propamocarb (Previcur N®, Bayer Crop Science). No further pesticides were used. Four plants were placed on two pieces of rock wool substrate on each table. The rock wool mats were
continuously immersed in a nutrient solution that was automatically supplied twice a day. One day after planting, the plants were infested with spider mites by adding two cucumber leaf discs (2 cm diameter), each containing ten females that were collected from the culture. These leaf discs were put on the fourth and fifth leaf of each plant, counted from the lowest leaf. Predatory mites were released seven days after introducing the spider mites. Female predatory mites were collected in the laboratory with a fine paintbrush and placed on leaf discs of sweet pepper (*Capsicum annuum* L.) (2-cm-diameter) containing cattail pollen. One leaf disc with 15 mites was introduced onto each cucumber plant on the seventh leaf from below. Each table was considered as a single replicate. Hence, there were four replicates.

The experiment lasted eleven weeks, roughly corresponding to the standard cropping period for modern glasshouse cultures. Cucumbers were harvested as soon as they reached the standard fruit size, but fruit yield was not measured. The numbers of predatory mites and the percentage of leaf surface with spider mite damage were assessed five, seven, nine and eleven weeks after introducing the pest species. Assessment of the populations was done on six leaves of each replicate. The leaves were collected by randomly choosing three shoots of which the sixth and eighth leaf, counted from the tip, were collected. Each leaf was put in a separate plastic bag and transported to the laboratory, where it was cut into strips of 5 cm wide. The predatory mites were counted on both sides of the leaves using a stereomicroscope (40x). Spider mite damage was assessed by estimating the percentage of leaf damage by persons which were trained for these observations with the computer program ‘Distrain’ (Tomerlin and Howell 1988).

**Effects of prey diversity on pest control**

The combined control of whiteflies, thrips and spider mites by *A. swirskii* was studied on plants in greenhouse compartments as explained above and with various combinations of pests: (1) spider mites plus thrips; (2) spider mites plus whiteflies; (3) spider mites plus thrips and whiteflies; (4) thrips only. Combinations of whitefly and thrips were examined in an earlier experiment, described elsewhere (Messelink et al. 2008). Experiments were carried out in eight compartments simultaneous with the experiment on spider mite control. Each treatment was replicated four times.

For all treatments with thrips, female thrips were collected with an aspirator from a culture on chrysanthemum and introduced at a rate of 10 per plant. For all treatments with whiteflies, adult greenhouse whitefly (sex ratio 1:1.27 male: female) were collected with an aspirator from a culture on tobacco plants and released at a rate of 40 per plant. All pest species except spider mites were released one day after planting. Predatory mites (15 females per plant) were released seven days after introducing the pests, at the start of the second week (thus, exactly at the same time and same num-
ber as in the experiment on spider mite control). Spider mites were introduced three weeks later than the predatory mites, in order to evaluate the effects of an already established predator population. A period of three weeks was chosen to allow the predator populations to increase on the food present in the crop (thrips, whiteflies or both). Labelled cucumber leaves of young side shoots were provided with small cucumber leaf discs (2 cm diameter), each containing 20 female spider mites collected from the culture on bean plants. These discs with spider mite colonies were applied to four leaves per replicate. During the next six weeks, we assessed the percentage of spider mite damage on four younger and four older leaves, next to the leaves on which the spider mites were released. Assessments per leaf were done as described in the experiment on spider mite control by the same trained person, as it was impossible to count mites on so many plants using a non-destructive method. The typical leaf tissue damage caused by spider mites (Park and Lee 2002) could easily be distinguished from thrips damage. Heavily infested and wilting leaves were considered as 100% damaged by spider mites. A control treatment with spider mites added to plants with only predatory mites could not be included, because predatory mites do not survive on plants in a period of three weeks without any prey. The numbers of predatory mites, thrips and whiteflies were assessed five, seven, nine and eleven weeks after starting the experiment (and introducing the pest species). Assessment of the populations was done on six leaves in each treatment, as described above. The number of predatory mites, thrips and whiteflies were counted on both sides of the strips of leaf using a stereomicroscope (40x). Predatory mites were regularly slide-mounted for identification to species with the aid of a phase contrast microscope (400x). Only the juvenile stages of thrips and whiteflies were counted because adults fly away when leaves are collected. When the densities of whiteflies exceeded 500 individuals per leaf, densities were assessed only on representative parts of the underside of each leaf, and then extrapolated to the whole surface of the leaf. The average temperature and relative humidity were comparable among greenhouse compartments (22 °C and 74% RH).

For statistical analyses, a repeated measures ANOVA was performed on the arcsine square-root transformed fractions of estimated leaf damage by spider mites. The time since introduction of the pest organisms was chosen as the repeated measure variable. The same repeated measures analyses were performed for densities of thrips, whiteflies and predators after a log(x+1) transformation. Differences between treatments were tested at a 5% confidence level using Fisher’s LSD method.

Effects of predator density on spider mite control
A third greenhouse experiment was carried out to test the effects of densities of A. swirskii on the establishment and population dynamics of spider mites in a situation
where the predators were introduced one week prior to spider mite infestation. Differences in spider mite densities among treatments in the former experiment might not only be caused by predator densities, but also by interactions of thrips and whiteflies with spider mites, such as resource competition, induced plant resistance or predation by thrips. We therefore released predators in two densities prior to spider-mite infestations in the absence of the other two pest species. In this way, the effects of thrips and whitefly presence on spider mite densities through the shared predator population was mimicked, while excluding the other interactions between spider mites and the other pests. In one greenhouse compartment of 24 m², we placed eight cucumber plants (cv. Filia, powdery mildew resistant) on rockwool mats on each of three tables (1.5 × 3m). These plants were treated once with Abamectine when they were two weeks old, to keep them free of thrips. Plants were grown as in the experiments described above. Each plant was isolated and did not touch other plants and was allowed to grow up to a 2 m high wire that supported the plant. When the plants were four weeks old, with 7-8 full-grown leaves, we divided the plants into three groups, and treated them with (1) no predatory mites (control), (2) a low density of 50 predatory mites per plant and (3) a high density of 500 predatory mites per plant. The predatory mites were released as a mixture with bran and the sugar mite C. lactis (Koppert Biological Systems, Berkel en Rodenrijs, The Netherlands), and they were deposited on top of the rockwool blocks near the base of the plants, from where the mites were able to walk up the plants. The few sugar mites present were not observed to walk onto the plants. We released the predators one week prior to the spider mite infestations to allow them to colonize the plant. Because the plants were devoid of prey, and predatory mites do not survive on plants without food, we added 20 µg of pollen of T. latifolia as food for the predators onto all plants on the 7th leaf from below. Plants with the same treatment were placed on one table to avoid contamination among treatments. The dispersal of mites among plants was impeded by placing sticky plates around the rockwool slabs on which the plants were standing. Each plant was considered as one replicate. One week after the predator releases, all plants were infested with spider mites on the 9th leaf (counted from below) by adding one cucumber leaf disc (2 cm diameter), each containing 20 females, collected from a culture on bean plants. The numbers of spider mites and predatory mites were assessed on these leaves two weeks later by cutting them and counting the mites using a stereo microscope (40x) as described in the greenhouse experiments above. The average greenhouse temperature was 22 °C and the average relative humidity 74%. Effects of treatments on spider mite densities were analysed using an ANOVA on the log(x+1)-transformed numbers of the sum of eggs and mobile stages. Differences between treatments were tested at a 5% confidence level using Fisher’s LSD method.
Results
Effect of webbing on predation
Spider mite webbing had a significant effect on the predation of spider mite eggs by *A. swirskii* \((F_{5,64} = 20.68; P<0.001)\); predation was reduced by 57% (Figure 4.1). The presence of webbing had no impact on predation by the predatory mites *P. persimilis* and *N. californicus*. In the absence of webbing, the predation rate of *A. swirskii* was also lower than that of *P. persimilis* (Figure 4.1). The maximum predation rate of *P. persimilis* might even be higher than observed here, because spider mite eggs were almost depleted in some replicates. In the *N. californicus* and *A. swirskii* treatment, ample amounts of eggs were available throughout the experiment.

Spider mite control by *Amblyseius swirskii*
*Amblyseius swirskii* was not able to control spider mites on cucumber plants without populations of other pest species (Figure 4.2). At the end of the experiment, the plants were completely covered by spider mite webbing. Crop growth was poor and there were many desiccated leaves. Though *A. swirskii* was able to establish, densities remained low \(<1.3/leaf\), at least until week 9 (Figure 4.2). At the end of the experiment, a light contamination with thrips was observed in all replicates (average 0.2 and 0.3 larvae/leaf in respectively week 9 and 11), which may explain the increase in predator densities (Figure 4.2).

![Figure 4.1](image-url) Predation rates of three predatory mite species on two-spotted spider mite eggs on cucumber leaf discs with (grey bars) or without (white bars) spider mite webbing. Shown are average numbers of spider mite eggs consumed (± SE) per female predatory mite in 24 h. Different letters indicate significant differences among treatments (Fisher’s LSD test, \(P<0.05\)).
Effects of prey diversity on pest control

When spider mites were released on plants with thrips, whiteflies or thrips plus whiteflies, there was a strong effect of pest treatment on leaf damage by spider mites (Figure 4.3), resulting in significant differences among treatments ($F_{2,42} = 19.97; P<0.001$). The highest levels of spider mite damage were observed in the treatment

![Graph](image1.png)

**Figure 4.2** The dynamics of two-spotted spider mite damage and of numbers of predatory mites (*A. swirskii*) during an 11 week greenhouse experiment. Spider mites were added at the start of the experiment (week 1), predators were added in the second week. No other pest species were released. Shown are average percentages ($\pm$ SE) of leaf damage and average densities ($\pm$ SE) of the predatory mite *A. swirskii*.

![Graph](image2.png)

**Figure 4.3** Leaf damage by two-spotted spider mites in a greenhouse in the presence of the predatory mite *A. swirskii* and the pest species Western flower thrips, greenhouse whiteflies or Western flower thrips plus greenhouse whiteflies. Shown are average percentages ($\pm$ SE) of leaf damage on eight marked leaves that neighboured the leaves where spider mites were released. Different letters indicate significant differences among treatments through time (Fisher’s LSD test, $P<0.05$).
with whiteflies, spider mites and *A. swirskii*. Damage was lower in the treatment with thrips, spider mites and *A. swirskii*, and the lowest levels of spider mite damage were found when both thrips and whiteflies and *A. swirskii* were present (Figure 4.3). There was a significant effect of pest species diversity on the densities of predators (Figure 4.4) ($F_{3,92} = 88.45; P < 0.001$). The highest predator levels were found in the treatments with thrips, whiteflies and spider mites, where predator levels were at least 11 times higher than in the other treatments at the population peak in week 9 (Figure 4.4). Two replicates of the whitefly treatment were slightly contaminated with thrips at the end of the experiment (on average 0.8 larvae/leaf in week 11), but this was ignored in the statistical analyses.

