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Management of the western flower thrips, Frankliniella occidentalis (Pergande) (Thysanoptera: Thripidae) in the greenhouse

presented by

Andi Nasruddin

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## MANAGEMENT OF THE WESTERN FLOWER THRIPS, Frankliniella occidentalis (Pergande) (Thysanoptera: Thripidae) IN THE GREENHOUSE

Вy

Andi Nasruddin

A THESIS

Submitted to
Michigan State University
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MASTER OF SCIENCE

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#### **ABSTRACT**

MANAGEMENT OF THE WESTERN FLOWER THRIPS, Frankliniella occidentalis (Pergande) (Thysanoptera: Thripidae) IN THE GREENHOUSE

By

#### Andi Nasruddin

The relationship of the time interval between insecticide applications to Frankliniella occidentalis (Pergande) damage to gloxinia (Sinningia speciosa Benth & Hook) flowers, population density of F. occidentalis, and flower phytotoxicity was determined through greenhouse experiments. Application of abamectin and chlorpyrifos at 5- and 10-day intervals and application of cyfluthrin at 5-day interval held thrips flower injury to a tolerable level. For abamectin and cyfluthrin, the number of thrips per flower decreased as the interval between applications decreased. The amount of insecticide phytotoxicity to flowers increased as the application interval decreased. The potential of gloxinia seedlings as indicator plants for tomato spotted wilt virus (TSWV) was evaluated. Two thrips per seedling were sufficient to transmit the virus to gloxinia seedlings. Yellow cards increased attractiveness of gloxinia seedlings to F. occidentalis. However, gloxinia without yellow cards developed TSWV symptoms at the same time and at the same rate as the gloxinia with yellow cards.

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# CHAPTER I

IDENTIFICATION AND CHARACTERISTICS OF

WESTERN FLOWER THRIPS, Franklniella occidentalis

(Pergande), AND TOMATO SPOTTED WILT VIRUS

#### Introduction

The western flower thrips, Frankliniella occidentalis

(Pergande), was originally found from sea level to subalpine
altitude in western North America (Bryan & Smith 1956), while
eastern flower thrips, F. tritici, was found primarily in the
eastern U.S. and Canada (Robb & Parella 1986,
Broadbent et al. 1987). Currently F. occidentalis is
considered the most prevalent species of thrips attacking
floriculture crops and a major greenhouse plant pest in
North America (Allen & Broadbent 1986, Firth 1986,
Linguist 1986, Parella & Robb 1987, Robb 1990).

F. occidentalis has a wide plant host range of 139 species in California (Bryan & Smith 1956). It damages plants directly by feeding on flowers and indirectly by transmitting tomato spotted wilt virus (TSWV) (Sakimura 1962). Like F. occidentalis, TSWV has also a tremendous number of host species; some 236 species in 34 families, including vegetables and ornamentals grown in subtropical and temperate climates (Cho et al. 1986, Matteoni et al. 1987).

Greenhouse crop growers rely heavily on insecticide use to control F. occidentalis and to prevent the spread of TSWV. However, the thrips has shown resistance to carbamate, organophosphate, pyrethroid, and macrocyclic lactone

(Robb 1990).

Since the genus Frankliniella consists of 145 species (Moulton 1948), accurate identification of species to distinguish F. occidentalis from other Frankliniella species that do not transmit TSWV is important.

A scanning electron microscope study was undertaken to investigate morphological characters of the species.

### Identification of F. occidentalis

Taxonomic characters used to identify species, genera, and families of thrips (Thysanoptera) range from the very obvious, such as color and body shape, to the very obscure, such as internal structures or fine details of body sculpture (Palmer et al. 1989). Characters currently used in the identification of <u>F. occidentalis</u> are discussed below. Suborder Terebrantia. Forewings, when present, usually have distinct longitudinal veins bearing setae (Fig. 1). Females have saw-like ovipositors (Fig. 2). Abdominal segment X of thrips in this suborder are usually conical (Fig. 3) (Palmer et al. 1989).

Family Thripidae. Antenna has 8 segment and sensory area of antennal segments 3 and 4 are developed into forked or simple sensecones (Fig. 4) (Bryan and Smith 1956, Stannard 1968, Palmer et al. 1989).

Genus Frankliniella. Thrips in this genus have a head that is wider than it is long, eyes normal, occili always present, interocellar and postocular setae normally well developed (Fig. 5), antennae with 8 segments. Antennal segments 3 and 4 are with forked sensecone, 7 and 8 are smaller and forming a style (Fig. 4) Mouthcone is moderately stout and rounded. Prothorax is wider than long. Pronotum has a pair of long anteromarginal setae and an addition of small setae between the median posteromarginal setae. There are usually 5 minor setae on either side along the posterior margin (Fig. 5). Wings are nearly pointed with fringes on both anterior and posterior margins. Each forewing has longitudinal veins (costa, first and hind veins), which have regularly and uniformly placed setae (Fig. 1) (Stannard 1968, Palmer et al. 1989).

The position of the interocellar setae may be determined by an imaginary triangle formed by lines connecting the centers of three ocelli. Position 1 indicates that the interocellars are placed laterad of the anterior ocellus and entirely outside the triangle. Position 1-2 indicates that the interocellars are on the line connecting the outer margins of the anterior and posterior ocelli. Position 2 means the interocellars are located within the triangle. Position 2-3 indicates that these setae are approximately on the line connecting anterior margins of the posterior ocelli;

and position 3 means they are located immediately between the posterior ocelli. Abdominal tergite VIII is with or without complete comb of setae on posterior margin (Fig. 3) (Moulton 1948, Palmer et al. 1989).

Species occidentalis. Interocellar setae are in the position 2. Interval between posterior ocelli is more than 2 times than their diameter. Major postocular setae are very long, often almost as long as ocellar seta III (Fig. 5). Length of antennal segment III, VII and VIII are 48-50, 10, and 13 microns, respectively. Pedicel of the antennal segment III is without a sharp angulation (Fig. 4). Eighth tergum comb is absent or extremely weak and usually visible only at sides (Fig. 3). Pronotal anteroangular setae are almost as long as anteromarginals (Fig. 5). Immature forms are clear to yellow in color. Adult thorax is usually shaded with orange; each tergum with line along anterior margin or there may be a grayish band sometimes heavier in the middle; legs concolor with head; wings washed with yellow, antennal segment 1 concolor with head, antennal segment 2 brownish yellow, segments 3-5 mostly yellow, shaded brown in apical portions, or segment 5 only at extreme apical end, segments 6-8 grayish brown. Length of body totals 1.0-1.2 mm (Moulton 1948, Stannard 1968, Bryan and Smith 1956, Allen & Broadbent 1986, Palmer et al. 1989).

Biology of E. occidentalis. Eggs are deposited by adults within leaf and petal tissue. At temperature fluctuating between 16.6-36.6 OC the eggs hatch in 2.5-4 days (Parella & Robb 1987). Small translucent first instar of nymphs begin to feed immediately after the eggs hatch. The nymphs molt to the second instar after 1-2 days. The second instar is yellow. It is an active feeder. Both instars generally occur in tight, protected areas such as the petals and developing terminal foliage. The second instar turns white just before molting and moves down the plant to pupate in soil or leaf debris. The second nymphal stadium lasts 2-4 days at the fluctuating temperature (Parella & Robb 1987).

Thrips are unique among other insects in that they pass through two pseudopupal stages (prepupa and pupa). In the prepupal stage antennae are short and positioned toward of the head while the wing buds are moderately short and along the side of the body. This is followed by the pupal stage characterized by antennae over the head dorsally and the wing buds elongate (Stannard 1968, Palmer et al. 1989). The prepupal and pupal stages, respectively, last 1-2 days and 1-3 days. Therefore, 7.5-13 days are required by F. occidentalis to develop from eggs to adults in fluctuating temperature (Robb & Parella 1987).

Females of F. occidentalis are larger than males.

The adult females may live 30-45 days and can deposit

150-300 eggs during her lifetime. The haploid males are
derived from unfertilized eggs. In the greenhouse 25-30%

of F. occidentalis are males (Mound 1976, Robb & Parella 1987).

