Introduction

The two-spotted spider mite *Tetranychus urticae* Koch (Acari: Tetranychidae) (TSSM) and the western flower thrips *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) (WFT) are widespread over an array of geographical range (CAB International, 2003). WFT is a native insect of the western part of North America and was first reported in 1895. In the 1970s and early 1980s, this species spread throughout North America (Beshear 1983). Soon thereafter it was found in Europe in Dutch greenhouses and since then it has become a major exotic pest of greenhouse production in many countries throughout the world (Tommasini and Maini, 1995, van Lenteren and Loomans 1998, Kirk and Terry, 2003). TSSM is also a cosmopolitan pest. It has been recorded from most countries in Europe, Asia, Africa, Australasia, the Pacific and Caribbean islands, America (CAB International, 2003). In many field crops both species often occur simultaneously on the same plants in the field (Trichilo and Leigh, 1986; Wilson et al., 1996; Colfer et al., 1998, 2000) just as in the greenhouse (Brødsgaard and Enkegaard, 1997; Sterk and Meesters, 1997).

TSSM and WFT are notorious pests particularly of ornamental plants and vegetables. While feeding, both pests penetrate the plant foliage/leaves/flowers with their mouth stylets and suck out the cell contents. Photosynthesis and metabolism of plants are seriously affected. In addition, WFT causes indirect damage by vectoring tomato spotted wilt (TSWV) and impatiens necrotic spot viruses (INSV) (Tommasini and Maini, 1995; van de Wetering et al., 1996). Moreover, in ornamentals even low pest densities can affect the external quality. Quantitative and qualitative damage often results in important economic losses in crops and ornamentals (Rabbinge, R., 1995; Lewis, 1997; Nachman and Zemek, 2002).

Chemical control of both pests is extremely problematic, particularly that of WFT. The core difficulties are reflected by the facts that the organisms are minute and can live sheltered on or in leaves and/or flower buds (Brødsgaard, 1989), where they are often not reached by contact insecticides. More efficient are systemic insecticides with multiple modes of actions, but particularly with WFT even these pesticides are unable to control non-feeding stages like eggs of WFT and TSSM or prepupa and pupa of WFT. Since all insecticides are unable to control these soil-dwelling developmental

stages of WFT, an important source for recolonisation of the plant cannot be eliminated. Moreover, both TSSM and WFT achieve high reproduction rates and short generation cycles especially under warm conditions and under high pesticide load both pests tend to fast selection of resistant biotypes. It is reported that WFT (Gillespie, 1989; Brødsgaard, 1994; Zhao et al., 1995) and TSSM (Cranham and Helle, 1985; Eilenberg et al., 2000) have become resistant to numerous insecticides or acaricides registered for their control. Systemic insecticides are often very persistent which requires long waiting periods after application. Moreover, some efficient pesticides can have undesired side effects on important natural enemies, influencing systems of biocontrol in multiple pest associations where other pests should be controlled by predators or parasitoids (van Lenteren and Loomans, 1998), or they kill bumblebees, which are used in greenhouse tomatoes for pollination (Shipp and Wang, 2003). Highly demanded are safe and environmentally harmonious alternatives to the use of synthetic insecticides like the implementation of biological control as a management tactic.

The important natural enemies of TSSM include phytoseiid mites (Hussey et al., 1965; Oatman and McMurtry, 1966) and Stethorus spp. (Coleoptera: Coccinellidae) (Chazeau, 1985). Other useful beneficials include anthocorids (mainly Orius spp.), larvae of chrysopids, predatory thrips (e.g. Scolothrips spp.) as well as larvae of cecidomyiid midges, in particular Feltiella acarisuga Kieffer (i.e., Therodiplosis persicae) (Diptera: Cecidomyiidae) (Chazeau, 1985) and fungi, such as *Neozygites* spp. (Klubertanz et al., 1991). However, the most effective natural enemies of TSSM are predatory mites from the family Phytoseiidae. These mites, belonging to a number of genera, such as Phytoseius, Amblyseius, Euseius and Neoseiulus, have been shown to regulate populations of TSSM on a range of crops (CAB International, 2003). The most frequently used predator has been the phytoseiid mite *Phytoseiulus persimilis* Athias-Henriot, which was first used in glasshouses on various crops in the 1960s (for example, Hussey et al., 1965), and has been used ever since successfully on a wide variety of crops in a range of protected and unprotected environments. However, P. persimilis shows a high degree of specialization to tetranychid mites as prey (McMurtry and Croft, 1997; Cross et al., 2001). Its high efficiency can lead to an overexploitation of the prey and the predator will die out because of starvation, the inability of subsistence on alternate food, especially in confined spaces, as in a greenhouse (Wysoki, 1985). This