Not only spider mites, but also whiteflies were controlled significantly better in the presence of thrips (Figure 4.5) ($F_{1,43} = 40.77; P < 0.001$), confirming the results of a similar experiment to which no spider mites were added (Messelink et al. 2008). Thrips densities did not differ significantly among treatments ($F_{2,66} = 0.01; P = 0.991$) and were always controlled adequately (Figure 4.6), as in experiments reported elsewhere (Messelink et al. 2008).

### Effects of predator density on spider mite control

The release of predatory mites prior to spider mite infestation significantly affected densities of spider mites (Figure 4.7) ($F_{2,20} = 32.77; P < 0.001$). The establishment of spider mites was even prevented on some plants on which high densities of predatory mites were released. The average predatory mite densities in the treatments with low and high predator releases were 7.3 and 11.1 mites/leaf respectively.

![Figure 4.4](image_url) The dynamics of predatory mites on cucumber plants during an 11-week greenhouse experiment. Shown are average densities (± SE) of the predatory mite *A. swirskii* on plants with only Western flower thrips, Western flower thrips plus two-spotted spider mites, greenhouse whiteflies plus two-spotted spider mites, and Western flower thrips plus greenhouse whiteflies plus two-spotted spider mites. Different letters indicate significant differences among treatments through time (Fisher’s LSD test, $P < 0.05$).
We tested the hypothesis that higher diversity of pest species enhances biological control with generalist predators. Indeed, in the presence of the generalist predatory mite *A. swirskii*, spider mite leaf damage was significantly lower in the presence of both thrips and whiteflies, than when there were either thrips only or whiteflies only. Spider mite leaf damage was reduced more in the presence of predatory mites plus thrips than with predatory mites plus whiteflies. In the absence of other pest species or other alternative food such as pollen, *A. swirskii* was clearly not able to control spider mites and there was hardly any growth of the predator populations.
The exact mechanisms responsible for this strong reduction in spider mite leaf damage in the presence of other pest species cannot be inferred from our experiment; a combination of direct and indirect interactions among the pests can be involved. Direct effects of whiteflies on spider mites are not likely to occur (e.g. reciprocal predation has never been observed), but competition for resources among whiteflies and spider mites might have reduced the growth of spider mite populations. However, because of the high availability of undamaged cucumber leaf tissue, we assume this to be of minor importance. If resource competition did strongly affect the population growth of spider mites, we would expect higher levels of spider mite leaf damage in the treatment with lower whitefly levels. However, the opposite occurred: lower levels of spider mite leaf damage were found at lower levels of whiteflies. This suggests that some other mechanism suppressed spider mites, such as indirect interactions of whiteflies with spider mites via the plant or via the predator. Unlike whiteflies, thrips do not only act as herbivores, but also as predators of spider mite eggs (intraguild predation, Trichilo and Leigh 1986). These direct effects on spider mites might explain the lower densities of spider mites in the presence of thrips than in the presence of whiteflies. As predator densities did not differ significantly between these two treatments (Figure 4.4), it is less likely that predator densities are responsible for the strong difference in effects on spider mites. Competition for food between the spider mites and the thrips is also not likely to have occurred, because

Figure 4.7 Two-spotted spider mite densities on cucumber plants on which no, low or high densities of the predatory mite *A. swirskii* were released prior to spider mite infestation. Shown are average densities (± SE) of mobile stages and eggs of spider mites per leaf, two weeks after the plants were infested with 20 females of spider mites per leaf. Different letters indicate significant differences among treatments for the sum of egg and mobile stages (Fisher’s LSD test, P<0.05).
thrips levels were quite low (<5 larvae/leaf). However, other studies have shown that thrips hide inside the webbing produced by spider mites when predators are present (Pallini et al. 1998, Venzon et al. 2000), so that local competition between thrips and spider mites might have played a role. Nevertheless, it is likely that the lower levels of spider mite leaf damage were a result of predatory mites as well as thrips feeding on spider mites. The hiding of thrips in the spider mite webbing could also result in reduced control of thrips (Magalhães et al. 2007), but we found no evidence for this, possibly because the amount of spider mite webbing was too low. Whiteflies and thrips might have reduced population growth rates of spider mites indirectly via the plant, as attacks of plants by one pest species can induce resistance mechanisms in the plant, which can subsequently slow the population growth of a second pest species (Karban and Carey 1984). This so-called induced resistance has potential for improving biological control (Karban et al. 1997). Further experiments are needed to clarify if such induced resistance occurs among the pest species in this study. Induced resistance might even have affected the consumption of spider mite eggs by thrips. On cotton, it was shown that induced plant resistance caused thrips to shift more towards predation than herbivory (Agrawal et al. 1999), but the reduced density and quality of spider mites on induced plants may antagonize this shift towards increased predation (Agrawal and Klein 2000).

The second indirect interaction between the pests that possibly resulted in lower levels of spider mite leaf damage is mediated by the shared predator. Our laboratory experiment shows that, despite the predators experiencing hinder from the spider mite webbing, predation of spider mite eggs still occurred. We decided to use spider mite eggs because they cannot escape from the web, but in the greenhouse, all stages of spider mites were present. We assume that the effects of the predators on spider mites in the greenhouse were mainly based on consumption of mobile stages outside or near the edges of spider mite webbing.

Reduction of spider mite leaf damage in the presence of other pests may well arise as a consequence of the strong numerical response of the predator when a second or third pest species was present in addition to the spider mites. Indeed, predator densities were higher when thrips and whiteflies were present together with spider mites, with up to 50 times higher densities when both thrips and whiteflies were added. The experiment where two densities of predatory mites were released prior to spider mite infestations clearly shows that higher predator densities reduce spider mite densities more than lower predator densities. So besides all other possible direct and indirect pest interactions, we suggest that predator densities are to a large extent responsible for the improved suppression of spider mites in the presence of other pest species. Not only spider mites, but also whiteflies were better controlled at higher predator densities due to the presence of both thrips and whiteflies. These predator-mediated
interactions among the three pest species can be classified as apparent competition, with the addition of thrips to a system of spider mites and whiteflies resulting in lower levels of spider mite leaf damage and better control of whiteflies. In addition to these density effects, several trait-mediated effects might have occurred as well. These occur when one species modifies the interaction between a pair of species by changing the behaviour of individuals of one or more of these species (not their numbers) (Prasad and Snyder 2006). We suggest that thrips larvae inside spider mite webbing may have caused spider mites to move out of the webbing, thereby making them more susceptible to predation.

Our results suggest that generalist phytoseiid mites such as *A. swirskii* can play an important role in reducing the colonization of a crop by spider mites, even when they are incapable of controlling spider mites alone. To which extent *A. swirskii* can control starting colonies of spider mites depends, at least partly, on the predator densities at the time of infestation, and thus of the presence of food for sustaining predator populations. The sequence of crop infestation by different pest species is therefore very important for the control of spider mites by *A. swirskii*, at least in crops where alternative food sources such as pollen are not available. Once spider mites have formed colonies, generalist predators such as *A. swirskii* cannot control them, and more specialized spider mite predators, such as *P. persimilis*, will be needed for spider mite control.

The higher predator densities in the treatments with more than one pest species may not have been merely caused by increased prey availability alone. Previous experiments showed that juveniles of *A. swirskii* survive and develop better on a mixed diet of thrips and whiteflies than on a single diet of either of these species. These effects of a mixed diet were suggested to be responsible for strong increases in predator densities in greenhouses in which both thrips and whiteflies were present, and consequently, for lower densities of whiteflies in the presence of thrips (Messelink et al. 2008). The high predator densities observed in the treatment with thrips, whiteflies and spider mites together support this idea. In theory, the addition of spider mites to a menu of thrips or whiteflies could have had the same effect, but the present results show no evidence for this: the addition of spider mites to treatments with thrips did not result in an increased predator population compared to a treatment with only thrips. Maybe spider mite densities were too low for such an effect to occur. We suggest that both the higher availability of prey and the effects of a mixed diet contributed to a high predator population. These high predator densities, in turn, contributed to improved control of spider mites. Although *A. swirskii* is not an efficient spider mite predator, it nevertheless reduced spider mite damage when the predator-prey ratio was sufficiently high.

In summary, we provide evidence that diversity of pest species enhances biological control of whiteflies and spider mites with a generalist predatory mite. Similar effects
might also be achieved by adding a non-pest alternative food source, such as pollen. Several studies have shown the benefit of pollen in terms of enhancing pest control (Nomikou et al. 2002, 2009, van Rijn et al. 2002), but so far, this has not resulted in large-scale applications. Our results furthermore suggest that it might be advantageous to allow or create some pest species diversity in a crop, rather than to try to exterminate all phytophages present. Further experiments have to be done to determine whether the total crop damage of three or two pest species is less than the damage inflicted by one of the pest species. Releasing pest species in a crop is considered risky, but is accepted in some cases, such as in sweet pepper, where some growers use the ‘pest-in-first’ strategy with spider mites to enhance control by P. persimilis (Hussey et al. 1965; K. Bolckmans, R&D Department, Koppert BV, personal communication). Avoiding total eradication of all pest species, thereby maintaining some pest diversity, might be more acceptable for the growers than introducing a new pest. For example, for cotton it has been suggested to leave a ‘pest residue’ as food for predators early in the season in order to enhance biological control of pests that occur later in the season (Luckmann and Metcalf 1975, Gonzalez and Wilson 1982). The demonstrated effects of pest diversity on a generalist predator in this study might furthermore be useful for evaluation programs of ‘new’ generalist predators by assessing their performance not only on the target prey alone, but also in the presence of other relevant pest species.