## Characters of Tomato Spotted Wilt Virus

Tomato spotted wilt virus (TSWV) (R/1:  $\Sigma$ 7.4/\*:S/S:S/Th) (Den Hurk et al. 1977) has some uniquenesses. It has one of the widest known host range of any plant virus; it is the only plant virus transmitted in a persistent manner by certain species of thrips; it is highly unstable in vitro; and its particles are covered by lipoprotein envelope (Best & Katekar 1964, Best & Palk 1964, Ie 1964, Kammen et al. 1966, Best 1968, Francki & Hatta 1981). Another uniqueness of TSWV among other plant viruses is that of the only enveloped virus with a positive-stranded and segmented RNA genome (Verkleij et al. 1982).

Particle Structure and Properties. TSWV particles are approximately isometric with 70-90 nm in diameter, apparently bounded by a membrane. The outer layer of the membrane seems to consist of a nearly continuos layer of projection ca. 5 nm thick, which stains more densely than the membrane itself. Particle sedimentation coefficient (S20,w) is 530 (Best 1968).

Stability in Plant Sap. Physically and chemically, TSWV is one of the most unstable plant viruses. In plant sap the thermal inactivation point (10 min.) is 40-46 °C. Longevity in vitro at room temperature is 2-5 h. Dilution end point

is between  $2x10^{-2}$  and  $1x10^{-3}$ . Infectivity rapidly falls at values below pH 5 and a pH value near 7 is the best. Stability of TSWV in plant sap can be enhanced by adding a reducing agent such as Na<sub>2</sub>SO<sub>3</sub> (Best 1968, Ie 1971). Transmission of TSWV. TSWV is transmitted by six species of thrips (Order:Thysanoptera): Frankliniella occidentalis (Pergande), F. fusca (Hinds), F. schultzei (Tribom), Scirtothrips dorsalis Hood, Thrips setosus Moulton and T. tabaci Lindeman (Best 1968, Paliwal 1976, Amin et al. 1981, Cho et al. 1989). As F. occidentalis became abundant in commercial greenhouses, reports of damage caused by TSWV increased drastically in North America (Allen & Broadbent 1986, Broadbent et al. 1987, Allen & Matteoni 1988). TSWV is transmitted by adults that have fed on diseased plant as nymphs (Sakimura 1962). F. occidentalis requires 5-15 minutes for acquisition-access feeding and 2-10 days for latent period. They need 15-30 minutes for inoculation feeding (Robb et al. 1988). This species is able to transmit TSWV throughout its adult lifetime; however, they do not pass the virus to their progeny (Ananthakrishnan 1980).

Tomato spotted wilt virus can also be transmitted through seeds. Jones (1948) reports that 96% seed transmission occurred on cineraria and tomato. There is no report showing that the virus is transmitted by dodder (Ie 1971). Another way that TSWV can be transmitted is mechanically with infected plant sap.

Observations on some virus isolates and their epidemiological behavior suggest that TSWV may have evolved to become adapted to different groups of host species. For example, some strains of the virus have become adapted to solanaceous hosts, others to beans and yet others to cucurbits (Francki & Hatta 1981). Recently, strains referred to as the Impatiens (I) and Lettuce (L) strains have become increasingly important because of their damage potential and distribution throughout the country. An ELISA kit for serological identification of both strains is commercially available by Agdia, Inc. (Mishawaka, Indiana).

TSWV has often been isolated from diseased plants that are also infected by other viruses, sometimes two viruses have successfully replicated in the same cell. Although in some cases it has been observed that the symptoms on such plant are similar to those infected by TSWV alone, on others, the co-infections may alter the appearance of symptoms associated with TSWV (Francki 1981).

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Figure 1. Forewing of Frankliniella occidentalis with longitudinal vein bearing setae (arrow). Bar=49  $\mu m$ .



FIGURE 1

Figure 2. Saw-like ovipositor of adult female of E. occidentalis (arrow). Bar=100  $\mu m$ .



FIGURE 2

Figure 3. Adult Frankliniella occidentalis with conical abdomen segment X (arrow). Bar=0.17  $\mu m$ .



FIGURE 3

Figure 4. Antenna of Frankliniella occidentalis with forked sensory area of antennal segment 3 and 4 and segment 7 and 8 forming style-like structure. Bar=40  $\mu m$ .



FIGURE 4

Figure 5. Dorsal view of head and prothorax of Frankliniella occidentalis, showing important taxonomical characteristics: a) interocellar setae(ocellar setae III) in the position 2 b) ocellus c) post ocular setae d) pronotal anteromarginal setae e) pronotal anteroangular setae. Bar=48  $\mu$ m.



FIGURE 5

## CHAPTER II

RELATIONSHIP OF <u>Frankliniella occidentalis</u>

(Thysanoptera: Thripidae) POPULATION DENSITY AND

FEEDING INJURY TO THE FREQUENCY OF INSECTICIDE

APPLICATIONS TO GLOXINIA

#### **ABSTRACT**

Abamectin, chlorpyrifos and cyfluthrin applied at 5-, 10- or 15-day intervals reduced the amount of feeding injury to gloxinia (Sinningia speciosa Bent and Hook) flowers from western flower thrips, Frankliniella occidentalis (Pergande). Less than 9 mm<sup>2</sup> of feeding injury per flower sample was found on flowers from all insecticide treatments compared with 74 mm<sup>2</sup> on control flowers. Five- and 10-day intervals between applications of abamectin and chlorpyrifos, and 5-day intervals between applications for cyfluthrin held thrips flower injury to a tolerable level. For abamectin and cyfluthrin, the number of thrips per flower and the amount thrips injury to flowers decreased as the length of time between applications decreased. The frequency of chlorpyrifos applications did not correlate well with thrips populations. The amount of flower injury attributable to insecticide phytotoxicity increased as the interval between applications decreased  $(r^2=0.79-0.86)$ , and approached unacceptable levels (>4.0% of petal tissue necrotic) at 10-day intervals for chlorpyrifos, and at 5-day intervals for abamectin, cyfluthrin, and chlorpyrifos.

## Introduction

Western flower thrips, Frankliniella occidentalis

(Pergande) is now the most prevalent species of thrips

attacking floricultural crops in the United States and Canada

and is considered by many to be the most important greenhouse

plant pest in North America (Allen & Broadbent 1986, Firth

1986, Linquist 1986, Parella & Robb 1987, Robb 1990).

Recently, F. occidentalis has also been reported to cause

serious injury to variety of flower crops in the Netherlands

(Cevat 1989).

On gloxinia (Sinningia speciosa Benth & Hook)

E. occidentalis may feed on young seedlings, causing distorted growing point. F. occidentalis also infests gloxinia flowers and may render a crop unusable by leaving white streaks and patches where they feed on the petals. Although F. occidentalis feeding injury is undesirable, the greatest threat to gloxinia crops is from the transmission of tomato spotted wilt virus (TSWV). The virus is spread only by a few species of thrips vectors: F. occidentalis (Pergande), F. fusca (Hinds), F. schultzei (Trybom), Scirtothrips dorsalis Hood, Thrips setosus Moulton, and T. tabaci Lindeman (Best 1968, Paliwal 1976, Amin et al. 1981, Cho et al. 1989). As F. occidentalis became widespread in

North America greenhouses, reports of damage from TSWV increased dramatically (Allen & Broadbent 1986, Broadbent et al. 1987, Allen & Matteoni 1988, Cho et al. 1989). Epidemics of TSWV in tomato, Lycopersicum esculentum; impatiens, Impatiens wallerana; Dahlia x hybrida; calceolaria, Calceolaria herbeohybrida; cyclamen, Cyclamen persicum; marigold, Tagetes erecta; cineraria, Senecio cruentes; begonia, Begonia semperflorans and other greenhouse crops have caused serious economic losses for greenhouse growers (Allen & Broadbent 1986, Allen & Matteoni 1988, Baker & Jones 1988). Some of the most devastating loses were in gloxinia crops (Baker & Jones 1988). Some greenhouse managers avoid growing gloxinia plants adjacent to impatiens or other plants that are susceptible to <u>F</u>. <u>occidentalis</u>. Other strategies for managing F. occidentalis infestation and TSWV infection include complete weed management inside and adjacent to greenhouses, inspection of incoming plant material and insecticide suppression of F. occidentalis to the lowest level possible (Nameth et al. 1988).

