THE CORRELATION OF SOIL TYPE, MOISTURE, VARIETY AND RATES OF APPLICATION ON UPTAKE OF SYSTEMIC INSECTICIDES IN FIELD BEANS

Thesis for the Degree of M. S.
MICHIGAN STATE UNIVERSITY

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1961

ABSTRACT

THE CORRELATION OF SOIL TYPE, MOISTURE, VARIETY
AND RATES OF APPLICATION ON UPTAKE
OF SYSTEMIC INSECTICIDES IN FIELD BEANS

by Thomas Rosario Castro

Soil applications of 10% granular Thimet (phorate), (0,0-diethyl S-ethylthiomethyl phosphorodithioate) and 10% granular Di-Syston, (0,0-diethyl S-2(ethylthio) ethyl phosphorodithioate) were evaluated for the effects of soil type, moisture, variety and rates of application on plant growth and uptake of toxicant in field beans. Bioassays to determine effectiveness of the treatments were conducted with the two-spotted spider mite, Tetranychus telarius (L.); bean aphid, Aphis fabae Scop.; Mexican bean beetle, Epilachna varivestis Muls.; and vinegar fly, Drosophila melanogaster Meig. The results of the experiments are outlined as follows:

1) A greenhouse culture of two-spotted spider mites was found resistant to Thimet when the insecticide was applied in bands in soil at rates up to 12 pounds per acre.

2) Thimet applied in bands in the soil at rates as low as 0.25 pounds per acre was effective in controlling a non-resistant strain of the two-spotted spider mite.

3) Thimet applied at 1, 2 and 4 pounds, and Di-Syston at 2 pounds per acre in bands, were effective against Mexican bean beetle larvae and adults for 44 days after planting. These rates did not control infestations of whiteflies.

4) Better control of Mexican bean beetle larvae was obtained from the chemical 20 days after seeding than 14 days after seeding.

5) There was no apparent toxic effect on adult Mexican bean beetles, fed on green pods from plants grown in muck, loam and loamy sand soils treated with Thimet at a rate of 2 pounds per acre.

6) Thimet residues from 3 (1 pound broadcast) to 5.8 (3 pounds broadcast) p.p.m. were present in bean leaves 15 days after seeding. Thirty-five days after seeding most residues had decreased to 1 p.p.m.

7) Comparable amounts of Thimet applied to beans in-row with seed or mixed in the soil (broadcast) resulted in more phytotoxicity than when applied in bands 2 inches to the side and 1 inch below the seed.

8) Excessive moisture reduced the effectiveness of systemic application and increased phytotoxicity symptoms.

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By

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A THESIS

Submitted to the College of Science and Arts,
Michigan State University
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Entomology

1961

ACKNOWLEDGMENTS

My grateful appreciation is extended to Professor Ray Hutson, Head, Department of Entomology, for making this study possible.

I wish to express special thanks to Dr. Gordon Guyer, under whose direction this investigation was made. His constant help and enthusiasm never failed me during this research.

My grateful appreciation is extended to Mr. Arthur Wells for his invaluable assistance throughout the project. I also wish to thank Dr. R. Hoopingarner for his assistance with statistical analysis and in conducting the bioassay tests.

My sincere appreciation is also extended to Doctors E.C. Martin, Axel Andersen and J.W. Butcher for their critical reading of the manuscript.

I wish to thank Messrs. Alfred Borgatti and Robert McClanahan for their help and suggestions during this study.

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INTRODUCTION

Insects and insect transmitted diseases are serious problems of the field and snap bean industry in the United States. The Mexican bean beetle, Epilachna varivestis Muls., is one of the most serious bean pests. It has been present in the southwest for more than a century and appears to have migrated from Mexico, where it was widely distributed. It was first recorded from Alabama in 1920. Records of its presence in the central and eastern bean growing regions followed in rapid succession. In Michigan the bean aphid, Aphis fabae Scop., the six-spotted leafhopper, Macrosteles fascifrons (Stal.), and the potato leafhopper, Empoasca fabae (Harris), are also important bean pests. The aphids produce injury both by feeding and disease transmission.

Numerous insecticides have been applied as foliar sprays or dusts for control of the insect complex on beans. A more recent development has been the use of systemic insecticides for this purpose. Way and Needham (1957) reported that beans could be protected from bean aphid infestations by the application of demeton directly to seeds or as soil treatments. Also, Guyer et al (1960) reported satisfactory control of sucking insects on field and snap beans with soil applications of Thimet (phorate) and Di-Syston (Bayer 19639).

The objectives of the investigations reported here were:

1) To study the movement of systemic materials (Thimet and Di-Syston) through the bean plant.

2) To determine by bioassays the quantity of insecticides in the plant at different stages of growth, when applied at various rates.

3) To evaluate the effects of soil moisture and texture on phytotoxic symptoms in treated plants.

4) To evaluate the response of different varieties of field beans to treatments.

LITERATURE REVIEW

Bennett (1949) defined a systemic insecticide as: "a substance which is absorbed and translocated to the other parts of the plant, thus rendering untreated areas insecticidal". The systemic behaviour of chemicals was observed as early as the fifteenth century, when Leonardo da Vinci injected arsenic into a peach tree trunk, killing the pests on the tree and rendering the fruits toxic (USDA 1960). One of the first known uses of a systemic insecticide in the United States was the injection of potassium cyanide into a Spanish broom shrub for control of cottony-cushion scale, *Icerya purchasi* Maskell (USDA 1960).

One of the most fully explored systemic insecticides was sodium fluoroacetate, which was present in a South African plant, *Dichapetalum cymosum* (Bennett, 1957). David and Gardiner (1951) stated that sodium fluoroacetate was an extremely effective systemic insecticide, when applied to the leaves or roots of beans. However, they indicated that it was not of practical importance because of its high mammalian toxicity.

In 1936, Hurd-Karer and Poos studied the systemic action of selenium. They observed that wheat growing on seleniferous soils was not injured by aphids. In 1937

Hurd-Karer demonstrated that there was a correlation between the rates of absorption of sulphur and selenium by different plants, and that the sulphur-selenium ratio of the plant increased as the sulphur content of the seleniferous soil was increased. Fulton and Mason (1937) were the first workers to experimentally demonstrate the systemic property of derris. They found that derris when applied to the first two leaves of bean plants reduced the attack of the Mexican bean beetle on the new growth. Extracts from the first and second trifoliate leaves added to water resulted in 100 percent mortality to goldfish.

