BIOLOGICAL CONTROL OF THE TWO-SPOTTED SPIDER MITE, <u>TETRANYCHUS</u> <u>URTICAE</u> KOCH, IN RASPBERRIES USING THE PREDATORY MITE, <u>PHYTOSEIULUS</u> <u>PERSIMILIS</u> ATHIAS-HENRIOT

by

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THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF PEST MANAGEMENT in the Department of Biological Sciences

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BIOLOGICAL CONTROL OF THE TWO-SPOTTED SPIDER MITE, TETRANYCHUS URTICAE KOCH, IN RASPBERRIES USING THE PREDATORY MITE, PHYTOSEIULUS PERSIMILIS

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ABSTRACT

The two-spotted spider mite, <u>Tetranychus</u> <u>urticae</u> Koch (Acari: Tetranychidae), may be a major pest of raspberries in commercial fields, particularly if pesticides have been used to control other insects or fungi. <u>Tetranychus urticae</u> has become resistant to many organophosphate and carbamate insecticides, as well as to most acaricides. In the Fraser Valley of British Columbia, raspberry growers have no effective method of control. Phytoseiids have been used to control <u>T</u>. <u>urticae</u> in greenhouse crops since the 1960's, and <u>Phytoseiulus persimilis</u> Athias-Henriot (Acari: Phytoseiidae) in particular is an effective predator. It is also capable of regulating <u>T</u>. <u>urticae</u> populations in orchards and on such field crops as strawberries.

I investigated the potential of <u>P</u>. <u>persimilis</u> to control populations of <u>T</u>. <u>urticae</u> in field raspberries at Agassiz, British Columbia. The predator was introduced at three different release rates and populations were monitored over a period of 8.5 weeks. <u>Phytoseiulus persimilis</u> became established in the three treatment plots in low numbers, and as a result <u>T</u>. <u>urticae</u> numbers were lower in the treatment plots than in the controls. Significant differences in numbers of <u>T</u>. <u>urticae</u> between the treatments and the controls were detected on two dates. <u>Phytoseiulus persimilis</u> dispersed or was carried into the control subplots and was present there at very low numbers throughout the experiment. In these subplots <u>T</u>. <u>urticae</u> numbers followed the same trend as in the treatment subplots.

<u>Phytoseiulus persimilis</u> is evaluated as a potential biological control agent of <u>T</u>. <u>urticae</u> in raspberries, and compared to two other potential phytoseiid predators. Release rates, timing of release, and insecticide resistance in both predator and prey are discussed in relation to biological control.

I conclude that <u>P. persimilis</u> is potentially effective as a biological control agent of <u>T. urticae</u> if strains of <u>P. persimilis</u>

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resistant to the pesticides used in raspberries are available, if it is introduced when <u>T</u>. <u>urticae</u> numbers are low, and if the cost of rearing can be reduced. However, further studies are needed with early infestations of <u>T. urticae</u> to determine if season-long control can be achieved. In addition, other native predators should be compared with <u>P. persimilis</u>.

ACKNOWLEDGMENTS

I would like to thank Ward Strong for first suggesting that twospotted spider mites in raspberries were a local problem that needed research, and for putting up with my many phone calls; Dave Raworth for generously and patiently giving his time and advice throughout my research; Dave Gillespie for lending me his mite-brushing machine and assisting me in getting started; Stan Freeman for allowing me the use of his research plots; Dr. Mackauer for his encouragement, advice, and patience with the protracted schedule of a part-time student; and finally my family, especially my two children, Lizzie and Robbie, for cheering me on.

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<u>CHAPTER 1</u>

INTRODUCTION

1.1 THE RASPBERRY INDUSTRY IN B.C.

Raspberries are an important commercial crop in B.C. In 1991, 26.3 million pounds were produced with a value of approximately \$15.4 million. Raspberry yields can range from 3 tons per acre to 7 tons per acre depending on the variety. The current total costs of production are about \$0.60 per pound for hand-picked berries, and about \$0.40 per pound for machine-picked berries (B. Peters, pers. comm.). Raspberry growers receive from \$0.30 per pound (1990) to \$1.00 per pound (1986) for handpicked berries, and \$0.22 to \$0.47 per pound for machine picked, depending on market conditions. Thus, in some years growers have little or no profit margin, and therefore would like to minimize crop loss due to pest damage.

Major pest problems in raspberries include root weevils, fruitworms, cutworms, leafrollers and two-spotted spider mites (D.Henderson, pers. comm.). Weevils are currently controlled by spraying carbofuran (Furadan) along the base of the plants, with the aim of minimizing detrimental effects on predators higher up in the plant foliage (B. Peters, pers.comm.). Leafrollers, fruitworms, and cutworms are controlled with azinphosmethyl (Guthion) of diazonon. In the last five years, growers have been relying more on mechanical harvesting rather than handpicking because it is more economical. Because it is difficult to avoid contamination of the fruit with insects during mechanical harvesting, some insects and arthropods (such as spiders), which are harmless to the plants, have become "pests". Non-chemical methods of control are needed for all these insects if long-term biological control of two-spotted spider mite is to be successful.

The two-spotted spider mite, <u>Tetranychus</u> <u>urticae</u> Koch (Acarina: Tetranychidae), can cause considerable damage to raspberry plants. In a

severe infestation, plants become covered with white webbing containing hundreds of <u>T</u>. <u>urticae</u> per leaf, and can be almost completely defoliated by late summer. The only miticide registered for raspberries is dicofol (Kelthane) (Berry Production Guide 1992-1993), and it is minimally effective because resistance has built up in the <u>T</u>. <u>urticae</u> population.

Economic thresholds have been developed for <u>T</u>. <u>urticae</u> on greenhouse vegetables using leaf damage indices (Hussey and Scopes 1985). When the index reached 0.4 on a scale of 1 to 5, <u>P</u>. <u>persimilis</u> successfully controlled <u>T</u>. <u>urticae</u>. In almond orchards in California, decisions are made based on accumulated mite-days and the predator/prey ratio (Hoy 1985). In strawberries in British Columbia, an economic threshold of 5 mites per leaflet was developed by Raworth (1986). However, an economic threshold for <u>T</u>. <u>urticae</u> in raspberries has not yet been determined. Raworth (1989) found no yield reductions from a spring infestation of <u>T</u>. <u>urticae</u> that attained levels as high as 300 mites per leaf, even though leaves showed visible damage by harvest time. In addition, yields in the following year were not related to accumulated mite-days of the previous year. Compared to strawberries, raspberries are apparently much more tolerant of <u>T</u>. <u>urticae</u> infestations.

Despite the lack of an economic threshold at present, <u>T</u>. <u>urticae</u> is nevertheless considered to be a pest of raspberries. Severe foliage damage caused by <u>T</u>. <u>urticae</u> can reduce plant vigour. Weak plants leaf out earlier in the spring, and are thus more susceptible to late frosts than healthy plants (B. Peters, pers.comm.) A fall infestation can result in reduced bud survival over the winter (Doughty et al. 1972). Further research may show that yields are reduced under such circumstances.

1.2 BIOLOGY OF TETRANYCHUS URTICAE

The life cycle of <u>Tetranychus</u> <u>urticae</u> consists of egg, larva, protonymph, deutonymph, and adult. Developmental time varies with

temperature, except where extremes of temperature and/or humidity cause the mites to cease activity and perhaps enter diapause. The minimum temperature for development is 12°C, the maximum is 40°C, and the optimum is 30-32°C (Huffaker et al. 1969). Development from egg to adult takes 6-12 days at 26-31°C (Huffaker et al. 1969). The average duration of each stage of the male and female respectively, under a diurnal temperature cycle of 15-28.3°C, was determined by Laing (1969) to be as follows: egg, 6.7 days for both ; larva, 3.6 and 3.7 days; protonymph, 2.7 and 3.0 days; deutonymph, 3.1 and 3.5 days. Total developmental times for all immature stages was 16.1 days for males and 16.9 days for females. The preovipositional period for females was 2.1 days, and the ovipositional period was 15.7 days. Females laid an average of 2.4 eggs per day for a total of 37.9 eqgs per female. Since the developmental rate is strongly influenced by temperature, and there can be variations among local populations of <u>T</u>. urticae , it is important to establish these developmental times before investigating, and particularly before modelling, predator/prey relationships (Brandenburg and Kennedy 1987).