References
CHAPTER 4 | PEST SPECIES DIVERSITY ENHANCES BIOCONTROL


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Intraguild predation among plant pests: western flower thrips larvae feed on whitefly crawlers

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Omnivores obtain resources from more than one trophic level, and choose their food based on quantity and quality of these resources. For example, omnivores may switch to feeding on plants when prey are scarce. Larvae of the western flower thrips *Frankiniella occidentalis* Pergande (Thysanoptera: Thripidae) are an example of omnivores that become predatory when the quality of their host plant is low. Western flower thrips larvae usually feed on leaf tissue and on plant pollen, but may also attack eggs of predatory mites, their natural enemies, and eggs of the two-spotted spider mite *Tetranychus urticae* Koch (Acar: Tetranychidae), one of their competitors. Here, we present evidence that western flower thrips larvae prey on *Trialeurodes vaporariorum* Westwood (Hemiptera: Aleyrodidae), another competitor for plant tissue. We tested this on two host plant species, cucumber (*Cucumis sativa* L.), considered a host plant of high quality for western flower thrips, and sweet pepper (*Capsicum annuum* L.), a relatively poor quality host. We found that WFT killed and fed especially on whitefly crawlers and that the incidence of feeding did not depend on host-plant species. The developmental rate and oviposition rate of western flower thrips was higher on a diet of cucumber leaves with whitefly crawlers than on cucumber leaves without whitefly crawlers, suggesting that thrips do not just kill whiteflies to reduce competition, but utilize whitefly crawlers as food.

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Omnivory, the phenomenon of species consuming resources at different trophic levels (Pimm and Lawton 1978), is widespread in natural and managed communities (Rosenheim et al. 1995, Agrawal et al. 1999). A special case of omnivory is intraguild predation, i.e. the killing and eating of species that use similar, often limiting, resources and are thus potential competitors. In such systems, one species is an omnivore (the intraguild predator), a second species is its prey (the intraguild prey) and they share a third species as their food (Polis et al. 1989, Coll and Guershon 2002, Janssen et al. 2007, Lucas et al. 2009). Intraguild predation can occur between predators that share a prey or between an omnivore and herbivore that share a host plant (Arim and Marquet 2004). In the context of biological control, the consequences of intraguild predation among natural enemies that share the same pest as prey have been studied frequently (Rosenheim et al. 1995, Harmon and Andow 2004, Janssen...
et al. 2006, Rosenheim and Harmon 2006, Bampfylde and Lewis 2007) and generally predict no synergism in reducing the pest (Janssen et al. 2006, Rosenheim and Harmon 2006). However, little is known of intraguild predation between different herbivores (see for example Trichilo and Leigh 1986, Wilson et al. 1996), let alone its consequences for biological control.

One of the examples of an intraguild predator is the western flower thrips (WFT) *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae), which mainly feeds on leaf parenchyma and plant pollen, but also on eggs of its natural enemies, the predatory mites *Iphiseius degenerans* Berlese (Acari: Phytoseiidae) and *Amblyseius cucumeris* Oudemans (Acari: Phytoseiidae) (Faraji et al. 2001). Western flower thrips also feed on the eggs of another predatory mite, *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) (Faraji et al. 2002, Janssen et al. 2002), which is not an important enemy of WFT, as well as on the eggs of another herbivore, the two-spotted spider mite *Tetranychus urticae* Koch (Acari: Tetranychidae) (Trichilo and Leigh 1986, Pallini et al. 1998, Agrawal and Klein 2000). Because spider mites and WFT both feed on cucumber plants (*Cucumis sativa* L.) and WFT also feed on spider mites, these thrips are omnivores and intraguild predators (Janssen et al. 1998). Western flower thrips is a major pest species of various crops such as cucumber, sweet pepper and eggplant in Northern Europe and North America (Byrne et al. 1990, Lewis 1997). However, thrips can be potential biological control agents of spider mites on cotton, *Gossypium hirsutum* L. in California and Australia (Trichilo and Leigh 1986, Wilson et al. 1996).

In this study, we investigate whether WFT are potential intraguild predators of yet another plant pest, the greenhouse whitefly *Trialeurodes vaporariorum* (Westwood), which frequently infests the same greenhouse crops as WFT (Byrne et al. 1990). We investigated predation on two stages of whiteflies that are likely to be most vulnerable on a priori grounds (small size, lack of defence and immobility) and preliminary observations (G Broufas and M van Maanen, unpubl.). We furthermore measured developmental rate and oviposition of WFT on cucumber plants with and without whiteflies as an additional food source.

Diet choice often depends on food quality (Agrawal et al. 1999, Eubanks and Denno 1999, Agrawal and Klein 2000, Janssen et al. 2003). For example, omnivores may consume more herbivores if the host plant is of poor quality (Agrawal et al. 1999, Eubanks and Denno 1999, Agrawal and Klein 2000, Magalhães et al. 2005b). We therefore studied the consumption of whitefly instars on two host plants that differ in quality to the WFT in terms of developmental and reproductive success. One host plant, cucumber (*C. sativa*) is a good quality host plant for WFT, whereas sweet pepper (*Capsicum annuum* L.) is a relatively poor quality host (Janssen et al. 2003, Magalhães et al. 2005a), which is nevertheless colonized by WFT (Funderburk et al. 2006, Rosenheim and Harmon 2006, Bampfylde and Lewis 2007) and generally predict no synergism in reducing the pest (Janssen et al. 2006, Rosenheim and Harmon 2006). However, little is known of intraguild predation between different herbivores (see for example Trichilo and Leigh 1986, Wilson et al. 1996), let alone its consequences for biological control.

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We hypothesized that WFT consume more whitefly individuals on a poor quality host plant than on a high quality host plant.

Materials and Methods
Cucumber plants (cv. Aviance RZ) were grown from seeds in plastic pots (2 l) with soil (Jongkind b.v. Aalsmeer, Hol03/No.3). Potted sweet pepper plants (cv. Plukpaprika) were bought at a local garden centre. All plants were kept in a walk-in climate room (25 °C and 65% humidity, 16 h daylight), free of herbivores. Western flower thrips were reared on cucumber leaves, placed on wet cotton wool in a Petri dish (14 cm diameter), and were provided cattail pollen (*Typha latifolia* L.) twice a week. In order to obtain cohorts of WFT of the same age, females were allowed to oviposit on cucumber leaves with cattail pollen during 24 h. Whiteflies on tobacco leaves were obtained from Koppert BV (Berkel en Rodenrijs, The Netherlands) and used directly for the experiments. All insects were kept in separate walk-in climate rooms (25 °C and 65% humidity).

Intraguild predation
Predation on whitefly eggs and crawlers was measured by keeping thrips larvae (6-seven days old since egg hatching) individually on a leaf disc of sweet pepper or cucumber (16 mm diameter) for 24 h with either 9-50 whitefly eggs, or 15 whitefly crawlers. Whitefly eggs were obtained by confining 20-25 adult whiteflies in a clip cage (2.5 cm diameter) on the underside of a cucumber or pepper leaf. One day later, the leaf area under the clip cage was punched out, and the whitefly eggs were counted. Whitefly crawlers were collected from tobacco leaves with a fine brush and placed on clean leaf discs of pepper or cucumber. After 16 h, predation per individual thrips larva was recorded as the number of whitefly crawlers or whitefly eggs consumed (as judged by the presence of prey remains). In total there were four treatments and 19-33 individual WFT larvae (replicates) were tested per treatment. Leaf discs were taken from two to four plants per day, and were randomly distributed over treatments. In a control treatment without thrips larvae, we incubated 15 whitefly crawlers or 9-50 whitefly eggs per leaf disc for 16 h to assess their natural mortality rate.

Predation on whitefly crawlers and eggs was compared among plant species with a generalized linear model (GLM) with a quasi-binomial error distribution. The proportion of thrips larvae feeding on eggs or crawlers was compared between plant species with a GLM with a quasi-Poisson error distribution. All statistical analyses were done using R (R Development Core Team 2010).
Development
Development and survival of thrips larvae were measured on leaf discs (24 mm diameter) of cucumber that were placed in a Petri dish (18 cm diameter) on wet cotton wool (four discs per Petri dish). Cucumber leaf discs, taken randomly from two to four plants, were either without additional food, supplied with 15 whiteflies crawlers, or supplied with ample Typha pollen, which is a good food source for thrips larvae (Janssen et al. 2003). One newborn thrips larva was added to each leaf disc with a fine brush. Food was added every day, and all thrips larvae were transferred to new leaf discs with the same diet every two days, until the thrips reached adulthood. Hence, there was always a surplus of food available. The transition from one stage to another was determined from the occurrence of a moulting skin on the leaf disc. Developmental time and mortality were monitored daily with a stereoscopic microscope. In total, 25-27 individual thrips larvae (replicates) were tested per treatment. Nonparametric survival analysis (the log-rank test, Hosmer and Lemeshow 1999) was used to test for differences in development and survival among days and differences between the numbers of whitefly crawlers eaten on cucumber leaf discs compared to those eaten on pepper leaf discs.

Reproduction
To measure reproduction, newly emerged adult WFT females were individually placed on a cucumber leaf disc (24 mm diameter) in a Petri dish (18 cm diameter) on wet cotton wool. Cohorts of newly emerged adult females were obtained from the WFT culture. Leaf discs were either without additional food, supplied with 20 whitefly crawlers, or with ample Typha pollen. Females were transferred to new leaf discs with the same food each day during a period of four days. After 24 h, the number of whitefly crawlers consumed per individual thrips adult was recorded and the adult female WFT was removed. Subsequently, the Petri dishes with leaf discs were incubated in a climate room until the WFT larvae emerged from the eggs. The number of first instars hatching on each disc was used as an estimate of the reproductive rate. In total, 20-22 individual adult female thrips (replicates) were tested per treatment. Because oviposition rates are affected by the recent dietary history of the adult thrips, we discarded data from the first day to reduce effects of food sources consumed before the experiment. To correct for repeated measures, reproduction was compared among treatments with a linear mixed effects model with a quasi-Poisson error distribution.

Results
Intraguild predation
Western flower thrips larvae were observed feeding on whitefly crawlers (Figure 5.1), leaving only their shrivelled exoskeletons behind. They consumed per capita on aver-
age seven whitefly crawlers on cucumber or sweet pepper leaf discs during the first 16 h (Figure 5.2). There was no difference in consumption of whitefly crawlers between host plants (GLM: $F_{1,50} = 0.22$, $P = 0.64$). Western flower thrips larvae killed low numbers of whitefly eggs and the difference in predation rates on the two host plants was not significant (GLM: $F_{1,45} = 0.29$, $P = 0.59$). No mortality of whiteflies was recorded in the control treatments. The fractions of WFT larvae preying on whitefly crawlers or eggs did not differ between the two host plants ($F_{1,2} = 0.302$, $P = 0.64$ and $F_{1,2} = 0.004$, $P = 0.96$, respectively) (Table 5.1).