The discovery by Schrader of systemic insecticidal activity in widely separated types of compounds initiated and gave impetus to recent advancements in this field (Bennett 1957). Systemic insecticides have been investigated in fruit and forest trees as well as in field and garden crops. In 1950 Metcalf and Carlson showed that trunk and foliar applications of octamethyl pyrophosphoramide and demeton on citrus were effective in controlling the citrus red mite, Panonychus citri (McG.). Jepson et al (1954) studied seasonal influences on the efficiency of foliar sprays, trunk and soil applications of demeton against citrus red mite and citrus bud mite, Aceria sheldoni (Ewing). They indicated that summer trunk treatment was the most effective.

Haynes <u>et</u> <u>al</u> (1958) evaluated systemic insecticides for the control of the European pine shoot moth, <u>Rhyacionia</u> <u>buoliana</u> (Schif.), on red pine, and indicated that Thimet as a foliar spray, soil drench or granular soil application provided significant reductions in the larval populations. In 1959 Al-Azawi and Norris Jr. demonstrated that it was possible to reduce the transmission of <u>Ceratocystis</u> <u>ulmi</u> (Buis.) in elm trees by treating with Chipman R-6199 at 10, 20 and 30 grams per tree. They observed that the insecticide at 20 grams per tree was also effective in reducing the length of feeding niches of the bark beetle, <u>Scolytus</u> <u>multistriatus</u> (Marsh.).

Wilcox and Howard (1957) conducted tests with Thimet on strawberries and turnips and reported excellent control of aphids and spider mites by band treatment near the plants. In 1958 Guyer <u>et</u> <u>al</u>, in their investigations of systemic insecticides for control of Hessian fly, <u>Phytophaga</u> <u>destructor</u> (Say) on wheat, reported that Thimet and Bayer 19639 (Di-Syston) were effective in the control of fly larvae on winter wheat. They observed phytotoxicity in Thimet seed treatments in the early stages of plant development. Andres <u>et</u> <u>al</u> (1959) demonstrated that Thimet and Bayer 19639 (Di-Syston) applied as seed or furrow treatments controlled aphids for over a month on cabbage and cauliflower at 1/2 pound toxicant per acre. Applications of Thimet and Bayer 19639 (Di-Syston) on granules gave good control for only 5 days. Reynolds (1957) protected seedlings of alfalfa, cotton and sugar beets by applying systemic insecticides at the time of planting. They applied demeton, Systox thiol isomer and Bayer 19639 (Di-Syston) as coatings on alfalfa seed, and obtained satisfactory results in the control of the spotted alfalfa aphid, Therioaphis maculata (Buck.). In radio-active tracer studies with Bayer 19639, demeton and Thimet applied as seed treatments on sugar beets, cotton and alfalfa, these same authors found the highest concentrations of the toxicant in the cotyledons. The insecticide apparently was not translocated in substantial amounts to other parts of the plant. The highest concentrations were found in the oldest leaves and the lowest in the youngest leaves.

Systemic insecticides have been investigated extensively as seed treatments. Chao (1950) protected plants of cotton, peas, beans and nasturtiums from aphid attack by preplanting seed treatment. Ashdown and Cordner (1952) protected peas from aphid infestations with diethoxy-thiophosphoric acid ester of 2-ethyl mercaptoethanol, applied both as a soil and seed treatment. Schradan and demeton were also used successfully for the treatment of peas and beans (David and Gardiner 1955). These authors did not observe preferential absorption of water or thiol isomer of demeton by the seed. These findings were in agreement with the conclusion of Atkins (1909), that the membrane of bean seeds becomes semipermeable only after germination. Chao (1950), however, observed selective absorption of water from a solution of schradan by broad beans.

A summary of the many studies associated with soil applications of systemic insecticides indicates that differential absorption of the toxicant by the plant does take place in various soil types. Getzin and Chapman (1959) studied the effect of soil type on uptake of systemic insecticides and found the highest translocation rates of Thimet and dimethoate in sand; lesser rates in clay loams and lowest in muck. They also studied the binding of the insecticide to soil by measuring leaching rates of radioactive Phosdrin through columns of twelve soils. They correlated binding of the insecticide with the base-exchange capacity, organic matter, and nitrogen content of the soil, and concluded that organic matter content was the factor primarily responsible for insecticide binding.

The translocation of systemic insecticides within the plant is not clearly understood. Bennett (1957) suggested that the process of translocation would vary according to the insecticide and the site of absorption. Chao (1950) demonstrated that schradan, when applied as a seed treatment, tended to accumulate more in the older parts of the plant than in the young leaves. David and Gardiner (1955) found that the distribution of the thiol isomer of demeton in the plant was fairly uniform after seed treatment. In 1952 Metcalf and March demonstrated that labelled schradan and phosphoric acid were translocated uniformly throughout lemon seedlings. Metcalf <u>et al</u> (1954) found that movement of the thiono isomer of demeton in lemon seedlings was similar in amount and direction to that of schradan, though it tended to accumulate more in the terminal leaves.

Due to the high mammalian toxicity of most systemic insecticides, detoxification is essential in edible crops. However, for the insecticide to be efficient, it should not break down too rapidly. Detoxification has been reported as occurring by actual loss of toxicant from the plant surface or by the breakdown of the toxicant by the plants (Bennett 1957). In 1949 Bennett showed that vapor loss of dimefox and bis (2-fluoroethoxy) methane occurred from leaves following root absorption. Again, Thomas <u>et al</u> (1955) demonstrated that the volatile thiol isomer of demeton was translocated to, and given off from the leaves, following root absorption. Guyer <u>et al</u> (1958) found that Thimet residues in wheat seedlings decreased from 4.9 p.p.m., 13 days after planting to below a detectable amount 57 days after planting. Hartley and Heath (1951) suggested that the breakdown of schradan was by enzymic oxidation.