Although some information on insecticide management of F. occidentalis is available for roses (Parella & Robb 1989), chrysanthemums (Hatta & Hara 1989) and some other greenhouse plants, there are no reports on management of F. occidentalis on gloxinia. Most previous studies of F. occidentalis management have been with 5- or 7-day intervals between insecticide applications. This intense schedule is undesirable for greenhouse managers because of the labor

required for spraying, the potential hazard to greenhouse workers and insecticide phytotoxicity to flowers. More information is needed to define the trade-off between the frequency of insecticide applications and control of E. occidentalis.

The purpose of this study was to determine how time interval between insecticide applications effects thrips damage to gloxinia flowers, population density of E. occidentalis and flower phytotoxicity.

## Materials and Methods

Studies were conducted from April to June 1989 in a greenhouse at the Pesticide Research Center, Michigan State University. Five-cm tall gloxinia 'Red Velvet' seedlings were planted in pots (6 cm-diam.) filled with a commercial soil mix (Pro plant mix 2, Michigan Peat, Co., Houston, TX). Gloxinia plants were irrigated three times per week and fertilized once every 2 weeks with approximately 0.4 g of Peter<sup>TM</sup> 20-20-20 (W.R. Grace & Co., Fogelsville, PA) applied through the irrigation system. F. occidentalis collected from a commercial greenhouse in Lansing, Michigan, were released in our research greenhouse in April. These thrips colonized unsprayed marigold, african violet and gloxinia plants grown on benches adjacent to the benches used for this study. Thrips adults continuously emerged from these extra plants during the experiment to infest gloxinia flowers in

our test. Three insecticides were selected for this study: cyfluthrin (Tempo 2C, Mobay Chemical Corp., Kansas City, Mo.), a synthetic pyrethroid; abamectin (Avid 0.15 EC, Merck & Co., Inc., Rahway, N.J.), a macrocyclic lactone; and chlorpyrifos (Dursban 20 ME, Dow Chemical Co., Midland, Mich.), an organophosphate. Dilute aqueous sprays were prepared from commercial formulation at concentration recommended for use on ornamental plants. Abamectin was applied at 0.011 g (AI)/liter, cyfluthrin at 0.020 g (AI)/liter, and chlopyrifos at 0.410 q (AI)/liter. Insecticides were applied through a hand-held CO2 sprayer with a single nozzle at 50 psi. Each plant was sprayed with approximately 6 ml of formulation. Each insecticide was applied at 5-, 10- and 15-day intervals during the 35- day test period. The first application was made to all plants except the controls two weeks after initiation of flowering. Gloxinia plants were arranged in a completely randomized design with 5 replications of two-plant units for a total of ten plants per treatment. A two-plant unit consisted of two individual pots, each with a gloxinia plant, pushed together for sampling purposes. This was necessary to obtain enough flowers each week to determine thrips population density. Plants were removed for insecticides application, and returned to their original location when the spray dried.

Four flowers from each two-plant unit were sampled one day before the first insecticide application and once every

seven days thereafter. Thrips were extracted from each flower sample by swirling the flowers in 200 ml of 70% ethanol, and filtering this solution through a cloth screen. Thrips were washed off the filter cloth into a 10 ml dish for counting under a dissecting microscope. Thrips feeding damage and insecticide injury to the flowers were quantified from weekly samples by measuring the area of petal tissue damaged from thrips or necrotic from insecticide injury. The area of petal tissue damage from thrips or insecticide phytotoxicity was mapped onto an outline of a gloxinia flower on a piece of paper with a superimposed 1.0 mm grid. The entire flower area was determined by cutting open the flower and running the flattened petals through a LI-3000 leaf area machine (Lambda Instrument Corp., Lincoln, Neb.).

Thrips population density in flowers were determined for each treatment from the mean number of thrips per flower across all five sample dates. Each replicate was sampled once per week by removing four flowers, for a total of 20 flowers per replicate and 100 flowers per treatment over a five-week period. Flower injury from thrips feeding was totaled over the last three sample dates, and is expressed as the total petal area scraped clean of pigment for 12 flowers per replicate and 60 flowers per treatment. The number of thrips per flower and the amount of thrips injury to flowers was transformed as the natural log of x + 1 before statistical analysis. Insecticide phytotoxicity to flowers was determined from flowers collected each week as described

for thrips extraction. Insecticide phytotoxicity is expressed as the percentage of petal tissue that was discolored or necrotic in a pattern of circular spots or marginal burning. Insecticide phytotoxicity was transformed to the square root of percent damage before analysis. Interactions among insecticide treatment and the length of intervals between insecticide applications was investigated by a two-way analysis of variance (ANOVA) (Wilkinson 1989). Comparison of thrips density, thrips flower injury and insecticide phytotoxicity among the three insecticides tested were made for treatments applied at the same frequency. Abamectin, cyfluthrin and chlorpyrifos means were then compared with Tukey's test (Wilkinson 1989). The influence of time interval between applications on thrips density, thrips flower injury and insecticide phytotoxicity was determined by regression analysis.

## Results

Mean daily temperatures in the greenhouse were 26.7, 22.1, 28.6, 31.3, 29.1, and 30.6°C, respectively for weeks 1-6 of the experiment. One day before insecticide applications were initiated there were no significant differences in the number of thrips per flower among treatments (F=15; df=9, 40; p>0.10). At the end of the 35-day experiment, all insecticide treatments had less thrips per flower than did the control (Table 1). Interactions among insecticide product effects

and application frequency effects were investigated for thrips population density, thrips injury, and insecticide phytotoxicity with a two-way ANOVA (Wilkinson 1989). Insecticide product and application frequency were significant factors in all three analyses (P<0.01 in all cases), and interactions among these two factors were insignificant (p>0.45, P>0.25, and P>0.65, respectively for thrips density, thrips injury, and insecticide phytotoxicity). Abamectin applied at 5-day interval suppressed thrips density in flowers to levels lower than cyfluthrin or chlorpyrifos applied at 5-day interval. differences in thrips density were found attributable to insecticide product applied at 10- and 15-day intervals (Table 1). Similarly, few differences were found in the amount of thrips injury to gloxinia flowers due to insecticide product. At 10-day interval between applications, flowers treated with chlorpyrifos or abamectin had less thrips injury than flowers treated with cyfluthrin. Chlorpyrifos was the most toxic to gloxinia flowers and caused more damage than abamectin or cyfluthrin when insecticides were applied at 10 or 15-day intervals (Table 1). Chlorpyrifos also caused more injury than cyfluthrin when both were applied at 5-day intervals, although at 5-day intervals all three insecticides caused enough injury (>4%) to be considered unacceptable by many growers.

Thrips density in flowers was positively correlated with the length of time between applications for abamectin  $(r^2 = 0.68)$  and cyfluthrin  $(r^2 = 0.39)$  but not for chlorpyrifos  $(r^2 = 0.10)$ . When frequency of application was regressed against thrips density the model for abamectin  $(y = 0.10x + 0.15, r^2 = 0.68, P < 0.001)$  explained more of the variation in thrips density than the model for cyfluthrin  $(y = 0.09x + 0.84, r^2 = 0.39, P < 0.02)$ . However, slopes for these models were very similar (Table 2, Fig. 1). thrips injury to flowers also increased with an increase in the time interval between applications. For thrips injury the strongest correlation to application frequency was for cyfluthrin model ( $r^2 = 0.52$ , P < 0.01). Thrips injury to flowers was not explained as well by application frequency in models with abamectin ( $r^2=0.28$ , P < 0.05) and chlorpyrifos  $(r^2 = 0.17, P < 0.24, Table 2)$ . Slopes of regression models for abamectin and cyfluthrin were similar (0.21 and 0.25, respectively) (Fig. 2). The amount of flower injury caused by insecticides was highly correlated with the frequency of applications for abamectin, cyfluthrin and chlorpyrifos  $(r^2 = 0.83, 0.79, and 0.86, respectively)$  (Table 2). When the interval between applications was regressed against thrips density, the slopes of the regression models were similar for abamectin (-0.24), cyfluthrin (-0.20), and chlorpyrifos (-0.28) (Fig. 3).