<table>
<thead>
<tr>
<th>Host plant</th>
<th>Whitefly stage</th>
<th>Ratio feeding thrips$^1$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cucumber</td>
<td>Eggs</td>
<td>4/23</td>
</tr>
<tr>
<td>Sweet pepper</td>
<td></td>
<td>10/24</td>
</tr>
<tr>
<td>Cucumber</td>
<td>Crawlers</td>
<td>15/19</td>
</tr>
<tr>
<td>Sweet pepper</td>
<td></td>
<td>29/33</td>
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</tbody>
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$^1$The number of thrips larvae that preyed on whitefly eggs or crawlers/total number of thrips larvae tested.

Figure 5.1 Thrips larvae feeding on whitefly crawlers.
CHAPTER 5 | THRIPS LARVAE FEED ON WHITEFLY CRAWLERS

Figure 5.2 Predation of whitefly eggs or crawlers by second-instar larvae of *F. occidentalis*. Shown are mean numbers (± SE) of whitefly eggs and crawlers eaten per thrips larva per 16 h on sweet pepper and cucumber leaf discs. The consumption of whiteflies crawlers or eggs did not differ with host plant.

Figure 5.3 Development and survival of juvenile *F. occidentalis* on a diet of cucumber leaf tissue plus pollen (Pollen: closed circles), cucumber leaf tissue plus whitefly crawlers (Whitefly: open circles) or cucumber leaf tissue without other food added (Leaf: squares). Shown are cumulative proportions (± SE) of juveniles that developed into adults. The final proportion equals the survival of the thrips larvae. Different letters indicate significant differences among treatments.
Development
The development of WFT juveniles was significantly affected by diet (survival analysis: $\chi^2_2 = 41.1$, $P<0.0001$). WFT larvae preying on whitefly crawlers developed slower than WFT feeding on pollen but faster than WFT feeding on cucumber leaf tissue only (Figure 5.3, all $P$'s<0.0001). This suggests an advantage for thrips to feed on pollen or on whiteflies as an additional food source to leaf tissue. Juvenile survival was not affected by diet ($\chi^2_2 = 0.4$, $P = 0.80$); all thrips survived in all three treatments.

Reproduction
Overall, reproduction of WFT differed significantly among the three treatments (Figure 5.4, all $P$'s<0.0001). However, there was a significant interaction between time (days) and diet ($\chi^2_4 = 34.4$, $P<0.0001$), indicating that there were differences in reproduction on different diets through time (Figure 5.4). This is caused by the reproduction on a diet of cucumber leaf tissue and pollen being constant over the four day period, whereas reproduction on the two other diets decreased on the 4th day (Figure 5.4). Adult thrips consumed per capita on average two (SE = 0.4) whitefly crawlers on cucumber leaf discs during the first 24 h.

Figure 5.4 Reproduction of thrips adults on different diets. Shown are the average (± SE) numbers of newborn larvae per adult thrips on a diet of cucumber leaf and pollen (Pollen: closed circles), cucumber leaf and whitefly crawlers (Whitefly: open circles) or cucumber leaf (Leaf: squares) per day. Different letters indicate significant differences among treatments.
Discussion

We show that western flower thrips larvae and adults prey on crawlers of greenhouse whiteflies. The predation rate by WFT larvae on whitefly eggs was close to zero. Perhaps the egg shells are difficult to pierce by thrips (G Broufas and R van Maanen, pers. obs.).

In contrast to earlier research showing that thrips larvae fed more on non-plant food when their host plants were of low quality (Agrawal et al. 1999, Agrawal and Klein 2000, Janssen et al. 2003, Magalhães et al. 2005a), we found no significant effects of host plant quality on predation rates of whiteflies by thrips larvae. Moreover, the proportion of WFT larvae killing whiteflies did not differ between cucumber and sweet pepper leaf discs. We suggest three explanations. First, thrips may prefer whitefly crawlers to plant material on both plant species because of their higher nutritional value. The latter is suggested by the oviposition rate and developmental rate being higher on cucumber leaves provided with whitefly crawlers. Hence, even on this high-quality host plant, it might be advantageous for the thrips larvae to feed on whitefly crawlers. It should be noted that we did not control for such effects on the nutritional quality of the plant. Second, thrips may consider whiteflies as competitors and therefore kill them regardless of the quality of the host plant. Thrips larvae are known to kill eggs of their predators not because it offers food, but because it reduces predation (Janssen et al. 2002, 2003). It is therefore conceivable that they kill crawlers of whiteflies because it reduces competition. The third explanation is that we used another cucumber variety, which may not be of the same high quality as that used by Janssen et al. (2003).

Thus, western flower thrips is an omnivore as well as an intraguild predator, competing with and feeding on spider mites and whiteflies. Given the intraguild predation of western flower thrips on greenhouse whiteflies, one may wonder how this will affect the dynamics of the whiteflies and thrips and ultimately the overall damage of the host plant. Theory on intraguild predation at equilibrium conditions predicts that intraguild predators will always exclude intraguild prey in environments with high productivity (Polis et al. 1989). Productivity is likely to be high in cropping systems, but the period of cropping is so short that equilibria may not be reached (Briggs and Borer 2005), i.e. intraguild prey may not go extinct in this period. Little is known about the effects of intraguild predation between herbivores on biological control. Intraguild predation may cause the intraguild prey (whiteflies in our case) to avoid co-occurrence with the intraguild predator (thrips in our case) in space, as was found for other species (Magalhães et al. 2005b). However, other interactions between the herbivores may also affect their population dynamics, such as the induction of plant resistance by either of the two species (Zarate et al. 2007, Zhang et al. 2009) or other changes in plant quality caused by herbivory. For example, the phytophagous mite *Tetranychus evansi* Baker & Pritchard (Acari: Tetranychidae) suppresses the main...
pathways involved in induced plant defenses in tomato (Sarmento et al. 2011a), resulting in increased host plant quality for the closely related spider mite *T. urticae* (Sarmento et al. 2011b). Likewise, it has been shown that feeding stages of the silverleaf whitefly *Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) suppress induced plant resistance (Zarate et al. 2007), which could improve the host plant quality for WFT. The same whitefly species was found to interfere with induced plant defences against spider mites in Lima bean (Zhang et al. 2009). Further research should clarify the effects of intraguild predation and interactions through induced plant defences on the dynamics of WFT and greenhouse whiteflies.

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**References**


CHAPTER 5 | THRIPS LARVAE FEED ON WHITEFLY CRAWLERS


Predators disguised with chemical cues from one prey species have improved attack success on another prey

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In the presence of predators, prey alter their behaviour in order to reduce the risk of being eaten. To tune this behaviour to the current danger, prey need to assess the risks based on the available information. Chemical cues associated with predators may be helpful indicators. Many prey species can distinguish chemical cues from predators that fed on conspecific prey from those that fed on heterospecific prey, and react stronger to the first. We therefore hypothesize that generalist predators have an increased chance to capture a given prey species when they contaminated with chemical cues from another prey species. We tested this hypothesis using a generalist predatory mite that feeds on thrips larvae as well as on whitefly eggs and crawlers. Predatory mites were marked with cues (i.e. body fluids) of one of these two prey species and were subsequently offered the same or the other prey species. We found that predators marked with thrips cues killed significantly fewer thrips than predators marked with whitefly cues, even though the predator’s tendency to attack was the same. In addition, we show that more thrips seek refuge in the presence of a predatory mite marked with thrips cues than when marked with whitefly cues or other cues serving as controls. Thus, generalist predators may experience improved attack success in mixtures of prey species.

Submitted

Many prey species are able to recognize chemical cues associated with the presence of predators (e.g. reviews by Chivers and Smith 1998, Kats and Dill 1998, Dicke and Grostal 2001). These cues usually induce anti-predator behaviour in the prey, such as fleeing, hiding, remaining motionless or aggregating (Lima and Dill 1990). Because not all predators are equally dangerous and do not forage in the same way, this behaviour has to be tuned to the current danger. Therefore, prey need to discriminate between various predator species and assess the risks based on the available information (Sih 1987, Lima and Dill 1990, Chivers and Smith 1998, Venzon et al. 2000). Specialist predators that attack only a particular prey should be avoided by this prey. However, prey may perceive generalist predators as harmless when the predators have switched from feeding on conspecific prey to another prey species (Venzon et al. 2000). For example, in many aquatic systems, prey can distinguish chemical cues from predators that fed on conspecific prey from those of predators.
that fed on heterospecific prey, and react stronger to the first (Wilson and Lefcort 1993, Chivers et al. 1996, Laurila et al. 1997). In terrestrial systems there are few examples of prey responding more strongly to volatiles associated with predators that fed on conspecific prey than to volatiles from predators that have fed on other prey (Venzon et al. 2000, Persons et al. 2001, Meng et al. 2006). Interestingly, until now only three papers highlighted the idea that if diet-related chemical cues enable prey to discriminate between harmless and dangerous predators, predators might be able to ‘chemically disguise’ themselves by eating different prey species alternatingly (Venzon et al. 2000, Lima et al. 2003, Stabell et al. 2003), thereby reducing antipredator behaviour and increasing predation rates. To the best of our knowledge, our study is the first to experimentally assess whether generalist predators have increased success in seizing a prey when they are marked with cues of a prey distantly related to the target prey.

We used a system consisting of the generalist predatory mite *Amblyseius swirskii* Athias-Henriot (Acari: Phytoseiidae) and two of its prey species; the western flower thrips *Frankliniella occidentalis* (Pergande) and the greenhouse whitefly *Trialeurodes vaporariorum* (Westwood) (Nomikou et al. 2001, Messelink et al. 2008). In earlier experiments, we found that juvenile survival and the developmental rate of *A. swirskii* was highest on a mixed diet of thrips and whiteflies, intermediate on a diet of thrips larvae, and lowest on a diet of whitefly eggs (Messelink et al. 2008). Therefore, we consider thrips as a prey of higher quality than whiteflies.

Juvenile thrips are able to defend themselves in various ways, for example, by counter-attacking vulnerable stages of the predator (Janssen et al. 2002), and by swinging their abdomen at the attacking predator (Bakker and Sabelis 1987, 1989), which is sometimes accompanied by the secretion of an anal droplet. These droplets contain a mixture of decyl acetate and dodecyl acetate, which acts as an alarm pheromone for conspecific thrips (Teerling et al. 1993a,b, de Bruijn et al. 2006). When predators are contaminated with such droplets, they often cease attack and start cleaning themselves (Bakker and Sabelis 1987, 1989). Thrips can also seek refuge in the web produced by spider mites (*Tetranychus urticae* Koch) (Pallini et al. 1998, Venzon et al. 2000), another herbivore species, often found on the same plants as thrips. Thrips larvae prefer clean plant parts without web, but move into the web upon perceiving volatile cues associated with predators (Pallini et al. 1998). This behaviour leads to a reduction of predation risk, but also to a reduced juvenile developmental rate due to competition with spider mites (Pallini et al. 1998). Juvenile whitefly stages are not able to actively defend themselves.