Reproduction in <u>T</u>. <u>urticae</u> is by arrhenotokous parthenogenesis, that is, unfertilized females produce only males while fertilized females may produce both males and females (Jeppson et al. 1975). Eggs are laid on either upper or lower leaf surfaces. The female deutonymph spins a web under which it moults, attracts a male by means of pheromones, and then lays its eggs in the webbing. The webbing is added to by the male, and by the larval and nymphal stages as well, creating a nest for the colony (Gerson 1985). The webbing protects <u>T</u>. <u>urticae</u> from non-specific predators but attracts specific ones such as <u>P</u>. <u>persimilis</u> and <u>Stethorus</u> sp. which lay their eggs in the webbing (Takafuji and Chant 1976).

Tetranychus urticae develops faster and produces more eggs under conditions of low (25-30%) than high (85-90%) relative humidity (Nickel 1960). Survival of immature stages, adult longevity, and percentage egg hatch are all reduced under conditions of high relative humidity

(Boudreaux 1958).

Tetranychus urticae has sedentary and dispersal phases. Dispersal occurs in two ways, aerially or by ambulation over foliage or the ground (Gerson 1985). Dispersal is stimulated by food scarcity or low humidity. Under conditions of food shortage or dessication, spider mites on plants exhibit a positive phototactic response, and crawl up the plant (Suski and Naegele 1966). They may then crawl over the foliage to a new plant. Krainacker and Carey (1990) found that 46% of female T. urticae and 16% of males were able to crawl horizontally a distance of 240 cm in 48 h and a few went as far as 480 cm. Vertically, 22% of females and 8% of males travelled 240 cm. If spider mites are exposed to wind they will exhibit aerial dispersal behaviour, which involves orienting themselves away from the light, raising their forelegs and forebodies, and facing downward so that they are carried along on updrafts (Smitely and Kennedy 1985). All stages except for adult males disperse, but female adults, deutonymphs and protonymphs are the primary dispersers (Smitely and Kennedy 1985). Certain insecticides and acaricides stimulate dispersal, and thus enhance population growth by producing new colonies with high reproductive potential (Kennedy and Smitely 1985). Gerson and Aronowitz (1981) (cited in Gerson 1985) observed pesticide-induced dispersal in T. cinnabarinus (Boisduval) in the presence of the acaricides cyhexatin and dienochlor. Penman and Chapman (1983) found the same behaviour occurred in T. urticae on plants treated with the synthetic pyrethroid Fenvalerate.

Diapause is initiated in adult females by shorter daylength, decreased temperature, and dwindling food supplies (Jeppson et al. 1975), although of these three factors, photoperiod is the most important (van de Vrie et al. 1969). There is a sharply defined critical day length of 14 h for the induction of diapause (Veerman, 1985). If the temperature is above 25° C, <u>T</u>. <u>urticae</u> does not enter diapause regardless of day length. If the food supply is marginal, diapause may begin earlier; if the food supply is adequate, photoperiod and temperature are the only factors influencing

diapause (Veerman 1985). Overwintering females are orange, and hibernate on the ground under leaves or in any protected place. Eggs may also overwinter. Diapause is terminated by higher temperatures and the presence of food but, in some strains, only after a requisite cold period has occurred (van de Vrie et al. 1969). Photoperiod and the cold period interact so that an early cold period might result in diapause terminating at a shorter photoperiod; diapause may already be over by the beginning or middle of winter (Veerman 1985). Females have already been fertilized when they arrive on plants in the spring.

Mite outbreaks occur during hot, dry weather due to a combination of shorter developmental time and greater plant susceptibility. On the other hand, heavy rains can decimate mite populations.

Host plants show a wide range of variation in their response to mite infestation. Tetranychid mites damage the palisade and spongy mesophyll layers of plant cells (Jeppson et al. 1975) by piercing them with their stylets and extracting the cell contents. Puncturing of cells in a circle leads to the small chlorotic spot typical of mite injury, and bronzing may result due to damage to the mesophyll cells. Photosynthetic rates are decreased and transpiration rates are increased (Hall and Ferree 1975) so that leaves dry out and drop off. Sances et al. (1979) observed a decrease in transpiration due to deformation of stomata in strawberry plants infested by spider mite at levels as low as 2.5 mite-days/cm². Photosynthetic rates also decreased linearly with increasing infestation levels, resulting in a 60% reduction at 50 mite-days/cm². Tomczyk and Kropcznska (1984) observed that young chrysanthemum and cucumber plants exhibited a decrease in the basic elements N,P,K and Ca, but older plants did not. A decrease in the sugar content of cucumber fruits, affecting the quality of the yield, was also observed. It is thought that mites inject toxins or growth regulators into plants (Jeppson et al., 1975), but the mechanism is not known. Avery and Lacey (1968) found higher levels of gibberellin-like promoters in infested than in control plants. Why some

plants respond to T. urticae infestations at even low levels (e.g. pears) by becoming severely scorched, while others, such as raspberries and apples, may only have stippling is not well understood. Injury levels depend on the state of vigour of the crop in question, and are also related to environmental factors. Some plants are genetically more tolerant than others. Tomczyk and Kropcznska (1984) found that some cultivars of chrysanthemums had the ability to compensate for mite feeding and to repair damage by increasing their photosynthetic rates. Severe infestations early in the season can result in complete destruction of a crop, whereas high mite levels later in the season which appear to have caused major plant injury may not affect crop yield the next year. In raspberries, however, late season injury can be significant because fruit is produced on canes grown in the previous year. Doughty et al. (1972) found that severe defoliation of raspberries in the fall by \underline{T} . urticae resulted in freeze injury to the buds, reduced starch and sugar content in the canes, and reduced bud survival and growth. The leaves produce a photoperiodic translocatable factor which is necessary for cold acclimation, so if they fall before this process occurs, acclimation is delayed and freeze injury results. Photosynthetic reserves necessary for cold hardiness are also lost if premature defoliation occurs (Jennings et al. 1964). Secondary growth of buds in the autumn can occur when mite damage is severe, resulting in fewer live primary buds in the spring. Primary laterals can start to grow but suffer premature death due to a lack of sufficient strength to support leaves and fruit.

1.3 BIOLOGICAL CONTROL OF TETRANYCHUS URTICAE BY PHYTOSEIIDS

Historical data indicate that plant-feeding mites are seldom an economic problem on crops that are not treated with chemicals, including acaricides, insecticides, and fungicides. Mites became a problem after the

introduction of DDT and other synthetic insecticides after World War II (Huffaker et al. 1969; Huffaker and Flaherty 1966) .The conclusion many people have reached is that natural enemies were and are effective in keeping mite populations in check, as long as they are not affected either directly or indirectly by pesticides. Exclusive reliance on chemical control to reduce mite populations has two main detrimental effects (Stern et al. 1959): 1) Natural enemies are reduced or eliminated, resulting in resurgence of target pests and, 2) Potential or minor pests, once released from control by their natural enemies, can become major pests. In the case of <u>T</u>. <u>urticae</u>, the latter effect seems to have resulted from the broad use of pesticides, in that prior to the use of DDT they were not major pests (Huffaker 1971).

Interest in using predatory mites to control pest mites was greatly stimulated after the success of Huffaker & Kennett (1956) in controlling cyclamen mites in strawberries. Much work was done in this field (Huffaker et al. 1969, McMurtry 1983). In California, the predator mite <u>Metaseiulus</u> <u>occidentalis</u> Nesbitt was found to have resistance to some organophosphates (Huffaker & Kennett, 1953). Selection and genetic improvement of these strains, as well as mass production and inoculative releases, led to a highly successful IPM program in almonds in California (Hoy 1983).

Much research has been conducted on phytoseiids as biological control agents of spider mites on such greenhouse crops as cucumber (Mori et al.1989, Tonks and Everson 1977), tomatoes (French et al. 1976, Dixon 1973), roses (Gough 1991, Simmonds 1972, Burnett 1979), and strawberries (Cross 1984). Greenhouse crops are particularly vulnerable to damage caused by <u>T</u>. <u>urticae</u> because of the favourable environment; a population of <u>T</u>. <u>urticae</u> can double in three days at optimum temperature (Sabelis 1985). A lack of effective pesticides combined with a desire to limit their use in greenhouses led to the development of phytoseiids, particularly <u>P</u>. <u>persimilis</u>, as an alternate method of control, and they have been very successful (Tanigoshi 1982). In British Columbia, <u>P</u>.

persimilis is widely used in greenhouses (Tonks and Everson 1977, Costello and Elliot 1981).

Integrated mite control programs have reduced costs in many tree fruit areas of North America. The programs use a combination of resistant predators, selective pesticides for other pests, and acaricides which are used when the predators are not abundant enough to provide effective biological control. Acaricide use was reduced by 75% in Pennsylvania where an integrated program using <u>Stethorus punctum picipes</u> Csy to control <u>Panonychus ulmi</u> Koch (European red mite) was used for 8 years (Croft et al. 1975).