## Discussions

Approximately half of the research greenhouse section used for this test was filled with gloxinia test plants. The remaining space held marigold and other plants susceptible to F. occidentalis. During the 35-day experiment, the mean number F. occidentalis extracted from four unsprayed gloxinia flowers climbed from 14.0 to 271.0. In a greenhouse where all plants are sprayed, these treatments would have had a greater effect on the population of F. occidentalis.

Therefore, if all plants in a greenhouse are sprayed the same way as the plants in any given treatment of this study, the thrips population reduction should be at least as large as we observed.

If flower protection was the only concern, applications once every 10 days would be adequate for most growers.

However, because F. occidentalis is a vector of TSWV, gloxinia growers must reduce population of thrips to extremely low level if there is a source of virus in the greenhouse. In transmission tests Allen & Broadbent (1986) found that 20-30% of Lycopersicum esculentum test plants became infected with TSWV when they were caged with 2-6 thrips per plant, and 80% of Gomphrena globosa became infected with the virus when they were caged with 4 thrips per plant. Because it is not known what population density

of E. occidentalis is required to spread tomato spotted wilt virus, and epidemic have been observed in greenhouse with low populations, it is desirable to suppress E. occidentalis population to a low level when susceptible plants are being grown. Parella & Robb (1987) have tested many different insecticides for thrips control using 5-day interval between applications and provide some guidelines for determining which insecticide have the best activity against E. occidentalis. However, little information is available on how the frequency of insecticide applications affect E. occidentalis population or the amount of flower injury they cause. We found a linear relationship between application interval and thrips density in flowers. When the time of interval between applications doubled, thrips density also doubled for abamectin and cyfluthrin (Fig. 1).

This study indicates that control of F. occidentalis population can be improved by shortening the length of time between insecticide applications. It also demonstrates that the time interval between insecticide applications necessary to minimize flower damage could be as long as 15 days, depending on the insecticide used. At the same time, we found that the amount of flower injury increased directly with an increase in spray frequency. Although gloxinia growers can obtain better control of F. occidentalis by increasing the frequency of insecticide applications, thrips populations must be under good control before flowering is

initiated, because flower injury may become unacceptable after several applications at 5-day intervals.

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Table 1. Protection of gloxinia flowers with abamectin, cyfluthrin, or chlorpyrifos applied at 5-, 10-, or 15-day intervals.

Treatment		Number of	F. occidentalis	Insecticide phytotoxicity to flowers+SD	
	D	F. occidentalis	injury to		
		per flower+SD	flowers+SD		
			(mm <sup>2</sup> )	mm <sup>2</sup>	
Abamectin	5	0.68 ± 0.13a	0.73 ± 1.02a	5.14 ± 0.37ab	
Cyfluthrin	10	$1.31 \pm 0.50b$	1.65 ± 1.56a	4.84 ± 0.39a	
Chlorpyrifos	15	$1.53 \pm 0.32b$	1.45 <u>+</u> 1.35a	$5.97 \pm 0.28b$	
Abamectin	5	1.10 <u>+</u> 0.45a	1.28 ± 1.75a	4.54 ± 0.35a	
Cyfluthrin	10	1.72 <u>+</u> 0.60a	$3.68 \pm 0.64b$	$4.52 \pm 0.55a$	
Chlorpyrifos	15	1.49 <u>+</u> 0.39a	$0.44 \pm 0.98a$	$5.52 \pm 0.11b$	
Abamectin	5	1.67 <u>+</u> 0.27a	2.87 ± 1.77a	$0.00 \pm 0.00a$	
Cyfluthrin	10	2.22 ± 0.48a	$4.20 \pm 0.73a$	$0.00 \pm 0.00a$	
Chlorpyrifos	15	1.88 <u>+</u> 0.60a	3.19 <u>+</u> 1.80a	$3.20 \pm 0.41b$	
Control	-	3.00 ± 0.51	$5.19 \pm 0.08$	$0.00 \pm 0.00$	

D=interval between insecticide applications (day). Insecticide treatments are compared with other treatments applied at the same time intervals. Data are expressed as the  $\ln x + 1$ .

Treatments applied at the same application interval are not different from each other if followed by the same letter (Tukey's test, P=0.05; Wilkinson [1989]).

Table 2. Relationship of the time interval between insecticide applications to population of F. occidentalis, thrips damage to flowers, and phytotoxicity to flowers (n=5).

Modela	Dependent variable	Insecticide	ANOVA F	ANOVA P	Model
Y=0.10x+0.15	Thrips per flower <sup>b</sup>	Abamectin	27.2	<0.001	0.68
Y=0.09x+0.84	Thrips per flower	Cyfluthrin	8.3	<0.2	0.39
Y=0.04x+1.30	Thrips per	Chlorpyrifos	1.5	>0.24	0.10
Y=0.21x-0.52	flower Thrips damage <sup>C</sup>	Abamectin	5.0	<0.05	0.28
Y=0.25x+0.61	Thrips damage	Cyfluthrin	13.8	<0.01	0.52
Y=0.13x-0.03	Thrips damage	Chlorpyrifos	2.7	>0.10	0.17
Y=-0.28x+4.07	Phytotoxicityd	Abamectin	65.3	<0.001	0.83
Y=-0.20x+3.23	Phytotoxicity	Cyfluthrin	48.5	<0.001	0.79
Y=-0.24x+4.63	Phytotoxicity	Chlorpyrifos	81.0	<0.001	0.86

ax= Interval between applications

b = The number of F. occidentalis per gloxinia flower was determined for each replicate by averaging the weekly samples during entire experiment. Data were transformed ( $\ln x + 1$ ) before analysis.

c = The area of gloxinia flowers  $(mm^2)$  damaged by thrips feeding was combined for all sample dates and transformed (ln x + 1) before analysis.

d = The area of gloxinia flowers damaged by insecticides was converted to a percent of the total petal tissue. Percent damaged was square-root transformed before analysis.

Figure 1. Influence of the interval between insecticide applications on thrips infestation of gloxinia flowers. Regression statistics are in Table 2.

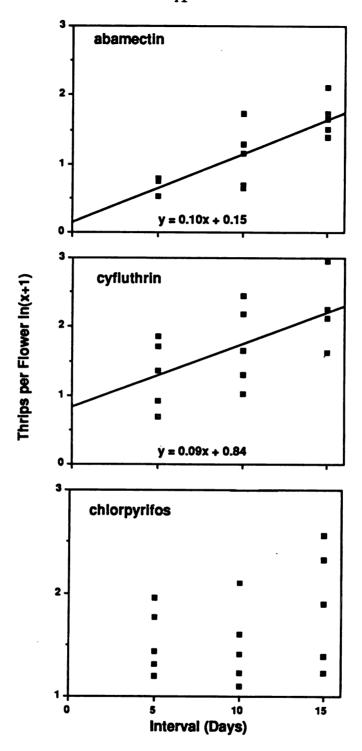


FIGURE 1

Figure 2. Thrips feeding injury to gloxinia flowers as a function of the time interval between insecticide applications. Regression statistics are in Table 2.

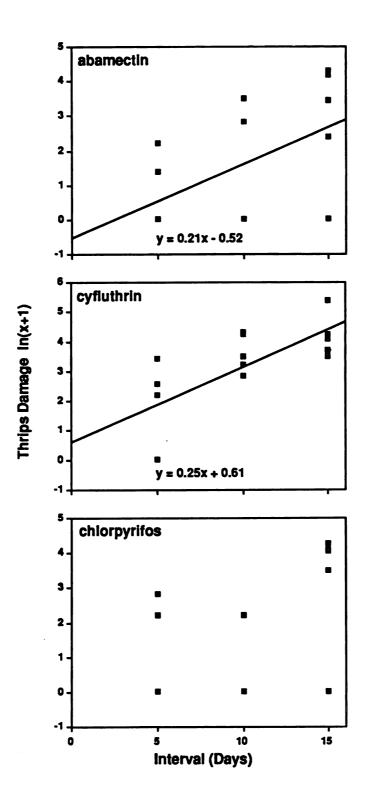


FIGURE 2

Figure 3. Relationship of interval between insecticide applications to the amount of insecticide injury to gloxinia flowers. Regression statistics are in Table 2.