We hypothesize that predatory mites that are marked with cues of thrips larvae have decreased predation success on thrips larvae compared to predators marked with cues from whitefly crawlers. Decreased predation success might be due to changes
in the foraging behaviour of the predator or changes in anti-predator behaviour of the prey. We performed a second experiment in which we quantified one type of antipredator behaviour of thrips larvae, i.e. the refuge seeking in the web of spider mites, as this is easier to interpret than abdominal swings with the associated production of anal droplets, which do not only serve as defence, but also for communication with other thrips larvae (PJA de Bruijn, pers. comm.). We expected more thrips to hide in the refuge in the presence of a mite marked with cues of thrips compared to the presence of a mite marked with cues of whitefly. We quantified attacks on whitefly eggs by predators with a mark of thrips or without such a mark as a control for changes in the foraging behaviour of the predators due to the mark. As these eggs cannot show antipredator behaviour, we hypothesize that changes in predation rate emerge from physical effects of the mark on predator behaviour.

Materials and Methods
Cucumber plants (Cucumis sativa L. var. Aviance Z, Rijk Zwaan, De Lier, The Netherlands) were grown from seeds in herbivore-free, walk-in climate rooms (25 °C, 60% RH, 16:8 light:dark). Western flower thrips and spider mites were both collected from cucumber plants in a commercial greenhouse in May 1994. The thrips stock was mixed with individuals that originated from chrysanthemum in experimental greenhouses of Wageningen UR Greenhouse Horticulture, Naaldwijk, The Netherlands, three weeks before the experiments started. Spider mites, thrips and whiteflies were reared on cucumber plants (Aviance) in separate climate rooms (25 °C, 60% RH, 16:8 light:dark). Whitefly crawlers were supplied by Koppert BV (Berkel en Rodenrijs, The Netherlands) on tobacco leaves. Predatory mites were reared on plastic arenas (8 × 15 cm), placed on wet sponges in a plastic tray containing water (Nomikou et al. 2003). They were kept in a walk-in climate room (25 °C, 60% RH, 16:8 light:dark) and were supplied with ample cattail pollen (Typha latifolia) twice a week. Small pieces of cotton wool were provided as oviposition substrates. Twice a week, cotton wool pieces with predatory mite eggs were transferred to a clean plastic arena. This ensured a steady supply of adult mites that were of the same age and of the same satiation level. To obtain deutonymph predators, larvae were transferred each to an individual leaf disc. Deutonymph predators were used because they experience more hinder from antipredator behaviour by the thrips larvae. Prior to measuring predation on thrips, deutonymph predatory mites (as judged by the presence of two exoskeletons on each individual leaf disc) were labelled with cues of whitefly crawlers or cues of thrips larvae. Labelling was done as follows: a whitefly crawler or thrips larva was killed with the aid of a needle and directly afterwards, we touched the dorsum of the predatory mite with this needle. Because the needle was sticky due to the body flu-
ids of the prey, the mite stuck to the needle and could be lifted from its leaf disc. Upon touching the experimental leaf disc, the mite pulled itself off the needle by grabbing the leaf. Sometimes thrips larvae produced an anal droplet just before being killed; therefore, the mark would have contained alarm pheromones in some cases. As there are no sensory structures on the dorsum, and predators cannot reach the dorsum with their legs to clean, predators do not try to remove the mark. Labelling resulted in an invisible (40× magnification) mark, which was too small to measure with a Sartorius supermicrobalance (d = 0.0001 mg).

Marked mites were introduced as described above, each on a separate cucumber leaf disc (24 mm diameter) with one late first instar thrips larva (5 days old since egg stage). After 16 and 72 h, we assessed whether the thrips larva was eaten or not, judged by the presence of remains. We tested respectively 48 and 49 predatory mites in the two treatments on five different days. Replicates where the thrips larva was lost, drowned, or dead but not preyed upon, were excluded from the analyses. Kaplan Meier survival analysis was applied to compare the survivorship curves of the different treatments, using a log-rank test for differences between replicates and differences among treatments (Hosmer and Lemeshow 1999).

To quantify refuge seeking as a type of antipredator behaviour of thrips larvae, we performed a second experiment, but now we added spider-mite web as a refuge for the thrips (Pallini et al. 1998, Venzon et al. 2000). Cucumber leaf discs (24 mm) were punched out in such a way that the main vein divided the discs in two halves. The leaf discs were placed on wet cotton wool in a Petri dish. A wet thread of cotton wool was placed on top of the vein (Pallini et al. 1998). Thirty adult spider mites were obtained from the rearing and added to one half of the leaf disc, where they were allowed to feed and produce web. The spider mites did not cross the wet cotton wool, thus, half of each leaf disc became damaged and covered with web by the spider mites. After two days, the spider mites and the cotton wool were removed carefully using a small needle, leaving the disc with spider-mite eggs, faeces and a thin layer of web on the leaf surface. In this experiment we used adult female predatory mites because their predation rate is three times higher than that of deutonymphs (personal observation, van Maanen 2009). We tested 12-31 mites per treatment on six different days. All mites tested were seven days old since egg hatching and of the same satiation level. Marked mites were introduced as described above, each on a separate cucumber leaf disc with one half covered with spider mite web and one late first instar thrips on the other half of the leaf disc. In the control, no predator was added to the leaf disc to verify whether refuge seeking was induced by predator cues or not.

The position and predation of the thrips larvae were scored after 24 h. The differences in numbers of thrips eaten among the two treatments were analyzed with Kaplan-Meier survival analysis as above. The proportion of all thrips (eaten plus alive)
found in the refuge was analyzed with a generalized linear model with binomially distributed errors (R Development Core Team 2009).

Differences in predation rates could be caused by the predators being hindered in some way by the mark, especially the acids from the anal droplet present in the mark from thrips larvae may hinder predatory mites (de Bruijn et al. 2006). Therefore, we performed a control experiment with immobile prey, which could not respond to the cues present in the mark on the predator’s dorsum, thus restricting the effect of the mark on the foraging behaviour of the predator only. Adult female whiteflies were confined overnight in clip cages on cucumber leaves. Subsequently, leaf discs were punched from the area that had been enclosed inside the clip cage, resulting in leaf discs containing between 20 to 70 whitefly eggs. Earlier experiments showed that adult A. swirskii consume c. 12 whitefly eggs per 24 h (Messelink et al. 2008), thus, ample prey was provided. The leaf discs were placed on wet cotton wool inside Petri dishes. Each Petri dish contained four leaf discs. In one treatment, we marked predatory mites with thrips cues as above, and in the other treatment, we marked them with water as a control for the handling the predator. We tested 36 predators per treatment. All predators tested were seven days old since the egg stage and of the same satiation level. After 24 h, the remaining whitefly eggs were counted. Leaf discs with whitefly eggs but without predatory mite served as control for mortality of whitefly eggs for reasons other than predation. Mean predation was compared between the two treatments and analyzed using a generalized linear model with quasi-binomially distributed errors (R Development Core Team 2009).

Results
Predation on thrips larvae by juvenile predatory mites in the absence of a refuge was significantly affected by the prey cues on the dorsum of the mites (Figure 6.1: survival analysis, $\chi^2 = 4$, df = 1, $P = 0.046$). There were no differences among replicates carried out on different days ($\chi^2 = 6.6$, df = 4, $P = 0.16$). Mites marked with whitefly cues preyed c. twice as many thrips larvae within 24 h than mites marked with thrips cues (Figure 6.1). Predatory mites did not spend time attempting to clean themselves when prey cues were applied on the dorsum.

In the experiment where a refuge for the prey was provided, similar results were found. The average proportion of thrips eaten by mites marked with cues of thrips was 0.36 (SE = 0.08) in 24 h and 0.63 (SE = 0.08) by mites marked with cues of whiteflies and this difference was significant ($\chi^2 = 3.9$, df = 1, $P = 0.049$).

Overall, there was a significant effect of treatment on the proportion of thrips in the refuge (Figure 6.2: GLM with binomial distributed errors, $\chi^2 = 7.6$, df = 2.65, $P = 0.022$). There was a significant difference between number of thrips hiding in the web in the presence of mites marked with cues of thrips or in absence of mites ($\chi^2 = 6.4$,
Chapter 6 | Disguised predators perform better

Figure 6.1 The mean proportion (± SE) of thrips killed through time by deutonymph predators either marked with cues from thrips (diamonds) or from whiteflies (squares).

Figure 6.2 The mean proportion (± SE) of thrips larvae found in the spider-mite web (refuge) after 24 h in the presence of predatory mites that were either marked with cues from thrips or from whiteflies and in the absence of predatory mites (Control). Different letters indicate significant differences among treatments.
df = 1.65, P = 0.011). Twice as many thrips moved into the web in presence of mites marked with cues of thrips than in presence of mites marked with whiteflies ($\chi^2 = 3.8$, df = 1.65, P = 0.052). A smaller effect was found between the treatment without predators and predators marked with whiteflies ($\chi^2 = 1.5$, df = 1.65, P = 0.23). This suggests that thrips responded more strongly to cues associated with predators that fed on conspecific prey than to cues from predators that had fed on other prey.

The average number of whitefly eggs eaten by mites marked with thrips was 7.8 (SE = 0.94) in 24 h and 9.6 (SE = 0.78) by mites marked with water this did not differ significantly (GLM with quasi-binomial distributed errors, P>0.15). This suggests that the mark did not hinder the mites in their predation behaviour. No mortality of whitefly eggs was observed in the control without predators.

Discussion
Juvenile and adult predatory mites marked with whitefly cues killed significantly more thrips than predators marked with thrips cues (Figure 6.1). In addition, we show that more thrips seek refuge in the presence of a predatory mite marked with thrips cues than when marked with whitefly cues (Figure 6.2). The predatory mites under test were shown not to be hindered by the mark and were all of similar age and well-fed. We conclude that the lower predation by predators marked with thrips was due to a response of the thrips larvae, not to differences in the motivation to attack thrips among predators with different treatments. Thrips larvae that were exposed to predators marked with thrips succeeded twice as often in escaping from predators by hiding in a refuge than thrips larvae exposed to a predator marked with cues of whitefly. These results suggest that predators can feed on inferior prey in order to chemically disguise themselves, and thereby reduce antipredator behaviour of the superior prey.