In B.C. apple orchards, the use of DDT, and subsequently organophosphates, to control codling moth, <u>Laspeyresia pomonella</u> (L.), was followed by increased problems with mites, particularly with strains resistant to acaricides. Fortunately, the native predator, <u>Metasieulus occidentalis</u> Nesbitt, developed resistance to some of the organophophates used, such as azinphosmethyl (Hoyt 1969). Downing and Arrand (1976) achieved integrated control of McDaniel spider mites, <u>Tetranychus</u> <u>mcDanieli</u> McGregor, apple rust mites, <u>Aculus schlechtendali</u> (Nalepa), and European red mite, <u>P. ulmi</u>, in Similkameen orchards using a resistant strain of <u>M. occidentalis</u> and oil sprays at the half-inch green bud stage instead of acaricides. Three organophosphate sprays per season were used for codling moth. Low numbers of the predator controlled mite populations from 1967 to 1974.

The phytoseiid <u>M.occidentalis</u> has been genetically improved by selecting for pesticide resistance. Natural strains that had acquired natural resistance to such organophosphates as azinphosmethyl, diazonon, and phosmet were selected in the lab for permethrin and carbaryl resistance. These lab-selected strains successfully established in apple, pear and almond orchards in California, Oregon and Washington (Hoy et al.1983, Hoy and Knop 1979, Hoy and Standow 1981).

In California, T. urticae has become a major pest affecting

strawberry production, largely because it developed resistance to most acaricides (Oatman and McMurtry 1966). Research on the use of P. persimilis as an introduced predator, both alone and in conjunction with other native predators, showed it was capable of significantly reducing T. urticae populations (Oatman and McMurtry 1966, Oatman et al.1967, Oatman et al.1968, Oatman 1970, Oatman et al.1977). It was considered to be the most efficient phytoseiid predator because of its high mobility, high prey consumption abilities, and high reproductive rate (Oatman et al., 1977). As a result of releases on strawberries, P. persimilis became established in Ventura County and, 4 years later, was found 20 km from the first release fields (McMurtry et al.1978). It was also found by these authors to be established in lima bean fields adjacent to the strawberry fields, and on a number of nearby weed species. Since mass releases of P. persimilis were too costly (Marsden and Allen 1980), conservation of weedy areas was recommended to maintain predator populations. In other areas of the U.S., however, biological control has not been used for mites in small fruit crops. A report by van Driesche and Hauschild (1987) on pest control in Massachusetts indicated that chemical control was the only method in use there.

In other countries, such as England, Australia and New Zealand, research has shown that <u>P</u>. <u>persimilis</u> is a potentially effective alternative to acaricides, and it is being used both in greenhouses and in outdoor strawberry crops (Charles et al.1987, Waite 1988, Cross 1984, Dixon 1973, French et al.1976, Easterbrook 1988).

<u>Phytoseiulus persimilis</u> has been used to a lesser extent on other outdoor crops (Wysoki 1985). In New Zealand, it was introduced in 1979 and is widespread throughout the country. It has been established in commercial raspberry gardens, and research is being done there using <u>P</u>. <u>persimilis</u> and <u>Stethorus bifidus</u> Kapur to control <u>T</u>. <u>urticae</u> (Charles et al.1985, Charles and White 1988).

1.4 **BIOLOGY OF PHYTOSEIULUS PERSIMILIS**

Phytoseiulus persimilis was originally described from Algeria by Athias-Henriot in 1957. Dosse (1958) reported a new species from Chile which he named <u>Phytoseiulus riegeli</u> Dosse. The Chilean species was variously referred to as either <u>P. riegeli</u> or <u>P. persimilis</u>. A third stock, from Sicily, was classified as <u>P. tardi</u> Lombardini in 1959. Based on taxonomic and cross-breeding studies of all three stocks, Kennett and Caltagirone (1968) concluded that the Sicilian, Chilean, and Algerian populations represented segments of one species, with the synonymy of <u>Phytoseiulus persimilis</u> Athias-Henriot. <u>P. persimilis</u> is also apparently indigenous in Libya, Tunisia, southern France, and Italy (Tanigoshi 1982). It has now been established in several other countries including Israel, Australia, and in the U.S.A. (McMurtry 1982).

Developmental times of a Chilean stock of <u>P</u>. <u>persimilis</u> at a diurnal temperature cycle of 58 to $83^{\circ}F$ (average temperature of $68.5^{\circ}F$) and humidity ranging from 65 to 95% were determined by Laing (1968) to be as follows for males and females respectively: egg - 3.1 days for both, larva - 1.0 day for both, protonymph - 1.7 and 1.6 days, and deutonymph - 1.7 days for both. Total developmental time from egg to adult was 7.5 days for males and 7.4 days for females. The preovipositional period for females was 3.0 days, and the ovipositional period was 22.3 days. Adult females lived an average of 29.6 days, and laid an average of 2.4 eggs per day for a total of 53.5 eggs on average during their lifetimes. The maximum for an individual was 101, the minimum 14. The immature stages of <u>P</u>. <u>persimilis</u> ate an average of 20.5 <u>T</u>. <u>urticae</u> eggs in total, and adult females ate an average of 25.5 eggs per day with the majority of these (14.3 eggs per day) being consumed during the ovipositional period. Table 1 compares the life history of <u>P</u>. <u>persimilis</u> with that of <u>T</u>. <u>urticae</u>.

Sex	Stage	Duration of stage in days		
		(mean ± SD)		
	- -	<u>T. urticae</u>	<u>P. persimilis</u>	
Male	Egg	6.7 ± 0.5	3.1 ± 0.2	
Female	Egg	6.7 ± 0.4	3.1 ± 0.2	
	•			
Male	Larva	3.6 ± 0.8	1.0 ± 0.1	
Female	Larva	3.7 ± 1.0	1.0 ± 0.1	
Male	Protonymph	2.7 ± 0.6	1.7 ± 0.2	
Female	Protonymph	3.0 ± 0.5	1.6 ± 0.2	
Male	Deutonymph	3.1 ± 0.6	1.7 ± 0.1	
Female	Deutonymph	3.5 <u>+</u> 0.6	1.7 ± 0.1	
Total:male	Egg-adult	16.1 ± 1.3	7.5 ± 0.1	
Total:female	Egg-adult	16.9 ± 1.4	7.4 ± 0.1	
Female	Preovipositional	2.1 ± 0.9	3.0 ± 0.6	
	•			
Female	Ovipositional	15.7	22.3	
	•			
Female	Adult		29.6	
Female	Ecos laid per dav	2.1 ± 0.7	2.4 ± 0.3	
Female	Total egos laid	37.9	53.5	
	,			

Table 1. Comparative developmental times of T. urticae and P. persimilis and number of eggs laid (after Laing, 1968 & 1969)

<u>Phytogeiulus persimilis</u> has a specific food requirement for tetranychid mites, and has not been found to consume any alternative foods (Dosse 1958, Mori and Chant 1966). Researchers have attempted to produce an acceptable artificial diet but have not succeeded (Kennett and Hamai 1980).

Phytoseiulus persimilis detects colonies of spider mites by responding to kairomones emitted by the prey and its products (Sabelis and van de Baan 1984). Using a Y-tube olfactometer, they found that extracts of exploited leaf surfaces, black fecal pellets, and eqgs of T. urticae attracted P. persimilis. Neither the silk webbing nor spider mite exuviae were associated with these kairomones (Sabelis and Dick 1985). Other kairomones may be involved in the arrestment response of P. persimilis to its prey, and in feeding stimulation. Hislop and Prokopy (1981) found that T. urticae webbing and its associated faeces caused an arrestment response in P. macropolis Banks and Amblyseius fallacis even when it was up to 7 days old. Jackson and Ford (1973) found that P. persimilis females preferred to consume unwashed T. urticae eggs rather than eggs washed in distilled water, and hypothesized the existence of a feeding stimulant on the eggs. These studies support the hypothesis put forth by de Moraes and McMurtry (1985) that the attractant kairomones are probably volatile chemicals, while the arrestment and stimulant kairomones may be more stable non-volatile chemicals.

Generally speaking, phytoseiids are not as tolerant of low humidities and high temperatures as are tetranychids (Sabelis 1985). Stenseth (1979) found that both egg viability and the ability of <u>P</u>. <u>persimilis</u> to control a population of <u>T</u>. <u>urticae</u> were reduced at 40% RH as compared to 80% at a constant temperature of 27° C.

Only fertilized phytoseiid females overwinter in temperate regions (McMurtry et al.1970), either in foliage or at the base of plants under debris. However, as <u>P</u>. <u>persimilis</u> is a subtropical species, it remains

active throughout the year in its native environment and does not survive the winter in temperate climates. This can be a problem if it is introduced in a temperate region, because permanent establishment is not possible.