creates unstable systems with often fast resurgence of the pest. On long-term basis it doesn't always succeed in suppressing TSSM (Sterk and Meesters, 1997). On the other hand, accompanying infestation with WFT cannot be suppressed by this specialized predator. Other phytoseiid mites, such as *Amblyseius (Neoseiulus) californicus* McGregor (Garciá-Marí and González-Zamora, 1999; Easterbrook et al., 2001) and *A. fallacis* Garman (Raworth, 1990; Cooley et al., 1996) are also used to control spider mites on some certain plants. They are, however, also selective predators of tetranychid mites (McMurtry and Croft, 1997).

The biocontrol agents of WFT include predatory mites (Ramakers, 1987; Gillespie, 1989; Bennison and Jacobson, 1991), anthocorid bugs (van den Meiracker and Ramakers, 1991; Sabelis and Rijn, 1997), nematodes (Ebssa et al., 2001, 2004; Premachandra et al., 2003), predatory thrips (Ramakers et al., 2000), parasitoids (van Lenteren and Loomans, 1998) and fungi (Butt and Brownbridge, 1997; Murphy et al., 1998). Among the most reliable biocontrol agents are phytoseiid mites of the genus *Amblyseius* and anthocorid flower bugs of the genus *Orius*, whereas *A. cucumeris* Oudemans and *O. insidiosus* Say are two often used species. *A. cucumeris* was first associated as a predator of WFT in Dutch greenhouses. It was later fully developed to control thrips and used on a commercial basis in the early 1980's. *O. insidiosus* is a nearctic species. It has been extensively used from the early 1980's in Europe (van den Meiracker and Ramakers, 1991; Sörensson and Nedstam, 1993).

Both *Amblyseius* (Acarina: Phytoseiidae) and *Orius* spp. (Heteroptera: Anthocoridae) are generalist predators. Besides feeding on thrips, they also consume other small arthropods like spider mites (Lewis, 1997; McMurtry and Croft, 1997). For *Orius* spp. thrips and mites are believed to constitute important parts of their diet (Wright, 1994). Both *Amblyseius* and *Orius* spp. are also sometimes used to control spider mites (Helle and Sabelis, 1985; McMurtry and Croft 1997; Colfer et al., 1998, 2000). Therefore, in a mix-infestation system of TSSM and WFT, *Amblyseius* or *Orius* spp. seem to offer attractive protagonists against both pests. However, the preference even of a generalist predator for different preys may be distinct (Salas-Aguilar and Ehler, 1977; Isenhour and Yeargan, 1981). Likewise, the densities of preys may also influence the preying efficiency of a predator. Therefore, the first objective of this study was to investigate

how TSSM and WFT at their different densities affected each other the control efficiency of *A. cucumeris* or *O. insidiosus* in their mixed infestations.

Amblyseius and *Orius* spp. have been extensively used to control WFT. However, satisfactory control of WFT cannot be easily and reliably achieved with each of the two predators. For instance, *Orius* spp. are only effective if sufficient prey is available. At a lower density of WFT, *Orius* spp. tend to leave the crop or even the greenhouse unless pollen as alternative food source is available (Ramakers and Meiracker, 1991). Likewise, *Amblyseius* spp. are able to prey only on the first instar larvae of thrips (Gillespie and Ramey, 1988; Bakker and Sabelis, 1989; Van der Hoeven et al., 1990), and their dispersal potential is lower than that of WFT (Jacobson et al., 2001b). Abiotic factors, such as temperature (Shipp et al., 1996; Shipp and van Houten, 1997) and humidity (van Houten et al., 1993) also restrict the impact of *Amblyseius* spp. on WFT.