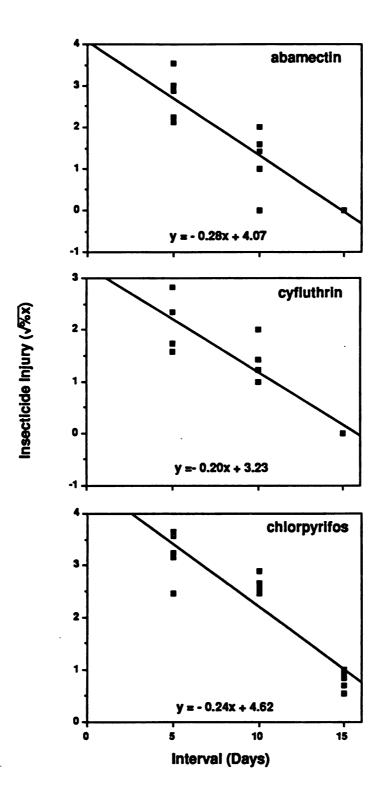


FIGURE 3

# CHAPTER III

INDICATOR PLANTS FOR DETECTION OF

TOMATO SPOTTED WILT VIRUS IN THE GREENHOUSE

## **ABSTRACT**

Enzyme-linked immunosorbent assay (ELISA) tests were performed on seven species of plants that had been freely exposed to Frankliniella occidentalis (Pergande) with tomato spotted wilt virus (TSWV)-infected plants in the same greenhouse for one year. Both strains of TSWV Lettuce (L) and Impatiens (I) were present in the greenhouse when this study was conducted. ELISA results showed that onion and dahlia were not infected; gomphrena and marigold were infected by both strains; and begonia, cyclamen, red impatiens, and white impatiens were infected by strain I alone.

Six species of plants were used in a thrips transmission of TSWV test. In caged and uncaged treatments, cucumber, tomato and lettuce did not react positively to ELISA test. Caged tobacco plants were infected by both strains of TSWV; however, uncaged plants were not infected by either strain of TSWV. Both strains were found in caged gomphrena plants but only strain I was found in uncaged plants. Symptoms on gloxinia seedlings were expressed two weeks after the experiment was initiated while other species of plants expressed symptoms one week later.

To determine the number of thrips required for a successful transmission of TSWV on gloxinia seedlings,

different numbers of F. occidentalis: 0, 2, 4, 6 or 8 per plant were tested. Two thrips per plant were sufficient to initiate infection, however, at the end of the 4-week experiment the intensity of TSWV infections with four or more thrips per plant were significantly higher than with two thrips per plant. Neither of TSWV strains was detected from control plants.

Preference of <u>F</u>. <u>occidentalis</u> to white, yellow and bluesticky cards with two different levels of heights, 25 cm and 50 cm above the greenhouse bench were tested. The highest count was obtained from yellow traps at 50 cm above the bench.

Yellow cards (not sticky) were added to gloxinia seedlings placed 50 cm above the bench in order to increase the attractiveness of the seedlings to F. occidentalis. The number of thrips collected from plants with yellow cards was significantly higher than from the plants without yellow cards. However, the intensity of TSWV infection were not significantly different among the treatments.

## Introduction

Tomato spotted wilt virus (TSWV) has a broad host range of 236 species in 34 families, including vegetable and ornamental plants grown in subtropical and temperate climates (Matteoni et al. 1988, Cho et al. 1989). However, TSWV has not caused economic losses on all host species. In many cases the virus causes lesions at sites of initial infections without causing systemic symptoms but they are still infected and may serve as virus reservoirs (Parella & Robb 1987, Matteoni et al. 1988).

Six species of thrips: Frankliniella occidentalis

(Pergande), F. fusca (Hinds), F. schultzei (Trybom),

Scirtothrips dorsalis Hood, Thrips setosus Moulton, and T.

tabaci Lindeman are capable of transmitting TSWV (Best 1968,

Ie 1970, Paliwal 1976, Cho et al. 1989). F. occidentalis is

now a dominant pest of floricultural crops in the United

States and Canada (Allen & Broadbent 1986, Parella & Robb

1987, Robb 1990). Widespread infestation of F. occidentalis

has resulted in an increased incidence of TSWV in susceptible

flowers and vegetables (Allen & Broadbent 1986, Broadbent et

al. 1987, Allen & Matteoni 1988, Cho et al. 1989).

Epidemics of TSWV have caused losses in greenhouse production of tomato, dahlia, gloxinia, begonia, chrysanthemum, marigold, cineraria, impatiens, calceolaria,

anemone and exacum (Allen & Broadbent 1986, Allen and Matteoni 1988, Baker & Jones 1988, Matteoni et al. 1988).

Greenhouse growers rely heavily on insecticides to control F. occidentalis and to prevent the spread of TSWV. However, insecticides registered for use to control thrips in greenhouse are only moderately effective. In many cases insecticide sprays are sufficient to suppress direct losses caused by F. occidentalis but insufficient to prevent the spread of TSWV (Francki & Hatta 1981, Cho et al. 1989).

Because many plants can serve as TSWV reservoirs without expressing symptoms, growers may not recognize the presence of the virus until a large proportion of plants become infected (Matteoni et al 1988). This is a potential problem for bedding plant growers because their major crops do not always express obvious symptoms when infected. Bedding plant growers need a method of detecting TSWV in their greenhouses. Leaf samples can be collected routinely and sent to a laboratory for serological identification but the large number of plant species and cultivars grown in bedding plant greenhouses makes this difficult. Another method used to monitor the presence of TSWV in greenhouse is the use of indicator plants (Bos 1978, Matteoni et al. 1988). A sensitive indicator plant that develops symptoms rapidly would be helpful to bedding plant growers for early detection of TSWV.

The purpose of this study was to determine:

1) which strains of TSWV (Lettuce [L] or Impatiens [I]) is found in bedding plant species 2) the sensitivity of seven greenhouse crops which had been reported susceptible to TSWV 3) the potential of the most sensitive plants for use as indicator plants for TSWV in greenhouse and 4) if colored cards placed in pots with indicator plants helped to attract E. occidentalis.

## Materials and Methods

Studies were conducted from March 1990 to February 1992 in a greenhouse at the Pesticide Research Center, Michigan State University.

Thrips colony and virus isolate. A colony of F.

occidentalis was established in the research greenhouse from individuals collected from a commercial greenhouse in Lansing, Michigan. The thrips were allowed to develop and disperse among plants, gloxinia, marigold, african violet, grown on the greenhouse benches. This colony was used in host sensitivity test, thrips transmission, color preference and indicator plant tests.

The original TSWV isolate was from an infected impatiens and gloxinia plants from a commercial greenhouse in Wayne County, Michigan. Thrips were allowed to feed on these plants and to infect additional impatiens and other plants.

This TSWV isolate was used in the thrips transmission and indicator plant tests.

## 1. Host Sensitivity

Plant species and plant maintenance. Six species of plant that had been reported seriously damaged by TSWV (cucumber, Cucumis sativus L.; tobacco, Nicotiana tabacum L.; "Dwarf Buddy" gomphrena, Gomphrena globosa L.; lettuce, Lactuca sativa cv. longifolia L.; "Red Velvet" gloxinia, Sinningia speciosa Benth & Hood; and "Glomour" tomato, Lycopersicum esculentum Mill.) were used in thrips inoculation test. All species of plant but gloxinia were grown from seeds which came from Stokes Seeds, Inc. (Buffalo, NY). Four leaf-stage of gloxinia seedlings were obtained from Earl Jay Small, Inc. (Florida).

Plant seedlings were grown individually in 6-cm diam. clay pots with a greenhouse soil mix (Pro plant mix, Michigan Peat Co., Houston, TX). Plants were irrigated three times a week. Approximately 0.4 g of Peters<sup>TM</sup> 20-20-20 fertilizer was used once in two weeks and applied through an irrigation system. Plants were kept in other part of the experimental greenhouse which was free of thrips and TSWV before the initiation of the experiment.