Before the development of systemic insecticides, rotenone, pyrethrins, piperonyl cyclonene, methoxychlor and other materials were employed for control of insects on beans. The organophosphorus insecticides, malathion, parathion, and demeton have been evaluated for control of bean insects by Brett and Brubaker 1955 and 1958, Moore 1950, Ditman and Wiley 1958. In 1957 Way and Needham systemically protected beans from aphid infestations by applying demeton as a seed treatment and soil application. Guyer et al (1960) reported satisfactory control of sucking insects and a reduction of Mexican bean beetle injury when 1 and 2 pounds of Thimet or Di-Syston were applied per acre. In 1960, Wilcox and Howland reported good control of two-spotted spider mites on lima beans for 2 to 3 months by soil applications of granular or liquid formulations of Thimet and Di-Syston as seed-furrow treatments.

PROCEDURE

All experiments in this study were conducted in the Michigan State University Plant Science greenhouses at East Lansing, Michigan. Eight-inch clay pots, sterilized greenhouse soil and dark red kidney beans (*Phaseolus vulgaris* L. var. Dark Red Kidney) were used except for Experiments I and VI, where different soils and bean varieties were substituted. All treatments were randomly arranged in 4 replications. Regular Thimet (phorate) 10% granular was used in all experiments. Di-Syston and Thimet C.S. both 10% granular were also used in Experiment II. The rates given throughout this presentation are amount of active toxicant.

Insects used for Testing

The Mexican bean beetles used for these investigations were collected from bean fields in Montcalm County, Michigan. They were then reared in the greenhouse by a modification of the methods described by Peterson (1959). The beetles were reared on tender bean plants grown in four-inch clay pots at temperatures from 75° to 85° F. and at relative humidities over 60 percent. A large, wooden framed, galvanized screen cage, measuring 25 x 30 x 30 inches was used for oviposition. Fluorescent bulbs were installed over the cage to furnish the light required to maintain 14 to 15 hours photoperiod. Egg masses were collected on alternate days. The excessive leaf material was removed, leaving about 1 square-inch-piece of leaf with the egg mass. These egg masses were then placed in a covered Petri dish on a disc of blotting paper, wetted with a saturated solution of sodium chloride. The eggs started to hatch after 5 to 7 days when kept at 70° to 75° F. The larvae, upon hatching, remained clustered on the empty egg shells for about 10 to 16 hours. Within this period the leaf pieces with the young larvae were placed on tender bean foliage. The plants with the young larvae were placed in another wooden framed, galvanized screen cage measuring 15 x 22 x 22 inches. Fresh foliage was supplied every 2 days. When the larvae pupated, the pupae were collected in a pint box and placed in the oviposition cage.

Bean aphids (*Aphis fabae* Scop.) were collected in August from field beans in Saginaw County, Michigan. The cultures were maintained on young bean plants in the greenhouse. Attempts to rear these aphids for an extended period failed.

Two different cultures of two-spotted spider mites, *Tetranychus telarius* (L.) were maintained. The culture used for Experiments I and II was from the Michigan State University greenhouse culture. These mites had been subjected to treatments of numerous miticides and organophosphoric insecticides. The mites used in Experiments V and VI were collected from a red clover field on the Michigan State University Farm, Crop Farm, East Lansing. This mite culture was reared on bean plants in the greenhouse, but was isolated from greenhouse infestations.

Vinegar flies, *Drosophila melanogaster* Meig., were obtained from a culture maintained in laboratory.

Methods of Seeding and Placement of Insecticides

In Experiments I to VI the insecticide was applied in bands in the soil. When this method was used an 8-inch clay pot was filled with 5 inches of well compacted soil. A thin band of the insecticide was applied and over this band one inch of soil was added. Five bean seeds were then planted and arranged so that the band of insecticide was 1 inch below and 2 inches to the side of the seeds. The seeds were covered with 1 inch of soil leaving still another 1 inch of space in the pot for holding water.

In the in-row application the insecticide was placed directly with the seed over 6 inches of soil in the clay pot and covered with 1 inch of soil.

For the broadcast treatment the insecticide was mixed with the upper 1-inch layer of soil around the seed.

Bioassay Tests

A modification of the floating leaf-disc method as described by Rodriguez (1953) and Eichmeier (1958) was used to test mites. Using this method, leaf discs were excised with a seven-eighth inch diameter cork borer from detached leaves. As the discs were cut, they were fastened to a piece of pencil eraser by a pin and floated in a Petri dish containing a 2 percent sucrose solution. The piece of eraser acted as an anchor and kept the discs in position. The floating discs were infested with 5 female two-spotted spider mites. The Petri dishes with the infested, floating discs were placed under fluorescent lights and watered regularly. Tests with aphids were conducted in the same manner using 5 aphids on each leaf disc. The leaf discs were maintained for a period ranging from 6 to 10 days after which the number of mites and aphids on the discs were counted.

The first instar Mexican bean beetle larvae tests on bean plants were conducted by placing a piece of leaf containing 50, 8 to 10-hour-old larvae on the foliage. Mortality counts were made every 2 days. The second and third instar larvae and adults of Mexican bean beetle were placed directly on the bean foliage for feeding and mortality studies.

Assessment of the feeding injury by the Mexican bean beetle larvae and adults on bean foliage was rated as follows: 1 = scattered feeding; 2 = 1/4 to 1/3 leaf destroyed; 3 = 1/3 to 1/2 leaf destroyed; 4 = more than 1/2 leaf destroyed.

Experiment I

The purpose of the first experiment was to evaluate the uptake of Thimet from widely different soil types. It was begun on July 25, 1960 using the following three soils:

1) muck (obtained from the Muck Experimental Farm, Clinton Co.).
2) loam (obtained from the Lutz Farm, Montcalm Co.).
3) loamy sand (obtained from the Hillman Farm, Montcalm Co.).

Each of the soils received a banded 2-pound Thimet treatment and together with an untreated control, made 6 pots in each of the 4 replications. On August 9 (15 days after planting) bioassays were conducted with 2 sets of leaf discs excised from the first true leaves of the treated and untreated bean plants, using two-spotted spider mites (greenhouse culture) and bean aphids. Counts of mites and aphids were made after 3 and 9 days respectively. On August 15 (21 days after planting) bioassays with 2 more sets of leaf discs from the third and fourth trifoliate leaves of the bean plants were conducted using mites and aphids. In this case counts of mites and aphids were made after 6 and 10 days respectively.