Though *A. cucumeris* is often used as a preventive method with inundative releases at very low infestation densities of WFT, occasions of failure in efficient control with *A. cucumeris* are reported. In particular, if sudden immigration of large WFT numbers from adjacent crops occurs, the predators are not able to limit built-up of damaging populations (Jacobson et al., 2001b). Therefore, sometimes both predators are released together on the same greenhouse ornamentals and vegetables to achieve additive or even synergistic effects and to improve the efficacy of control (Sörensson and Nedstam, 1993; Sterk and Meesters, 1997). During the last decade, because the availability of beneficials for biocontrol in greenhouses has substantially increased and production costs could be reduced (Eilenberg et al., 2000), the combined release of several biocontrol agents simultaneously to combat one or more pests has become a common phenomenon (Meyling et al., 2002).

However, with the combined release of beneficials, especially with generalist predators in same environment, the risk of competition increases. One predator may be consumed by the other, a general phenomenon, called intraguild predation (IGP) (Polis et al., 1989; Rosenheim et al., 1995). The predator, which preys on the other one, is called intraguild predator (IG-predator), and the victim is the intraguild prey (IG-prey). The extent of IGP can vary due to differences in the preference of the IG-predator for the IG-prey or the herbivorous pest, thereby possibly affecting the outcome of biological control. For example, if the IG-predator indiscriminately preys on both the IG-prey and the herbivorous pest, combined releases of the IG-predator and the IG-prey may lead to a decreased control efficiency of the herbivorous pest. For instance, Wittmann and Leather (1997) recorded that *O. laevigatus* (Fieber) did not distinguish between *A. cucumeris* and WFT as prey. Hence, a combination of *O. laevigutus* and *A. cucumeris* is not likely to result in efficient biological control of WFT. However, biological control should be enhanced if the IG-predator prefers preying on the herbivorous pest to on the IG-prey. In the mentioned study Wittmann and Leather (1997) observed that *O. laevigatus* preferred preying on WFT to on another predatory mite species *A. degenerans* (Berlese), which would improve WFT control. Gillespie and Quiring (1992) found that *O. tristicolor* (White) favoured WFT over *A. cucumeris*, indicating that a combination of these predators might also improve biological control of WFT.

Additionally anti-predation behaviour of the IG-prey in the presence of the IG-predator may affect the extent of IGP. For example, TSSM produce webbings, which can be used by other pests like WFT and also small predators to seek refuge from carnivory (Pallini et al., 1998). Finally the prey density may also influence the extent of IGP by reducing the competition for prey between the IG-predator and the IG-prey.

Thus, in the biological control system including multi-pests and predators, the relationships among them can be complicated. In this study, the outcome of combined releases of *A. cucumeris* and *O. insidiosus* on mixed infestations of TSSM and WFT at varying initial densities were investigated. The specific objectives were to study (1) whether TSSM and WFT can be effectively controlled by combined releases of both predators, and (2) if and how pest densities affect IGP of *O. insidiosus* on *A. cucumeris*.

Currently, the biological control of WFT focuses primarily on the foliar dwelling developmental stages, with little attention to the soil inhabiting stages. However, the late second instar larvae of WFT will leave the plant, prepupate and pupate in the soil (Palmer, 1989; Tommasini and Maini, 1995; Berndt, 2004), where they spend about one-third of their life cycle (Loomans and van Lenteren, 1995). This developmental stage of WFT in the soil is excluded from the prey spectrum of the common predators foraging on the plants. Soil predatory mites like *Stratiolaelaps (Hypoaspis) miles* (Berlese) and *Hypoaspis aculeifer* Canestrini (Acarina: Laelapidae) commercially