Thrips inoculation test. Three treatments were tested in this experiment. For the first treatment, F. occidentalis were used to inoculate the test plants. Adult thrips were collected from TSWV-infected gloxinia flowers by tapping the flowers on a piece of white paper. Three thrips were

transferred to each gloxinia seedling confined to a cage made from a 2-liter polyethylene bottle with four cloth-covered openings on its sides and one on the top, for a 3-day inoculation access feeding. At the end of the inoculation feeding, plants were sprayed with abamectin (Avid 0.15 EC, Merck & Co., Inc., Rahway, N.J.) at a concentration of 0.011 g (AI)/l. For the second treatment, the test plants were introduced into the greenhouse without cages to expose the plants to thrips colony and TSWV isolate that were present in the greenhouse. A final treatment consisted of plants caged without thrips (control). The experiment was arranged in a completely randomized design with five replications. Plants were observed once a week for four weeks for TSWV symptoms development. The symptoms of TSWV were recognized according to Ie (1970), Cho et al. (1987), Allen & Matteoni (1988), and Matteoni et al. (1988), (Table 1). At the end of the experiment, plants were sampled for ELISA analysis. Enzyme-linked immunosorbent assay (ELISA). ELISA tests were used to detect the strains of TSWV present in seven species of plant, begonia, Begonia hiemalis Fostch; cyclamen,

were used to detect the strains of TSWV present in seven species of plant, begonia, Begonia hiemalis Fostch; cyclamen, Cyclamen persicum Mill.; red and white impatiens (Impatiens wallerana Hook. f.), onion, Allium cepa L.; dahlia, Dahlia pinnata x coccinea Cav.; "Dwarf Buddy" gomphrena, Gomphrena globosa L.; and marigold, Tagetes erectus that were exposed to F. occidentalis and TSWV for one year in the greenhouse. The double antibody sandwich ELISA (Clark & Adams 1977) was

used to confirm TSWV infections and to determine the strains (Lettuce [L] or Impatiens [I]) of TSWV infecting the plants.

The ELISA test was also performed to determine the strain of TSWV infecting the plants in the thrips inoculation test. This serological test was run after the symptom observations were completed. ELISA kit used in this test was obtained from Agdia, Inc. (Mishawaka, IN). One half-gram of leaf sample taken from each test plant was placed in a plastic bag containing 5 ml ELISA extraction buffer, and triturated with a blunt end of a pen. The extraction buffer used was phosphate buffered saline containing 0.05 % Tween (PBS-Tween), which was added with 2 % Tween-20 (v/v), 2 % egg albumin (w/v), and 2 % polyvinylpyrrolidone (PVP) (w/v).

In this experiment two microtitre plates were used, one precoated with strain I and the other precoated with strain L antibody. One hundred microliters of sample solution was added into each microwell of the microtitre plate. The plates were then incubated at room temperature for 2 hours before they were washed 5 times with PBS-Tween. One hundred microliters of enzyme-conjugated antibody for atrain I or L was added into each assay well in the respective plate and incubated for 2 hours in room temperature. The plates were then washed 5 times with PBS-Tween and 100 microliters substrate solution (o-phenyleneDiamine) was added into each assay well. The plates were incubated for 20 minutes at room temperature for color development, attributable to positive reaction. The reaction between the enzyme-conjugated

antibody and its substrate solution was stopped with 2.5 M  $H_2SO_4$ . ELISA reactions were measured spectrophotometrically at  $490_{nm}$  with a "Series 700" microplate reader (Cambridge Technology, Inc.). Samples were considered TSWV-positive if  $490_{nm}$  reading was greater than twice the average of buffer readings.

# 2. The Potential of Gloxinia Seedlings As Indicator Plants

Thrips transmission test. From the thrips inoculation test, gloxinia was the most susceptible to TSWV among the test plants. Therefore, the plant was chosen to determine its potential to be an indicator plant for TSWV in the greenhouse. TSWV can only be transmitted by thrips that acquire the virus as nymphs (Sakimura 1962). To evaluate the number of F. occidentalis necessary to inoculate a gloxinia seedling with TSWV, a transmission test was conducted using adult thrips that had fed on infected gloxinia leaves as nymphs. An infected leaf was detached and placed on a piece of moist filter paper in a 15-cm diam. petri dish. Thirty nymphs were placed onto each leaf with a moistened brush and allowed to feed on the leaf for three days. The nymphs were then transferred back to gloxinia plants to complete their life cycle.

Gloxinia 'Red Velvet' seedlings were individually planted in a 6-cm pot at the four leaf-stage and confined to a cage made from 2-liter polyethylene bottle with four opening on its sides and one on the top, covered by fine

cloth. The cages were arranged in a completely randomized design with five replications. Different numbers of viruliferous F. occidentalis were placed into the cages (0, 2, 4, 6 or 8) for a 3-day inoculation feeding. After the inoculation feeding, test plants were sprayed with abamectin (Avid 0.15 EC, Merck & Co., Rahway, N.J.) at a concentration of 0.011 g (AI)/liter.

Test plants were observed weekly for four weeks. Tomato spotted wilt virus symptoms on gloxinia were characterized by line pattern, ringspot and chlorotic markings on the leaves (Cho et al. 1987, Matteoni et al. 1988). The severity of TSWV infection was estimated by the percentage of leaf area showing symptoms. A sample of two lowest leaves from each plant were observed for TSWV infection. Area of the sample leaves was measured by running the leaves individually through a LI-3000 leaf area machine (Lambda Instrument Corp., Lincoln, Neb.). All parts of the leaves showing TSWV symptoms were then trimmed with a razor blade. The trimmed parts were also run through the leaf area machine. Both figures, the area of the leaves and the area of the trimmed parts, were used to determine the percentage of leaf area showing TSWV symptoms.

Color preference test. Three colors reported to be the most attractive to F. occidentalis (white, yellow and blue) were tested (Yudin et al. 1987, Parella et al. 1989). Test cards were 7 x 12.5 cm long. Yellow and blue cards were made from white illustration board coated with spray enamel paint

number 1214 and 1208, respectively (The Testor Corp., III).

White cards were left unsprayed. Degree of greenness,
yellowness, and blueness of the colors are presented in
Table 2. Newly painted cards were allowed to dry for 24
hours and then coated with Tangle Foot (The Tangle Foot Co.,
Michigan). Two cards were mounted back to back on a wood
stake using staple gun and placed 25 cm or 50 cm above the
greenhouse bench. Cards were arranged in a randomized
complete block design with 8 replications. There were spaced
50 cm apart in row and 85 cm between rows. A second test was
conducted three weeks after the first test was finished. In
the second test, cards were coated with polyisobutilene (Tack
Trap, Animal Repellent, Griffin, GA). The Tack Trap was
heated to facilitate coating of cards.

Cards were collected 10 days after the experiment was started and the number of F. occidentalis was counted with a dissecting microscope. Three and five thrips were randomly removed from each treatment combination in test 1 and test 2, respectively, and placed into glass vials containing 70% ethanol for identification.

The addition of yellow cards to gloxinia indicator plants. Gloxinia seedlings were individually grown in 6-cm clay pots. One yellow card was slit down the center, and second, identical card, was mounted perpendicular to the first cards by sliding into the slit. The cards were made from white illustration board and painted as before. A stake with cards was placed in the pot so the yellow cards were

positioned just above the plant leaves. The plants were elevated 50 cm above the greenhouse bench by placing the pot on top of another pot. Another treatment was the gloxinia seedlings elevated 50 cm above the greenhouse bench without cards.

The experiment was arranged in a completely randomized design with six replications. Plants (six at each time for each treatment) were removed at 1, 2, 3, and 4 weeks after the initiation of the experiment for counting thrips and observing TSWV symptoms. The plants were covered with sealed plastic bags before being removed to prevent E. occidentalis from escaping. Thrips were extracted from the plants by swirling 70% ethanol and filtering through a fine cloth. The number of extracted thrips was determined by a dissecting microscope. The area of leaf expressing TSWV symptoms was determine with a LI-3000 leaf area machine (Lambda Instrument Corp., Lincoln, Neb.) as described before.

Data analysis. All percent data were transformed to the arcsine square root (x) before further statistical analysis. Means of TSWV infection for different number of thrips per plant were compared with Tukey's test (Wilkinson 1989). Interaction among color and height treatment was analyzed by a two-way ANOVA (Wilkinson 1989) and average number of F. occidentalis caught with each treatment were compared with Tukey's test (Wilkinson 1989). For the addition of yellow cards to gloxinia indicator plant tests, means were separated by ANOVA test (Wilkinson 1989).

