

DISSERTATION

IRIS YELLOW SPOT VIRUS IN COLORADO ONIONS: A SURVEY OF ITS
SPATIAL DISTRIBUTION AND TECHNIQUES TO MANAGE THE PEST

Submitted by

Scott M. Fichtner

Department of Bioagricultural Sciences
and Pest Management

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Committee on Graduate Work

Leisha Jankel
Michael Bartolo

Harold F. Schwartz
Advisor

Greg Hill
Co-Advisor

Thomas O. Holt
Department Head

ABSTRACT OF DISSERTATION

IRIS YELLOW SPOT VIRUS IN COLORADO ONIONS: A SURVEY OF ITS SPATIAL DISTRIBUTION AND TECHNIQUES TO MANAGE THE PEST

The *Iris yellow spot virus* (IYSV) is a new, and at times devastating pathogen of alliums throughout the U.S. as well as many other countries. Since its discovery in Colorado in 2001, IYSV has been identified in nearly all of the major onion producing regions within the state. The severity of this virus disease appears to fluctuate from one year to the next but incidence continues to increase with newly infested fields identified each year. With the number of outbreaks on the rise and the inadequacy of current control strategies, new management techniques as well as novel chemistry pesticides have become a major focus for the management of this virus disease and its thrips vector. Our trials demonstrated the use of reflective materials such as straw or silver reflective mulch can result in a reduction in thrips populations by as much as 69% on onions and a reduction of nearly 9% in IYSV disease incidence. Additionally, Entrust (Spinosad) and Aza-Direct (Neem extract) were found to work as well as or better than conventional materials such as Warrior (Pyrethroid) and Lannate (Carbamate). To better understand the epidemiology of the IYSV pathogen, we also conducted an extensive survey at several locations along the Colorado Front Range and Western Slope. In our surveys, we collected information including thrips populations and incidence of IYSV using 0.2 ha grids developed using mapping software (MapInfo) creating several randomly chosen plots in each field. With data collected on several sampling dates, we attempted to identify a spatial correlation of within field spread of the virus during the growing season. The levels of positive spatial autocorrelation from our survey locations were minimal,

this leads us to believe that secondary outbreaks of the disease are occurring in a random fashion across the field.

Scott Michael Fichtner
Department of Bioagricultural Sciences and Pest Management
Colorado State University
Fort Collins, CO 80523
Spring 2007

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Chapter I

Literature Review

Introduction:

Iris yellow spot virus (IYSV) is a new and devastating disease of alliums in several locations throughout the world (Gent et al., 2006). IYSV is an insect vectored tospovirus whose only known vector is the onion thrips, *Thrips tabaci* (Lindeman) (Gent et al., 2006). Symptoms of IYSV on onion appear as elongated, diamond-shaped necrotic lesions (Pozzer et al., 1999). In severe cases, lesions can coalesce and form large necrotic areas resulting in a reduced photosynthetic surface area (Pozzer et al., 1999). On onion seed crops, lesions can form on the scape leading to premature collapse and complete loss of the seed head (umbel) (du Toit et al., 2004). Bulb crops, on the other hand, still produce a viable crop, but market size is usually reduced (Gent et al., 2004). Because of the relationship between IYSV and *T. tabaci*, much of the current research has focused on controlling the vector in order to manage the disease. To effectively manage this disorder, we need to increase our understanding of the epidemiology associated with this vector-virus complex. The research reported in this dissertation focused on improving our understanding of both the vector and the pathogen, and how the disease develops at the field scale. Also, we looked at different management strategies to reduce losses incurred by the vector and the virus on the onion bulb crop in Colorado.

Literature Review

The Onion Crop:

Onion (*Allium cepa* L.), believed to be indigenous to Asia as far back as 3,000 B.C., is ranked as one of the top ten vegetables produced in the United States. Production of this crop in the U.S. dates back as far as the early 17th century (Hanelt, 1990). In recent years, growers have harvested upwards of 64,750 hectares of onions resulting in total production of about 3,175,147 kg (National Agricultural Statistics Service, 2006). A biennial monocot herb, the onion is primarily grown for its bulb. If seeds are required, the plants and/or bulbs must be carried over to a second season. The onion is a cool season plant that grows well under a wide range of temperatures. Onion seedlings are highly resistant to frost damage at temperatures as low as -2C (Boyhan et al., 2001). Temperatures ranging from 7 to 27C are optimal for seed germination, while plants that have emerged will grow best at temperatures from 13 to 24C (Boyhan et al., 2001). Onion plants produce a shallow root system with nearly all roots present in the upper 45 cm of soil, and 90% of the roots are present in the upper 30 cm of soil (Boyhan et al., 2001). Due to their shallow root system, onion plants are poor weed competitors, therefore, a good weed management program is necessary to obtain optimum yields. Timely application of irrigation water is necessary to obtain maximum yield potential. Cultivated onions are classified based on shape (flat, round, globe), color (red, yellow, white), pungency (sweet, pungent), and bulbing response due to day length (short, intermediate, long day) (Brewster, J.L., 1990). Depending upon the cultivar, long day type onions initiate bulbing in response to day length and mature in 100 to 140 days (Boyhan et al., 2001).

Colorado ranks as one of the largest onion producing states, with production occurring in four major regions including the western slope, along the Front Range, in the Arkansas Valley and in northeastern counties (Gent et al., 2004). A majority of the acreage produced in Colorado is devoted to long day storage onion produced from seed, although 10 to 15% of this acreage is grown from transplants intended for early and fresh market sales (Schwartz et al., 1995). Transplants are available locally but are primarily imported from Arizona, California and Texas (Schwartz et al., 1995).

Thrips as a Pest and Vector:

Insects belonging to the order Thysanoptera (Thripidae), commonly referred to as thrips, are serious pests in many economically important crops. Most species are about 1 to 2 mm in length, with varying colors from white to brown to dark black (Lewis, 1997). Under magnification, thrips can clearly be differentiated into several segments including the head, with forward facing antennae, a prothorax, a fused pterothorax with two pairs of fringed wings, and an 11-segmented abdomen (Lewis, 1997). Several species of thrips are known to transmit viruses belonging to the genus *Tospovirus*. Transmission of tospoviruses by thrips impacts many food, fiber and ornamental crops belonging to hundreds of plant species, and cause devastating losses in many crops with worldwide economic implications (Kritzman et al., 2001). Many tospoviruses can be mechanically transmitted from diseased to healthy hosts under experimental conditions, however, dispersal and survival of the virus typically is dependent upon the acquisition by and transmission through an insect vector (Kritzman et al., 2001).

The life cycle and feeding behavior of thrips are conducive to their role as vectors of tospoviruses. Female thrips lay their eggs on host tissues. Upon hatching, the first and

second instars will feed on the tissue using their rasping, sucking mouthparts. While feeding, the insects will tear open the leaf epidermis and ingest the cytoplasm from mesophyll cells using a stylet, either collapsing the individual cells or, at times, destroying several adjacent cells around the feeding site (Terry, 1997). The relationship between thrips and thrips-vectored tospoviruses is somewhat unusual among insect transmitted plant viruses in that adult thrips can only transmit tospoviruses when acquisition occurs during larval feeding on virus infected plants (Kritzman et al., 2002). Once acquired, tospoviruses become circulative in the insect and can be passed transtadially (Kritzman et al., 2001; Kritzman et al., 2002). Viral replication may occur in thrips cells (Ullman et al., 1992), and the virus is transmitted persistently by adult thrips (Ullman et al., 1992). Adult thrips that have not fed on infected plants during their larval stage cannot become viruliferous even after feeding on infected plant tissue (Ullman et al., 1989; Ullman et al., 1991; Ullman et al., 1992). Adult thrips are unable to acquire the virus because a midgut barrier prevents the virus particles from becoming established within the hemocoel and other organs where the virus may replicate (Ullman et al., 1992).

Many tospoviruses have more than one thrips vector. Several studies have been conducted to identify which thrips species are able to vector different tospoviruses (Kritzman et al., 2001; Kritzman et al., 2002). Thrips were allowed to feed on several species of plants infected with various tospoviruses for extended periods to acquire the virus. They were then moved to healthy plants and allowed to feed. Plants were analyzed using different serological techniques to identify whether each virus had been introduced to the healthy plant tissue. *Thrips tabaci* (onion thrips) is the only proven

vector of IYSV, thus far. Other tospoviruses can be transmitted by *T. tabaci*, as well as several other *Thrips* and *Frankliniella* spp. (Nagata et al., 2004).

An individual species of thrips can consume a wide range of plant tissue types (Trichilo and Leigh, 1988), and can even be predacious (Gonzalez and Wilson, 1982; Rude and Clark, 1984; Trichilo and Leigh, 1986). Attempts have been made to isolate compounds from specific tissues that are preferred feeding for thrips. Several lines of research suggest the importance of dietary nitrogen to some species of thrips (Slansky and Scriber, 1985). Studies involving an artificial diet have shown that the addition of pollen increases rates of growth, development, and fecundity for *F. occidentalis* (Trichilo and Leigh, 1988) and other thrips species (Lesky et al., 1997). Increased soil nitrogen also has been shown to increase thrips populations (Schuch et al., 1998; Vos and Frinking, 1997).

Tospoviruses and Relation to IYSV:

Tomato spotted wilt virus (TSWV) was the first phytopathogenic virus identified as a member of the family *Bunyaviridae* based on molecular studies (Cortes et al., 1998). A second plant pathogenic virus in the family *Bunyaviridae*, *Impatiens necrotic spot virus* (INSV), was identified in the United States in 1990 (Cortes et al., 1998). Subsequent research revealed the existence of two additional plant pathogenic viruses, *Tomato chlorotic spot virus* (TCSV) and *Groundnut ringspot virus* (GRSV), which were added to the family *Bunyaviridae*. In 1995, these viruses were placed into the genus *Tospovirus* within the family *Bunyaviridae*. Several additional tospovirus species have since been placed in the genus based on nucleoprotein (N) serology, N-protein sequence, and vector specificity (Cortes et al., 2002). Viruses belonging to the genus *Tospovirus*

have enveloped isometric particles (80-110 nm) containing virus-encoded glycoproteins (Cortes et al., 2002; Kitajima et al., 1992), and are transmitted by thrips in a persistent manner (Sakimura, 1962). The tospovirus genome consists of tri-partite RNAs: L (~8900 nucleotides), M (~5000 nucleotides) and S (~2900 nucleotides) (Mohamed et al., 1973; Tas et al., 1977). The L RNA has negative polarity and contains only one open reading frame encoding the virus replicase (De Haan, 1991). The M RNA has an ambisense expression strategy to encode the glycoproteins G1/G2 and the virus movement protein (NS_M) (Cortes et al., 2002; Kormelink et al., 1992). The S RNA segment is also ambisense, and encodes the nucleocapsid protein (N) and a non-structural protein (NS_S) (De Haan, 1990; Kormelink et al., 1991).

Iris yellow spot virus (IYSV), first described by Cortes et al. (1998), was identified from *Iris hollandica* Tub in the Netherlands, where occasional disease problems consisting of chlorotic spots that developed into necrotic lesions had been reported since 1992. Chlorotic and necrotic lesions were typically found on plants infested with thrips. When plants were chemically treated to control the thrips, no symptoms were observed. Analysis of the diseased tissue with an electron microscope revealed the presence of spherical particles, 80 to 120 nm in diameter, typical of a tospovirus (Cortes et al., 1998). Serological and nucleotide sequence data revealed this to be a new and distinct tospovirus species for which the name *Iris yellow spot virus* (IYSV) was assigned.

In 1993 a tospovirus was discovered associated with seed onions in the U.S. It was subsequently determined that this virus was IYSV (Hall, 1993). *Iris yellow spot virus* was a relatively new pathogen that had the potential to devastate the onion bulb and

seed industries. IYSV has since become widely established in all western onion growing states including Arizona (Gent et al., 2004), California (Gent et al., 2004), Colorado (Schwartz et al., 2002), Nevada (Gent et al., 2004), New Mexico (Creamer et al., 2004), Oregon (Crowe and Pappu, 2005), Utah (Gent et al., 2004), and Washington (du Toit et al., 2004). The virus has also been discovered in states further to the east including Georgia (Mullis, 2004), Texas (Miller et al., 2006) and New York (*personal communication*). The virus causing iris yellow spot disease has also been found to occur in other areas such as India, Slovenia, Australia, Italy, Japan, Chile, Peru, Spain, Tunisia, Reunion Island, and Guatemala (Gent et al., 2006).

IYSV is a member of the genus *Tospovirus* which includes eighteen recognized members: *Tomato spotted wilt virus* (TSWV), *Tomato chlorotic spot virus* (TCSV), *Groundnut ringspot virus* (GRSV), *Impatiens necrotic spot virus* (INSV), *Watermelon silverleaf mottle virus* (WsMoV), *Capsicum chlorosis virus* (CaCV), *Calla lily chlorotic spot virus* (CCSV), *Chrysanthemum stem necrosis virus* (CSNV), *Peanut bud necrosis virus* (PBNV), *Melon yellow spot virus* (MYSV), *Peanut chlorotic fan virus* (PCFV), *Peanut yellow spot virus* (PYSV), *Physalis severe mottle virus* (PSMV), *Tomato yellow fruit ring virus* (TYFRV), *Tomato yellow ring virus* (TYRV), *Watermelon bud necrosis virus* (WBNV), and *Zucchini lethal chlorosis virus* (ZLCV) (http://www.oznet.ksu.edu/tospovirus/tospo_list.htm).

IYSV Management Approaches:

A great deal of research has been done to study how thrips-vectored viruses move within and between fields throughout the growing season (Camann et al., 1995; Coutts et al., 2004; Gitaitis et al., 1998). Based on aggregation analysis, limited secondary spread

occurs at the field level (Camann et al., 1995; Coutts et al., 2004; Gitaitis et al., 1998; Puche et al., 1995). Primary infection appears to be the most significant means of spread of the virus.

A study, conducted in 2003 through Colorado State University, was aimed at identifying the spatial distribution of IYSV at the field level (Gent et al., 2004). Two fields were evaluated in this study, both having confirmed IYSV infections. Incidence of disease as well as yield components were spatially mapped and interpolated at both locations. Geospatial data layers were created using differential Global Positioning System to create a virtual grid at a 0.40 ha scale. Spatial variability of IYSV incidence, plant population and yield were interpolated using mapping software. Results from this study identified limited evidence of positive spatial dependence at a 0.40 ha sampling scale. It also was determined that this random systematic sampling grid may not have been sufficient to detect spatial dependence of disease incidence or yield at a scale less than 0.40 ha.

Trials have been conducted to identify possible resistance or tolerance to the IYSV and/or its vector, *T. tabaci* (*personal communications*). In Colorado, during the 2003 and 2004 growing seasons, trials were conducted to determine the ability of 43 and 46 cultivars, respectively, to resist/tolerate *Iris yellow spot virus* and its thrips vector. Disease incidence ranged from 16 to 100% among the 43 cultivars in 2003. In 2004, incidence ranged from 13 to 61% among the 46 cultivars evaluated. Susceptibility was greatest in cultivars with red bulbs and dark-green to blue leaf color. Cultivars having a greater ability to tolerate thrips feeding appeared to be less susceptible to *Iris yellow spot virus*. A similar trial was conducted in Washington in 2004 to test cultivar

resistance/susceptibility and tolerance to the virus/vector complex (*personal communications*). In this trial a severe outbreak of the virus occurred with incidence ranging from 58 to 97% on the 46 cultivars evaluated. Significant differences in susceptibility to the virus were observed among cultivars in this study.