Phytoseiids disperse short distances by walking, as do tetranychids. Bernstein (1983) found that P. persimilis could walk at a mean speed of 0.149 cm/sec, compared to \underline{T} . urticae which travelled at a mean speed of 0.107 cm/sec. He concluded it could easily disperse throughout a greenhouse at this rate. Outdoors, phytoseiid ambulatory and aerial dispersal methods are similar to those of tetranychids. Hoy et al.(1985) found that M. occidentalis females dispersed from almond orchards a distance of 200 m on prevailing winds, and that most dispersal occurred between 16 and 22 h when relative humidity and wind speeds had increased and temperature had decreased. Wind tunnel experiments with P. persimilis revealed that they do not lift up their first and second pair of legs for take-off as do tetranychids, so some other mechanism must be involved (Sabelis et al.1983). P. persimilis disperses in response to low prey density, but may sometimes remain in an area of spider mite webbing for a day or more after the spider mites have all been removed (Charles and White, 1988; Sabelis et al. 1983). This suggests that a nonvolatile arrestment kairomone still present in the webbing is delaying dispersal.

Desirable characteristics in a predator chosen for biological control include the following (Rosen and Huffaker 1983, Beckendorf and Hoy 1985):

- 1) Adaptation to the environment
- 2) Good dispersal and searching ability
- 3) Prey specificity
- 4) High power of increase
- 5) Pesticide resistance or tolerance
- 6) Is easily and economically reared

Biological control is defined as the suppression of a host or prey by its natural enemies (Rosen and Huffaker 1983). <u>Phytoseiulus persimilis</u> is considered to be an effective biological control agent because it possesses several of these characteristics.

Adaptation to the Environment

Phytoseiulus persimilis is adaptable to many crops and environments in its temperature range. It has become established in countries where it was not originally native, such as California and New Zealand. In the present experiment it established readily in raspberries. However, its inability to overwinter because of its subtropical origins is a disadvantage in this climate. It means reintroduction each year would be necessary, and it cannot be introduced too early in the spring when the weather is still cool. Perhaps this disadvantage could be overcome by selecting for cold tolerant strains. However, a cold tolerant strain from New Zealand was tried in strawberries in British Columbia and failed to overwinter (D. Raworth, pers. comm.).

Dispersal and Searching Ability

Because spider mites are a transient food source due to their tendency to overexploit their hosts, their phytoseiid predators have been selected for good dispersal abilities (Sabelis and Dicke 1985). <u>Phytoseiulus persimilis</u> is a very mobile predator and disperses readily within crops in greenhouses and fields and into fields from weed reservoirs (Oatman and McMurtry 1966; McMurtry et al. 1978). At low prey densities, dispersal ability needs to be combined with good searching ability for the predator to survive (Huffaker et al. 1970), and this is very well developed in <u>P. persimilis</u>. It is attracted by chemical cues produced by the prey, and once it has found an area of spider mite

colonies it tends to stay until the prey is eaten (Sabelis et al. 1983).

Prey Specificity

Specialized predators are superior in the their ability to control prey populations compared to generalists because they are more synchronized in their habits, life cycle and dispersion patterns with their prey. However, generalists are better able to maintain themselves when prey is scarce because they can eat alternate foods. Phytoseiulus persimilis is a specialized predator of tetranychids. It is attracted to and aggregates more intensively in areas of high prey density, where it oviposits selectively. The immature predators are positioned to eat prey as soon as they have hatched. It is also very voracious; ovipositing females consume 14 or more eggs a day (Laing, 1968). It has the ability to reduce pest populations very quickly, but this can also be a disadvantage because elimination of the prey leads to its own starvation (Takafuji, 1977). Huffaker and Flaherty (1966) considered a voracious predator to be less suitable because it requires more prey and so cannot prevent outbreaks. It may cause a faster decline in prey numbers, but then it cannot maintain its own numbers on few prey, so must disperse to another location. This kind of predator needs to be repeatedly introduced. At low prey density, a predator that could survive by just replacing itself and being able to find scarce prey would have an advantage, but P. persimilis cannot survive at low prey densities in greenhouses.

Power of Increase

A "good" predator must be able to respond quickly to increases in the prey population (Huffaker et al. 1969). For long-term control of a prey species to be successful, the numerical response of the predator is probably more important than the functional response (Huffaker et al.

1970). <u>Phytoseiulus persimilis</u> is superior in its ability to increase rapidly, due to its short generation time and high fecundity. It is thus able to overtake and suppress a population of <u>T</u>. <u>urticae</u> that is initially more numerous than its own.

Pesticide Resistance

In many agricultural systems, the use of chemicals must be harmonized with the use of natural enemies. Pesticides will have to be used selectively, along with genetically improved natural enemies (Hoy 1985). In the field, selection pressure from regular applications of insecticides usually leads to the development of resistance in the prev faster than in the natural enemies (Croft and Strickler, 1983), and this is true in mites. Tetranychus urticae is resistant to most of the acaricides that are frequently used, while the predators are still susceptible, and this is also the case with many organophosphates used for other pests. However, even though resistance is slow to develop in the field, some native predators such as Amblyseius fallacis in British Columbia and Metaseiulus occidentalis in California do possess some natural resistance to several organophosphate and carbamate insecticides as well as to acaricides (Raworth 1990, Hoy 1983). This resistance can be enhanced in the lab (Hoy 1983), although the use of genetically improved natural enemies requires the continued use of pesticides in order to maintain selection pressure.

The lag in resistance development in phytoseiids is hypothesized to be due not to any inherent differences in their capacity to develop resistance as compared to tetranychids, but rather to starvation due to a scarcity of prey. Morse and Croft (1981) found that <u>A</u>. <u>fallacis</u> developed resistance as quickly as <u>T</u>. <u>urticae</u> when adequate prey was available, but when it was not, resistance developed much later. Since heavy chemical use in the field usually leads to rapid development of resistance in prey and

extinction in predators, a program of reduced insecticide use combined with artificial augmentation of the food supply of the predator would result in decreasing the rate at which resistance develops in the prey while allowing it to develop in the predator (Tabashnik 1986).

Mass Rearing Technology

Large-scale rearing of P. persimilis is expensive because artificial diets have not yet been devloped (Kennett and Hamai 1980). Since P. persimilis has specific prey requirements, it must be raised with T. urticae as its food. Mass rearing methods usually involve production of large numbers of T. urticae on pinto bean plants in a greenhouse, and subsequent inoculation of these plant-prey systems with predators. Hoy et al. (1982a) described the production of 1.5 million M. occidentalis in three months in a greenhouse using this method. These authors also reared 62 million predators in a 0.2 ha soybean field in the same time period. Alternatively, phytoseiid predators can be mass reared using small enclosed containers, such as stacked cylinders, to which leaves infested with prey are added regularly. The predators are harvested by removing the cylinders and trapping the mites in bran. Fournier et al. (1985) reared 500-2000 adult P. persimilis per day using this method. With both methods, the main constraints that exist are the necessity for a continuous supply of healthy bean plants and tetranychid prey. In temperate climates, the cost can be high because greenhouses are required to rear subtropical species such as P. persimilis, as well as the T. urticae needed for food. In warmer climates, phytoseiids can be reared more economically (Koppert 1980).

1.4 COMPARISON OF P. PERSIMILIS WITH TWO OTHER PHYTOSEIID PREDATORS

Two other phytoseiid predators, namely Metaseiulus occidentalis and <u>Amblyseius</u> <u>fallacis</u> are potential biological control agents of <u>T.</u> <u>urticae</u> on raspberries. Laing and Huffaker (1969) compared P. persimilis and M. occidentalis with respect to their ability to control T. urticae on strawberries in a greenhouse. Developmental times were shorter for P. persimilis, - from egg to adult took 9 days compared to 13 days for M. occidentalis - and P. persimilis ate more prey - 7.3 eggs per adult per day compared to 3.8 eggs for M. occidentalis. Phytoseiulus persimilis laid slightly more eggs per day (2.4 compared to 2.2 for M. occidentalis), and the generation time was about the same for both. The combination of higher voraciousness, shorter developmental time, and greater fecundity resulted in <u>P. persimilis</u> being superior at controlling high T. urticae populations. Metaseiulus occidentalis allowed prey numbers to go much higher before it overtook them, and it also had a longer lag period. Metaseiulus occidentalis was considered to be better at long-term regulation of prey populations at low density; it survived self-generated prey population crashes whereas P. persimilis did not. This was due to the fact that M. occidentalis did not overexploit the prey as did P. persimilis, and because it could survive on alternate prey or plant foods.