Under natural conditions, predators with different previous diets will usually have different satiation levels, which can result in differences in their motivation to attack prey, as well as in different prey preference (Sabelis 1986, 1990). We mimicked a previous diet by marking the predators with prey cues. Mites were all reared on the same previous diet of pollen, so that differences in motivation and preference were avoided. Research on other systems has shown that cues emitted by damaged or crushed prey evoked less strong anti-predator behaviour than cues emanating from predators that consumed prey (Schoepfner and Relyea 2009). This suggests that the differences in predation rate may be larger when the predators would have been allowed to become marked in the process of feeding on different prey species.

In our experimental system, the marks of thrips and whiteflies may not only have differed in composition but also in quantity. However, we could not quantify marks since they were not visible with a 40× magnifying binocular and the mass of the mark could not be detected with a sensitive microbalance.
Thrips could also have expressed other types of anti-predator behaviour, such as swinging their abdomen (Bakker and Sabelis 1987, 1989, Teerling et al. 1993a, b) and producing acid droplets. We did not investigate whether this type of anti-predator behaviour contributed to escaping predation. Furthermore, when thrips larvae were killed with a needle to mark the predators, thrips might have produced an anal droplet containing alarm pheromone. Therefore, predators marked with thrips might also have been contaminated with the thrips alarm pheromone. De Bruijn and co-authors (2003) showed that alarm pheromone of thrips increases the vigilance of conspecific thrips. It would be interesting to assess whether such increased vigilance promotes survival of the thrips.

Lima and co-authors (2003) incorporated anti-predator vigilance into a classical model of diet choice. Such classical models (Charnov 1976, Stephens and Krebs 1986) consider the dietary choice of a predator when it encounters prey of high and low quality on the same patch, and predict that a predator should always accept the superior prey type, but should accept inferior prey as well when encounter rates with the superior prey fall below a certain threshold. Lima and co-authors (2003) assume that the vigilance of prey increases with the number of conspecific prey that are attacked by the predator, and predict that antipredator behaviour of the prey causes predators to select a more generalized diet than one might predict from classical theory on optimal prey choice. For one thing, this is caused by the predators experiencing a lower attack rate on the superior prey because of the increased vigilance, but for another, the predators can manipulate the antipredator behaviour of the superior prey by feeding on the inferior prey. These authors suggest that predators will often benefit from ‘managing’ the anti-predator responses of the prey. This scenario closely resembles our idea that predators may feed on inferior prey in order to chemically disguise themselves, thus preventing strong antipredator behaviour in the superior prey. However, if generalist predators feed on heterospecific prey species in order to disguise themselves, we expect selection to act on prey individuals to use other more permanent chemical cues of predators or use the cues from heterospecific prey as indicators of threat as well. In aquatic systems, it was found that, although prey often exhibit the strongest anti-predator behaviour when the predator consumes only conspecific prey (Wilson and Lefcort 1993, Schoeppper and Relyea 2005), prey indeed also continued to respond to predators that had consumed heterospecific prey (Smith 1992, Chivers and Mirza 2001).

In this article, we show that a generalist predator had a decreased predation rate on prey when they were experimentally marked with body fluids of heterospecics of this prey compared to predators marked with a species conspecific to the current target prey. This supports the hypothesis that generalist predators reach higher predation rates on mixtures of prey species. Perhaps generalist predators might have an
as yet overlooked advantage of eating different prey species, because their chemical disguise does not alert defensive prey. They could achieve this passively, by attacking prey as they are encountered, or actively, by switching between prey species. Classical foraging theory predicts that generalist predators should switch from eating only prey of high quality to eating a mixture of prey when the density of the high-quality prey falls below a threshold (Charnov 1976, Stephens and Krebs 1986). We suggest that predator switching might also have evolved because predators face reduced antipredator behaviour, thereby promoting the success rate of predation on the preferred prey.

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The aim of this thesis was to study indirect interactions among species in a relatively simple food web and how these interactions can be modified by behaviour of individuals. The insights obtained in this way may serve to improve biological control of greenhouse pests. I investigated direct interactions between herbivores and indirect interactions via a shared predator to gain insight into dynamics of the prey species. In particular, I investigated intraguild predation between herbivores and apparent competition (including apparent mutualism) between herbivores via their predator. I also studied the effects of behaviour of predator and prey on these interactions. I start with a summary of the results of each chapter separately, and end with a discussion to integrate the results of the thesis as a whole.

In Chapter 2, I studied predator-mediated interactions between the two prey species. In such a one predator - two prey system, indirect interactions can occur between the two pest species, such as apparent competition and apparent mutualism. The question was, whether the dynamics of this system is characterized by apparent competition or apparent mutualism. Whereas apparent competition is desired for biological control because it brings pest levels down, apparent mutualism is not, because it does the opposite. I evaluated the control efficacy of the predatory mites *A. swirskii* or *E. ovalis* in cucumber crops in greenhouse compartments with only thrips, only whiteflies or both herbivorous insects together. Each predator controlled thrips but *A. swirskii* reduced thrips densities the most. There was no effect of the presence of whiteflies on thrips densities. Whitefly control in the absence of thrips was not sufficient. However, whitefly densities were reduced dramatically in the presence of thrips, especially with the predator *A. swirskii*. Hence, the dynamics of the system resembles that of non-reciprocal apparent competition (Chaneton and Bonsall 2000). The densities of predators were up to 15 times higher in the presence of both pests than in the single-pest treatments. Densities of whiteflies in the treatment with predators and whiteflies alone were always high, hence, food was never limiting. I therefore suggest that the high predator densities observed in the presence of both prey were not only caused by higher food availability. Laboratory experiments with *A. swirskii* suggest that the higher predatory densities were due to a higher juvenile survival and developmental rate on a mixed diet. There was no juvenile mortali-
ty on a diet consisting of only thrips, whereas 34% of the juvenile predators that were feeding on whitefly eggs died and the developmental rate of predators was significantly higher on a diet of only thrips compared to only whiteflies. Hence, thrips are a superior food source for *A. swirskii* compared to whiteflies, but a mixture of the two is even better.

Overall there is very little knowledge on the effects of a mixed diet of two prey species on the dynamics of predator and prey populations. However, it is easy to see from simple models on apparent competition what would be the effect of such a mixed diet on population dynamics (Holt 1977). Equilibrium prey densities are inversely related to the growth rate of the shared predator (Holt 1977), hence, if the predators’ growth rate increases because of a mixed diet, prey equilibria go down. I suggest that the better biological control of whiteflies may have been achieved not only because of apparent competition, but also through a positive effect of mixed diets on predator population growth. I found no evidence for apparent mutualism, but this may possibly have occurred in the short term, for which no data were collected.

In Chapter 3, the occurrence of short-term apparent mutualism was further investigated. Many studies demonstrated effects of apparent competition, but studies showing longer-term apparent mutualism are scarce (Chaneton and Bonsall 2000). I present evidence for a short-term escape (i.e. apparent mutualism) of one pest species from control by the predator *A. swirskii* when a second prey species is present. Initially, I found significantly higher numbers of thrips larvae on cucumber plants in greenhouse compartments where both pests were present than when only thrips were present. More thrips larvae escaped predation on plants most distant from the release plants because the predators dispersed slower in compartments with two pests. After six weeks, this effect was overruled by a strong decrease in thrips densities through a higher number of predators in greenhouses with two prey species. Successful biological control of thrips was achieved in the end, despite temporarily higher thrips numbers. This shows that apparent mutualism is overruled by apparent competition within a few generations. This is likely to have been enhanced by the positive effects of a mixed diet on predator population growth.

In Chapter 4, I again studied the dynamics of pest species on greenhouse cucumber in combination with the predator species *A. swirskii*. Besides Western flower thrips and greenhouse whitefly, the two-spotted spider mites, *Tetranychus urticae* Koch was included in the investigation. The question was whether prey species diversity reduces populations of non-prey species because of the numerical response of generalist predators on populations of other prey species. This is especially interesting in the light of mixed-diet effects on predator population densities. The high predator densities resulting from a mixed diet may significantly affect prey populations on which the predators have small *per capita* effects. When plants were infested by spi-
der mites prior to predator release, predatory mites were not capable of controlling
spider mite populations in the absence of other pest species. A laboratory experiment
showed that the webbing of spider mites hindered the predators. In a greenhouse
experiment, spider mite leaf damage was lower in the presence of thrips and preda-
tors than in the presence of whiteflies and predators, but damage was lowest in the
presence of thrips, whiteflies and predators. The lower levels of spider mite damage
probably resulted from a strong numerical response of the predator (up to 50 times
higher densities) when a second and third pest species were present in addition to
spider mites. This shows that apparent competition effects can also affect species
that are not considered suitable for the predator. It also shows that diversity of pest
species can enhance biological control through increased predator densities.
In Chapter 5, I investigated the direct interaction between thrips and whiteflies. Larvae
of the Western flower thrips were indeed found to prey on eggs and larvae of green-
house whiteflies. In contrast to research on intraguild predation of thrips on spider
mites and predatory mites (Agrawal and Klein 2000, Janssen et al. 2003), I found that
the incidence of feeding did not depend on host-plant quality. Because the develop-
mental rate and oviposition rate of thrips was higher on a diet of cucumber leaves with
whitefly crawlers than on cucumber leaves without whitefly crawlers, I infer that thrips
do not just kill whiteflies to reduce competition, but utilize whitefly crawlers as food.

In Chapter 6, I investigated whether predators might be able to ‘chemically disguise’
themselves by eating several prey species alternatingly, thus preventing antipredator
behaviour in the prey. I marked predatory mites with cues of either whiteflies or
thrips, and subsequently offered the same or the other prey species. Predators
marked with thrips cues were found to kill significantly fewer thrips than predators
marked with whitefly cues, even though the predator’s tendency to attack was the
same. In addition, more thrips sought refuge in the presence of a predatory mite
marked with thrips cues than when marked with whitefly cues. I suggest that predato-
ors can indeed increase their predation rate on one prey species by selecting a
mixed diet, thus affecting anti-predator behaviour of the prey.