Conventional insecticide management for thrips involve broad-spectrum insecticide applications, namely pyrethroids, organophosphates and carbamates (Bauske et al., 1998; Stavisky et al., 2002). In some areas, insecticides are applied more than 10 times throughout a single growing season (Momol et al., 2004). Due to the intensity at which applications are made and the lack of rotation between insecticides with different modes of action, resistance to many insecticides is becoming more common (Allen et al., 2005). Due to the voraciousness of the thrips and the reduction in efficacy of many conventional pesticides, there is an overwhelming need for new insecticide chemistries and cultural techniques to more effectively manage this pest. Much of the research that is being conducted to manage this virus-vector complex stems from previous studies done with a related complex, *Tomato spotted wilt virus*, in combination with various genera and species of thrips vectors. Spinosad (Spintor, Dow Agro Sciences, Indianapolis, IN), a product classified as reduced-risk due to its low mammalian toxicity and low environmental risk (Jensen, 2003), has shown promising results in several trials (Jensen, 2003). This product has a unique mode of action when ingested by an insect causing an excitation of the nervous system, leading to involuntary muscle contractions, prostration with tremors, and paralysis (Sparks, 2001). Another reduced risk insecticide that has been introduced for thrips management is imidacloprid (Admire, Provado, Bayer CropScience, Research Triangle Park, NC). Imidacloprid is a systemic insecticide,

chemically related to the tobacco toxin, nicotine (Maienfisch et al, 2001). Its mode of action involves the blocking of acetylcholine from binding to receptors in the nervous system, and the consequent prevention of nervous impulse transmission which leads to modified feeding behavior and subsequent death of the insect (Joost and Riley, 2005). AZA-Direct (Azadirachtin, Gowen, Yuma, AZ), obtained from the neem tree, has found its use in the insecticide market alongside the previously mentioned products (Warnock, 2005). The principle active ingredient, azadirachtin, acts as an insect growth regulator (IGR) preventing exoskeleton development and impeding the molting process (Chiasson, 2004).

Another method for the control of thrips and other pests and pathogens is through the use of the plant's natural defense response. Plants can activate protective mechanisms upon detection of invading pathogens. Protection occurring locally at the site of primary inoculation and systemically in other distant tissues is known as systemic acquired resistance (Sticher, 1997). Acibenzolar-S-methyl (Actigard, Syngenta Inc., Greensboro, NC), an inducer of systemic acquired resistance, is effective against a broad range of pathogens (Louwe, 2001).

Techniques for thrips control using ultra violet (UV)-reflective mulch have been evaluated in several trials (Momol, 2004). The importance of color, affecting reflected UV light, has been monitored for its effect on host selection behavior by thrips (Momol, 2004). UV-reflective silver mulch has proven effective at reducing thrips colonization on several crops including tomato (Brown and Brown, 1992; Greenough et al., 1990; Scott et al., 1989). Silver reflective mulch also has been shown to reduce incidence of *Tomato spotted wilt virus* in tomato (Staub, 2001). The theory with UV-reflective mulch is that

thrips are unable to distinguish individual plants, and are therefore unable to land and colonize plant tissue (Staub, 2001). Straw mulching was introduced in 1985 as a means of improving irrigation water infiltration and reducing runoff (Jensen, 2003). Several growers using this technique found that their onions experienced reduced thrips pressure compared to fields lacking the mulch. At the time, it was believed that the mulch acted as a habitat for predators of the thrips.

Researchers at Oregon State University conducted several studies using mulch and other alternative methods to manage onion thrips (Jensen, 2003). Newly developed biological pesticides with reduced toxicity to beneficial predators were evaluated. The products included neem tree extracts (azadirachtin) and bacterial fermentation products (spinosad). In these studies, thrips control with soft chemistry alternative pesticides and mulch were equally as effective or slightly less effective than conventional treatments, while yield and quality of the crop were greatly improved. Thrips populations were significantly lower on treated compared to untreated plants (Jensen, 2003). As we become more aware of the epidemiology relevant to this virus-vector relationship, an integrated pest management program can be built to more effectively manage both the thrips vector and the iris yellow spot disease.

The objectives of this study was to gain a better understanding of the spatial and temporal distribution of IYSV in bulb onions, and to reduce the losses that occur as a result of this virus through improved management techniques. Data on virus incidence and thrips vector populations were collected and recorded throughout the season. A soil analysis also was completed for each location in several of the fields. Interpolation maps were created with this data and spatial analysis was conducted to identify relationships

between vector, virus and soil parameters. Additional trials were conducted to identify best management practices for the thrips vector as well as the IYSV. The information presented in this paper should strengthen our knowledge and understanding of this devastating vector-virus complex.

Chapter II

Studies to Manage Iris Yellow Spot Virus and Thrips (*Thrips tabaci*) in the Colorado Bulb Onion Crop

Introduction:

Iris yellow spot virus (IYSV) continues to devastate alliums throughout much of the U.S. and the world, and is being discovered in additional locations each year (Gent et al., 2006). IYSV is an insect-vectorred tospovirus and its only known vector is the onion thrips, *Thrips tabaci* (Cortes, 1998). Symptoms of IYSV on onions appear as elongated, diamond-shaped necrotic lesions. In severe cases, lesions can coalesce and form large necrotic areas resulting in a reduced photosynthetic surface area. On seed crops, lesions can form on the scape leading to premature collapse and complete loss of the seed head (umbel) (du Toit, 2004). Bulb crops, on the other hand, will still produce a viable crop, but size often is reduced. Because of the relationship between IYSV and *T. tabaci*, much of the current research has focused on controlling the vector to manage the virus. Prior research working with different formulations of mulch has proven promising in reducing thrips feeding on various crops (Brown and Brown, 1992; Greenough et al., 1990; Momol, 2004; Scott et al., 1989). Also, several novel chemistry insecticides have been evaluated with promising results at reducing thrips populations on several crops (Jensen, 2003; Joost and Riley, 2005).

The purpose of our research was to observe and evaluate the ability of several different management strategies, including new and novel pesticide chemistry, individually and in combination, for their ability to reduce thrips populations and incidence of IYSV on onion. We also evaluated several different mulch types for their

ability to reduce thrips populations and incidence of IYSV through either an increase in thrips predators or reflectance of light to disrupt thrips landing on individual plants.

Materials & Methods:

Mulch Study:

In 2005, two studies were conducted at the Colorado State University Agricultural Research, Development, and Education Center (ARDEC) in Fort Collins, CO. Plots were evaluated in both seeded cultivar (Vantage) and transplanted cultivar (Teton) onion fields. All fields were furrow irrigated. Seeded onions were planted into 102 cm beds (2 lines per bed) on 6 April, 2005. The transplanted onions were hand planted into 102 cm beds (2 lines per bed) on 24 April, 2005. All plots were 6 m in length by 8 beds wide and replicated four times in a randomized split-plot design, with mulch type as the main plot and insecticide treatment as the split-plot factor (Table 1). Wheat straw, at 1120kg/ha, was applied by hand (20 to 25 cm pieces) to the centers of each bed on the transplanted and seeded onions on 8 June and 27 June, respectively. Additional straw was applied on 5 July in both trials to cover exposed soil where straw was blown away during intense winds. Strips (15 cm wide) of silver reflective mulch were cut to fit the centers of each bed and held in place with the ends of 30 cm plastic-coated pieces of wire curved to cover 8 cm of the bed and pushed through the mulch into the soil every 1.5 to 2.0 m length of bed on the same dates as previously mentioned for the straw applications. The reflective mulch strips had to be refastened several times due to high wind conditions. Insecticide treatments were applied in 187 liters of water per hectare with a CO₂ backpack sprayer at 21,093 kg m², using 8002 Teejet flat-fan nozzles (2 nozzles per bed).

Insecticide treatments included two grower standard products, Lannate @ 1.754 l/ha (Methomyl, DuPont, Wilmington, DE) and Warrior @ 0.269 l/ha (Lambda-cyhalothrin, Syngenta Crop Protection, Greensboro, NC) applied on a weekly rotation compared to two novel chemistry products, AZA-Direct @ 2.338 l/ha (Azadirachtin, Gowen, Yuma, AZ) and Entrust @ 0.585 l/ha (Spintor, Dow Agro Sciences, Indianapolis, IN) (Table 1). A modified 2.4 m boom sprayer was utilized because of the large plot size design. Treatments were applied to the transplanted onions on 15, 22, and 29 June, and 7, 13, and 20 July. Applications on seeded onions occurred on 12, 20, and 26 July, and 3, 10, and 17 August. Sticky cup traps also were placed by the CSU entomology program of W.S. Cranshaw in straw mulch plots and untreated mulch plots in both trials to monitor thrips migration within the field. The sticky cup traps were constructed using plastic 0.58 L cups nailed to a 1 m long wood stake. Tanglefoot (The Tanglefoot Company, Grand Rapids, MI) was applied to the exterior of the cups using a sponge paintbrush. Two traps were placed in each replicated plot, on the east and west side, and left in the field for approximately two weeks after which they were evaluated for total thrips present.

Weed pressure was high during the 2005 growing season in the seeded crop requiring several herbicide applications. Hand weeding also was necessary to remove larger weeds unaffected by the herbicides. The transplanted field had fewer weeds requiring less maintenance. A 1.5 m section from each of the center two beds for each plot was collected and topped for yield estimation. Total yield as well as colossal, jumbo, and medium market class yields (Schwartz and Bartolo, 1995) were quantified using general linear model in SAS v. 9.1 (PROC GLM, SAS Institute Inc., Cary, NC).

Table 1. Insecticide treatments during 2005 for the onion IYSV mulch trial in cultivars Teton and Vantage at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment¹	Rate²	Mulch³
Untreated	--	No Mulch
Warrior -Sprays 1,2,5,6	0.269 l/ha	No Mulch
Lannate LV -Sprays 3,4	1.754 l/ha	
Azadirect -Sprays 1,2,5,6	2.338 l/ha	No Mulch
Entrust –Sprays 3,4	0.585 l/ha	
Untreated	--	Straw Mulch
Warrior -Sprays 1,2,5,6	0.269 l/ha	Straw Mulch
Lannate LV -Sprays 3,4	1.754 l/ha	
Azadirect -Sprays 1,2,5,6	2.338 l/ha	Straw Mulch
Entrust –Sprays 3,4	0.585 l/ha	
Untreated	--	Reflective Mulch
Warrior -Sprays 1,2,5,6	0.269 l/ha	Reflective Mulch
Lannate LV -Sprays 3,4	1.754 l/ha	
Azadirect -Sprays 1,2,5,6	2.338 l/ha	Reflective Mulch
Entrust –Sprays 3,4	0.585 l/ha	

¹ Treatments were applied as six weekly sprays, numbers represent the week that each treatment was applied.

² Rate in liters per hectare of each treatment applied.

³ Type of mulch present in each treatment (Straw, Reflective, No Mulch).

Actigard Studies:

Several studies were conducted at ARDEC in Fort Collins, CO to evaluate the effectiveness of Acibenzolar-S-methyl (Actigard, Syngenta Inc., Greensboro, NC), an inducer of systemic acquired resistance, and novel chemistry insecticides, against the virus causing iris yellow spot disease on onions and/or its insect vector. Plots were evaluated in a furrow-irrigated seeded (cv. Vantage) onion field. Onions were planted into 102 cm beds (2 rows per bed) on 6 April, 2005. Included in this study were trials to evaluate several formulations of various insecticide materials for their ability to reduce the incidence of IYSV on onions, and to identify possible phytotoxic effects, e.g., production of multiple centers, to the bulbs. Tables 2, 3 and 4 list all the treatments evaluated in this trial. Dates of application for the insecticide study were 13, 20 and 27 July and 3 August. Evaluation of IYSV took place on 12 September and harvest on 27 September. Application dates for the phytotoxicity study were 13, 19 and 26 July and 2 August. Incidence of IYSV was evaluated on 12 September and harvest occurred on 3 October. At time of harvest, twenty medium to jumbo bulbs were cut in half to evaluate for the presence of multiple centers, a phytotoxicity effect. Treatments for the Actigard rate and timing study were applied on 13, 19 and 26 July, and 2, 10 and 16 August. Plots were evaluated on 7 September for IYSV and harvested on 3 October. In addition, twenty medium to jumbo bulbs were placed in a drying room at 25 to 28C and evaluated on 12 December for percent incidence of storage rot by fungi such as *Botrytis allii*. Weed pressure was high during the 2005 growing season in the seed crop requiring several herbicide applications. Hand weeding also was necessary to remove larger weeds unaffected by the herbicides.

Table 2. IYSV insecticide treatments evaluating the effects of Actigard in reducing disease when included with conventional and novel chemistry insecticides

Treatment¹	Timing²
Untreated Control	
Provado 1.6F @ 0.262 kg/ha	4, 3, 2, and 1 week prebulb
Provado 1.6F @ 0.262 kg/ha + Actigard @ 0.053 kg/ha	4, 3, 2, and 1 week prebulb
Admire 2F @ 1.121 kg/ha	At planting
Admire 2F @ 1.121 kg/ha + Actigard @ 0.053 kg/ha	At planting/ 4, 3, 2, and 1 week prebulb
Admire 2F @ 1.681 kg/ha	At planting
Admire 2F @ 1.681 kg/ha + Actigard @ 0.053 kg/ha	At planting/ 4, 3, 2, and 1 week prebulb
Warrior 0.269 l/ha	4, 3, 2, and 1 week prebulb
Warrior 0.269 l/ha + Actigard @ 0.053 kg/ha	4, 3, 2, and 1 week prebulb
Actigard @ 0.053 kg/ha	4, 3, 2, and 1 week prebulb

¹ Insecticide application rates in either liters per hectare or kg per hectare.

² Timing of application based on pest presence or crop growth stage.

Table 3. Actigard treatments with different insecticides and adjuvants evaluated for phytotoxic effects and control of IYSV

Treatment¹

Untreated Control

Actigard @ 0.070 kg/ha

Actigard @ 0.070 kg/ha + Amistar @ 0.347 kg/ha

Actigard @ 0.070 kg/ha + Pristine @ 1.296 kg/ha

Actigard @ 0.070 kg/ha + A13842 @ 2.241 kg/ha

Actigard @ 0.070 kg/ha + Rovral @ 1.753 l/ha

Actigard @ 0.070 kg/ha + Guthion @ 3.507 l/ha

Actigard @ 0.070 kg/ha + Vydate @ 2.338 l/ha

Actigard @ 0.070 kg/ha + Warrior @ 0.283 l/ha

Actigard @ 0.070 kg/ha + Activator 90 @ 0.25% v/v

Actigard @ 0.070 kg/ha + Kinetic @ 0.125% v/v

Actigard @ 0.070 kg/ha + Bravo Weather Stik @ 3.507 l/ha

Actigard @ 0.070 kg/ha + Switch (62.5%) @ 0.613 kg a.i./Ha

¹ Treatment application rate in either kilograms per hectare or liters per hectare.

Table 4. Actigard treatments to evaluate different rates and timings of application to reduce the incidence of IYSV

Treatment¹	Timing²
Untreated Control	
Actigard @ 0.035 kg/ha	4, 3, 2, and 1 week pre bulb
Actigard @ 0.053 kg/ha	4, 3, 2, and 1 week pre bulb
Actigard @ 0.070 kg/ha	4, 3, 2, and 1 week pre bulb
Actigard @ 0.053 kg/ha	4 and 3 weeks pre bulb
Actigard @ 0.070 kg/ha	4 and 2 weeks pre bulb
Actigard @ 0.053 kg/ha	2 and 1 weeks pre bulb, 1 and 2 weeks post bulb
Actigard @ 0.053 kg/ha	1 and 2 weeks post bulb

¹ Treatment application rates in kilograms per hectare of product.

² Timing of application based on crop growth stage.

A 1.5 m section from each of the center two rows from each plot was collected for analysis. Total yield as well as colossal, jumbo, and medium market class yields (Schwartz and Bartolo, 1995) were quantified using the general linear model in SAS v. 9.1 (PROC GLM, SAS Institute Inc., Cary, NC).

Results:

Mulch Study:

Analysis of the transplant study validated prior research in Colorado during 2004 that straw mulch has a positive impact on reducing thrips populations in onion. A visual evaluation of thrips populations on transplanted onions was conducted on 14 and 27 June, and 5, 12 and 19 July. Evaluation of seeded onions took place on 12, 19, and 26 July, and 2 and 10 August. The evaluations consisted of visual counts of thrips on 5 randomly selected plants on the center two beds from each plot. Incidence of IYSV was determined by making visual counts of the two center beds to identify the total number of symptomatic plants. Evaluations were conducted on 22 August and 7 September, respectively. Yields were collected on 8 September and 3 October for transplanted and seeded onions, respectively.