Bioassay tests with first instar Mexican bean beetle larvae were conducted from August 19 - 23 on treated and untreated bean plants. Mortality counts were made every 2 days. On August 27 (33 days after planting) the plants in the 3rd and 4th replications were infested with 10 second and third instar larvae of the bean beetle. Larval mortality counts were made every 2 days. Feeding injury of the Mexican bean beetle larvae was assessed according to the rating described in the methods section.

On September 14 (50 days after planting) two green bean pods were picked from each of the treatments and the controls and placed separately in 1 pint paper boxes along with 3 newly emerged Mexican bean beetle adults. The feeding damage and mortality were evaluated after 5 days. All the plants were removed from the pots on September 21 so the roots could be examined for symptoms of phytotoxicity.

Experiment II

The second experiment was established on August 5 to correlate various rates of Thimet and Di-Syston applications with insect mortality in the aerial plant parts. The insecticide application, experimental design and planting methods were the same as those used in Experiment I. Sterilized greenhouse soil was used with insecticide treatments as follows:

Treatments	Dosage per acre
Granular Thimet	1 lb.
Granular Thimet	2 lbs.
Granular Thimet	4 lbs.
Granular Thimet C.S.	2 lbs.
Granular Di-Syston	2 lbs.
Control	--

On August 19 (14 days after planting) bioassays were conducted with mites (greenhouse culture) and aphids on leaf discs excised from the first true leaves. After one week, the same tests were run with discs from the third and fourth trifoliate leaves.

Bioassays with first instar larvae of the Mexican bean beetle were conducted on all bean plants, on August 27. The plants were also assessed for phytotoxicity symptoms and stand counts were made.

On September 11 (35 days after planting) mortality and feeding tests were carried out with 5 newly emerged Mexican bean beetle adults on 2 of the 4 replications. The plants in this experiment were heavily infested with mites and whiteflies, _Trialeurodes vaporariorum_ (Westwood) as a consequence of normal greenhouse migrations. The bean plants were assessed for the degree of infestation. Plants were rated from 1 to 4 according to the number of mites or whiteflies per infested leaf as follows: 1 = less than 20; 2 = 21-40; 3 = 41-60; 4 = 61 and more.

Experiment III

The third experiment was set up on August 23, to study the effect of different levels of soil moisture on the uptake of Thimet. The arrangement of the experiment was similar to Experiments I and II. The soil-moisture levels were designated as high, normal and low and each contained a 2-pound Thimet treatment and an untreated control. The high moisture level was obtained by placing pots in metal trays containing approximately one inch of water which was maintained throughout the experiment. The normal moisture treatments were watered in the regular manner and in the low moisture treatments just enough water was added for the plants to survive.

Fifteen days after planting, bioassays were carried out with first instar Mexican bean beetle larvae. At this stage the plants in the low moisture treatments had developed only 2 leaves while the plants in the other treatments had reached the first and second trifoliate leaf stage. On September 19 a second bioassay test was run again with first instar larvae. Feeding injury, stand and growth were recorded. The normal growth of the bean plants was adversely affected by the high and low moisture treatments.

Experiment IV

The fourth experiment was established on September 6 to study the effect of high rates of Thimet on bean plants and to evaluate the effect of excessive rates of Thimet on what appeared to be a resistant mite population. Ten percent granular Thimet was applied in soil bands at the rate of 6, 8 and 12 pounds per acre. Records were obtained on stand, growth and phytotoxicity.

Experiment V

This experiment was arranged on September 15 to ascertain the minimum rates of Thimet effective for the control of mites and Mexican bean beetles. The four treatments in this experiment were 0.25, 0.5, and 1.0 pounds of Thimet (10% granular) per acre and a control. The bean seeds and insecticides were arranged as in the preceding experiments.

On October 15 a bioassay was made with the first instar larvae of Mexican bean beetles in the same manner as in the previous experiments. Bioassays using the floating leaf disc method were started on October 11 and 27 using mites from the red clover field culture. By November 5 the bean plants were heavily infested with mites and whiteflies and were destroyed.

Experiment VI

On September 24 Experiment VI was arranged to evaluate the uptake of Thimet in four varieties of field beans. The varieties used were Michigan Dark Red Kidney, Michigan Yellow Eye, Michelite and Michigan Cranberry. Each variety of bean received a 1-pound Thimet treatment applied in the same manner as before and a control.

On October 25 a bioassay was carried out by the leaf disc method with mites from the red clover culture.

Experiment VII

The last experiment was begun on December 14 to compare in-row and broadcast treatments of Thimet, and consisted of 7 treatments. In the in-row application, the Thimet was placed with the bean seeds and in the broadcast treatment it was mixed with the upper one inch of soil. The three in-row treatments were 0.5, 1.0 and 2.0 pounds of Thimet per acre, and the broadcast treatments 0.5, 1.0 and 3.0 pounds per acre.

On December 30 (15 days after planting), bioassay tests were conducted with Drosophila melanogaster Meig. on leaf tissue media. The first leaves from replications I and II were removed from the plants. Ten grams of leaf material were collected from each treatment and placed in a 1-pint Mason jar with 20 grams of distilled water. The leaves were macerated in the water by a blending Osterizer. To the macerated leaf material, 70 grams of pumpkin media were added and mixed thoroughly to give a homogeneous mixture of pumpkin media and leaf matter, which made a dilution of 10 percent leaf matter. Further dilutions of 5, 2.5, 1.25 and 0.625 percent were made with pumpkin media. Thus there were 5 dilutions of leaf mixture for each treatment in the experiment.

Two shell vials were marked for each of the different treatment dilutions and small strips of paper towels were placed in each vial. Small amounts of the diluted leaf-pumpkin media were spread on the papers inside the vials according to their respective dilutions. About 25 to 50 flies were anaesthetized with ether and carefully inserted in each of the vials. The vials were plugged with cotton and maintained at 30° C. After 24 hours the mortality of flies was recorded for all the vials. A standard test employing pure Thimet dilutions with pumpkin media was also run to establish the ED_{50} dosage curve. ED_{50} values for different treatments were estimated by the "Nomograph Calculations of Dose-effect" method of Litchfield and Wilcoxon and were then compared with the ED_{50} dosage of the standard Thimet test. These values were then correlated with the amount of Thimet in leaves of the different treatments.