General discussion
The results show that the densities of a shared predator reaches much higher levels
in the presence of two prey species than with either prey species alone, and that this
occurred within a time span of 8 weeks. This resulted in lower densities of only one of
the two prey species, whereas the densities of the other prey were low, independent
of the presence of the alternative prey. This predator-mediated interaction can be
2004). Although apparent competition is usually defined as a reciprocal negative prey
interaction, most empirical studies show non-reciprocal indirect interactions between
prey (Chaneton and Bonsall 2000). The control of whiteflies was improved by the presence of thrips, but the control of thrips was not affected by the presence of whiteflies. Such asymmetric effects of prey species on each other through a shared predator have also been referred to as indirect amensalism rather than apparent competition (Chaneton and Bonsall 2000). The results of Chapter 3 support the theory that two prey species that share a predator may also affect each others’ densities positively because an increase in the numbers of one species may lead to predator satiation (Holt and Lawton 1994, Abrams and Matsuda 1996, Holt 1997). Such short-term apparent mutualism, which occurs within a generation, has typically been shown in studies with predators with a long generation time relative to that of predatory mites, such as carabid beetles (Koss and Snyder 2004, Symondson et al. 2006). In our study system, both the predators and prey species went through several generations before apparent mutualism effects were overruled by apparent competition, nevertheless, the theoretical prediction that long-term dynamics will be characterized by apparent competition and not by apparent mutualism (Holt 1977) was confirmed.

The developmental rate and juvenile survival of the predator A. swirskii on a mixed diet of whiteflies and thrips was higher than on thrips alone, the best of these two prey species, and this explains the high numbers of predators that were observed (see Chapters 2, 3 and 4). Positive effects of mixing high- and low- quality prey, such as increased survival or fecundity, have been demonstrated for several arthropods (Uetz et al. 1992, Toft 1995, Evans et al. 1999, Toft and Wise 1999, Harwood et al. 2009). There are several explanations for why mixed diets lead to higher survival and developmental rate of predators. The first explanation that comes to mind is the higher availability of food. In Chapter 2, I showed that food was not limiting. Thus, the high numbers of predators in the treatments with both pests must have had another cause than prey availability. The second explanation is that different food types are complementary resources (Waldbauer and Friedman 1991, Bernays et al. 1994). The most famous field study on this subject is probably that of the moose, which selects a nonrandom diet of aquatic plants to meet sodium requirements and foliage from woody plants for their caloric content (Belovsky 1978). In Chapter 5, I present evidence of such a positive effect of dietary complementarity on fitness: the developmental rate and oviposition rate of thrips was higher on a diet of cucumber leaves with whitefly crawlers than on cucumber leaves alone.

A third and so far underexposed explanation for why mixed diets lead to higher growth rates, is that predators can reach higher predation rates on a superior prey species by masking themselves by alternatingly feeding on the inferior and the superior prey species (Chapter 6). This advantage of a mixed diet for generalist predators sheds another light on existing theory on adaptive diet choice. I suggest that contrary to the density-dependent approach of most optimal foraging models, diet
choice might depend on behavior of predators and prey. Predators that face strong anti-predator behaviour might actively search for another prey species to avoid the induction of antipredator behaviour in their preferred prey (Lima et al. 2003). Prey switching in predators might have evolved because it results in reduced antipredator behaviour, leading to a higher predation rate on the preferred prey.

Lima and co-authors (2003) incorporated anti-predator vigilance into a classical model of diet choice. Such classical models (Charnov 1976, Stephens and Krebs 1986) consider the diet choice of a predator when encountering prey of high and lower quality on the same patch. These models predict that a predator should always accept the superior prey type, but should accept inferior prey only when encounter rates with the superior prey fall below a certain threshold. Lima et al. (2003) assumes that the vigilance of prey increases with the number of conspecific prey that are attacked by the predator, and predict that antipredator behaviour of the prey causes predators to select a more generalized diet than one might predict on the basis of classical theory. For one thing, this is caused by the predators experiencing a lower attack rate on the superior prey because of the increased vigilance, but for another the predators can manipulate the antipredator behaviour of the superior prey by feeding on the inferior prey. These authors suggest that predators will often benefit from 'managing' the antipredator responses of the prey. Hence, this closely resembles the scenario proposed in this thesis, i.e. predators feeding on inferior prey in order to chemically disguise themselves, thus preventing strong antipredator behaviour in the superior prey. However, if predators commonly feed on heterospecific prey species in order to disguise themselves, there might consequently be selection for prey individuals that distinguish chemical cues of predators that fed on heterospecific co-occurring prey species. In aquatic systems, it was found that although prey often exhibit the strongest anti-predator behaviour when the predator consumes a diet of conspecific prey (Wilson and Lefcort 1993, Schoeppner and Relyea 2005), prey also responds to predators that consumed heterospecific prey (Smith 1992, Chivers and Mirza 2001).

In conclusion, although predator-prey interactions have been studied for centuries, there are still unexplored areas, such as interplay between antipredator behaviour and diet choice of generalist predators. I think that this deserves more attention, especially the effects of this behavioural response race (Sih 1984) on population dynamics and predator prey co-evolution.

With respect to biological control, I show that the use of one species of natural enemy against two pests can result in reduced control in the short-term, but increased control in the long-term. In general, biological control strategies might be improved by tolerating acceptable levels of pests in order to stimulate population growth of generalist predators (Chapters 2 and 3). A new finding in this thesis is, that mixed diets can increase the numerical response of the predators. Hence, accepting low levels of
various pests in a crop can enhance biological control. A potential danger of accepting such low pest levels, is that pests could also profit from a mixed diet consisting of plants and other pests through intraguild predation (Chapter 5). Based on the positive effects of pest diversity on densities of a generalist predator and its subsequent negative effect on pest densities (Chapters 2, 3 and 4), I suggest that it is useful to evaluate natural enemies by assessing their performance not only on the target prey, but also in the presence of other relevant pest species. Moreover, the system with A. swirkii as a natural enemy of two prey species showed to be an excellent model organism for studying indirect interactions between prey on a relatively short timescale, because of their short generation time and strong numerical response.

References


Summary

In this thesis, I study the interactions among prey that share a predator. In such systems, indirect interactions mediated by the shared predator can occur between pest species, such as apparent competition and apparent mutualism. Theory predicts that adding a population of a new prey species to a system consisting of one predator and one prey results in a lower equilibrium density of the resident prey, even when the two prey species do not compete for resources. This is because the equilibrium density of the shared predator increases with the increased equilibrium density of the added prey species. This interaction can even lead to exclusion of the resident prey species. In the short term, before reaching equilibrium, two prey species that share a predator may also affect each others’ densities positively because an increase in the numbers of one species may lead to predator satiation, resulting in decreased predation on the other species (so-called apparent mutualism). In biological control systems, apparent competition is desired because it brings pest levels down; apparent mutualism is not, because it does the opposite. Moreover, biological control systems, especially those in greenhouses, consist of relatively simple food webs that are open to manipulation of the species composition, and therefore offer an ideal opportunity to study interactions among prey species. I used a system consisting of a biological control agent, the generalist predatory mite *Amblyseius swirskii*, and several pest species (greenhouse whitefly, Western flower thrips and two-spotted spider mites) and cucumber plants. I investigated direct interactions between herbivores and indirect interactions via a shared predator to gain insight into the dynamics of the prey species. In particular, I studied whether these dynamics can be characterized as positive or negative indirect interactions among the pest species, i.e. whether shared predation is positive or negative for biological control. I also studied the effects of behaviour of predator and prey on these interactions. For example a generalist predator might have a preference for one prey species and the other prey species may therefore temporarily escape predation, which can lead to apparent mutualism. Another example is that mixed diets are known to have positive effects on reproduction in some predator species and the effect of adding a new species to a system consisting of one prey species and one predator species would then surpass that of simply adding more prey items. A mixed diet can result in a high-
er growth rate of the predator population, resulting in the reduction of the prey species population (apparent competition).

In Chapter 2, I report that the densities of a shared predator reach much higher levels in the presence of two prey species than with either prey species alone, and that this occurs within a time span of eight weeks. This results in lower densities of one of the two prey species, whereas the densities of the other prey were low, independent of the presence of the alternative prey. This predator-mediated interaction can be classified as apparent competition. Hence, the control of whiteflies was improved by the presence of thrips, whereas thrips were adequately controlled in the presence as well as in the absence of whiteflies.

Laboratory experiments with *A. swirskii* (Chapter 2) suggested that the higher predator densities observed in the greenhouse were partly due to a higher juvenile survival and developmental rate on a mixed diet. Whereas thrips were a superior food source for *A. swirskii* than whiteflies, a mixture of the two is even better.

In Chapter 3, I show that two prey species that share a predator may also affect each others’ densities positively. Such, short-term apparent mutualism is undesired for biological control. In the system studied here, both the predators and prey species went through several generations before positive indirect effects between the pests were overruled by negative effects (apparent competition). In Chapter 4, I studied whether the high predator densities resulting from a mixed diet affected pest populations on which the predators have a small per capita effect. Besides the Western flower thrips and greenhouse whitefly, a marginally suitable prey, the two-spotted spider mite (*Tetranychus urticae* Koch) was included in the investigation. Lower levels of spider mite damage were found in the presence of the other two pest species, which probably resulted from a strong numerical response of the predator (up to 50 times higher densities) on thrips and whiteflies. This shows that apparent competition effects can also affect species that are not considered suitable for the predator. It also shows that diversity of pest species can enhance biological control through increased predator densities.

The increased control reported in Chapters 2, 3 and 4 are, thus, not only caused by the increased presence of prey for the predators, but also through a positive effect of mixed diets on predator population growth. A slightly different and so far underexplored advantage of a mixed diet for generalist predators is that predators can reach higher predation rates on a superior prey species. This is because many prey species are able to recognize chemical cues associated with the presence of predators and these cues usually induce anti-predator behaviour in the prey, such as counterattacking, hiding, remaining motionless or aggregating. To tune this behaviour to the current danger, prey need to assess predation risk. Many prey species can distinguish chemical cues from predators that fed on conspecific prey from those that fed on het-
erospecific prey, and react stronger to the first. The explanation for this is that predators that fed on conspecific prey pose a larger threat than predators feeding on other prey. However, the predator side of this story has been underexposed: if diet-related chemical cues enable prey to discriminate between harmless and dangerous predators, predators might be able to ‘chemically disguise’ themselves by eating different species alternatingly thereby reducing antipredator behaviour and increasing predation rates. In Chapter 6, I studied whether generalist predators indeed have an increased chance to capture a given prey species when they are contaminated with chemical cues from another prey species. I marked predatory mites with cues of either whiteflies or thrips, and subsequently offered the predators the same or the other prey species. Predators marked with thrips cues were found to kill significantly fewer thrips larvae than predators marked with whitefly cues, even though the predator’s tendency to attack was the same. In addition, more thrips larvae sought refuge in the presence of a predatory mite marked with thrips cues than when marked with whitefly cues. I suggest that the predator A. swirskii can indeed increase her predation rate on the superior prey (thrips) by selecting a mixed diet.