Disease pressure from *Iris yellow spot virus* was low in the transplanted field trial in 2005. Thrips populations were less, though not statistically significant on 27 June, and 5 and 12 July for the straw mulch plots without insecticide compared to bare soil untreated plots (Table 5). Silver reflective mulch alone had fewer thrips than bare soil untreated plots on 5 July (Table 5). Mulch type had no significant impact on incidence of IYSV. The bare soil and straw mulch treatments both had 17%, and the silver reflective

mulch had 21% disease incidence (Table 6). The greatest impact of mulch type was observed in the yield analysis of the transplanted onions. Total yield was greatest with the straw mulch (47,384 kg/ha) followed by the silver reflective mulch (41,598 kg/ha) (Table 6). The bare soil treatment had the lowest total yield with 40,792 kg/ha. Straw mulch yielded more colossal onions (3,955 kg/ha) than the silver reflective mulch (1,172 kg/ha) (Table 6). There were no colossal size onions harvested from the bare soil untreated control plots. Jumbo market class onion yields were greatest from the straw mulch plot (35,959 kg/ha) followed by the silver reflective mulch (31,858 kg/ha), with the bare soil treatment having the lowest jumbo market class yield (29,661 kg/ha) (Table 6). Medium market class yields were greatest with the bare soil treatment (7,616 kg/ha), followed by silver reflective (6,225 kg/ha) and straw (4,833 kg/ha) mulch (Table 6).

Disease pressure was greater in the seeded trial compared to the transplanted trial in 2005. The straw mulch had fewer thrips on 26 July and 2 August, while the silver reflective mulch had fewer thrips on 12 July and 10 August (Table 7). Although not significant, disease incidence was lowest for the straw mulch plots (39%), followed by the bare soil treatment (43%) and the silver reflective mulch (49%) (Table 8). The silver reflective mulch plots had the greatest total yield (54,488 kg/ha), with straw mulch following closely at 53,682 kg/ha and the bare soil treatment at 52,584 kg/ha (Table 8). There were no colossal sized onions harvested from any treatment. The jumbo market class yield was greatest from the straw mulch treatment producing 32,444 kg/ha, followed by the silver reflective mulch with 27,610 kg/ha and the bare soil treatment with 23,289 kg/ha (Table 8). Medium market class yields were greatest with the bare soil

Table 5. Evaluation of thrips populations from the IYSV mulch trial on transplanted cultivar Teton at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	No. of Thrips Present on				
	14 June ¹	27 June	5 July	12 July	19 July
Bare Soil Untreated	184.8 ²	372.5bc	1,023.8ab	611.8ab	263.0d
Bare Soil Conventional	184.8	453.0ab	1,055.5ab	566.5ab	530.0abc
Bare Soil Biological	184.8	347.5bcd	77.8c	352.3c	415.5bcd
Straw Mulch Untreated	218.8	244.3cd	710.8cd	517.0bc	267.0d
Straw Mulch Conventional	218.8	313.0bcd	915.0abc	532.0abc	633.3a
Straw Mulch Biological	218.8	191.0d	472.8d	375.0c	384.3cd
Reflective Untreated	195.3	376.5bc	926.8abc	756.3a	297.3d
Reflective Conventional	195.3	616.8a	1,195.5a	557.3abc	583.3ab
Reflective Biological	195.3	393.8bc	871.5bc	435.3bc	423.8bcd
Coefficient of Variation	25.05	30.55	22.61	29.80	27.54
LSD	<i>n.s.</i>	163.88	289.62	227.27	169.59
Prob. (≤ 0.05)	<i>n.s.</i>	0.0014	0.0014	0.0356	0.0005

¹ Date of thrips evaluation

² Number of thrips/5 plants; courtesy of the CSU entomology program of W.S. Cranshaw

Table 6. Evaluation of onion harvest from the IYSV mulch trial on transplanted cultivar Teton at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	Disease IYSV ¹	Total ²	Yield Component		
			Colossal	Jumbo	Medium
Bare Soil Untreated	16.5	40,792.0	0.0d	29,661.0ab	7,616.0
Bare Soil Conventional	16.5	37,058.0	952.0d	26,072.0b	8,275.0
Bare Soil Biological	10.0	45,260.0	5,713.0ab	30,320.0ab	7,690.0
Straw Mulch Untreated	16.5	47,384.0	3,954.0abcd	35,959.0a	4,833.0
Straw Mulch Conventional	12.5	48,190.0	6,372.0a	35,520.0a	3,589.0
Straw Mulch Biological	16.5	47,896.0	5,346.0abc	36,179.0a	3,809.0
Reflective Untreated	21.0	41,598.0	1,171.0cd	31,858.0ab	6,225.0
Reflective Conventional	15.0	39,035.0	2,051.0bc	26,439.0b	8,275.0
Reflective Biological	16.5	39,548.0	806.0d	28,416.0ab	8,569.0
Coefficient of Variation	38.91	14.16	110.81	19.86	47.65
LSD	<i>n.s.</i>	<i>n.s.</i>	4226.7	8058.7	<i>n.s.</i>
Prob. (≤ 0.05)	<i>n.s.</i>	<i>n.s.</i>	0.05	0.05	<i>n.s.</i>

¹ % incidence of IYSV per 3.3m section of each plot

² kg/ha of onions

Table 7. Evaluation of thrips populations from the IYSV mulch trial on seeded cultivar Vantage at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	No. of Thrips Present on				
	12 July	19 July	26 July	2 Aug	10 Aug
Bare Soil Untreated	116.8	91.5	286.3cd	346.8c	263.3bcd
Bare Soil Conventional	116.8	114.3	517.3b	883.8ab	334.8abc
Bare Soil Biological	116.8	84.0	261.0cd	329.0c	154.3d
Straw Mulch Untreated	116.3	154.5	149.0d	242.8c	229.3cd
Straw Mulch Conventional	116.3	120.3	372.8c	734.0b	391.5ab
Straw Mulch Biological	116.3	124.0	259.0cd	356.0c	221.5cd
Reflective Untreated	109.0	150.3	239.3cd	365.5c	178.8d
Reflective Conventional	109.0	108.3	709.0a	965.8a	468.3a
Reflective Biological	109.0	113.3	306.5c	334.0c	134.3d
Coefficient of Variation	27.05	43.01	27.80	28.96	38.76
LSD	<i>n.s.</i>	<i>n.s.</i>	139.77	214.00	149.33
Prob. (≤ 0.05)	<i>n.s.</i>	<i>n.s.</i>	<0.0001	<0.0001	<0.0012

¹ Date of thrips evaluation

² Number of thrips/5 plants; courtesy of the CSU entomology program of W.S. Cranshaw

Table 8. Evaluation of onion harvest from the IYSV mulch trial on seeded cultivar Vantage at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	Disease IYSV ¹	Total ²	Yield Component		
			Colossal	Jumbo	Medium
Bare Soil Untreated	43.3	52,584.0	0.0	23,289.0	24,461.0
Bare Soil Conventional	23.3	55,147.0	0.0	19,994.0	30,466.0
Bare Soil Biological	41.5	50,050.0	0.0	22703.0	22,997.0
Straw Mulch Untreated	38.5	53,682.0	0.0	32,444.0	18,089.0
Straw Mulch Conventional	36.3	50,534.0	0.0	24,022.0	23,436.0
Straw Mulch Biological	56.0	51,675.0	0.0	28,928.0	19,774.0
Reflective Untreated	49.3	54,488.0	0.0	27,610.0	23,656.0
Reflective Conventional	34.3	48,043.0	0.0	23,582.0	18,968.0
Reflective Biological	40.8	57,124.0	146.0	26,804.0	25,999.0
Coefficient of Variation	34.77	14.07	600.00	33.20	35.57
LSD	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
Prob. (≤ 0.05)	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>

¹ % incidence of IYSV per 3.3m section of each plot

² kg/ha of onions

treatment (24,461 kg/ha), followed by the silver reflective (23,656 kg/ha) and straw (18,089 kg/ha) mulch (Table 8).

The novel chemistry biological insecticides proved very effective at reducing thrips populations in both trials. In the transplanted trial, thrips counts were statistically reduced on 5 and 12 July on plots treated with Entrust and AZA-Direct (Table 5) in the absence of mulch compared to the untreated plots without mulch. Thrips populations were less on 27 June, and 5 and 19 July for untreated plots compared to the conventional treatment of Lannate and Warrior without mulch. In plots treated with straw mulch, the biological treatments were lowest on 27 June, and 5 and 12 July while the conventional treatment had equal or greater thrips numbers than plots with straw mulch alone on all other evaluation dates (Table 5). The reflective mulch plots with biological treatments had the lowest thrips populations on 5 and 12 July (Table 5).

The conventional treatment had fewer thrips than the untreated plot under reflective mulch on 12 July (Table 5). Incidence of IYSV was lowest on the biological treated plots in the absence of mulch (Table 6). In the straw mulch plots, the conventional treatment had a lower incidence of IYSV than both the biologically-treated and untreated plots (Table 6). In the reflective mulch plots, the conventional treatment had a lower incidence of IYSV followed by the biologically-treated and untreated plots (Table 6).

Total yield, as well as colossal and jumbo market class yields, were greatest with the biological treatment without mulch (45,260 kg/ha, 5,713 kg/ha and 30,320 kg/ha, respectively) compared to the conventional and untreated plots (Table 6). The conventional treated plots on bare soil had more colossal and medium market class

onions than untreated, bare soil plots. In the straw mulch plots, the conventional treatment had greater total and colossal yields (48,190 kg/ha and 6,372 kg/ha, respectively) than both the untreated and biologically-treated plots with straw mulch (Table 6). The biological treatment with straw had the greatest jumbo yield (36,179 kg/ha), and exceeded the untreated plots with straw mulch in total and colossal yields (Table 6). In the presence of silver reflective mulch, untreated plots had the greatest total and jumbo yields (41,598 kg/ha and 31,858 kg/ha, respectively) (Table 6). The biologically-treated plots with silver reflective mulch had the greatest medium yield (8,569 kg/ha) compared to untreated and conventional plots with silver reflective mulch (Table 6). The conventional treatment with silver reflective mulch exceeded the untreated and biologically-treated plots in colossal yield only (Table 6).

In the seeded trial, the biological treatment without mulch had the lowest thrips populations on 19 and 26 July, and 2 and 10 August (Table 7). The conventional treatment had equal or greater thrips populations than untreated plots in the absence of mulch on all evaluation dates (Table 7). In the presence of straw mulch, the biological treatment was lowest on the final evaluation date of 10 August. The conventional treatment with straw mulch was lowest on 19 July. Straw mulch alone had the least number of thrips on 26 July and 2 August (Table 7).

Disease incidence was least on conventionally treated plots in the absence of mulch (23%), followed by the biologically-treated (42%) and untreated (43%) plots (Table 8). On plots without mulch, total and medium yields were greatest with the conventional treatment (55,147 kg/ha and 30,466 kg/ha, respectively) (Table 8). The biological treatment without mulch had the lowest yield in all market classes. In the

presence of straw mulch, conventionally treated plots had the least disease (36%), while the biological treatment had the greatest incidence of disease (56%) (Table 8). Total and jumbo yields were greatest on straw mulch plots with no chemical treatment (53,682 kg/ha and 32,444 kg/ha, respectively) (Table 8). The conventional treatment had the greatest medium yield with straw mulch at 23,436 kg/ha. Straw mulch plots with the biological treatment had greater total and jumbo yields (51,675 kg/ha and 28,928 kg/ha, respectively) than straw mulch plots with conventional chemical treatments, but not the untreated plots with straw (Table 8). Plots with reflective mulch had the least disease when the conventional treatment was applied, followed by the biologically-treated and non-treated plots (34%, 41% and 49%, respectively) (Table 8). Total, colossal and medium yields were greatest for the biological treatment with UV reflective mulch (57,124 kg/ha, 147 kg/ha and 25,999 kg/ha, respectively) (Table 8). The reflective mulch plots with no chemical treatment had the greatest jumbo yield with 27,610 kg/ha.

Actigard Studies:

During the 2005 growing season, *Iris yellow spot virus* occurred at moderate levels throughout our trials. Our first study involved insecticide treatment combinations for the management of IYSV. The standard treatment with Warrior at 0.0269 kg/ha resulted in the lowest incidence of disease and was significantly better than the untreated control, both the low rate and high rate of Admire 2F, high rate of Admire with Actigard and Warrior with Actigard (Table 9). The low rate of Admire, high rate of Admire and Admire plus Actigard showed the least control of IYSV, all resulting in greater, although not significant, disease incidence than the untreated control (Table 9). High rate Admire

resulted in the greatest total yield (45,257 kg/ha) and was significantly better than the low rate Admire (33,030 kg/ha) (Table 9). The untreated control had the second greatest total yield (44,088 kg/ha) but was not significantly different than any other treatment. Jumbo market class yield was greatest with the high rate Admire plus Actigard (12,597 kg/ha), both Provado alone and Warrior alone resulted in the lowest, though not statistically significant, yields (4,980 and 5,639 kg/ha, respectively) (Table 9). The untreated control had the third highest jumbo market class yield, but was not significantly different than any other treatment (Table 9). Medium market class yields were greatest with the Provado plus Actigard treatment (27,610 kg/ha), but was not significantly greater than any other treatment (Table 9). The untreated control had the second highest medium market class yield (25,486 kg/ha), while the low rate Admire treatment had the lowest yield (17,430 kg/ha) (Table 9).

In our next study, we evaluated Actigard rates and timing of applications. Incidence of IYSV was the least (16%) at the 0.053 kg/ha rate applied weekly beginning four weeks pre-bulb for a total of four applications (Table 10). The highest incidence of IYSV (30%) occurred with the 0.070 kg/ha rate applied twice, at four and two weeks pre-bulb. Incidence of IYSV was significantly different for the previous two treatments, all other treatments were not statistically significant (Table 10). Total yield was greatest on the untreated plots (51,046 kg/ha), followed by the 0.053 kg/ha rate applied one week and two weeks post-bulb (48,702 kg/ha) (Table 10). Actigard applied at 0.053 kg/ha four weeks and three weeks pre-bulb resulted in the lowest total yield (37,277 kg/ha), although not significantly different than the highest yielding treatment (Table 10). Jumbo

Table 9. Evaluations of IYSV incidence and onion market class yields for the insecticide/Actigard study at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	Disease IYSV ¹	Yield Component		
		Total ²	Jumbo ²	Medium ²
Untreated	41.2abc	44,088.0	8,935.0	25,486.0
Provado 1.6F @ 0.262 kg/ha	27.3cd	38,962.0	4,980.0	23,656.0
Provado 1.6F @ 0.262 kg/ha + Actigard @ 0.053 kg/ha	27.4cd	43,063.0	7,104.0	27,610.0
Admire 2F @ 1.121 kg/ha	45.7ab	33,030.0	6,738.0	17,430.0
Admire 2F @ 1.121 kg/ha + Actigard @ 0.053 kg/ha	36.1abcd	42,257.0	6,884.0	22,410.0
Admire 2F @ 1.681 kg/ha	49.6ab	45,626.0	10,400.0	23,436.0
Admire 2F @ 1.681 kg/ha + Actigard @ 0.053 kg/ha	50.0a	39,328.0	12,597.0	20,946.0
Warrior @ 0.269 l/ha	24.8d	38,962.0	5,639.0	23,802.0
Warrior @ 0.269 l/ha + Actigard @ 0.053 kg/ha	39.8abc	39,548.0	8,276.0	22,118.0
Actigard @ 0.053 kg/ha	35.7bcd	40,500.0	7,763.0	21,056.0
Coefficient of Variation	26.17	19.93	3.71	4.30
LSD	14.34	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
Prob.(≤0.05)	<0.01	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>

¹ Percent incidence of IYSV per 3.3m section of each plot.

² kg/ha of onions.