#### Results and Discussions

## 1. Host Sensitivity and ELISA Tests

In this experiment TSWV infections were confirmed by ELISA. When the test plant were freely exposed to F. occidentalis and TSWV for one year, onion and dahlia were not infected. Begonia and marigold were infected, however, they did not show symptoms (Table 3 and 4). Gomphrena and marigold were infected by both strains (Lettuce [L] and Impatiens [I]) while the other plants, begonia, cyclamen, red and white impatiens were infected by strain I alone (Table 4). Onion seemed to be resistant to both strains of TSWV. Dahlia has been reported as a host of TSWV but in our experiment it did not show positive reaction to TSWV in ELISA test. It is possible that dahlia was less attractive to F. occidentalis than other test plants.

In the thrips inoculation test 20% of tobacco seedlings caged with viruliferous F. occidentalis expressed TSWV symptoms three weeks after the experiment was initiated and reacted positively to both strains of TSWV. However, there were no symptoms and negative ELISA results against both strains on uncaged seedlings (Table 5 and 6). This suggests that tobacco seedlings were less attractive to F. occidentalis, but still susceptible to both strains of the virus. Gomphrena placed in cages expressed necrotic lesions

symptoms three weeks after the thrips were introduced. of 20 % and 100% of the plants reacted positively to ELISA tests for strains L and I, respectively. All uncaged seedlings were infected by strain I alone. This suggests that strain I of TSWV was dominant in the greenhouse. tomato seedlings showed symptoms or gave positive reactions to ELISA tests (Table 5 and 6). Allen & Broadbent (1986) reported that TSWV transmission level with gomphrena was remarkably higher relative to those with tomato. However, in our experiment tomato seedlings indicated resistance to both strains of the virus. It seemed that three thrips per seedling were insufficient to initiate and develop infection on tomato seedling. In addition the tomato seedlings might have resistant genes to strains L and I. Sources of resistance had been found in Lycopersicum sp. and some attempts to introduce this resistance into commercial tomato cultivars had been reported (Best 1968).

Because the first TSWV symptoms were detected on 40 % of the gloxinia seedlings two weeks after the experiment was initiated (one week earlier than other test plants)

(Table 5), gloxinia seedlings were further tested for their potential as indicator plants for TSWV in greenhouse.

# 2. The Potential of Gloxinia Seedlings As Indicator Plants

Thrips transmission of TSWV. The symptoms of TSWV on gloxinia vary with the plant age and the stage of infection development. Young plants may die within six weeks of

transplanting. At this stage, TSWV is often confused with root rot disease caused by a fungal pathogen. Older plants are more tolerant to infections and virus symptoms appear as foliar markings such as line patterns and ringspots. This may look like damage caused by cold water or by pesticide deposits. Chlorotic markings become necrotics with age (Matteoni et al. 1988). In our experiment, chlorotic markings and ringspots were found 2 and 3, and 4 weeks, respectively, after the experiment was started (Table 5).

Development of TSWV symptoms on gloxinia seedlings depended on the number of thrips test plants were caged with. Two weeks after the experiment was started 20% and 100% of plants applied with 2-thrips treatment and other treatments, respectively, showed symptoms. However, the percentage mean of gloxinia leaf area covered by TSWV symptoms for all treatments were not significantly different. Three weeks after the initiation of the experiment 100% plants caged with viruliferous thrips showed symptoms and none of the control plants had any symptoms. The percentage mean of gloxinia leaf area with symptoms for plants caged with 2 thrips per plant was significantly lower than that plants caged with 4, 6, or 8 thrips per plant (Table 7). indicats that higher number of infected plants and symptoms development rate occur with 4 thrips or more per plant as opposed to two thrips per plant. It suggests that the amount of TSWV may be important to infection and development of symptoms.

In test with Emilia sonchifolia L. 24 - 32 % of the plants became infected after being caged with 4 thrips per plant (Sakimura 1962). Tomato plants caged with 2-6 thrips and gomphrena plant caged with 4 thrips per plant, respectively, developed 20-30% and 80% infection (Allen & Broadbent 1986). Apparently, gloxinia seedlings are more susceptible to TSWV than those plants.

Color preference test of <u>F</u>. <u>occidentalis</u>. All thrips collected from sticky cards were identified as <u>F</u>. <u>occidentalis</u>. Morphological characteristics used in the thrips identification were those described in Chapter I under species <u>occidentalis</u>.

The results of three color attractiveness experiments support the finding of Robb and Parella (1989) that yellow cards and blue cards were highly attractive to F. occidentalis. Our results also suggested that attractiveness of sticky cards was influenced by the height of the cards from the bench (F=16.1, P<0.001). The greatest number of thrips were found on yellow cards placed 50 cm above the greenhouse bench. However, In the experiment 1, it was not significantly different from the number of thrips caught on blue traps placed at the 25 cm height. The number of thrips caught by yellow traps at the level 25 cm above the bench was not significantly different from those caught with white traps at the same level (Table 8). One possible reason why little or no significant difference was obtained may have been the low number of thrips trapped in this experiment.

The addition of yellow cards to gloxinia indicator plant. One and two weeks after the initiation of the experiment there were no differences in the number of thrips collected from gloxinias with and without yellow cards. However, three and four weeks after the experiment was started the number of thrips collected from gloxinia seedlings with yellow cards was higher than from gloxinias without yellow cards (Table 9). The average number of thrips collected during this experiment was relatively low for both treatments compared to the number of thrips caught by yellow sticky cards (previous experiment).

Four weeks after the experiment was begun 10.61% of leaf area on indicator plant with cards showed TSWV symptoms compared to 5.30% on plants without yellow cards. However, these means were not significantly different by ANOVA (P>0.05, Table 10).

#### Conclusion

Gloxinia seedlings showed TSWV symptoms more rapidly than other test plants. This suggests that gloxinia seedlings are the most susceptible host to TSWV among test plants. However, the symptoms vary from line patterns to ringspots. Gloxinia seedlings may be useful for early warning of TSWV infection but the plant should be sampled for ELISA analysis to obtain a conclusive result.

As low as two viruliferous <u>F. occidentalis</u> caged with gloxinia seedling resulted in transmission of TSWV and symptom development. The number thrips caged with seedling influence the severity of TSWV symptoms.

Use of gloxinia seedlings as indicator plants to monitor the presence of TSWV in greenhouse appeared promising. Yellow cards increased attractiveness of gloxinia seedlings to F. occidentalis but gloxinias without yellow cards developed symptoms at about the same time and the same rate of infection as the gloxinias with yellow cards. Therefore, it is not clear from our results if adding yellow cards to gloxinia seedlings will enhance their use as indicator plants. Our results agree with previous studies that yellow cards are more attractive to F. occidentalis than blue or white cards.

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Table 1. Symptoms of TSWV on potential indicator plants.

Host	Symptoms
Cucumber <sup>1</sup>	Necrotic local lesions and systemic symptoms, chlorotic spots with necrotic centers on cotyledons.
Tomato <sup>3</sup>	Stunted, necrotic patches, sunken necrotis lesions along the stems and petioles.
Tobacco <sup>1</sup>	Necrotic local and systemic symptoms and leaf deformation.
Gomphrena <sup>1</sup> , <sup>3</sup>	Chlorrotic, yellowing and ringspots.
Lettuce <sup>4</sup>	Necrotic spots, one side-twisted plant, chlorotic and malformed leaves.
Gloxinia <sup>3</sup>	Wilting, foliar markings: line patterns and ringsports, chlorotic markings, and necrotic.
Cyclamen <sup>3</sup>	Stunted, irregular flower, necrotic patches, ringspots, and vein necrosis.
Impatiens <sup>3</sup>	Ringspots on young plant, premature senescence and yellowing.
Begonia <sup>3</sup>	Asteroid chlorotic lesions on the leaves, vein necrosis, and apical necrosis.
Dahlia <sup>5</sup>	Pale green spotting, concentric light green spots.

<sup>&</sup>lt;sup>1</sup>Ie (1970)

 $<sup>^{2}</sup>$ Cho et al. (1987)

<sup>&</sup>lt;sup>3</sup>Matteoni et al. (1988)

<sup>&</sup>lt;sup>4</sup>Allen & Matteoni (1987)

<sup>&</sup>lt;sup>5</sup>Drayson (1958)

Table 2. Color spectrum measurements with reference to the degree of greenness, yellowness, and blueness from color cards used in the color preference test.