McMurtry (1982), in a comparison of the same two species as well as <u>Amblyseius fallacis</u>, found that they all had high dispersal powers, ability to distribute themselves in relation to their prey, and high reproductive potential. <u>Phytoseiulus persimilis</u> was highly voracious, eating 14-23 eggs a day, while the other two ate about 8 eggs each. Both <u>M. occidentalis</u> and <u>A. fallacis</u> ate alternate food; <u>M. occidentalis</u> ate eriophyids, tydeids, thrips, and scale crawlers, while <u>A. fallacis</u> ate <u>P</u>. <u>ulmi</u> and pollen. <u>Phytoseiulus persimilis</u> did not survive when prey was scarce; the other two were more able to do so.

Gilstrap and Friese (1985) compared A. californicus with the other

two predators and considered <u>P</u>. <u>persimilis</u> to be superior. In addition to the traits listed above, they added its longer reproductive period, faster rate of movement, and higher ratio of prey killed to prey contacted. Oatman et al. (1977) came to similar conclusions regarding the same three predators in strawberries. Raworth (1990) found <u>A</u>. <u>fallacis</u> to be the predominant predator of <u>T</u>. <u>urticae</u> in strawberry fields in Abbotsford, B.C. It responded numerically to population increases of <u>T</u>. <u>urticae</u>, and was resistant to several pesticides. These qualities, in addition to its ability to overwinter and its presence with its prey early in the spring and late in the fall, make it a good candidate for a biocontrol agent.

In summary, <u>P</u>. <u>persimilis</u> would appear to be a better candidate for quick short-term control of a medium to high <u>T</u>. <u>urticae</u> population in a field situation, but might have to be reintroduced if the population crashes. This could be prevented, however, by careful monitoring of the predator and prey numbers, and augmentation of the prey before it is completely eliminated. The other major disadvantage of <u>P</u>. <u>persimilis</u>, that it is adapted to warm climates and does not tolerate the cooler temperatures here, could perhaps be overcome by a genetic selection program. On the other hand, it might be preferable to use either of the other two predators, which can provide longer-term control and will overwinter, thus requiring fewer introductions. However, at present <u>P</u>. <u>persimilis</u> is the most easily obtained predator from the suppliers that rear phytoseiids, and it was the only available one when the present experiment was conducted.

1.5 **OBJECTIVE**

The objective of this research was to examine the efficacy of <u>Phytoseiulus</u> <u>persimilis</u> as a biological control agent of <u>Tetranychus</u> <u>urticae</u> on raspberry crops in the Fraser Valley. The null hypothesis is the following: in a raspberry field infested with <u>T</u>. <u>urticae</u>, there will

be no difference in <u>T</u>. <u>urticae</u> numbers between treatment plots to which <u>P</u>. <u>persimilis</u> have been introduced and control plots with no introduced <u>P</u>. <u>persimilis</u>.

<u>CHAPTER 2</u>

METHODOLOGY DEVELOPMENT

2.1 ESTABLISHMENT OF TETRANYCHUS URTICAE

2.1.1 Introduction

Tetranychus urticae populations tend to adapt to a particular host plant so that, if transplanted to another host, they often require a generation or more to adapt fully to the new host and to achieve the optimum reproductive rate (Dabbour 1977). Jesiotr (1979) observed that \underline{T} . urticae, when moved from roses to beans, took six generations to achieve the same reproductive abilities it had on roses. He concluded that the ability to adapt to a new host depended on its relative food quality compared to the original host, and on genetic characteristics (Jesiotr 1980). Since the \underline{T} . urticae used in this study were originally raised on pinto bean plants and then transplanted to raspberries, it was necessary to find out whether they would require a period of adjustment when inoculated onto a new host before proceeding with the field experiment.

2.1.2 <u>Materials</u> and <u>Methods</u>

A preliminary trial was undertaken to compare the establishment in the field of <u>T</u>. <u>urticae</u> raised on raspberry leaves with those raised on pinto bean (<u>Phaseolus vulgaris</u>) leaves. Potted raspberry cuttings from a garden shop were infested with <u>T</u>. <u>urticae</u> and kept in a growth room at 22° C. When the population of <u>T</u>. <u>urticae</u> reached 10-15 mites per leaflet, eight clip cages were set up (D.R. Gillespie)¹. Four clip cages contained leaf pieces of raspberries with 5-8 mites each, and four contained pieces

¹ Agriculture Canada Research Station, Agassiz, B.C. Canada

of pinto bean leaves with similar numbers of mites. These cages were clipped onto fresh raspberry leaves in the field. The original leaf piece dried up within 1-2 days. Because the mites could not escape from the cages they had a choice of either colonizing the fresh raspberry leaf or dying. One clip cage was set up per row, in 8 rows, on randomly selected plants. The cages were removed after 9 days along with the enclosed raspberry leaf, and the number of eggs and active stages on each leaf were counted under a dissecting microscope.

2.1.3 <u>Results and Discussion</u>

Tetranychus urticae established as well on raspberries when the original host was pinto bean plants as they did when the original host was potted raspberry plants (Table 2). The number of eggs and active stages produced by mites that had come from pinto beans was higher than that produced by counterparts coming from raspberries, although the differences were not significant (t-test, p>0.05). This indicates that this change in the host plant did not adversely affect the reproductive potential of <u>T</u>. urticae. One reason for this might be that the potted raspberries, which were rather stunted in their growth, were not an adequate source of food for <u>T</u>. urticae, whereas field grown raspberries were. Alternatively, this population of the mite may be genetically non-selective in its host preferences, and able to adapt quickly to a wide range of potential hosts.

2.2 CALIBRATION OF THE MITE-BRUSHING MACHINE

2.2.1 Introduction

Direct counting of mites under a dissecting microscope can be more time consuming than indirect methods, especially on raspberry leaves which have many ribs and hairs in which mites can be hidden . A mite-brushing

Table 2. Numbers of T. urticae on clip-caged raspberry leavesat Agassiz, 9 days after inoculation with stockcolonies reared on two different host plants

	<u></u>	<u>Original h</u>	ost		
	Ras	oberry	Bea	n	
<u>Cage #</u>	Eggs	<u>Active</u> stages	Eggs	<u>Active</u> stages	
1	28	13	67	9	
2	2	3	24	1	
3	16	14	12	19	
4	5	7	21	46	
Mean	12.8	9.2	31.0	18.8	
SE	5.9	2.6	12.3	9.8	

machine (Henderson and McBurnie 1943) is widely used to estimate mite populations. Leaves are passed through rotating brushes, and the mites that are brushed off fall on to a revolving disc coated with a thin layer of vaseline to immobilize any active mites. The disc is divided into black and white sections of equal area, any number of which may be counted depending on the number of mites. Brushing has the advantage of removing mites from leaves without damaging them, and of permitting easier identification of mites than some of the other methods. Information on the validity of counting mites using the mite-brushing machine was needed for the subsequent field experiment, in which mites were to be counted using this method. If there were not a linear relationship between counts obtained by this method and direct counting, then an adjustment of brushed counts would have to be made according to the mathematical relationship that existed between counts obtained with the two methods. The objective of this experiment was thus to determine whether the mite-brushing method gave a good estimate of the sample population by comparing it with direct counting.

2.2.2 <u>Materials and Methods</u>

Leaflets were picked randomly from raspberry plants at Agassiz that had been infested with <u>T</u>. <u>urticae</u>. These leaves were placed in a plastic bag and taken to the lab where they were stored at 5° C until they were counted, usually within one or two days. <u>T</u>. <u>urticae</u> active stages on each leaflet were counted directly using a dissecting microscope. After counting, each leaflet was passed through the mite-brushing machine according to the method of Henderson and McBurnie (1943), and the number of <u>T</u>. <u>urticae</u> on the glass plate was recorded. A total of 26 leaflets was examined. Regression analysis was performed on the data.

2.2.3 Results and Discussion

The regression of brushed mite counts on direct counts was linear (Table 3; Figure 1a, $r^2=0.958$, p<0.01, $y = 0.85x \pm 8.14$). When a quadratic factor was added to the equation, it was not significant (p = 0.50). The x-intercept was not significantly different from zero (p = 0.56). Because of the linearity of the relationship, there was no need to calibrate the brushed mite counts in the subsequent field release experiment. Regression analysis was also done on the same data but omitting the top five points because these were well out of the range of mite counts encountered in the field releases (Fig 1b). The relationship was still linear ($r^2=0.77$, p<0.01, $y = 0.82x \pm 7.74$).