Table 10. Evaluations of IYSV incidence and onion market class yields for the Actigard rate and timing study at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	IYSV Disease	Yield Components			% Bulbs w/ Storage rot
		Total	Jumbo	Medium	
Untreated Control	26.6	51,046.0	26,072.0	19,774.0b	26.3
Actigard @ 0.035 kg/ha	22.0	45,216.0	18,749.1	22,073.0b	35.0
Actigard @ 0.053 kg/ha	16.2	41,789.0	11,620.0	22,366.0b	33.8
Actigard @ 0.070 kg/ha	21.4	43,253.0	14,940.0	23,143.0ab	20.0
Actigard @ 0.053 kg/ha	28.1	37,277.0	11,352.0	20,653.0b	33.8
Actigard @ 0.070 kg/ha	30.4	40,866.0	16,698.0	19,188.0b	26.3
Actigard @ 0.053 kg/ha	23.0	45,992.0	23,143.0	17,870.0b	21.3
Actigard @ 0.053 kg/ha	26.4	48,702.0	13,769.0	29,148.0a	27.5
Coefficient of Variation	39.22	22.13	47.75	19.22	60.20
LSD	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	6,570.0	<i>n.s.</i>
Prob. (≤ 0.05)	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	0.04	<i>n.s.</i>

¹ Percent incidence of IYSV per 3.3m section of each plot.

² kg/ha of onions.

³ Primary storage rot problem encountered was *Botrytis allii*.

market class yield was also greatest on the untreated plots (26,072 kg/ha) (Table 10). Actigard applied at 0.053 kg/ha beginning four weeks pre-bulb for a total of four weekly applications and the 0.053 kg/ha rate applied four and three weeks pre-bulb were the lowest yielding (11,620 kg/ha and 11,352 kg/ha, respectively) (Table 10). The medium market class yield was greatest in the 0.053 kg/ha plots treated one week and two weeks post-bulb (29,148 kg/ha) (Table 10). All other treatments resulted in significantly lower yields except the 0.070 kg/ha rate applied four times beginning four weeks pre-bulb (23,143 kg/ha) (Table 10). The lowest yielding treatment for the medium market class was the 0.053 kg/ha rate applied one and two weeks pre-bulb and one and two weeks post-bulb initiation (17,870 kg/ha) (Table 10). Our final evaluation with regards to this study involved an analysis of bulb storage rot caused by fungi such as *Botrytis allii*. The 0.070 kg/ha rate of Actigard applied four time pre-bulb resulted in the lowest amount of *Botrytis* bulb rot (20%), while the lowest rate of Actigard at 0.035 kg/ha had the greatest incidence of bulb rot (35%); no treatment was significantly different (Table 10).

In our final study, we evaluated the effects of different insecticides applied with the 0.070 kg/ha rate of Actigard, and their ability to cause a phytotoxic effect on the onion plant or bulb. The incidence of IYSV was least when Actigard was applied with Pristine (27%) and was better, though not significant, than the Actigard treatment alone, Actigard + Amistar, Actigard + A13842, Actigard + Kinetic, and Actigard + Switch (Table 11). The Actigard treatment alone had the greatest incidence of IYSV (50%) and was greater than the untreated plots, Actigard + Pristine, and Actigard + Guthion (Table 11). The Actigard + Vydate treatment had the fewest number of double center bulbs (3.75%) followed by the Actigard + Guthion treatment (5.0%) (Table 11). The previous

two treatments had fewer double centers than the Actigard + Amistar and Actigard + Pristine treatments (17.50% and 18.75%, respectively) (Table 11). The greatest total yield occurred with the Actigard + Rovral treatment (56,978 kg/ha), but was not significantly better than any other treatment. Actigard + A13842 was the lowest yielding treatment (46,212 kg/ha) in this study (Table 11). Jumbo market class yield was greater with the Actigard + Rovral treatment (34,348 kg/ha) when compared to the treatments of Actigard + Pristine, Actigard + A13842, Actigard + Guthion, Actigard + Vydate, Actigard + Warrior, Actigard + Activator 90, Actigard + Bravo Weather Stik, and Actigard + Switch (Table 11). The Actigard + Guthion treatment produced the lowest jumbo market class yield (18,602 kg/ha) (Table 11). Lastly, the medium market class yield was greatest with the Actigard + Bravo Weather Stik (26,292 kg/ha), but was not significantly better than any other treatment. Actigard + Pristine produced the lowest yield in the medium market class.

Discussion:

Management of *Thrips tabaci* and *Iris yellow spot virus* using mulch ground cover and novel chemistry insecticides appear to show promise in reducing thrips populations and feeding, thereby reducing losses incurred by both the insect and the virus. Whether directly or indirectly, the straw mulch had a significant impact on yield in the transplanted cultivar Teton. Significant reductions in thrips populations were observed but, more importantly, total yield, colossal, and jumbo grade onions were largely increased. Thus far it is unclear whether the straw mulch is acting as a natural habitat for

Table 11. Evaluations of IYSV incidence and onion market class yields for the Actigard phytotoxicity study at Colorado State University's Agricultural Research Development and Education Center (ARDEC), Fort Collins, CO

Treatment	Disease IYSV ¹	Doubles ²	Yield Component		
			Total ³	Jumbo ³	Medium ³
Untreated Control	34.1	7.5	54,561.0	33,103.0	19,261.0
Actigard @ 0.070 kg/ha	50.5	13.8	49,435.0	24,900.0	20,653.0
Actigard @ 0.070 kg/ha + Amistar @ 0.347 kg/ha	40.0	17.5	48,995.0	25,120.0	19,261.0
Actigard @ 0.070 kg/ha + Pristine @ 1.296 kg/ha	27.5	18.8	47,750.0	20,067.0	18,089.0
Actigard @ 0.070 kg/ha + A13842 @ 2.241 kg/ha	44.8	12.5	46,212.0	23,289.0	19,554.0
Actigard @ 0.070 kg/ha + Rovral @ 1.753 l/ha	35.3	11.3	56,978.0	34,348.0	18,822.0
Actigard @ 0.070 kg/ha + Guthion @ 3.507 l/ha	31.2	5.0	50,020.0	18,602.0	24,681.0
Actigard @ 0.070 kg/ha + Vydate @ 2.338 l/ha	39.0	3.8	46,285.0	21,239.0	22,410.0
Actigard @ 0.070 kg/ha + Warrior @ 0.283 l/ha	40.9	10.0	45,040.0	21,312.0	20,433.0
Actigard @ 0.070 kg/ha + Activator 90 @ 0.25% v/v	37.6	8.8	50,387.0	19,408.0	25,633.0
Actigard @ 0.070 kg/ha + Kinetic @ 0.125% v/v	45.9	12.5	48,922.0	27,932.0	19,627.0
Actigard @ 0.070 kg/ha + Bravo Weather Stik @ 3.507 l/ha	39.9	11.3	52,291.0	21,458.0	26,292.0
Actigard @ 0.070 kg/ha + Switch (62.5%) @ 0.613 kg a.i./Ha	45.4	10.0	48,190.0	22,264.0	22,337.0
Coefficient of Variation	28.09	68.70	18.41	30.84	36.51
LSD	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>

Prob. (≤ 0.05)	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
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¹ Percent incidence of IYSV per 3.3m section of each plot.

² Percent incidence of double center onions.

³ kg/ha of onions.

predators and parasites of thrips as mentioned in prior research (Jensen, 2003), or if some measure of reflectance from the straw is impacting the ability of thrips to distinguish individual plants. A light meter was used to obtain readings of reflectance from the different mulches used and straw mulch did show an increase in the amount of light reflected, although much less than silver UV reflective mulch, when compared to bare soil (data not shown). Predatory thrips, mites and minute pirate bugs were monitored, but no statistically significant differences were observed between treatments (data not shown). As expected, the straw mulch became less effective once the plant canopy covered the rows. We also observed greater moisture in soil beneath the straw mulch (data not shown), and this may have contributed to the increased yields within these treatments. Further work is needed to attempt to identify a more economical means of applying the straw mulch to a commercial field. One possibility is to use a modified manure spreader that could shred the straw and disperse it evenly throughout the field. Where furrow irrigation is used, this technique may prove problematic due to clogging of the furrows with excess straw; although we did not encounter any difficulties with our production system and relatively short irrigation run of 150 minutes on 150 meters of row length.

The UV reflective mulch has proven effective at reducing thrips populations and incidence of viral diseases in other crops such as tomato, but does not appear to be economical or practical in a densely-spaced crop such as onion. In our study, we applied the mulch in strips between the two lines of onions on each bed. This technique was extremely labor intensive, and due to high winds and occasional intense weather it was very difficult to maintain. The silver reflective mulch had variable results in reducing

thrips populations. Much of the variability may be due to the mulch being blown loose from the soil surface and rolling over, exposing its black underside and thereby reducing its reflective benefit. Settling of rainwater on the mulch also led to deterioration of the delicate, silver reflective surface. Most importantly, the results were mixed and, at times, there appeared to be a negative impact on thrips numbers with the UV reflective mulch when compared to the bare soil treatment.

Actigard at the 0.070 kg/ha rate did not appear effective at reducing incidence of IYSV under the low disease pressure encountered in our research trial in 2005. In all trials where this rate was evaluated, performance was poor resulting in higher incidence of virus when compared to many other treatments. Yield estimates were low in all market classes when the 0.070 kg/ha rate of Actigard was used alone. In our phytotoxicity trial, the third highest incidence of phytotoxicity occurred with the 0.070 kg/ha rate of Actigard. In our rate and timing study, both treatments with the high rate of Actigard had limited control of IYSV and yield estimates were poor in all classes. The 0.053 kg/ha rate of Actigard performed better than both the 0.035 kg/ha and the 0.070 kg/ha rate for both disease control and yield response, although, yields were even greater on the untreated plots. The conventional treatment Warrior and the novel chemistry treatment Provado did better at reducing disease incidence than the 0.053 kg/ha rate of Actigard alone. In several instances, yields were greater where insecticides were used in the absence of Actigard, although, for the insecticide treatment with Provado, the Warrior treatment and the treatment with a low rate of Admire, the addition of Actigard improved yield and with the low rate Admire it reduced the incidence of IYSV. With such a broad range of activity, Actigard continues to show promise as a multi-edged sword in the

management of disease complexes. Although our results did not show huge improvements with this material, other research has shown promise on a multitude of pathogens in various cropping systems.

Chapter III

Spatial and Temporal Distribution of IYSV in Colorado Onion Fields

Introduction

Onion producers in Colorado are one of the most recent groups impacted by this virus which causes iris yellow spot disease. Since its introduction into Colorado, IYSV has been isolated from an increasing number of fields, and continues to expand to new areas each year. Due to the speed at which the virus has moved within the state, surveys were carried out to study the spatial patterns and spread of this new pest. Thus far no known alternate host has been confirmed in Colorado, although, several weed species have tested positive by double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) (D.H. Gent and H.F. Schwartz, *unpublished*). Currently, studies are ongoing to verify these previously identified weed hosts and to identify other as yet unknown hosts of the virus. Additionally, studies have found that the virus does arrive each year on transplants brought in from outside the state, primarily from those areas where the virus is already known to exist (S.M. Fichtner and D.H. Gent, *unpublished*). Several of our surveys have focused on those areas receiving transplants that tested positive for the *Iris yellow spot virus*. The only known vector of IYSV, *Thrips tabaci*, is prevalent in the state of Colorado and was often found in significantly high numbers on many of the imported onion transplants (L. Mahaffe, *unpublished*). Our research focused on intensive surveys of several commercial onion fields in areas of the state where the virus had been isolated. These surveys were designed to expand our understanding of the spatial and temporal distribution of *Iris yellow spot virus* and to learn more about the

means by which the virus is able to be disseminated so quickly to areas and fields previously free of this disease.

Materials & Methods

2004 Season:

During the 2004 season three seeded commercial onion fields along the Colorado Front-Range were chosen. Sites were named based on the nearest town (e.g., Ault, Eaton, and Peckham). Ault and Peckham sites were furrow-irrigated, and Eaton was drip irrigated. Ault was planted with a yellow cultivar Ranchero having four seeded lines per bed. Eaton was planted with a yellow cultivar Granero with five lines per bed. Peckham was planted to two cultivars, Sterling (white) and Tango (red), with three lines and two lines per bed, respectively. Field perimeters were mapped by walking the border using a differentially corrected Trimble Ag114 backpack GPS unit (Trimble Navigation Limited, Sunnyvale, CA) equipped with a handheld pen-top-based Fujitsu computer (Tokyo, Japan) and MapInfo Professional v. 6.5 FarmGPS software (MapInfo Corp., Troy, NY). Fields were then divided into grids 0.2 ha in size, and a point was created within each grid based on the Random Systematic Unaligned sampling technique using the FarmGPS software. The Ault field was 13.8 ha in size and had 69 sample locations (Figure 1), Eaton was 11 ha in size and had 53 sample locations (Figure 2), and Peckham was 10.1 ha (6.5 ha Tango, 3.6 ha Sterling) in size with 51 sampling locations (Figure 3). Ten plants were removed from each plot three times early in the season (Table 12). Plant samples were analyzed using double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) to identify the timing of IYSV infection. Visual ratings in the field

were initiated once symptoms were observed on field borders. The ratings were based on incidence of IYSV over three meters from a single row at each flagged plot. Soil samples were collected from each flagged plot at all three locations on 9 June. A complete soil analysis was conducted on each sample collected. The Ault field was harvested 13 September, just as symptoms began to appear on the borders of the field so no visual ratings were performed. The Eaton field was completely destroyed by hail before any symptoms were observed so no visual ratings were possible. Visual ratings for IYSV at Peckham began in early August once symptoms were observed and continued until harvest (6, 13 and 23 August and 1, 6 and 10 September). Total yield and market class yield (colossal, jumbo and medium) were recorded (Schwartz and Bartolo, 1995) from a three-meter section at each sampling location. Several samples displaying symptoms of IYSV were collected at each site to confirm the presence of the virus using ELISA (Gent et al., 2004). Samples were considered positive if absorbance values were greater than three times the average of the negative controls, which consisted of healthy onion tissue. All the data collected during our survey was analyzed using simple linear regression in SAS v. 9.1 (PROC REG, SAS Institute Inc., Cary, NC) and general linear model in SAS v. 9.1 (PROC GLM, SAS Institute Inc., Cary, NC).

Figure 1. Perimeter and plot locations (V) for the 2004 Ault site used in our IYSV survey.

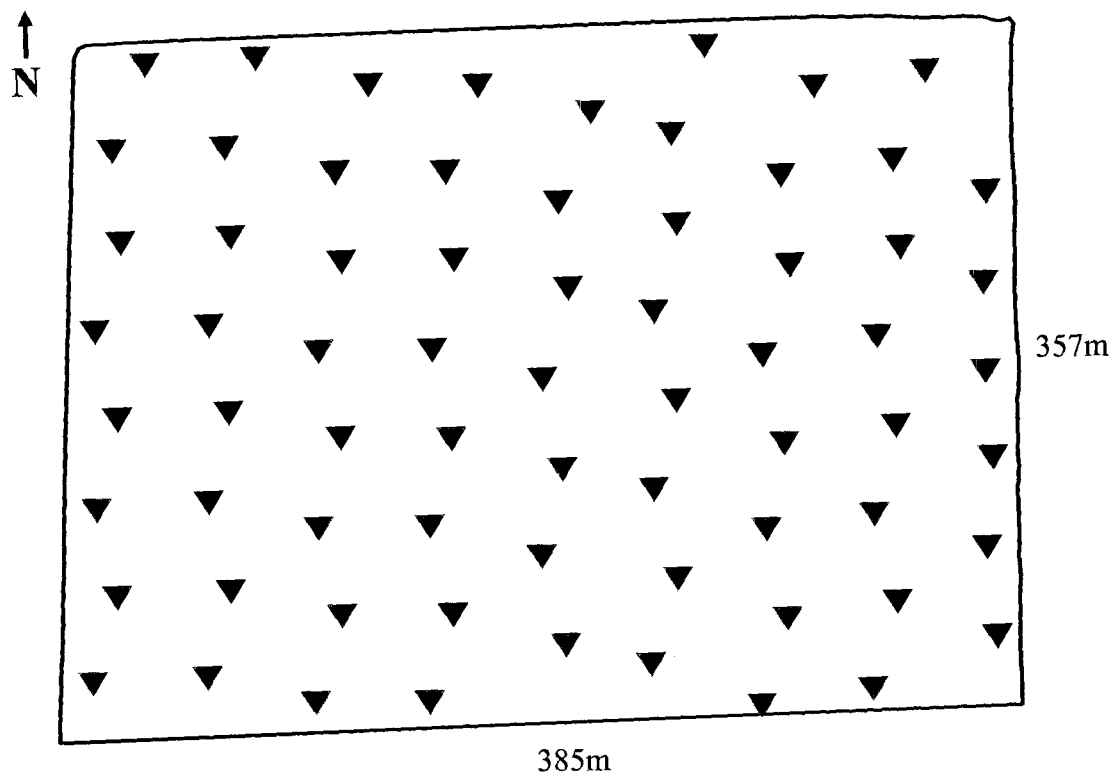


Figure 2. Perimeter and plot locations (V) for the 2004 Eaton site used in our IYSV survey.