On January 9 and 19, 1961 two similar bioassay tests were carried out. Leaves from the replications III and IV were used for one test on January 9, and leaves from all the replications for the test on January 19. The leaf matter concentration in pumpkin media for these tests ranged from 10 to 0.625 percent for the January test and 20 to 1.25 percent for the test on January 19.

PRESENTATION AND DISCUSSION OF RESULTS

Experiment I

Results of bioassays conducted on August 9 and 15 on detached leaf discs with two-spotted spider mites (greenhouse culture) and bean aphids are presented in Table 1. After 6 days there were no significant differences between mite populations on the treated and untreated plants. This was in contrast with the findings of Wilcox and Howland (1960) who reported 2 to 3 months' control of two-spotted spider mites with Thimet dosages of less than 1 pound per acre applied to the soil. It was postulated that the greenhouse mite culture used for this assay was resistant to Thimet. There was a significant reduction in numbers of bean aphids on discs from the first true leaves removed from the Thimet treated plants, grown in loam and loamy sand soils. Aphid populations on leaf discs from treated plants grown in muck were not significantly different from those of control plants, in fact, there was a trend toward higher populations on discs from treated plants. Aphid populations on discs from the top trifoliate leaves which were removed from both treated and untreated plants grown in loam and loamy sand soils were significantly lower than those from the plants grown in muck. However, there were no significant

Table 1. Numbers of two-spotted spider mites (from greenhouse cultures) and bean aphids on leaf discs, removed from the (a) first true leaf and (b) top trifoliate leaves of plants grown in three different soils.

Soil	Pounds of actual Thimet per acre	Mean(i) number of mites or aphids per leaf disc(ii)			
		from first true leaves 15 days after planting		from top trifoliate leaves 20 days after planting	
		Mites	Aphids	Mites	Aphids
Muck	2	9 a	33 bc	33 a	10 b
Muck	-	22 a	14 ab	23 a	20 c
Loam	2	13 a	2 a	32 a	0 a
Loam	-	25 a	24 bc	37 a	4 a
Loamy sand	2	19 a	7 a	31 a	1.0 a
Loamy sand	-	22 a	45 c	43 a	5.0 a

(i) Means with a common letter are not significantly different from each other at the 5 percent level (Duncan 1955).

(ii) Mites and aphids were maintained on the discs for 6 and 9 days respectively.

differences in the populations on discs from treated and untreated plants grown in these two soils. The variation in results in this part of Experiment I was thought to be due to the difficulty in maintaining a strong bean aphid culture as well as problems in developing a consistent technique for disc culture.

Results of the feeding studies conducted with a) first, and b) second and third instar Mexican bean beetle larvae are summarized in Tables 2 and 3. There was a maximum reduction of only 65 percent of beetle larvae on treated plants grown in muck, in contrast to 100 percent mortality of larvae on plants grown in loam and loamy sand soils. In both tests, however, there was a significant reduction in larval populations 2 and 4 days after infestation on treated plants grown in muck.

The rating of feeding injury on the leaves is presented in Table 4. All control plants were almost completely destroyed 24 days after infestation. Treated bean plants grown in the loam and loamy sand soils were injured the least, and the injury was significantly less than that on the treated plants grown in muck.

Mexican bean beetle adult feeding damage on the green bean pods was evaluated and the results are given in Table 5. The degree of feeding damage to pods did not

Table 2. Mortality of first instar Mexican bean beetle larvae fed on foliage of bean plants grown in three different soils. Plants were infested with larvae 26 days after planting.

Soil	Pounds of actual Thimet per acre	Percent larval mortality (i)	
		2 days after larval infestation	4 days after larval infestation
Muck	2	55 b	65 b
Muck	-	20 c	25 c
Loam	2	100 a	100 a
Loam	-	25 c	33 c
Loamy sand	2	100 a	100 a
Loamy sand	-	25 c	30 c

(i) Means with a common letter are not significantly different from each other at the 5 percent level (Duncan 1955).

Table 3. Mortality of second and third instar Mexican bean beetle larvae fed on foliage of bean plants grown in three different soils. Larvae placed on plants 53 days after planting.

Soil	Pounds of actual Thimet per acre	Percent larval mortality (i)	
		2 days after larval infestation	4 days after larval infestation
Muck	2	35 b	55 b
Muck	-	2 c	12 c
Loam	2	67 a	87 a
Loam	-	7 bc	20 c
Loamy sand	2	95 a	100 a
Loamy sand	-	5 c	17 c

(i) Means with a common letter are not significantly different from each other at the 5 percent level (Duncan 1955).

Table 4. Assessment of feeding injury of Mexican bean beetle larvae on bean plants grown in three different soils. Rating of feeding injury: 1 = scattered feeding; 2 = 1/4-1/3; 3 = 1/3-1/2; and 4 = more than 1/2 of leaf consumed.

Soil	Pounds of actual Thimet per acre	Mean feeding injury(i)
Muck	2	2.9 b
Muck	-	4.0 a
Loam	2	1.1 c
Loam	-	3.8 a
Loamy sand	2	1.0 c
Loamy sand	-	4.0 a

(i) Means with a common letter are not significantly different from each other at the 5 percent level (Duncan 1955).

Table 5. Evaluation of Mexican bean beetle adult feeding injury on green bean pods removed from plants grown in three different soils. Feeding injury rating: 1 = no feeding; 2 = less than 1/4; 3 = 1/4 to 1/2; and 4 = more than 1/2 of the outer pod surface.

Soil	Pounds of actual Thimet per acre	Mean feeding injury(i)
Muck	2	4 a
Muck	-	4 a
Loam	2	4 a
Loam	-	4 a
Loamy sand	2	3.8 a
Loamy sand	-	4 a

(i) Means with a common letter are not significantly different from each other at the 5 percent level (Duncan 1955).

indicate any significant difference among treatments and controls. This indicated that the toxicant, if present in the pods, was in very minute quantities and had no effect on the feeding of adult Mexican bean beetles.