Table 3. Regression analysis of T. urticae numbers comparing direct counts with mite-brushing machine counts

Dependent variable: direct counts , n = 26								
<u>Variable</u>	DF	<u>Estimate</u>	SE	I	<u>e</u>			
Intercept	1	8.14	5.35	1.52	0.14			
Slope	1	0.85	0.04	23.48	0.00			
r2= 0.958, SE	r2= 0.958, SE= 23.45							
		Analysis of Va	nance					
Source	DF	<u>Sums of</u> Squares	<u>Mean</u> square	<u>F value</u>	P			
Regression	1	303083.62	303083.62	551.10	0.0001			
Residual	24	13199.04	549.96					
Total	25							

Figure 1. Relationship between direct mite counts and counts obtained using the mite-brushing machine.





CHAPTER 3

FIELD RELEASE OF PHYTOSEIULUS PERSIMILIS

2.3.1 <u>Introduction</u>

Information from the literature on the release rates of predators and the timing of release was used to design this experiment. Laing and Huffaker (1969) modelled different introduction ratios of <u>P</u>. <u>persimilis</u>; with an introduction ratio of 1 predator to 10 <u>T</u>. <u>urticae</u> the prey would theoretically be eradicated in 9 days, whereas a ratio of 1:100 would produce eradication in 30 days. Croft (1976) advised the release of phytoseiids when prey mites were present at a density of 1-5 per leaf. In addition, he felt that predators should be released on the same leaves, or very close to the same leaves, as the prey because dispersal could be a problem. Hoy et al. (1982a) stated that they considered optimal spider mite/predator ratios to be between 20 and 40 spider mites per predator in order for the predator to achieve unlimited growth.

Most research supports the view that early introduction of predators is more effective than waiting until damage is visible. Oatman and McMurtry (1966) found that it was more effective in strawberries to release the predator before the prey reached an average of 1 per leaflet. Pickett and Gilstrap (1986) found that releases of <u>P</u>. <u>persimilis</u> made earlier in the season regulated spider mite populations better than those made a few weeks later, and reasoned that the ability to reproduce and disperse from release areas before spider mite populations reach significant densities gives them the advantage they need to exert efficient control. French et al. (1976) used a leaf damage index scale from 0 to 5 to describe <u>T</u>. <u>urticae</u> damage on tomatoes in a greenhouse. If <u>P</u>. <u>persimilis</u> was introduced when this index was 1.0 it failed to control spider mites; however, good control was achieved when the damage index was 0.1 at introduction.

The objective of this experiment was to release three rates of \underline{P} . <u>persimilis</u> into raspberry subplots containing <u>T</u>. <u>urticae</u> in order to determine its effectiveness as a predator.

2.3.2 <u>Materials and Methods</u>

A raspberry plot at the Agassiz Research Station of Agriculture Canada was used for this experiment. The plot consisted of 9 rows of red raspberries, Rubus idaeus L. ; each row was divided into 4 subrows, each 8 m long and separated from each other by an alley 2 m wide. There were 2 m between rows. Alternate rows were used, thus leaving a buffer of a row of untreated plants between each row of treated plants. The experiment was set up in a randomized complete block design. Within each row, each of 4 treatments was randomly assigned to one of the subrows by picking numbers out of a hat. Treatments were replicated 5 times, once in each of the 5 rows. T. urticae were introduced into the plot because there was no natural population. They were obtained from Applied Bionomics² and were introduced on May 11, 1989. The mites were supplied on pinto bean leaves, and were distributed by placing pieces of bean leaves directly onto raspberry leaves. One week later sampling resulted in a count of 3 Т. urticae per 500 leaflets. The weather was cold and wet and the mites did not establish well. They were reintroduced on July 9 and again on July 27.

Phytoseiulus persimilis predators were introduced on August 2 when sampling indicated that the <u>T. urticae</u> population was 0.70 mites per leaflet. A total of 8225 female predators were distributed at three different predator/prey ratios, namely 1:50 (Treatment A), 1:100 (Treatment B), and 1:200 (Treatment C). The fourth experimental unit (Treatment D) was a control and received no predators. <u>Phytoseiulus</u> persimilis was supplied by Applied Bionomics² at a cost of \$30 per

². Box 2637, Sidney, B.C. Canada V8L 4C1

thousand, and came on pinto bean leaves together with <u>T. urticae</u>. Numbers of predators released were verified by counting the <u>P. persimilis</u> on five leaf pieces and averaging the counts. They were released by stapling pieces of the pinto bean leaves onto raspberry leaves. Calculations were done as follows: at 0.70 mites per leaflet and 5600 leaflets per stool, there were 3920 <u>T. urticae</u> per stool. At a ratio of 1:50, 78 predators were needed per stool; at 1:100, 39 were needed; at 1:200, 20 were needed. There was an average of 30 <u>P. persimilis</u> on each pinto bean leaf piece, so 1:50 required 2.5 pieces, 1:100 required 1.33 pieces, and 1:200 required 0.67 of a piece. The leaf pieces were placed midway up each plant in the centre of the stool.

Tetranychus urticae and P. persimilis populations were sampled on August 13 and at weekly intervals thereafter until October 1, at which time many T. urticae females had attained the orange colour that signals the onset of diapause. Each sample consisted of 30 leaflets picked from each treatment replicate in each row, so that 600 leaflets were picked on each sample date. Sampling was done so that 10 leaflets came from the top third of the plants, 10 from the middle, and 10 from the bottom, but they were not counted separately. Leaves on which P. persimilis had been released were not taken as samples. Each 30-leaflet sample was bagged separately in plastic bags, transported to the lab, and maintained at 5° C until the mites were counted. Counting was usually done within 2 days. Active stages of both the predator and the prey were removed from the leaflets with the mite-brushing machine. They were then counted under a dissecting microscope, using the black and white grid under the plate to facilitate counting. When counting T. urticae, only the black sections were counted; when counting P. persimilis, the whole plate was counted. The data were expressed as mites per leaflet and analyzed using the SAS GLM Anova procedure with repeated measures, and linear and multiple regression. Parametric statistics were permitted since the data were determined to be normally distributed.

Observations were also made on native predators present in the plot. A few (3 per 600 leaflets) <u>Stethorus punctum</u> larvae were observed before the predators were added, and to avoid any influence they might have on the <u>T</u>. <u>urticae</u> population (since they are mite predators), the field was sprayed on July 25 with Malathion to eliminate these predators.

2.3.3 Results

Population Trends

In the three treatments and the control, the population trends of <u>T. urticae</u> followed a similar pattern (Figure 2). There was a peak at 4.5 weeks for the three treatments, and a week later for the control. Numbers of both <u>T. urticae</u> and <u>P. persimilis</u> remained at low levels throughout the experiment. Means of the three treatments and the control for <u>T. urticae</u> over time are given in Table 4. Mite counts were similar in all three treatments and the control 2.5 weeks after introduction of the predators, and then they diverged. The controls had a maximum population of <u>T. urticae</u> per leaflet in week 4.5 (September 3) of 6.77 ± 1.52 mites per leaflet, while Treatments A, B, and C peaked at 3.57 ± 1.21 , 3.47 ± 0.49 , and 3.99 ± 1.31 respectively in week 3.5 (August 27).

<u>Phytoseiulus persimilis</u> became established in the treatment subplots at low levels, and followed a pattern of population fluctuations similar to those of <u>T</u>. <u>urticae</u> (Table 5, Figure 3). Peak predator numbers in week 3.5 (August 27) for the three treatments were 0.093 ± 0.025 , 0.100 \pm 0.060, and 0.080 \pm 0.023 respectively, and 0.013 \pm 0.008 for the

<u>Predator/prey</u> <u>ratio</u>	<u>Treatment A</u> 1:50	<u>Treatment B</u> 1:100	<u>Treatment C</u> 1:200	Control
<u>Time</u> (weeks)	<u>Mean ± SE</u>	<u>Mean ± SE</u>	<u>Mean ± SE</u>	<u>Mean ± SE</u>
1.5	2.29 ± 0.54	1.62 ± 0.35	2.07 ± 0.27	2.41 ± 0.72
2.5	2.23 ± 0.93	1.73 ± 0.19	1.84 ± 0.26	2.14 ± 0.41
3.5	4.02 ± 1.52	4.87 ± 1.26	6.11 ± 2.32	5.89 ± 1.38
4.5	3.57 ± 1.21	3.47 ± 0.49	4.91 ± 1.37	6.77 ± 1.52
5.5	3.12 🗴 1.39	2.37 ± 0.54	3.99 ± 1.31	4.77 ± 1.18
6.5	3.28 ± 1.44	2.91 ± 0.68	3.67 ± 0.70	4.55 ± 0.94
7.5	3.32 ± 1.35	3.07 ± 0.40	3.85 ± 0.73	4.98 ± 0.99
8.5	2.56 ± 1.02	1.93 ± 0.50	3.24 ± 1.18	4.19 ± 1.23

Table 4. Mean numbers of T. urticae per leaflet over time.Means are an average of five replicates.