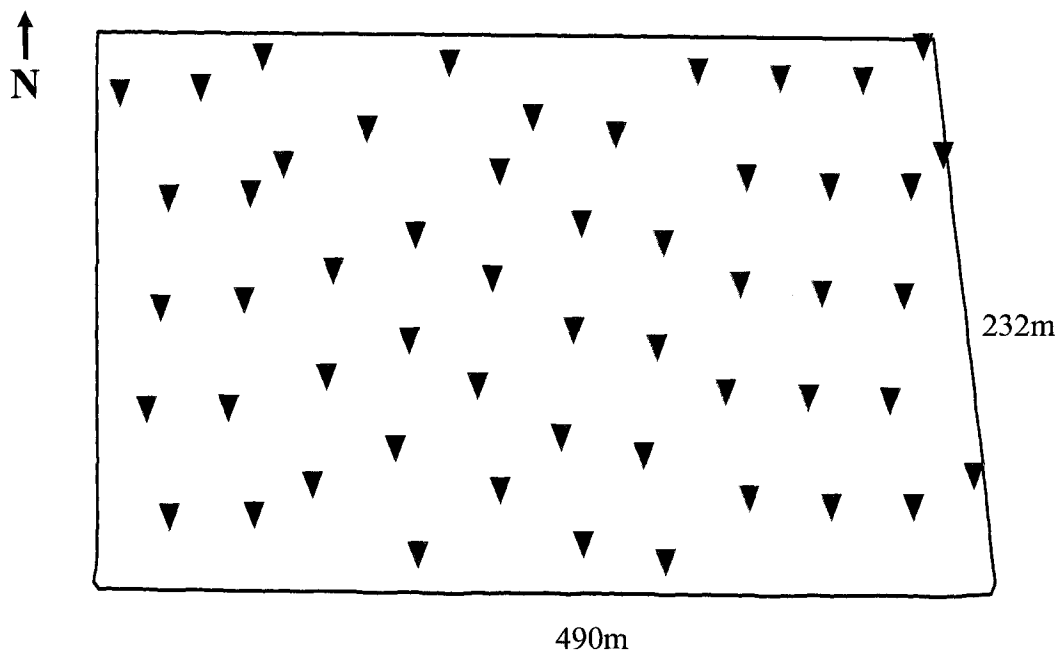


Figure 3. Perimeter and plot locations (V) for the 2004 Peckham site used in our IYSV survey.

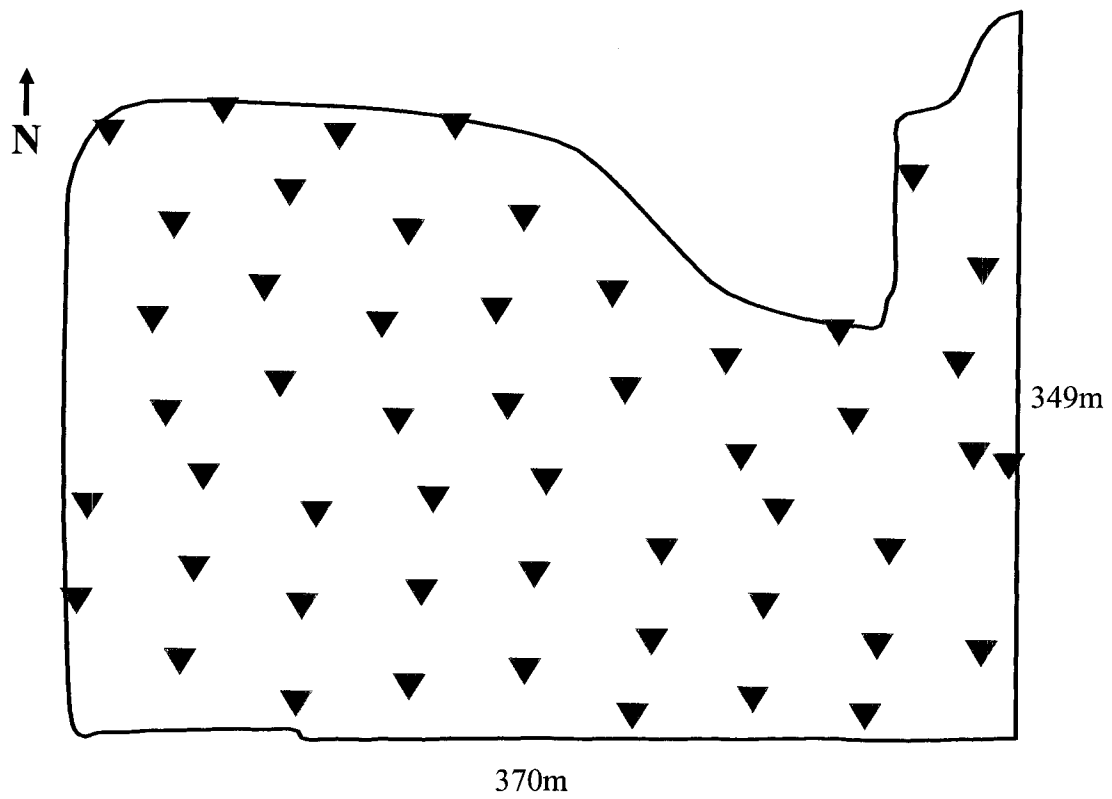


Table 12. Sampling dates for the collection of onions for analysis
using *Iris yellow spot virus* specific ELISA

Sample:	Location		
	Ault	Eaton	Peckham
1	05/31/2004	06/02/2004	06/07/2004
2	06/24/2004	06/24/2004	06/25/2004
3	07/14/2004	07/12/2004	07/14/2004

2005 Season:

Early in the 2005 season eight fields were identified and evaluated as potential sites to repeat the 2004 IYSV survey. Field perimeters were mapped by walking the border using a differentially corrected Trimble Ag114 backpack GPS unit, as described previously. Fields were then divided into grids 0.2 ha in size, and a point was created within each grid based on the Random Systematic Unaligned sampling technique using the FarmGPS software. Of these eight potential sites, three were on the Western Slope and five along the eastern Front Range of Colorado. On the Western Slope, two fields were located near Loma, CO and identified as Loma-north seeded (4 ha with 21 sampling sites) (Figure 4) and Loma-south seeded (4.2 ha with 24 sampling sites) (Figure 5). The third field was located near Delta, CO and was identified as Delta-seeded (13 ha with 64 sampling sites) (Figure 6). All three fields were furrow irrigated; with the Loma-north seeded site planted to Vaquero (yellow), Loma-south seeded site planted to Talon (yellow), and Delta-seeded site planted to Vaquero (yellow).

The five fields located along the Front Range also were identified by the nearest town; Ault-seeded (4 ha with 21 sampling sites) (Figure 7), Peckham-seeded (13.3 ha with 67 sampling sites) (Figure 8), Peckham-transplanted (3 ha with 16 sampling sites) (Figure 9), Brighton-north seeded (14.4 ha with 74 sampling sites) (Figure 10) and Brighton-south seeded (6.5 ha with 32 sampling sites) (Figure 11). All fields were furrow-irrigated. The Ault field was planted to Teton (yellow), Peckham-seeded planted to Tango (red), Peckham-transplant planted to Redwing (red), Brighton-north planted to Redwing (red), and Brighton-south planted to Teton (yellow). As the season progressed,

two fields were dropped from the survey (Delta-seeded and Brighton-north seeded) because no disease was observed. No data was collected from either location.

Visual ratings in the field were initiated once symptoms were observed on field borders. The ratings were based on incidence of IYSV over three meters from a single row at each flagged plot. Also, a count was conducted on five individual plants within each plot to identify the number of thrips present at the different sample locations. A field survey at the Ault site began 7 July with additional ratings on 22 July and 3 August. Due to the low disease incidence across the field (less than 1% incidence), no harvest data was collected at this site. At the Peckham-seeded site the survey, which included thrips counts and ratings for IYSV, was conducted on 12 July, 8 and 15 August. Soil samples were collected for each plot on 15 August. A complete soil analysis was conducted on each sample collected. At the Peckham-transplanted site, the survey was performed on 24 June, 7, 15 and 23 July and 8 August. Soil samples were collected on 12 August from this site. The Brighton-south seeded site was surveyed on 28 June, 15 and 22 July, 11 and 19 August. Soil samples were collected on 16 August from the Brighton-south seeded site. The survey at Loma-north seeded site began 11 July with additional surveys on 25 July, 9, 18 and 31 August. Soils were collected on 18 August for analysis. The Loma-south seeded site was surveyed on 11 and 25 July, 9 and 31 August and 13 September. Soils were collected from each plot on 18 August at this site.

Figure 4. Perimeter and plot locations (∇) for the 2005 Loma-north seeded site used in our IYSV survey.

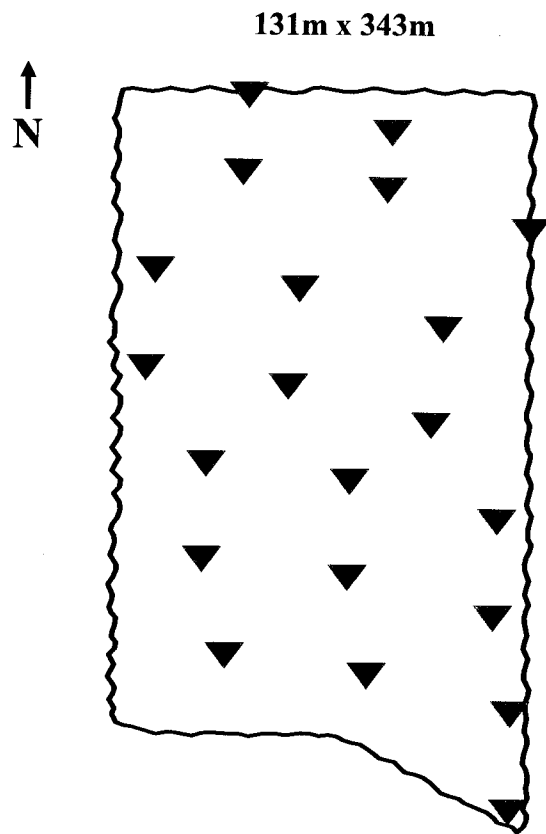


Figure 5. Perimeter and plot locations (∇) for the 2005 Loma-south seeded site used in our IYSV survey.

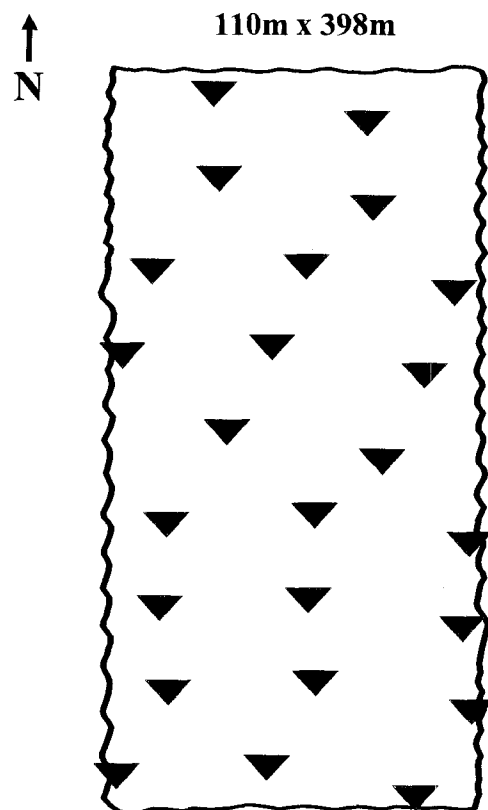


Figure 6. Perimeter and plot locations (▽) for the 2005 Delta-seeded site used in our IYSV survey.

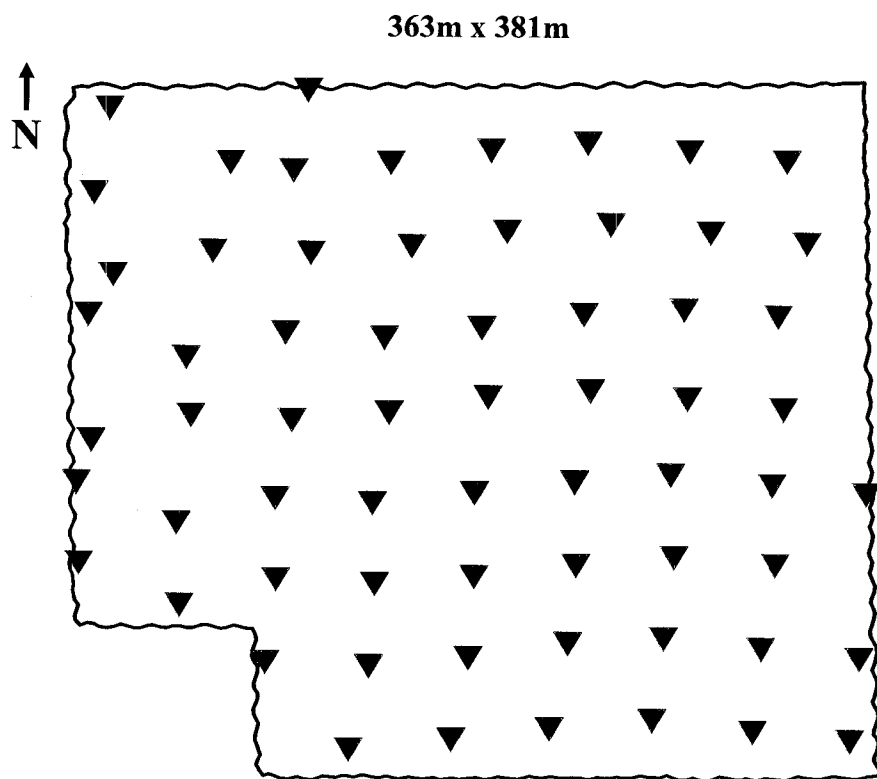


Figure 7. Perimeter and plot locations (V) for the 2005 Ault-seeded site used in our IYSV survey.

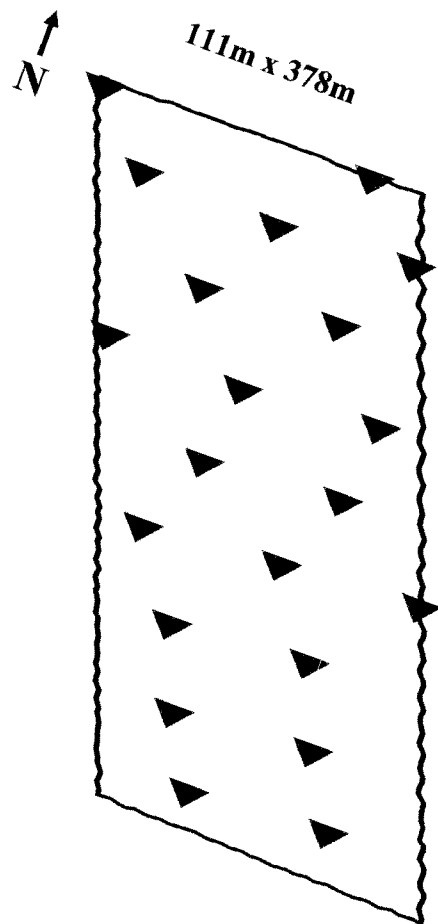


Figure 8. Perimeter and plot locations (∇) for the 2005 Peckham-seeded site used in our IYSV survey.