There was no indication of phytotoxicity symptoms in any of the soil-insecticide treatment combinations in Experiment I. Results of this experiment indicated that Thimet was less effective in muck than in the loam and loamy sand soils. Getzin and Chapman (1959) reported that Thimet uptake was greater in sand and clay loams than in muck. They indicated that organic matter bound the insecticide in the soil. This binding effect of Thimet on muck may have influenced the results in Experiment I.

Experiment II

The results of the bioassays carried out on August 19 and 24 (14 and 19 days after planting) with two-spotted spider mites and bean aphids on excised leaf discs are summarized in Table 6. Mite counts made 6 days after infestation indicated that there were no significant differences in populations of mites on the leaf discs removed from the treated and control plants. These results give additional support to the hypothesis that the mites from the greenhouse culture were resistant to Thimet. Aphid

Table 6. Number of mites and aphids on detached leaf discs from the first true and top trifoliate leaves of bean plants grown in treated and untreated soil.(i)

Treatments	Pounds of actual insecticide per acre	Mean number per disc(ii)		
		First true leaf disc		Top trifoliate leaf disc
		Mites	Aphids	Aphids
Thimet	1	61 a	24 a	4 a
Thimet	2	46 a	8 a	3 a
Thimet	4	46 a	2 a	11 a
Thimet	2	55 a	7 a	17 a
Di-Syston	2	50 a	37 a	12 a
Control	-	57 a	67 a	18 a

(i) Mites and aphids were counted 6 and 10 days respectively after infestation.

(ii) Means with common letter are not significantly differently from each other at the 5 percent level (Duncan 1955).

counts were taken 10 days after infestation and in both tests, on discs from the first true leaves and upper trifoliate leaves, the aphid populations appeared to be higher in the controls. Differences were not statistically significant, however. The lack of significant differences between the treatments and controls may have been due to the variable size of the populations in the different replications.

The mortality of first instar Mexican bean beetle larvae was significantly higher on the treated plants than on the controls (Table 7). All treatments had 100 percent larval mortality four days after infestation. Table 8 shows the results of the mortality studies with Mexican bean beetle adults on bean plants. The 2 and 4-pound applications of Thimet resulted in 100 percent kill of the beetles after 2 days. All other treatments gave 100 percent beetle mortality in 4 days, in contrast to no mortality in the control.

Results of the assays with second and third instar Mexican bean beetle larvae on bean plants are presented in Table 9. The larval mortality on all treated plants was high as early as 2 and 4 days after feeding was initiated. Thimet applications of 2 and 4 pounds per acre gave 100 percent mortality within 2 days. There were no significant differences in mortality among the different treatments. Assessment of feeding damage by the Mexican bean beetle

Table 7. Mortality of first instar Mexican bean beetle larvae placed on bean plants 32 days after planting; grown in treated and untreated soil.

Treatments	Pounds of actual insecticide per acre	Percent larval mortality (i)	
		2 days after larval infestation	4 days after larval infestation
Thimet	1	87 a	100 a
Thimet	2	100 a	100 a
Thimet	4	100 a	100 a
Thimet C.S.	2	87 a	100 a
Di-Syston	2	75 a	100 a
Control	-	14 b	24 b

(i) Means with common letter do not differ significantly from each other at the 5 percent level (Duncan 1955)

Table 8. Mortality of Mexican bean beetle adults fed on plants grown in treated and untreated soil.(i)

Treatments	Pounds of actual insecticide per acre	Percent Mexican bean beetle mortality(ii)	
		2 days after larval infestation	5 days after larval infestation
Thimet	1	70 a	100 a
Thimet	2	100 a	100 a
Thimet	4	100 a	100 a
Thimet C.S.	2	0 b	100 a
Di-Syston	2	0 b	100 a
Control	-	0 b	0 b

(i) Beetles placed on plant 35 days after planting.

(ii) Means with common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

Table 9. Mortality of second and third instar Mexican bean beetle larvae fed on bean plants grown in treated and untreated soil. Larvae were placed on plants 44 days after planting.

Treatments	Pounds of actual insecticide per acre	Percent larval mortality[i] 2 days after larval infestation	Percent larval mortality[i] 4 days after larval infestation
Thimet	1	75 a	87 a
Thimet	2	100 a	100 a
Thimet	4	100 a	100 a
Thimet C.S.	2	71 a	83 a
Di-Syston	2	71 a	79 a
Control	-	0 b	0 b

(i) Means with common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

larvae and adults (Table 10) indicated that the control plants were nearly destroyed after the tests, with little injury to the treated plants. No significant variations in feeding injury were indicated among the different insecticide treatments. Variations in natural mite and whitefly infestations in the greenhouse are given in Table 11. Intensive mite infestations were present on plants from both the treatments and control, however, there were significantly more mites on the Di-Syston treatment and control. There was a relatively high whitefly infestation with no significant differences among treatments and controls.

With the exception of the 4-pound Thimet treatment, the insecticide applications appeared to have no adverse effect on normal germination and growth of the bean plants. Plants treated with 4 pounds of Thimet per acre showed slight yellowing of leaves during the early period of plant growth.

Experiment III

The results of feeding studies of first instar Mexican bean beetle larvae on high, normal and low soil moisture treatments are presented in Table 12. Two days after infestation the larval mortality did not vary significantly in any of the treatments or control. Larval

Table 10. Assessment of feeding injury by Mexican bean beetle (36 days after planting) on bean plants grown in treated and untreated soil.

Treatments	Pounds of actual insecticide per acre	Mean of feeding injury(i)
Thimet	1	1.25 b
Thimet	2	1.00 b
Thimet	4	1.00 b
Thimet C.S.	2	1.00 b
Di-Syston	2	1.75 b
Control	-	4.00 a

(i) Means with common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

Table 11. Number of naturally occurring mites and white-flies on bean plants grown on treated and untreated soils. Ratings of infestation estimated as number of mites and whiteflies per infested leaf: 1 = less than 20; 2 = 21 to 40; 3 = 41 to 60; and 4 = 61 and above.