In conclusion, the results show that the densities of a shared predator reached much higher levels in the presence of two prey species than with either prey species alone. This can partly be explained by an increased developmental rate and juvenile survival of the predator on a mixed diet. Another explanation might be that predators can reach higher predation rates on a superior prey species by masking themselves by alternatingly feeding on the inferior and the superior prey species.

With respect to biological control, I show that the use of one species of natural enemy against several pests can result in reduced control in the short-term, but increased control in the long-term. In general, biological control strategies might be improved by using generalist predators that can feed and reproduce on several pest species.
Samenvatting

In dit proefschrift bestudeer ik de interacties tussen twee prooien en hun gezamenlijke rover. In systemen met twee prooien en één rover kunnen indirecte interacties via deze gezamenlijke rover plaatsvinden. Theorieën voorspellen dat, wanneer een populatie van een prooisoort wordt toegevoegd aan een systeem dat bestaat uit één roversoort en één prooisoort, de evenwichtsdichtheid van de al aanwezige prooisoort zal dalen, zelfs als de twee prooisoorten niet elkaars concurrenten zijn. Dit komt doordat de evenwichtsdichtheid van de rover-populatie zal stijgen met het toenemen van de dichtheid van de prooipopulatie die aan het systeem werd toegevoegd.

Deze interactie, die in het Engels apparent competition genoemd wordt (er is geen goede Nederlandse term voor), kan zelfs leiden tot zo’n sterke afname van de dichtheid van de oorspronkelijke prooi dat deze soort verdwijnt uit het systeem. Op korte termijn, als een evenwichtsituatie nog niet bereikt is, kunnen twee prooisoorten die een gezamenlijke rover hebben, elkaar ook positief beïnvloeden, omdat een stijging in de dichtheid van de populatie van de ene prooi kan leiden tot verzadiging van de rovers, waardoor die minder kunnen eten van de andere prooisoort. Deze interactie wordt apparent mutualism genoemd (ook hier is geen goede Nederlandse term voor). In de biologische bestrijding is een interactie als apparent competition wenselijk, omdat het resulteert in lagere plaagdichtheden. Dit in tegenstelling tot apparent mutualism, waarbij plaagdichtheden juist omhoog gaan. Biologische bestrijding, met name in kassen, biedt de ideale mogelijkheid om interacties tussen soorten te bestuderen, omdat deze bestaan uit relatief weinig soorten waarbij de soortensamenstelling makkelijk te beïnvloeden is. Het systeem waar ik het meest onderzoek aan heb verricht, bestaat uit een generalistische roofmijt Amblyseius swirskii, verscheidene plaagsoorten (kaswittevlieg, Californische trips en spintmijt) en komkommerplanten. Ik heb zowel de directe interacties tussen de plagen onderzocht als de indirecte interacties via de rover, om inzicht te krijgen in de populatiedynamica van plagen. In het bijzonder heb ik onderzocht of deze dynamica positief dan wel negatief is voor biologische bestrijding. Ik heb ook naar het effect van het gedrag van de rover en de prooi op deze interacties gekeken. Een generalistische rover kan namelijk een voorkeur hebben voor een van de twee prooisoorten, waardoor de andere prooisoort tijdelijk kan ontsnappen (apparent mutualism). Een
ander voorbeeld is dat van gemengde diëten over het algemeen bekend is, dat zij
een positief effect hebben op de reproductie van sommige roversoorten, wat maakt
dat het toevoegen van een nieuwe soort aan een systeem dat bestaat uit één rover
eén plaag, dan niet meer simpelweg staat voor het toevoegen van meer voedsel.
Een gemengd dieet kan dan namelijk leiden tot een hogere groeisnelheid van de
roverpopulatie en dientengevolge zullen de plaagpopulaties afnemen (apparent com-
petition).
In hoofdstuk 2 laat ik zien dat de dichtheid van de roverpopulatie veel hoger wordt in
aanwezigheid van twee prooisoorten dan in aanwezigheid van één prooi en dat
dit al gebeurt in acht weken. Dit leidt tot de daling van de dichtheid van één van de
twee prooisoorten (de kaswittevlieg), terwijl de dichtheid van de andere prooi al laag
was, onafhankelijk van de aanwezigheid van de nieuwe prooi. Kortom, de bestrijding
van kaswittevlieg gaat beter in aanwezigheid van de andere plaagsoort (Californische
trips), terwijl de laatste in alle gevallen succesvol bestreden wordt.
Laboratoriumexperimenten met de rover A. swirskii (Hoofdstuk 2) laten zien dat de
hogere dichtheden van de roverpopulatie die waargenomen werden in de kassen,
gedeeltelijk veroorzaakt worden door hogere juveniele overleving en hogere ontwik-
kelingsnelheid van de roversoort op een gemengd dieet van trips en wittevlieg.
Bovendien bleek dat trips een betere voedselbron vormen dan wittevlieg, maar dat
een mengsel van de twee plaagsoorten het beste diet voor de rovers is.
In hoofdstuk 3 laat ik zien dat twee prooisoorten die een gezamenlijke rover hebben,
ekaars dichtheden ook positief kunnen beïnvloeden. Deze interactie (apparent mutu-
alism) is onwenselijk voor biologische bestrijding. De rover en de prooi moesten een
aantal generatiecycelen doorlopen voordat de positieve effecten op de prooidichtheid
teniet werden gedaan door de negatieve effecten op de prooidichtheid (apparent
competition).
In hoofdstuk 4 heb ik onderzocht of de hoge roverdichtheid die veroorzaakt werd
door het gemengde diëet ook invloed had op de populatie van een plaagsoort
waarop de rover een klein per capita effect heeft. Ik heb daartoe naast trips en witte-
vlieg ook spintmijt (Tetranychus urticae), een minder geschikte plaagsoort,
toegevoegd. In aanwezigheid van wittevlieg en trips werd er minder schade door
spintmijt waargenomen, wat waarschijnlijk veroorzaakt wordt door de hoge aantallen
rovers in aanwezigheid van trips en wittevlieg (meer dan 50 keer hogere popu-
latiedichtheid). Dit laat zien dat effecten van een interactie zoals apparent competi-
tion ook van invloed zijn op de populaties van soorten die beschouwd worden als
ongeschikt voor de rover. Het laat ook zien dat diversiteit aan plaagsoorten de bio-
logische bestrijding verbetert door de toename van de dichtheid van de populatie
van de rover. De goede bestrijding van plagen die werden gevonden in zowel hoofd-
stuk 2 en 3 als in hoofdstuk 4, wordt dus niet alleen veroorzaakt door de stijging van
De hoeveelheid aanwezige prooi, maar ook door het positieve effect van een gemengd dieet op de groei van de roverpopulatie.

Een ander, en minder voor de hand liggend, voordeel van een gemengd dieet voor generalistische rovers is dat zij een hogere predatiesnelheid kunnen bereiken op de kwalitatief beste prooi. Veel prooisoorten kunnen namelijk chemische signalen, afkomstig van rovers, herkennen en deze signalen induceren gedragsveranderingen in de prooi. Deze gedragsveranderingen verminderen het predatierisico voor de prooi. Voorbeelden van zulk anti-rovergedrag zijn: een tegenaanval inzetten, schuilen, stil blijven zitten of aggregeren. Prooien moeten het predatierisico in kunnen schatten om zo hun gedrag af te stemmen op het aanwezige gevaar. Veel prooisoorten kunnen onderscheid maken tussen chemische signalen van verschillende rovers, maar ook bijvoorbeeld tussen rovers die soortgenoten van deze prooi gegeten hebben en rovers die een andere soort gegeten hebben; de prooien reageren sterker op de eerstgenoemde rovers. Dit is begrijpelijk want rovers die soortgenoten van de prooi gegeten hebben, vormen een groter gevaar dan rovers die een andere soort hebben gegeten. Echter, in de literatuur is tot nu toe onderbelicht gebleven dat rovers hier ook hun voordeel mee zouden kunnen doen. Gesteld dat prooien onderscheid kunnen maken tussen gevaarlijke en niet gevaarlijke rovers door middel van prooigerelateerde signalen van een rover, dan zouden rovers zichzelf kunnen ‘vermommen’ door tijdelijk een andere prooisoort te eten. Hiermee reduceren ze het anti-rovergedrag, en dus kan de predatiesnelheid op die plaag stijgen.

In hoofdstuk 6 bestudeer ik of een rover daadwerkelijk een grotere kans heeft om een prooi te pakken als hij in aanraking is geweest met chemische signalen van een andere prooisoort. Ik heb roofmijten gemerkt met lichaamsvloeistoffen van wittevlieg danwel van trips en daarna heb ik deze gemerkte rovers dezelfde prooisoort aangeboden of juist de andere prooisoort. Rovers die gemerkt waren met trips, aten significatief minder tripslarven dan rovers die gemerkt waren met wittevlieg. Bovendien schuilden meer tripslarven in aanwezigheid van rovers die gemerkt waren met trips, dan wanneer de rovers gemerkt waren met wittevlieg. Ik veronderstel daarom dat rovers inderdaad hun predatiesnelheid op een prooi kunnen verhogen door een gemengd dieet te selecteren.

Concluderend: de resultaten van dit onderzoek laten zien dat de dichtheid van een gezamenlijke rover veel meer kan toenemen in aanwezigheid van twee plaagsoorten dan wanneer er maar één van de twee aanwezig is. Dit kan behalve de grotere hoeveelheid aanwezig voedsel, ook verklaard worden door een hogere ontwikkelingsnelheid en juveniele overleving van de rover op een gemengd dieet. Een ander verklaring kan zijn dat rovers hogere predatiesnelheid kunnen bereiken op de kwalitatief betere prooi door afwisselend twee prooisoorten te eten, en zich daarmee chemisch te vermommen.
SAMENVATTING

In het licht van biologische bestrijding veronderstel ik dat het gebruiken van één soort natuurlijke vijanden tegen verschillende plaagsoorten op korte termijn kan leiden tot een minder goed effect op de bestrijding van deze plagen, maar op de langere termijn (acht weken) een goed effect heeft op de bestrijding van de plagen. Omdat de meeste productiecycli van gewassen langer is dan deze periode, denk ik dat biologische bestrijding verbeterd kan worden door juist generalistische natuurlijke vijanden te gebruiken.
Publications


Submitted papers