Figure 2. Numbers of <u>T</u>. <u>urticae</u> per leaflet as a function of time. Each point is a mean of five replicates. Average SE = 4.97. Initial predator/prey ratios were: Treatment A - 1:50, Treatment B -1:100, Treatment C - 1:200, Treatment D - control.



Time in weeks

<u>Predator/prey</u> ratio	<u>Treatment A</u> 1:50	<u>Treatment B</u> 1:100	<u>Treatment C</u> 1:200	<u>Treatment D</u>
<u>Time</u> (weeks)	<u>Mean±SE</u>	<u>Mean ± SE</u>	Mean ± SE	<u>Mean ± SE</u>
1.5	0.010 ± 0.010	0.000 ± 0.000	0.010 ± 0.010	0.020 ± 0.020
2.5	0.040 ± 0.012	0.033 ± 0.000	0.013 ± 0.013	0.000 ± 0.000
3.5	0.093 ± 0.025	0.100 ± 0.060	0.080 ± 0.023	0.013 ± 0.008
4.5	0.053 ± 0.029	0.067 ± 0.024	0.060 ± 0.027	0.040 ± 0.016
5.5	0.053±0.031	0.033 ± 0.021	0.047 ± 0.025	0.013 ± 0.008
6.5	0.027 ± 0.019	0.013 ± 0.008	0.020 ± 0.008	0.000 ± 0.000
7.5	0.047 <u>+</u> 0.023	0.000 ± 0.000	0.047 ± 0.031	0.033 ± 0.033
8.5	0.033 ± 0.033	0.007 ± 0.007	0.013 ± 0.008	0.013±0.013

Table 5. Mean numbers of P. persimilis per leaflet over time.Means are an average of five replicates.

Figure 3. Numbers of <u>P</u>. <u>persimilis</u> per leaflet as a function of time. Each point is a mean of five replicates. Average SE = 0.011. Initial predator/prey ratios were: Treatment A - 1:50, Treatment B - 1:100, Treatment C - 1:200, Treatment D control.



Time in weeks

Figure 4. Relationship between <u>T</u>. <u>urticae</u> and <u>P</u>. <u>persimilis</u> numbers in three treatments and the control (a-d). Each point is an average of five replicates.



Mean T. urticae per leaflet

controls (Table 5). <u>Phytoseiulus persimilis</u> were detected in the control subplots early in the experiment, and were present at very low levels until the end.

Numerical Response

The numerical response of <u>P</u>. <u>persimilis</u> to <u>T</u>. <u>urticae</u> for the three treatments and the control was determined using regression analysis (Figure 4 a-d). Each treatment was tested separately. For Treatment A, r^{2} = 0.604, p= 0.023; for Treatment B, r^{2} = 0.608, p= 0.023; for Treatment C, r^{2} = 0.857, p= 0.001; for the control, r^{2} = 0.309 , p= 0.153 . These results indicate that there was a numerical response by <u>P</u>. <u>persimilis</u> to <u>T</u>. <u>urticae</u> at all treatment levels, but not in the control.

Predator/prey introduction ratios

Since there is usually a lag period in the response by prey to a predator introduction, and that period appeared to be two weeks in this experiment (Figure 2), the data from the first two samples were ignored in the statistical comparisons of the <u>T</u>. <u>urticae</u> numbers between treatments and control. Repeated-measures analysis of variance (Table 6) showed overall differences between treatments were not significant (p= 0.11). However, this low p value indicated that differences probably existed at certain times, but did not appear in the overall analysis because it combined all time periods in one analysis. Therefore multiple comparisons were performed to look at differences between treatments and controls at each time period. Contrast analysis using Bonferroni's method (an a *posteriori* multiple comparison method) showed there were significant differences between Treatment A and the control (p=0.05), and between Treatment B and the control (p= 0.032) (Table 6).

Source	DF	<u>SS</u>	MS	£	<u>P</u>
Treatment	3	83.541	27.847	2.50	0.1090
Block	4	355.663	88.916	8.00	0.0020
Time	5	67.622	13.524	6.250	0.0001
Block*trt	12	133.423	11.118		
Time*block	20	61,210	3.061	1.410	0.1519
Time*trt	15	14.737	0.982	0. 450	0.954
Time*block*trt	60	129.903	2.165		

Table 6. Analysis of variance of numbers of T. urticae. The SAS GLM procedure with repeated measures was used, and multiple comparisons were performed using Bonferroni's method.

n=6, SD= 11.118, SE= 4.972

Contrast analysis using Bonferroni's method:

Contrast	DF	<u>SS</u>	MS	E	<u>P>F</u>
Trt A vs B	1	0.649	0.649	0.06	0.813
Trt A vs C	1	14.484	14.484	1.30	0.276
Trt A vs D	1	53.091	53.091	4.77	0.050*
Trt B vs C	1	21.265	21.265	1.91	0.192
Trt B vs D	1	65.480	65.480	5.89	0.032*
Trt C vs D	1	12.114	12.114	1.09	0.317

Analysis of contrast variables by time and treatment: P values:

<u>Contrast</u>	DF	<u>P>F</u> <u>3.5 wk</u>	<u>P>F</u> <u>4.5 wk</u>	<u>P>F</u> <u>5.5 wk</u>	<u>P>F</u> <u>6.5 wk</u>	<u>P>F</u> <u>7.5 wk</u>	<u>P>F</u> 8.5 wk
Trt A vs B	1	0.649	0.933	0.434	0.759	0.790	0.572
Trt A vs C	1	0.271	0.260	0.368	0.745	0.569	0.542
Trt A vs D	1	0.323	0.015*	0.102	0.300	0.090	0.157
Trt B vs C	1	0.506	0.229	0.106	0.530	0.408	0.250
Trt B vs D	1	0.584	0.013*	0.024*	0.186	0.056	0.058
Trt C vs D	1	0.904	0.127	0.419	0.464	0.232	0.400

Figure 5. Relationship between introductory predator/prey ratios and \underline{P} . persimilis numbers.



Predator-prey introduction ratios

At 4.5 weeks there was a significant difference between Treatment A and the control (p=0.015) and between Treatment B and the control (p=0.013). At 5.5 weeks, Treatment B was also significantly different from the control (p=0.024). Differences between Treatment A and the control and between Treatment B and the control continued in the same pattern through the rest of the experiment but were not significant (p>0.05).

There was considerable variability between blocks in the experiment, as indicated by the blocks mean square in the ANOVA table. This could reflect differences in abiotic environmental factors such as humidity levels, temperature, wind factors or soil type.

The relationship between introduction ratios and subsequent numbers of <u>P</u>. <u>persimilis</u> during the course of the experiment was examined. <u>P</u>. <u>persimilis</u> means averaged over time for each treatment were regressed on introduction ratios (Figure 5). The results suggest that the introduction ratios were generally successful, but the relationship was not statistically significant ($r^2 = 0.749$, p = 0.14).

Other Predators

Thrips (not identified) were observed at a rate of 5-9 per 600 leaflets until June 29, after which they were absent. They were thus not present during the field release experiment. Larvae of <u>Stethorus punctum</u> were observed at a rate of 3-4 per 600 leaflets before predators were added. On August 13, one <u>S. punctum</u> larva was observed in 600 leaflets, and on August 27, 6 were observed. Subsequently, none were observed for the rest of the experiment.

2.3.4 Discussion

Population Trends

The graphs of population trends (Figs.2 and 3) show a classical predator/ prey response (Huffaker 1976). Two and a half weeks after the introduction of <u>P. persimilis</u>, prey numbers began to increase. After reaching a peak at 4.5 weeks, numbers of both predators and prey declined slowly with small fluctuations until they reached a plateau that lasted until the termination of the experiment. The population trends for the three treatments and the controls are similar. This conclusion was reached by noting that there was no treatment*time interaction (Table 6), indicating that the four trend lines are parallel to each other. For the control subplots, this pattern is not what would be expected; rather, the population would be expected to continue to increase in the controls for some time after the treatment populations had declined. This population decline in the control subplots probably occurred because of dispersal of predators into them.

Both predator and prey were established at low levels; <u>T</u>. <u>urticae</u> never reached more than 15 mites per leaflet in any single sample. This kind of predator/prey interaction has been shown to be effective in keeping spider mite numbers low for long periods of time, and preventing economic losses. If achieved early in the season, this could lead to season-long prey suppression. Occurring late in the season, as in this experiment, it can prevent fall damage to new canes and an overwintering population of <u>T</u>. <u>urticae</u> from building up (Doughty et al. 1972).