Treatments	Pounds of actual insecticide per acre	Mean degree of infestation[(i)]	
		Mites	Whiteflies
Thimet	1	2.75 bc	1.25 a
Thimet	2	2.75 bc	1.00 a
Thimet	4	2.25 b	1.25 a
Thimet C.S.	2	2.50 b	1.25 a
Di-Syston	2	3.50 ac	1.25 a
Control	-	4.00 a	1.75 a

(i) Means with a common letter do not differ significantly from each other at the 5 per cent level (Duncan 1955).

Table 12. Mortality of first instar Mexican bean beetle larvae fed on bean plants grown at three soil moisture levels.

Moisture levels	Pounds of actual Thimet per acre	Percent larval mortality(i)				
		Larval infestations				
		14 days after planting			22 days after planting	
		(ii) 2 days	(ii) 4 days	(ii) 6 days	(ii) 2 days	(ii) 4 days
High moisture	2	17.5a	62.5bc	65.0c	25.0b	70.0b
High moisture	-	10.0a	17.5d	22.5d	12.5c	20.0d
Normal moisture	2	12.5a	50.0c	95.0a	95.0a	100.0a
Normal moisture	-	10.0a	20.0d	25.0d	17.5bc	22.5d
Low moisture	2	35.0a	100.0a	100.0a	100.0a	100.0a
Low moisture	-	22.5a	75.0b	75.0b	22.5bc	45.0c

(i) Means with a common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

(ii) Days after larval infestation.

mortality was significantly higher in the treatments than in the controls after 4 and 6 days. The second test, conducted a week after the first, resulted in higher larval mortality within 2 days in the treated plants than in the control plants. The delayed effect of the toxicant in the treated bean plants on Mexican bean beetle larvae in the first test may have been due to an insufficient uptake of toxicant in the plant. At the initiation of the first assay, the plants had completed 7 to 8 days of growth following emergence and may not have taken up enough insecticide from the soil to have an immediate effect on the Mexican bean beetle larvae. Treated plants from low and normal soil moisture levels produced significantly higher kill than did treated plants grown at the high moisture level. The significantly higher mortality in the low moisture controls may have been due to stunted growth and lack of succulent foliage for larval feeding.

Assessment of the feeding injury of adult Mexican bean beetles on bean plants is presented in Table 13. All treated plants at the different soil moisture levels had significantly less feeding injury than the plants in the control. The relationships between height and spread of the plants grown at various soil moisture levels are given in Table 14. The plants in the low moisture level were significantly shorter and had less growth than the plants

Table 13. Assessment of feeding injury of Mexican bean beetle larvae on Thimet treated and untreated plants grown at three soil moisture levels.

Moisture Levels	Pounds of actual Thimet per acre	Mean feeding injury(i)
High moisture	2	1.8 bc
High moisture	-	2.8 d
Normal moisture	2	1.3 ab
Normal moisture	-	2.3 cd
Low moisture	2	1.0 a
Low moisture	-	2.3 cd

(i) Means with a common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

Table 14. Mean height and spread (crown width) of the Thimet treated and untreated bean plants grown at three soil moisture levels.

Moisture levels	Pounds of actual Thimet per acre	Mean height in inches	Mean spread in inches(i)
High moisture	2	15 b	12 b
High moisture	-	17 a	15 a
Normal moisture	2	16 a	17 a
Normal moisture	-	16 a	16 a
Low moisture	2	6 c	10 b
Low moisture	-	6 c	8 b

(i) Means with a common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

in the other moisture levels, with the exception of the plant spread in the high moisture level. In the high moisture level the treated plants were significantly shorter and had less growth than the controls. Factors that may have affected the bioassay results of this experiment were: (a) high temperatures (over 100° F.) for short periods and (b) excessively high and low soil moisture levels.

Experiment IV

The bioassay tests with two-spotted spider mites (greenhouse culture) showed normal mite reproduction on detached leaf discs, from plants treated with as high as 12 pounds per acre of Thimet soil band treatments. This added additional weight to the hypothesis that the mites from the greenhouse culture were highly resistant to Thimet. Seed germination in all treatments was satisfactory. However, symptoms of phytotoxicity were observed in all Thimet treatments.

Experiment V

Table 15 presents the results of the bioassays conducted with first instar Mexican bean beetle larvae on treated and untreated bean plants. The larval mortality

Table 15. Mortality of first instar Mexican bean beetle larvae fed on bean plants treated with three rates of Thimet. Plants were infested 23 days after seeding.

Treatments	Pounds of actual Thimet per acre	Percent larval mortality(i)
		2 days after infestation
Thimet	1.00	100.0 a
Thimet	0.50	81.5 ab
Thimet	0.25	63.5 b
Control	-	15.0 c

(i) Means with a common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

on the treated plants was significantly higher than the controls. One pound Thimet per acre gave 100 percent kill in 2 days. Results of the bioassays conducted on October 11 and 27 (26 and 42 days after planting) with two-spotted spider mites (red clover culture) gave significant reductions in the mite populations on the discs from plants treated with 1 pound Thimet (Table 16). In the first assay, discs from the 0.25 and 0.5 pound Thimet treatments produced significant reductions in the mite populations.

Results of these bioassays indicated that Thimet at dosages as low as 0.5 pounds per acre produced mortality of the Mexican bean beetle larvae and two-spotted spider mites for 42 days after planting.

Experiment VI

Experiment VI was designed to study the effects of Thimet in terms of insect mortality and phytotoxicity to different bean varieties. Bioassay results with two-spotted spider mites (red clover culture) on excised leaf discs in this experiment are presented in Table 17. The leaf discs from the Thimet treated plants in the four different bean varieties gave significantly lower mite populations as compared to their controls. There were no significant differences between mite populations on leaf discs from

Table 16. Populations of the two-spotted spider mites (red clover culture) on detached leaf discs from plants treated with three rates of Thimet.

Treatments	Pounds of actual insecticide per acre	Mean number of mites after 9 days (i)	
		Leaf discs infested 26 days after planting	Leaf discs infested 42 days after planting
Thimet	1.0	2.5 a	13.0 b
Thimet	0.5	3.0 a	41.0 a
Thimet	0.25	13.0 b	30.0 ab
Control	--	58.0 c	47.0 a

(i) Means having a common letter do not differ significantly from each other at the 5 percent level (Duncan 1955).