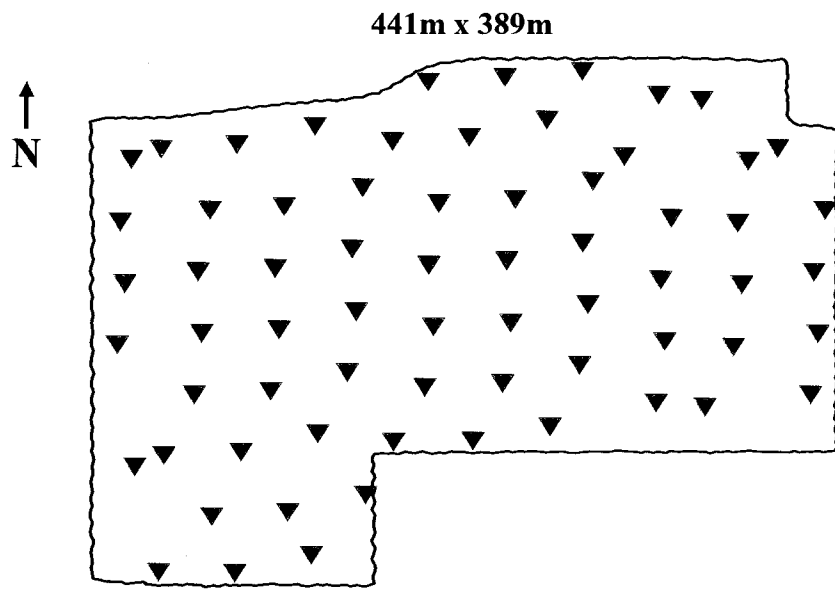


Figure 9. Perimeter and plot locations (V) for the 2005 Peckham-transplanted site used in our IYSV survey.

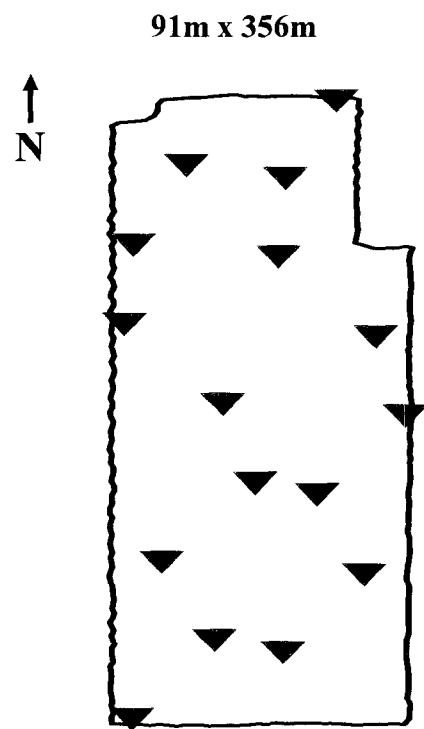


Figure 10. Perimeter and plot locations (V) for the 2005 Brighton-north seeded site used in our IYSV survey.

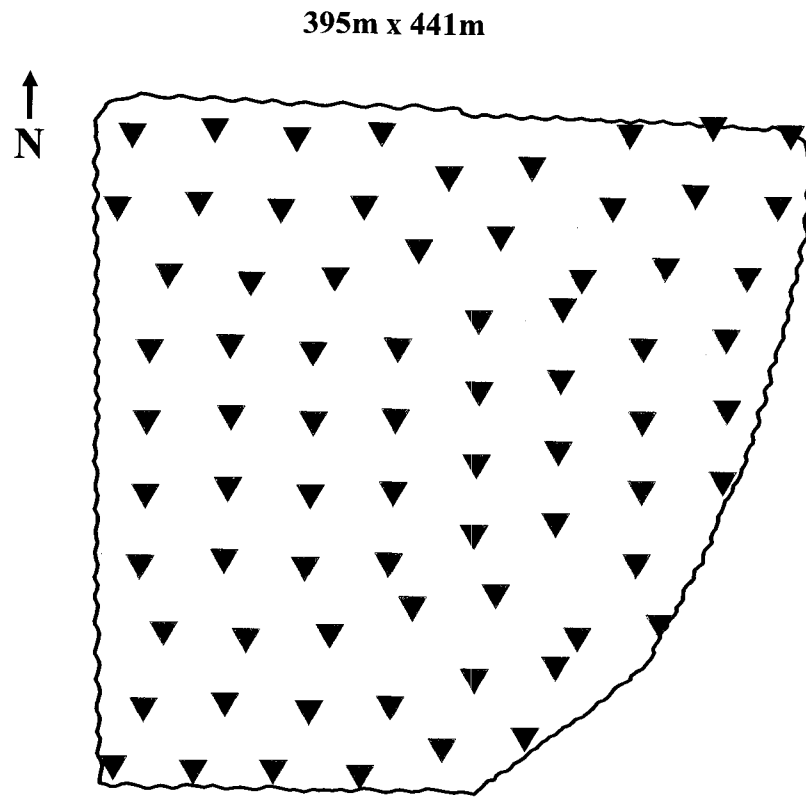
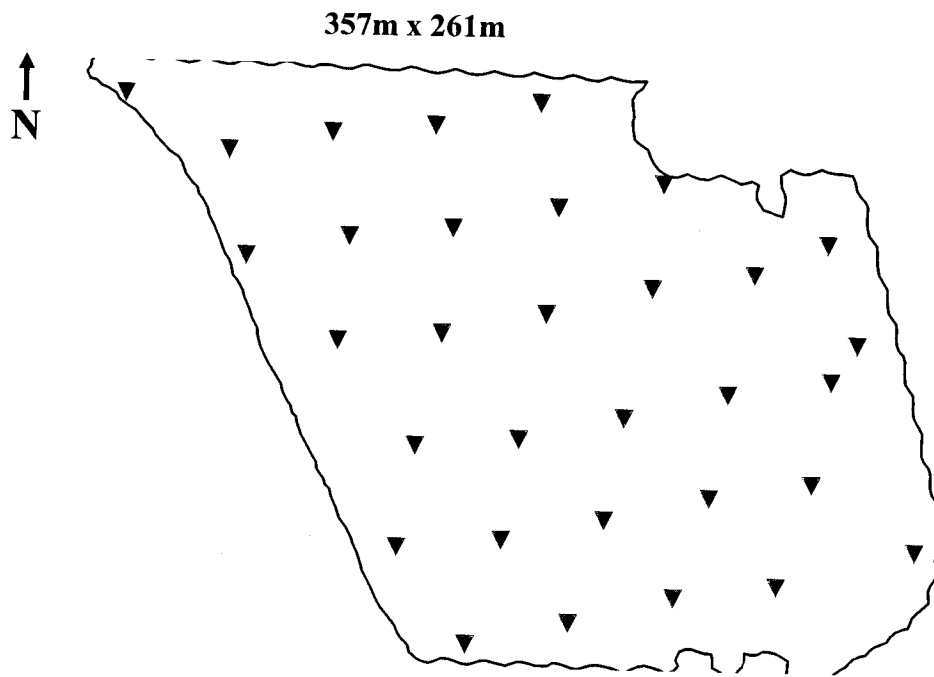


Figure 11. Perimeter and plot locations (∇) for the 2005 Brighton-south seeded site used in our IYSV survey.



Results:

2004 Season:

The early season ELISA results for all three fields yielded few positive IYSV samples (Table 13). Although some plants were positive in the serological tests, symptoms were not observed until early August. Visual symptoms were first observed 1 September on field border plants at the Ault site, but no symptoms were detected within our plots. Symptoms were never detected at the Eaton site. Ault and Eaton sites had intensive insecticide spray programs that may have contributed to the late appearance of IYSV symptoms. The Ault site was harvested on 9 September. Yield data collected from the Ault site were analyzed and correlated with the soil data. Independent variables were chosen using stepwise analysis (SAS Institute, Cary, NC). Three variables were significantly correlated to total yield explaining 41% of the variability observed (ppm potassium, $P<0.0001$; ppm phosphorus, $P=0.0323$; ppm calcium, $P<0.0001$). Twenty-nine percent of the variability in colossal market class yield was correlated to ppm magnesium and percent silt ($P<0.0001$ and $P=0.0001$, respectively). Jumbo market class onion yield was significantly correlated to ppm copper present in the soil explaining 23% of the variability ($P<0.0001$). Medium market class yield and plant population both had significant negative correlation to ppm potassium ($R^2=0.30$, $P<0.0001$; $R^2=0.39$, $P<0.0001$, respectively). Eaton was not harvested because bulbs were not yet mature and were severely damaged by hail. Peckham proved to be the most useful of all three locations. Incidence of IYSV was high and a border affect could be seen visually from our survey maps (Figure 12). Peckham was harvested on 17 and 20 September (Sterling

Table 13. Results of early onion plant samples analyzed using ELISA

Sample:	Ault	Location	
		Eaton	Peckham
1	11 ¹ (16%)	1 (2%)	4 (8%)
2	2 (3%)	3 (6%)	1 (2%)
3	3 (4%)	2 (4%)	0 (0%)

¹Number of plots with positive ELISA samples. No more than two of 10 plants were positive from any one plot.

²Percent of plots with positive ELISA samples.

and Tango, respectively). The Moran's I values, although significant, were very small for both cultivars (0.03, $P=0.003$ and 0.09, $P<0.001$ for Sterling and Tango respectively). Yield of the red cultivar 'Tango' was significantly impacted by IYSV incidence. Total yield, as well as jumbo and medium market class onions, were negatively correlated with IYSV incidence ($R^2=0.3137$, $P=0.0024$; $R^2=0.3923$, $P=0.0005$; $R^2=0.1910$, $P=0.0227$, respectively). Medium market class onion yields of white cultivar 'Sterling' were negatively correlated with disease incidence ($R^2=0.1693$, $P=0.0457$). Plant population of this cultivar also was negatively correlated to incidence of IYSV ($R^2=0.2566$, $P=0.0115$). Total yield and jumbo market class yield were not significantly correlated to disease incidence for this cultivar. Intensive thrips counts were only conducted at the Eaton site that was hailed out before the onset of virus symptoms, so no geo-spatial correlation was possible between thrips populations and the presence of IYSV.

2005 Season:

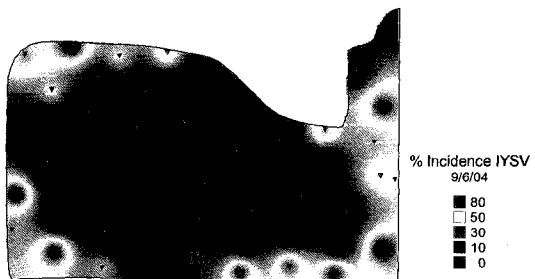
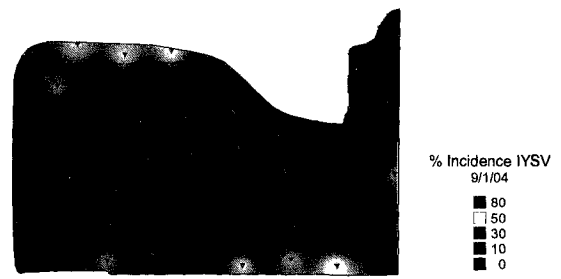
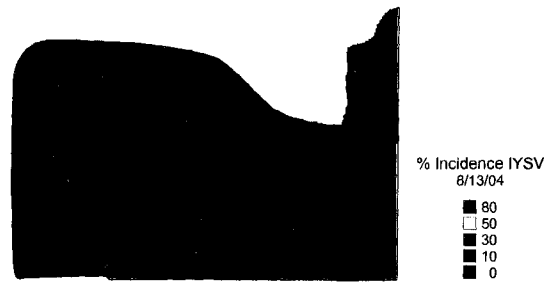
Iris yellow spot virus was identified and confirmed on volunteer onions sampled 2 March, 2005 in the vicinity of the Peckham field that was surveyed in 2004. This same area had the highest incidence of IYSV in 2005, possibly a result of over-wintering of the thrips vector or the virus on an as yet unidentified alternate host or within over-wintering thrips surviving in the soil or onion culls. All 2005 survey locations outside of the Peckham area had low levels of virus present. Several fields in the vicinity of Peckham had incidences approaching 100 percent resulting in severe necrosis of leaf tissue.

The Ault-seeded site had very low levels of IYSV present within the field. Although thrips populations did exceed 100 per plant in some areas throughout the field, damage was minimal and overall the field looked very healthy. This field was watered

on a continuous basis and no visual signs of water stress were apparent throughout the survey. Few weeds were present at this site, once again minimizing stress to the crop. The Ault-seeded field was approximately 32 kilometers from the Peckham site where we observed the highest incidence of IYSV in both 2004 and 2005. IYSV was only identified in two plots, consisting of one plant in each plot, at this location during our survey. The Moran's I values for IYSV at the Ault-seeded site was -0.065 for the final evaluation date signifying a near zero level of correlation between sample locations. Neither soil nor yield data was collected from this site due to the low level of disease present by the end of the season.

The Peckham sites, as mentioned previously, had very high levels of IYSV throughout the survey (Figure 13 and 15). The Peckham-seeded field also was one of the largest in the 2005 survey. Although this field was watered on a continuous basis, the high weed pressure and sandy soils resulted in extended periods of water stress for the crop. In much of the field, plants could not be seen through the thick canopy of weeds present. Plants appeared stunted and severe necrosis was apparent as a result of IYSV, severe thrips infestations (Figure 14) and unusually high levels of leaf miner (*Liriomyza* species) that was observed in several fields in the Peckham area. The analysis of soil collected from this site revealed significant correlations between several soil parameters and the visual observations we collected from each sampling location. The variability in plant stand had a significant and positive correlation to pH ($p=0.0038$) and ppm Cu ($p=0.0031$), and a significant and negative correlation to ppm Mg ($p=0.0408$). These correlations explained 26% of the variability in plant stand observed at this site

Figure 12. Percent incidence of IYSV, across 3 meters in each plot at the 2004 Peckham site (Figure 3) on 6, 13 and 23 August and 1, 6 and 10 September



($R^2=0.26$). The final incidence of IYSV was correlated to several soil parameters explaining 52% of the variability in disease across the field ($R^2=0.52$). Significant negative correlations were observed with ppm Ca ($p=0.0004$), ppm NH_4 ($p=0.0630$) and percent clay ($p=0.0235$). A positive correlation was observed with ppm Zn ($p=0.0027$) and ppm Fe ($p=0.0149$). The final observation of thrips number revealed a significant negative correlation with ppm NH_4 ($p=0.0433$), representing only 6% of the variability within the field ($R^2=0.06$). The yield analysis revealed a significant positive correlation between total yield and ppm B ($p=0.0223$), and a negative correlation between total yield and ppm Fe ($p=0.0015$), explaining 19% of the variability ($R^2=0.19$). Two soil parameters, ppm B ($p=0.0087$) and ppm Fe ($p=0.0360$) explained 15% of the variability ($R^2=0.15$) in jumbo yield with a positive and negative correlation, respectively. Lastly, medium yield had a significant negative correlation to ppm Fe ($p=0.0050$) and a significant positive correlation to percent organic matter ($p=0.0174$) with 19% of the variability explained ($R^2=0.19$). The second thrips population count had a significant correlation with the final IYSV incidence rating ($p=0.0005$) explaining 53% of the variability ($R^2=0.53$). No other parameter had a statistically significant impact on incidence of IYSV or yield at this survey location. Using the final IYSV incidence evaluations, we obtained a Moran's I value of 0.141 which signifies a low level of positive spatial autocorrelation; but due to the near zero value the correlation is not of any major biological significance.