Contamination of controls with predators is a common occurrence in field studies using <u>P</u>. <u>persimilis</u> (Oatman et al. 1968, D. Raworth, pers comm.), and is difficult to prevent even when anticipated. The predator is very mobile and can disperse quite far. It has even been found moving from one greenhouse to another (Dixon 1973), presumably through cracks in the

plastic or carried along by people going from one house to another. In this experiment, each row was separated from the next by 4 m and an intervening row of raspberries; however, the subrows were only separated from each other by 2 m, which perhaps was not enough. In addition, there were other people in the field picking and pruning the raspberries, and they could have carried mites between subplots.

The ability of P. persimilis to disperse in a continuous system is well-known, and can lead to instability in predator/prey intreactions. Takafuji (1977) found that low prey density led to P. persimilis dispersal out of those patches in search of more prey; if a few prey eggs were left behind, the prey could build up to damaging levels again in the absence of the predator. Preventing dispersal led to a longer period of stabilization. In a field situation, however, dispersal of the predator from areas of low prey density to adjacent areas of higher density would be an advantage, and tend to stabilize populations for much longer periods of time.

Numerical Response

A successful outcome of a predator/prey interaction depends on the ability of the predator to distribute itself in the same areas as the prey, and thus to increase its reproductive rate in areas of high prey abundance (Nachman 1981). A high correlation coefficient between predator numbers and prey numbers in a regression analysis indicates that the predator has the ability to increase its numbers in response to an increase in prey numbers. The results of this study indicate that <u>P</u>. <u>persimilis</u> responds very well to the density of <u>T</u>. <u>urticae</u> at low population levels in a raspberry field. Even though the control subplots were not free of <u>P</u>. <u>persimilis</u>, there were significant differences between two of the treatments and the control at two points in time, indicating

that introduction of the predators resulted in lower <u>T</u>. <u>urticae</u> numbers at these times. In Figure 5 it can be seen that numbers of <u>T</u>. <u>urticae</u> peaked a week earlier in the three treatments than in the control, a trend that might have been much more significant had predators been absent from the controls.

Introduction Ratios and Timing

This experiment shows that <u>P</u>. <u>persimilis</u> can establish and survive for at least 8.5 weeks in a raspberry field at an introductory ratio as low as 1:200. Significant treatment effects on two dates indicate that <u>P</u>. <u>persimilis</u> can be effective at both a 1:50 and a 1:100 predator/prey ratio in reducing <u>T</u>. <u>urticae</u> numbers when the population is at its peak. Overall significance treatment effects might have been achieved with a larger sample size and a control plot free of predators. The results are in agreement with much of the research done on <u>P</u>. <u>persimilis</u> which states that, in general, predators should be introduced at a ratio no higher than 1:100 (Hoy et al.1982a, Laing and Huffaker 1969, Croft 1976, Mori et al.1989).

The timing of predator introduction is an important factor in the success of biological control. Greater success is obtained when predators are introduced early, before prey numbers are high (Dixon 1973, Pickett and Gilstrap 1986, Oatman and McMurtry 1966). Theoretically, if the initial prey population density is very high, the numerical response of the predator may reach a plateau before control is achieved, and the prey will escape (Holling 1961). As an action threshold, 0.70 T. <u>urticae</u> per leaflet appeared to work in this experiment, since T. <u>urticae</u> populations never reached high numbers, and there were enough prey to sustain the predators for 8.5 weeks. However, since no other times of introduction were investigated, it cannot be determined whether others might have been equally successful. Oatman and McMurtry (1966) found <u>P</u>. <u>persimilis</u>

succeeded in controlling <u>T</u>. <u>urticae</u> on strawberries when released at 320,000 per acre before the prey reached 1 mite per leaflet. Waite (1988) also achieved good control on strawberries releasing <u>P</u>. <u>persimilis</u> when <u>T</u>. <u>urticae</u> was at 5 mites per leaf.

Economics

On the basis of this experiment, if the ratio of 1 predator to 100 prey is considered optimal, then approximately 80,000 predators per hectare (33,500 per acre) would be needed in a raspberry field with a <u>T</u>. <u>urticae</u> population of 0.70 mites per leaflet. The cost of this would be \$2000 per hectare (\$810 per acre) at a predator cost of \$25 per 1,000. Growers paid \$74 per hectare (\$30 per acre) in 1989 for Kelthane (Ward Strong, pers. comm.), but treatment with it is not effective. Predators are still expensive compared to miticides but their cost is decreasing. This year (1992), Applied Bionomics will offer predators at \$10 per 1,000, which would lower the cost to \$800 per hectare (\$324 per acre). Another supplier (Koppert's) sells predators for \$7 per 1,000 (D. Raworth, pers. comm.). If a predator such as <u>Amblyseius fallacis</u>, which overwinters in this climate, were chosen then further savings could be realized by spreading the cost over several years, as long as reintroduction was not necessary.

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

The factors that produce a stable predator/prey system are complex and not all well understood. Modelling predator/prey relationships can provide a background of information and predictive hypotheses against which experimentation can be done, but ultimately a biological agent has to perform in the field. The results of this study show that inoculative releases of <u>P</u>. <u>persimilis</u> on raspberries is a viable approach to controlling <u>T</u>. <u>urticae</u>. Reductions in the cost of rearing predators will make them more affordable to commercial growers.

The aim of biological control is to reduce or eliminate the need for pesticides. It has been indicated by this study, and supported by much of the literature referred to, that predatory mites can control twospotted spider mites, as they did before spider mites became pests. However, the need for pesticides to control the other pests of raspberries interferes with the use of mite predators, which are susceptible to many of these chemicals. One solution to this problem is to develop and use pesticide resistant enemies and integrate their use with the use of pesticides by the careful manipulation of application rates, timing, and methods, and the monitoring of population levels. Another solution is to develop alternative methods of biological control for these other pests. Work is currently underway in the Fraser Valley on control of other pests using appropriate biological agents (D. Henderson, pers.comm.).

If phytoseiids are used commercially to control two-spotted mites, it is important to maintain them in the crop all season long to ensure control in the next year. As was shown by Cross (1984), the presence of predators throughout a season can result in greatly reduced spider mite populations the following year, even if the predator dies over the winter, because the overwintering population of spider mites will be lower. For this reason, if a grower cannot release predators in the spring because of

their incompatibilty with the pesticides he is using, he could still achieve good control on raspberries with a post-harvest release.

The method of sampling <u>T</u>. <u>urticae</u> used in this study would not be efficient in a commercial release of predators. More efficient methods have been used by other researchers, and consist of either counting the number of leaves with a certain threshold number of mites (Cross 1984), or observing leaflets for the presence or absence of mites and referring to a table that gives the corresponding density (Raworth 1986).

In addition to the sampling method, the method of releasing predators must also be done more efficiently. Putting predators out on pieces of bean leaves by hand is very time consuming, but it could be combined with another spring raspberry management operation such as pruning. On a larger scale, Pickett et al. (1987) achieved a uniform to random distribution of <u>P. persimilis</u> by releasing it into a corn field using a conventional light aircraft.

In this climate, both <u>Amblyseius fallacis</u>, which is native to the Fraser Valley, and <u>Metaseiulus occidentalis</u>, which is native to the Okanagan, would survive the winter and the cool spring better than <u>P</u>. <u>persimilis</u>. Whether or not either of them could successfully control a rapidly increasing <u>T</u>. <u>urticae</u> population in raspberries here in the Fraser Valley is not known. Research is currently being done with <u>A</u>. <u>fallacis</u> on strawberries (D. Raworth, pers.comm.), but not in raspberries.

In conclusion, <u>Phytoseiulus persimilis</u> has a number of qualities that make it suitable as a biological control agent of <u>Tetranychus</u> <u>urticae</u> on raspberries in the Fraser Valley. It is very voracious, is superior in its abilty to increase rapidly, has good searching ability for the prey for which it is specialized, and disperses well in the field. Its main disadvantages are its tendency to reduce prey to low levels so quickly that it dies itself from lack of food, and its inability to overwinter in this climate. In this study, <u>P. persimilis</u> showed a potential ability to lower <u>T. urticae</u> populations in an unsprayed field at

a release rate of 1 predator to 100 prey, when the initial prey population was less than 1 mite per leaflet. The lack of controls that were completely free of predators prevents making definitive conclusions. Conclusions can also not be drawn about the ability of <u>P</u>. persimilis to control an early infestation of <u>T</u>. <u>urticae</u> or an initially high prey population without further field studies , but my study indicates that further research in the use of <u>P</u>. persimilis, or other phytoseiid predators, to control two-spotted spider mites in raspberries would be productive.

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