Color	Degree of greenness <sup>a</sup>	Degree of Yellowness <sup>b</sup>	Degree of Blueness <sup>C</sup>
White	2,015	2,212	362
Yellow	3,610	3,610	3,620
Blue	1,469	1,265	2,665

abc Measurements were conducted in day light at the greenhouse with a Minolta Chroma Meter CS-100.

Table 3. Host reactions to TSWV after the plants were freely exposed to F. occidentalis and TSWV for one year.

Host species	Symptoms
Gomphrena	leaf necrosis and ringspots
Cyclamen	vein necrosis and ringspots
Red Impatiens	leaf necrosis and ringspots
White Impatines	necrosis and ringspots
Onion	none
Dahlia	none
Marigold	none
Begonia	none

Table 4. Transmission of TSWV "Lettuce" or "Impatiens" strain to test plants, freely exposed to a culture of  $\underline{F}$ . occidentalis and TSWV for one year. Data are the proportion of test plants that became infected (n=3).

	Serological reactions				
Test plant	Strain L (%)	Strain I (%)			
Marigold	66.66	100.00			
Gomphrena	33.33	33.33			
Red Impatiens	0.00	100.00			
Begonia	0.00	100.00			
White Impatiens	0.00	66.66			
Cyclamen	0.00	66.66			
Dahlia	0.00	0.00			
Onion	0.00	0.00			

Table 5. Host reactions to TSWV at 1, 2, 3, or 4 weeks after the plants were caged with viruliferous

F. occidentalis.

		Symptom						
Host	Week 1	Week 2	Week 3	Week 4				
Gloxinia	_	Chlorotic markings <sup>1</sup>	Chlorotic markings	ringspots <sup>3</sup>				
Tobacco	-	-	necrotic lesions <sup>2</sup>	necrotic lesions <sup>3</sup>				
Gomphrena	-	-	necrotic lesions <sup>2</sup>	necrotic lesions <sup>3</sup>				
Lettuce	-	· -	-	-				
Tomato	-	-	-	-				
Cucumber	-	-	-	-				

<sup>140%</sup> of the test plants expressed symptoms

<sup>220%</sup> of the test plants expressed symptoms

<sup>3100%</sup> of the test plants expressed symptoms

Table 6. Transmission of TSWV "Lettuce" or "Impatiens" strain after the plants were caged with viruliferous F. occidentalis. Data were the proportion of test plants that became infected (n=5). None of the control plants became infected.

	Serological reactions			
Test plant	ca	Seedlings+thrips caged		s without
	L(%)	I(%)	L(%)	I(%)
Tobacco	20	20		-
Gomphrena	40	100	-	100
Cucumber	-	-	-	-
Tomato	<b>-</b> ·	-	_	-
Lettuce	-	-	-	-

Table 7. Symptom of TSWV on gloxinia leaves at 2, 3, and 4 weeks after exposure to viruliferous
F. occidentalis (n=5). Data were transformed to arcsine √x before statistical analysis.

	Week 2		Week 3		Week 4	
No. of	Arcsine	¥	Arcsine	ક	Arcsine	8
thrips	Vx ± SD		√x ± SD		√x ± SD	
0	0.00 <u>+</u> 0.00a	0.00	0.00 <u>+</u> 0.00a	0.00	0.00 <u>+</u> 0.00a	0.00
2	1.82 <u>+</u> 4.07ab <sup>1</sup>	0.50	7.65 <u>+</u> 1.07b	1.80	10.76 <u>+</u> 2.26b	3.60
4	6.27 <u>+</u> 4.13a <sup>2</sup>	2.50	11.53 <u>+</u> 0.02c	4.00	12.52 <u>+</u> 2.19b	8.00
6	6.07 <u>+</u> 2.70b <sup>2</sup>	4.00	11.82 <u>+</u> 0.62c	4.20	15.02 <u>+</u> 3.67bc	7.00
8	5.06 <u>+</u> 2.25ab <sup>2</sup>	4,.50	10.46 <u>+</u> 3.64c	3.60	17.77 <u>+</u> 2.04c	7.24

 $<sup>^{1}20</sup>$ % of the test plants showed TSWV symptoms  $^{2}100$ % of the test plants showed TSWV symptoms Week 3 and 4, 100 % of the test plants showed TSWV symptoms Means within column with the same letter are not significantly different by Tukey's HSD test (P=0.05, Wilkinson 1989).

Table 8. Mean number of <u>F</u>. <u>occidentalis</u> per card found on white, yellow, or blue sticky cards, placed 25 or 50 cm above the greenhouse bench (n=8).

Color	Height	x <u>+</u> SD	<del>x</del> ±sd
White	25	2.00 <u>+</u> 1.85a	5.38 <u>±</u> 1.33a
White	50	0.50 <u>+</u> 0.75a	3.25 <u>+</u> 1.10a
Yellow	25	0.75 <u>+</u> 1.16a	22.75 <u>+</u> 1.55b
Yellow	50	20.38 <u>+</u> 3.33c	35.25 <u>+</u> 2.25d
Blue	25	19.50 <u>+</u> 2.27c	29.75 <u>+</u> 6.13c
Blue	50	13.62 <u>+</u> 2.56b	25.87 <u>+</u> 5.91c

In column, means with the same letters are not significantly different at 0.05 level according to Tukey's HSD test (Wilkinson 1989).

Table 9. Mean number of  $\underline{F}$ . occidentalis collected from gloxinia plants with or without yellow cards at 1, 2, 3, or 4 weeks after the initiation of the experiment (n=6).

	Numbe	Number of F. occidentalis per plant					
Treatment	Week 1 ₹±SD	Week 2 <del>≅±</del> SD	Week 3 <del>x</del> ±SD	Week 4 ₹±SD			
Gloxinia with card	1.83 <u>+</u> 0.73a	1.80 <u>+</u> 1.14a	2.16 <u>+</u> 0.74b	3.25 <u>±</u> 0.85b			
Gloxinia without card	1.33 <u>+</u> 1.02a	2.00 <u>+</u> 0.03a	1.33 <u>+</u> 0.53a	2.33 <u>+</u> 1.04a			

Means within columns with the same letters are not significantly different by ANOVA test (P=0.05; Wilkinson 1989).

Table 10. Tomato spotted wilt virus symptom on gloxinia with or without yellow cards. Data were transformed to arcsine  $\sqrt{x}$  before further statistical analysis (n=6).

	Gloxinia	leaf	covered	bу	TSWV	symp	toms
	W	leek 3		-	7	Week	4
Treatment	Arcsine √x+SD		8		Arsine VX+SD		<b>%</b>
Gloxinia with card	8.77+10.37	7a	2.32	19.	01+7.9	7a	10.61
Gloxinia without card	10.48+5.79	a	3.31	13.	10+4.1	0a	5.30

There was no differences between means in column by ANOVA test (P=0.05, Wilkinson 1989).

#### APPENDIX 1

### Record of Deposition of Voucher Specimens\*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 1992-01
Title of thesis or dissertation (or other research projects):
Management of the western flower thrips, Frankliniella
occidentalis (Pergande) (Thysanoptera: Thripidae) in

the greenhouse Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Inves	tigator's Name (s	s) (typed)
An	di Nasruddin	
Date	March 20, 199	12

\*Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24:141-42.

Deposit as follows:

Original: Include as Appendix 1 in ribbon copy of thesis or

dissertation.

Copies: Included as Appendix 1 in copies of thesis or dissertation.

Museum(s) files.

Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

# APPENDIX 1.1

# Voucher Specimen Data

Page 1 of 1 Pages

		Number of:
Species or other taxon	Label data for specimens collected or used and deposited	Museum where depos- ited Other Adults of Adults P Pupae Nymph Larvae Eggs
Frankliniella <u>occidentali</u> (Pergande)	June 17, 1989 Andi Nasruddin Sinningia speciosa (Benth & Hook)	50 15 MSU
(Use additional sheets if necessary	sary)	
Investigator's Name(s) (typed)	Voucher No. 1992-	
Andi Nasruddin	Received the above 11:	listed specimens for
	Entomology Museum.	all state ulliveisity
	Sand L. File	- 25 march 1992
Date <b>March 20, 1992</b>	Curator	Date