A transplanted field near the previous seeded field in the Peckham area was the next used in our survey. This site had the highest incidence of IYSV of all fields surveyed in this study (Figure 15). Similar problems to those at the Peckham-seeded

field with high weed pressure and continuous water stress were observed at this site. Plants appeared stunted and severe necrosis was apparent as a result of IYSV, severe thrips infestations (Figure 16) and unusually high levels of leaf miner. The soil analysis of samples collected from this location revealed several correlations with factors including plant stand, incidence of IYSV, thrips pressure and yield. A positive correlation explaining 37% ($R^2=0.37$, $p=0.012$) of the variability in plant stand was found using percent of Na in the soil. High levels of S in the soil correlated to increased populations of thrips, explaining 25% of the variability ($R^2=0.25$, $p=0.048$). The final incidence of IYSV had a significant positive correlation with percent Na present in the soil describing 26% of the variability ($R^2=0.26$, $p=0.046$). Several soil parameters had an effect on total yield harvested from this location. A positive correlation was found between yield and percent Mg and soil pH ($p<0.0001$ and $p=0.007$, respectively), and a negative correlation with soluble salts ($p<0.0001$) explaining 89% of the yield variability ($R^2=0.89$). Jumbo market class yield was positively correlated with percent Mg ($p=0.004$) with 46% of the variability explained ($R^2=0.46$), while 60% ($R^2=0.60$) of the variability in medium market class yield was explained by percent Na in the soil ($p=0.0004$) with a positive correlation between these two parameters. A significant level of correlation was observed between plant stand and medium market class yield ($p=0.049$), resulting in 94% of the variability explained ($R^2=0.94$). The final incidence of IYSV revealed a Moran's I value of 0.042, a near zero value, signifying a non-significant level of spatial autocorrelation in disease across this field.

Figure 13. Percent incidence of IYSV, across 3 meters in each plot, at the 2005 Peckham-seeded site (Figure 8) on 12 July and 8 and 15 August.

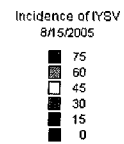
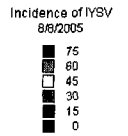
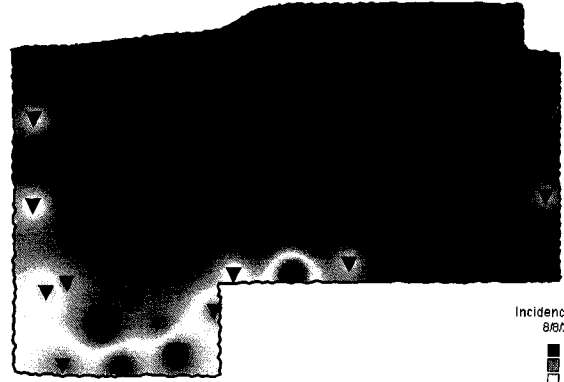
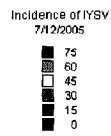


Figure 14. Thrips per plant, on 5 plants in each plot, at the 2005 Peckham-seeded site (Figure 8) on 12 July and 8 and 15 August.

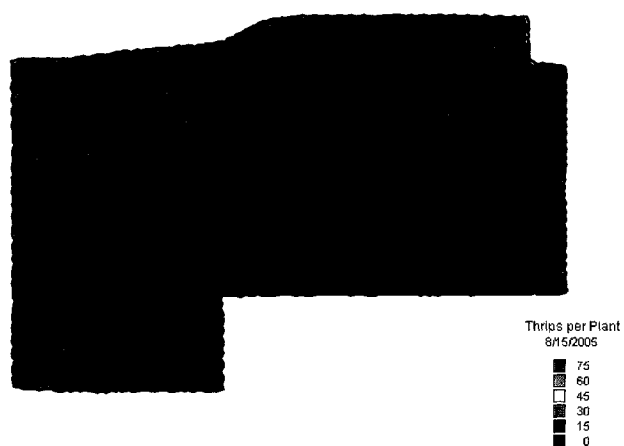
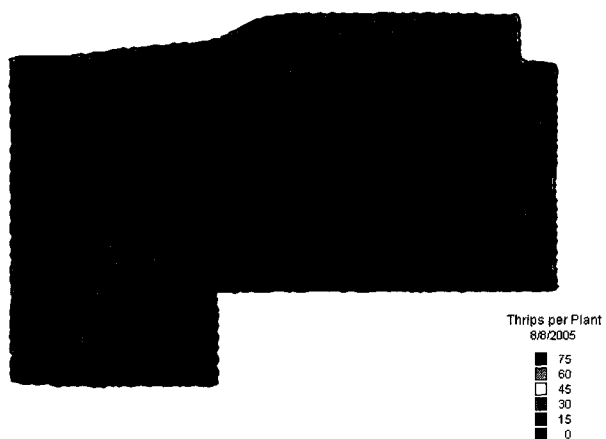
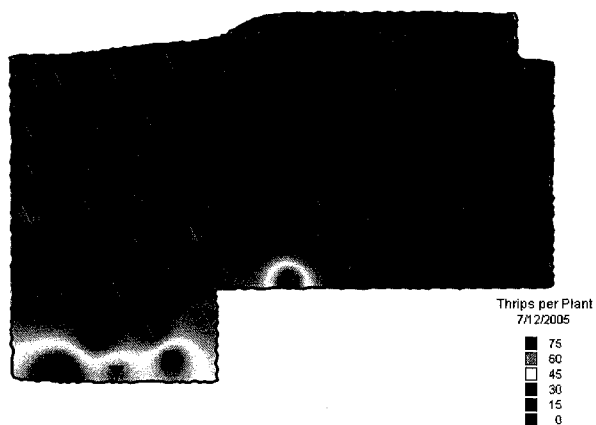


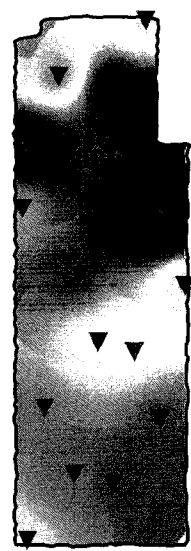
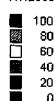
Figure 15. Percent incidence of IYSV, across 3 meters in each plot, at the 2005 Peckham-transplanted site (Figure 9) on 24 June, 7, 15 and 23 July and 8 August.



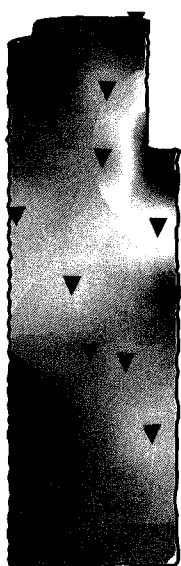
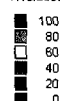
Incidence of IYSV
6/24/2005



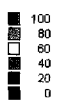
Incidence of IYSV
7/7/2005



Incidence of IYSV
7/15/2005



Incidence of IYSV
7/23/2005



Incidence of IYSV
8/8/2005

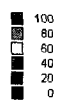
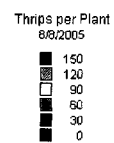
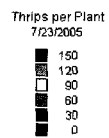
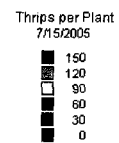
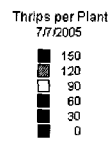
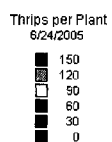
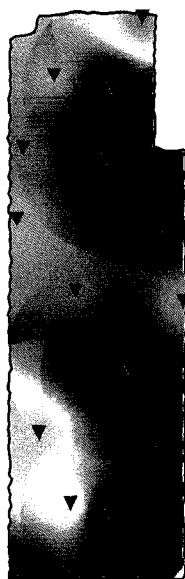


Figure 16. Thrips per plant, on 5 plants in each plot, at the 2005 Peckham-seeded site (Figure 9) on 24 June, 7, 15 and 23 July and 8 August.



The Brighton-south seeded site had very low levels of IYSV early and began to rise slightly later in the season (Figure 17), but did not reach damaging levels before harvest. The field was watered continuously and weeds were well controlled. Thrips levels were light to moderate throughout the season (Figure 18). The final incidence of IYSV, although low, did have a significant level of negative correlation with ppm Cu ($p < 0.0001$) and positive correlation with ppm Mg ($p = 0.0009$) present in the soil, describing 46% of the variability in disease ($R^2 = 0.46$). The final thrips count had a significant negative correlation with the percent silt of soil ($p = 0.0013$) explaining 29% of variability in thrips at this site ($R^2 = 0.29$). Incidence of IYSV at the time of our final survey had a positive significant correlation with plant stand ($p = 0.0019$), describing 84% of the variability in disease across the field ($R^2 = 0.84$). The Moran's I value for disease incidence at the Brighton site was 0.136, this implies a lack of spatial autocorrelation of disease incidence at this location. Plot flags were removed and the field harvested early by the grower, so no yield data was obtained.

Iris yellow spot virus was observed on only a few plants at our Western Slope survey locations (Figures 19 and 21). The Loma-north seeded site had minimal weed pressure was low. Moderate water stress was apparent throughout the season on the plants at this site. Thrips populations were low for much of the season with levels beginning to climb late (Figure 20). Analysis of the soils collected found a negative correlation between plant stand and ppm K ($p = 0.0056$), describing 34% of variability in stand ($R^2 = 0.34$). Nearly 74% of the variability in yield ($R^2 = 0.74$) could be explained by a significant negative correlation with ppm Zn ($p = 0.0034$) and ppm Cu ($p = 0.0243$), and a negative correlation with percent silt ($p = 0.0005$). Jumbo market class variability was

positively correlated with percent silt ($p=0.0037$) and negatively correlated with ppm Zn ($p=0.0114$), explaining 56% of the variability in jumbo yield ($R^2=0.56$). Medium market class yield was negatively correlated to both ppm Zn and soluble salts ($p=0.0308$ and $p=0.0484$, respectively), explaining 38% of the medium yield variability ($R^2=0.38$). Final incidence of IYSV was positively correlated with ppm Fe ($p=0.0181$), explaining 26% of the variability in disease ($R^2=0.26$). On our final day of observations, the thrips counts had a significant correlation with the incidence of IYSV ($p=0.0228$), with 90% of the variability being explained ($R^2=0.90$). The Moran's I value for the final IYSV observation was -0.019 indicating no spatial autocorrelation in incidence of IYSV based on our 0.2 ha plot grids.

The final site used in our survey was approximately two km south of the previous field and identified as Loma-south seeded. This field also had minimal weed pressure and had no signs of water stress throughout the season. Thrips populations were very low until late in the season (Figure 22), and incidence of IYSV was minimal (Figure 21). The soil analysis from this location revealed a negative effect on stand with increased levels of ppm S ($p=0.0285$) and a positive effect with increased levels of percent sand ($p=0.029$), with 41% of the variability explained ($R^2=0.41$). The final thrips observation was positively correlated with organic matter content ($p=0.0003$) in the soil with 45% of the variability explained ($R^2=0.45$). A total of 32% of the variability in total yield ($R^2=0.32$) was explained by a positive correlation with percent sand ($p=0.0041$). Colossal market class onions had a significant positive correlation with ppm S ($p<0.0001$) present in the soil, explaining 64% of the observed variability ($R^2=0.64$). Medium market class yield variability had a significant negative correlation with ppm Mn

and soluble salts ($p=0.0014$ and $p=0.0181$, respectively), explaining 43% of this variability ($R^2=0.43$). The first, second and fifth thrips observations were significantly correlated with plant stand ($p=0.0001$, $p=0.0319$ and $p=0.0265$, respectively), explaining 99%, 93% and 94% of the variability in thrips populations ($R^2=0.99$, $R^2=0.93$ and $R^2=0.94$, respectively). No correlation between IYSV incidence or thrips population and yield were observed at this site. The Moran's I value of -0.044 indicated no spatial autocorrelation with the *Iris yellow spot virus* at this survey location.

Discussion:

The results from our trials have led us to believe that positive spatial autocorrelation of IYSV is limited and that any form of secondary spread of the virus that is occurring is doing so in a random fashion. These results are similar to those observed by Gent et al. (2004) and suggest that the 0.2 ha grid sampling is still too large to observe spatial correlation between sampling locations. To identify the role of secondary spread of IYSV within the field, we would need to sample more intensively, less than 0.2 ha, to identify the scale at which spatial dependency can be recognized. Having used the Random Systematic Unaligned sampling technique to layout our sampling sites actually resulted in several plots being within a few feet of one another. Even at this close distance we were unable to observe any significant level of spatial dependence. In 2004, the Ault and Eaton locations both had very extensive pest control programs that may have resulted in the delay in appearance of IYSV symptoms. Management practices leading to reduced stress of the crop was likely a major factor in the reduced levels of IYSV at this site even in the presence of high thrips populations. High incidence of

Figure 17. Percent incidence of IYSV, across 3 meters in each plot, at the 2005 Brighton-south seeded site (Figure 11) on 28 June, 15 and 22 July and 11 and 19 August.

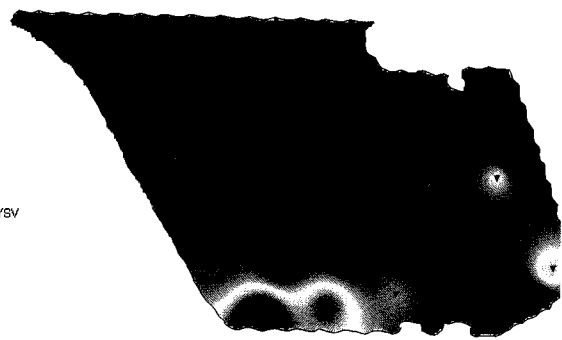
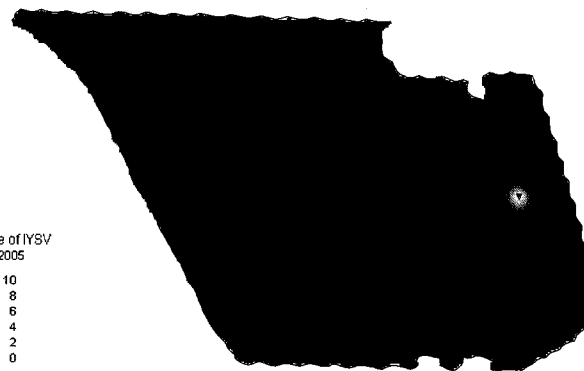


Figure 18. Thrips per plant, on 5 plants in each plot, at the 2005 Brighton-south seeded site (Figure 11) on 28 June, 15 and 22 July and 11 and 19 August.

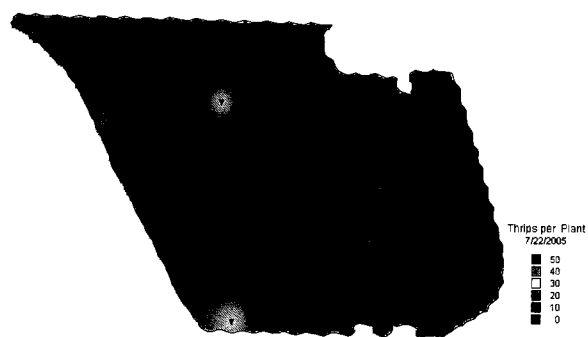
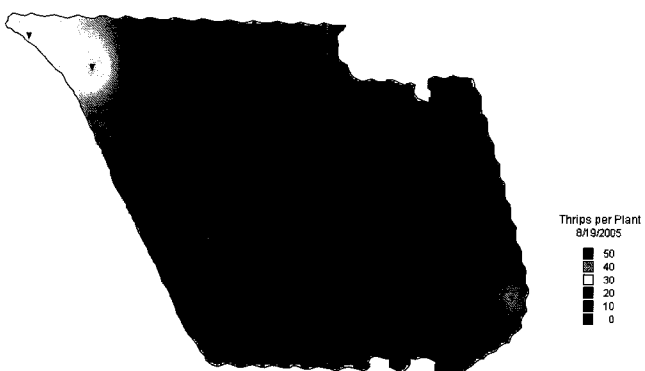
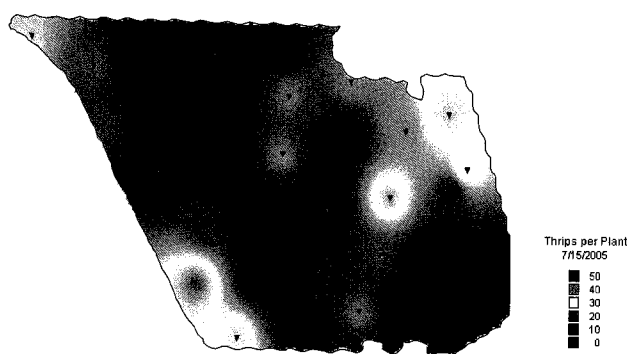
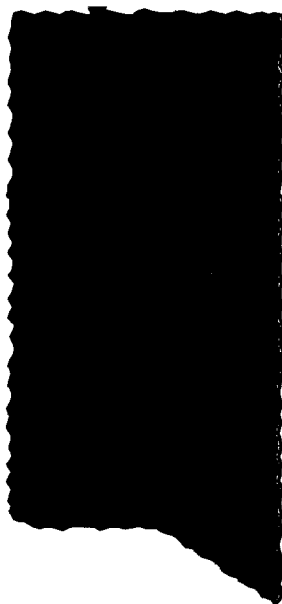


Figure 19. Percent incidence of IYSV, across 3 meters in each plot, at the 2005 Loma-north seeded site (Figure 4) on 11 and 25 July and 9, 18 and 31 August.



Incidence of IYSV
7/1/2005

10
8
6
4
2
0



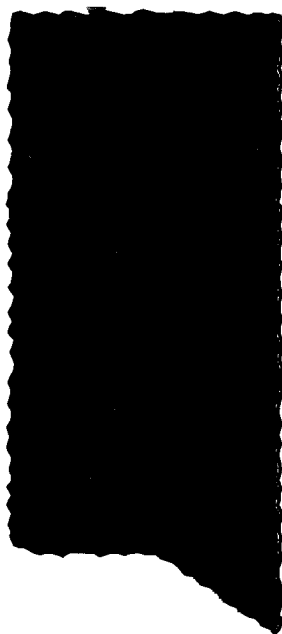
Incidence of IYSV
7/25/2005

10
8
6
4
2
0



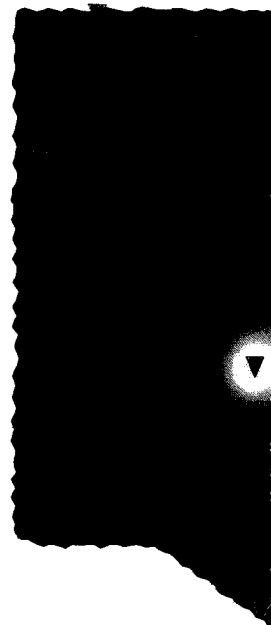
Incidence of IYSV
8/8/2005

10
8
6
4
2
0



Incidence of IYSV
8/18/2005

10
8
6
4
2
0



Incidence of IYSV
8/31/2005

10
8
6
4
2
0

Figure 20. Thrips per plant, on 5 plants in each plot, at the 2005 Loma-north seeded site (Figure 4) on 11 and 25 July and 9, 18 and 31 August.

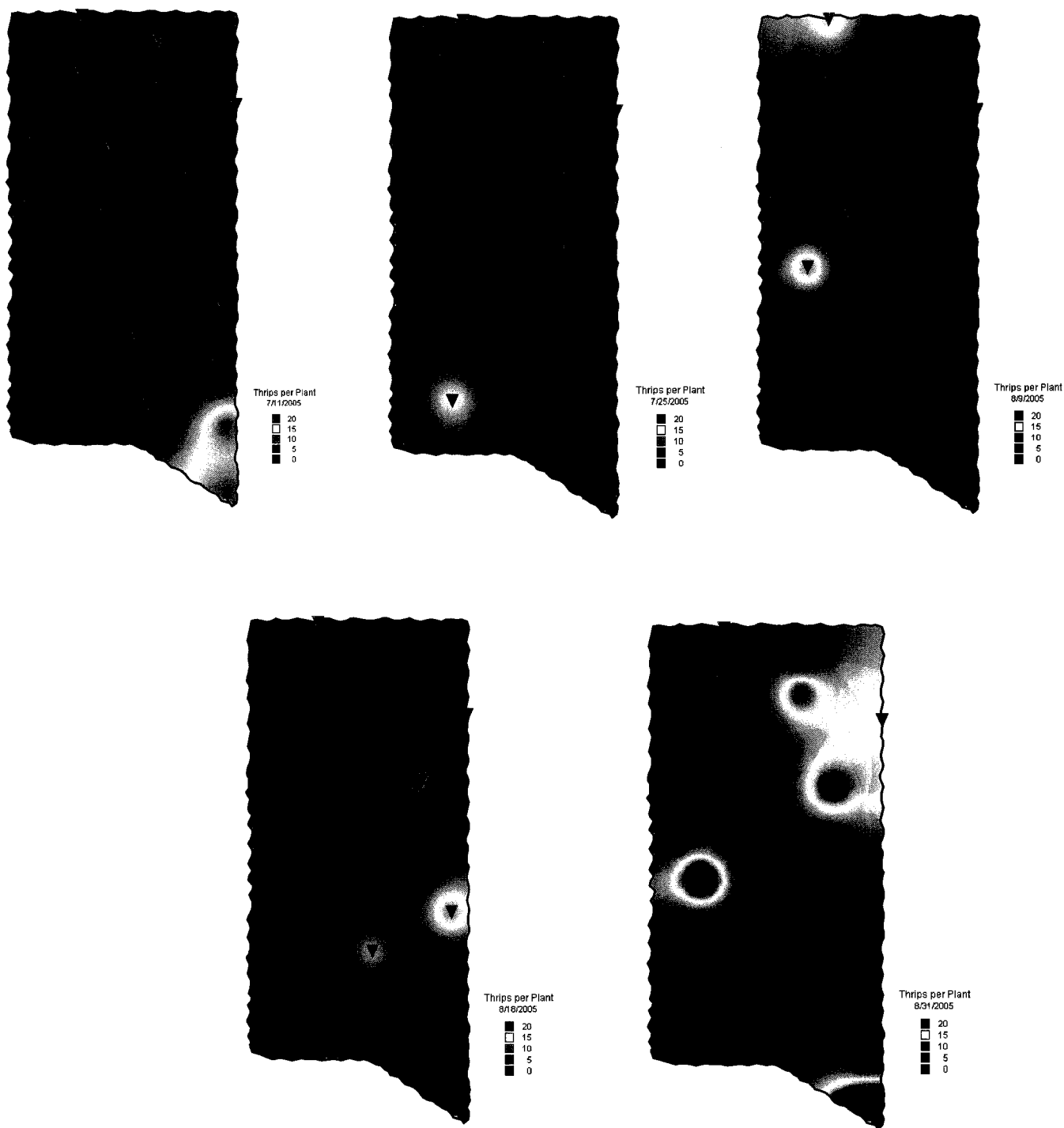
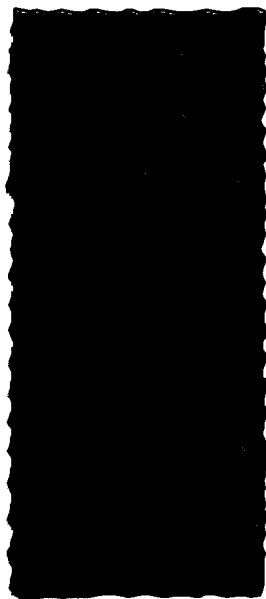
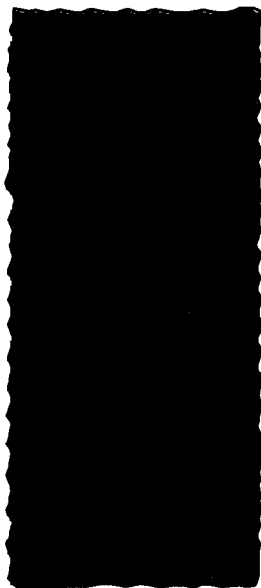


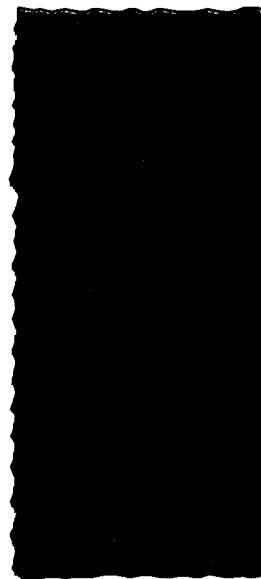
Figure 21. Percent incidence of IYSV, across 3 meters in each plot, at the 2005 Loma-south seeded site (Figure 5) on 11 and 25 July, 9 and 31 August and 13 September.



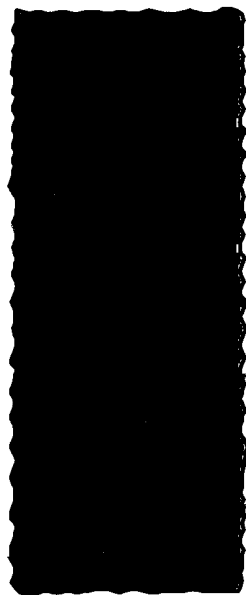
Incidence of IYSV
7/11/2005



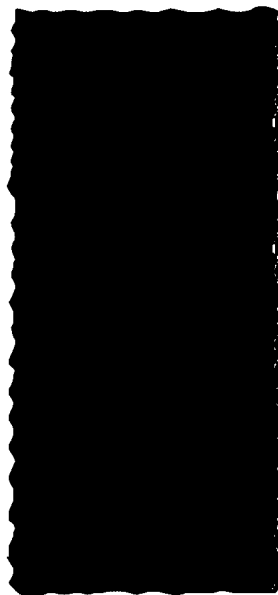
Incidence of IYSV
7/25/2005



Incidence of IYSV
8/9/2005



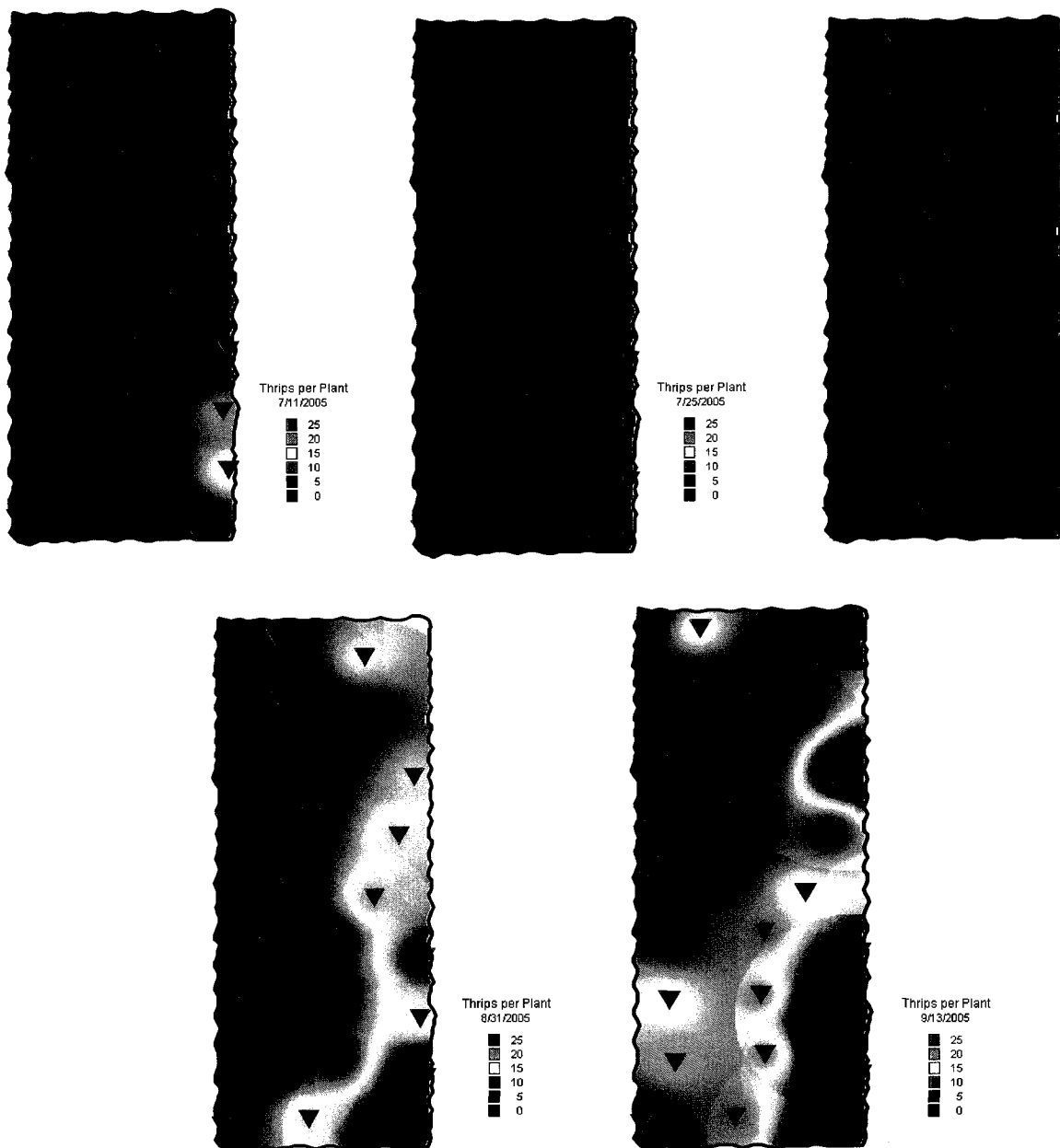
Incidence of IYSV
8/31/2005



Incidence of IYSV
9/13/2005



Figure 22. Thrips per plant, on 5 plants in each plot, at the 2005 Loma-south seeded site (Figure 5) on 11 and 25 July, 9 and 31 August and 13 September.



IYSV at our 2004 Peckham site may have been attributed to greater disease pressure in the region. This site was in the vicinity of several transplanted onion fields that tested positive earlier in the season for IYSV using ELISA. Harvest of these early transplanted fields occurred through the month of July. It is possible that the thrips vectors migrated and became established in nearby seeded fields during transplant field harvest. This corresponds with the onset of symptoms in our field. High weed pressure and continuous drought stress also were likely factors leading to increased incidence and/or expression of IYSV at this location.

In 2005, disease pressure was once again high in the two fields in the Peckham area. Volunteer onions showing symptoms of the *Iris yellow spot virus*, and testing positive using ELISA, were obtained from the 2004 Peckham site in early March 2005. On the west slope, we did not observe symptoms of IYSV until late in the season, around the time that an increase in the population of thrips began to take place. If the season had gone on longer, the incidence of IYSV would likely have increased to more damaging levels.

CHAPTER IV

Summary of Trials

The *Iris yellow spot virus* has proven to be a major player in production losses throughout much of the U.S. and the world onion market (Gent et al., 2006). With the advent of new technologies and novel chemistry pesticides, our ability to combat pest outbreaks has improved. In the several years that we have conducted our surveys, we have, thus far, been unable to identify the importance and pattern of secondary spread of IYSV. At this time, it appears that dissemination of IYSV from a source outside the field is the principal means of infection.

In the preceding chapters, I have introduced several materials that we have tested, both novel and conventional, for the ability to manage the IYSV pathogen and its thrips vector. Research has proven that the best technique to manage a pest most often involves an integrated pest management (IPM) program. Our research also validates this as was shown by the improved control of thrips with various forms of ground cover (mulch) in combination with an insecticide treatment. Prior research conducted by Gent et al. (2004) has shown that different onion cultivars show varying degrees of resistance and susceptibility to the IYSV complex. Based on our work and previous studies, the best recommendation for an effective management program would involve incorporating several techniques such as cultivar selection, groundcover, an effective rotation of novel chemistry and conventional insecticides, and cultural and sanitation practices to reduce vector populations and stress upon the host crop. This research also identified several correlations of both disease incidence and yield that appeared related to different soil parameters such as Na and soluble salts. It is probably a good idea to know the chemistry

of your soils before planting onions when IYSV is an issue. Stress is an important component when it comes to IYSV, and no matter how well you take care of your crop on the surface, what lies below the soil can still limit your yield capabilities.

More information is continuously being revealed about the virus that causes iris yellow spot disease and its vector *T. tabaci*. There is ongoing research to identify the effectiveness of different pesticide chemistries for their ability to control IYSV. Also, work is underway to identify possible alternate hosts that may act as an over-wintering source of the virus or its thrips vector. As we learn more of this pest complex, new techniques can be developed and tested to improve our ability to manage these pests.

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