Table 17. Mean number of mites per detached leaf disc from the Thimet treated and untreated plants of four different bean varieties.

Variety	Pounds of actual Thimet per acre	Mean number of mites 10 days after infestation
Michigan Dark Red Kidney	1	14.5 *
Michigan Dark Red Kidney	-	32.5
Michigan Yellow Eye	1	6.0 *
Michigan Yellow Eye	-	65.0
Michelite	1	13.5 *
Michelite	-	61.0
Michigan Cranberry	1	4.5 *
Michigan Cranberry	-	64.0

*Differs from controls at the 5 percent level of significance (L.S.D. test, Snedecor 1957).

any of the treated plants. There was no evidence of phytotoxicity to any of the varieties.

Experiment VII

Dilutions of 95 percent technical Thimet with pumpkin media were bioassayed, using Drosophila melanogaster Meig. as a test animal. The results of this assay are presented in Table 18. The ED_{50} of Thimet, as determined by the "Nomograph Calculations of Dose-effect" method of Litchfield and Wilcoxon (1949), was found to be 0.153 p.p.m. (slope of line was 1.84). Results of the three bioassays conducted with drosophila, using macerated leaves in pumpkin media, are summarized in Tables 19, 20 and 21. The leaves used were removed at different stages in growth of bean plants growing in various soil treatments of Thimet. ED_{50} values for drosophila in each treatment of the three assays were determined as described before in this experiment; however, the ED_{50} values were expressed as percent leaf matter concentrations. Table 22 gives the conversion values of percent leaf matter concentrations for ED_{50} into parts per million (p.p.m.) of toxicant, comparable to Thimet in the leaves. On the basis that ED_{50} for drosophila was 0.153 p.p.m. of Thimet, it was postulated that all medium effective doses (ED_{50}) of leaf matter concentrations had 0.153 p.p.m.

Table 13. Mortality[(i)] of drosophila maintained in different dilutions of Thimet in pumpkin media for 24 hours.

Thimet in p.p.m. in pumpkin media	Percent mortality	ED_{50}[(ii)] (Median effective dose)
0.500	100.0	
0.250	100.0	
0.125	84.0	0.155 p.p.m.[(iii)]
0.100	28.0	
0.075	15.6	
0.063	16.6	

(i) Flies dead and unable to move counted as affected by Thimet.

(ii) Median effective dose was determined by following the method of Litchfield & Wilcoxon (1949).

(iii) At 5 percent level ED_{50} confidence limits were 1.75 and 1.53 and slope of line 1.84.

Table 19. Mortality of Drosophila maintained on bean leaf-pumpkin media for 24 hours, utilizing bean leaves removed from plants 15 days after planting.

Treatments	Pounds of actual Thimet per acre	Percent mortality(i): Percent leaf matter in media					ED_{50}(ii) as percent leaf matter	Slope of line
		10	5	2.5	1.25	0.625		
In row with seed	2.0	89.7	62.5	21.5	15.9	1.6	4.2	1.95
In row with seed	1.0	91.2	61.8	57.6	3.5	0	3.5	1.96
In row with seed	0.5	63.6	30.7	27.3	3.0	0	6.6	2.46
Broadcast in soil	3.0	100.0	87.5	34.8	20.7	1.9	2.6	2.04
Broadcast in soil	1.0	82.3	57.1	12.9	0	0	5.0	1.68
Broadcast in soil	0.5	100.0	54.4	30.1	0	4.2	4.4	1.64
Control	-							

(i) Flies which were moribund and unable to walk.

(ii) ED_{50} determined by Litchfield and Wilcoxon (1949) method.

Table 20. Mortality of drosophila maintained on bean leaf-pumpkin media for 24 hours, utilizing bean leaves removed from plants 25 days after planting.

Treatments	Pounds of actual Thimet per acre	Percent mortality(i) — Percent leaf matter in media: 10	5	2.5	1.25	0.625	ED_{50}(ii) as percent leaf matter	Slope of Line
In row with seed	2.0	100.0	19.0	32.0	17.8	3.7	3.9	2.68
In row with seed	1.0	34.6	16.1	0	0	0	10.75	1.77
In row with seed	0.5	31.5	0	0	0	0	--	
Broadcast in soil	3.0	100.0	50.0	51.5	12.0	2.6	3.45	2.36
Broadcast in soil	1.0	100.0	66.6	0	7.6	6.2	4.2	1.50
Broadcast in soil	0.5	84.6	40.0	20.0	2.9	0	5.25	1.11

(i) Flies which were moribund and unable to walk.

(ii) ED_{50} determined by Litchfield and Wilcoxon (1949) method.

Table 21. Mortality of drosophila maintained on bean leaf-pumpkin media for 24 hours, utilizing bean leaves removed from plants 35 days after planting.

Treatments	Pounds of actual Thimet per acre	Percent mortality[i]					ED_{50}[ii] as percent leaf matter	Slope of Line
		Percent leaf matter in media						
		20	10	5	2.5	1.25		
In row with seed	2.0	92.3	40.7	26.9	15.9	0	7.0	2.32
In row with seed	1.0	84.1	61.1	5.7	0	0	10.0	1.75
In row with seed	0.5	96.7	3.1	2.7	0	0		
Broadcast in soil	3.0	100.0	100.0	71.8	23.0	0	3.5	1.43
Broadcast in soil	1.0	52.0	21.4	0	0	0	16.0	1.49
Broadcast in soil	0.5	75.0	26.6	9.1	0	0	14.2	1.99

(i) Flies which were moribund and unable to walk.

(ii) ED_{50} determined by Litchfield and Wilcoxon (1949) method.

Table 22. Amount of toxicant estimated to be in the leaf tissues of the bean plants treated with soil applications of Thimet.

Treatments	Pounds of actual Thimet per acre	P.p.m. of toxicant		
		Days after planting		
		15 days	25 days	35 days
In-row	2.0	3.9	3.9	2.1
In-row	1.0	4.3	1.4	1.5
In-row	0.5	2.3	-	-
Broadcast	3.0	5.8	4.4	4.3
Broadcast	1.0	3.0	3.6	1.0
Broadcast	0.5	3.4	2.9	1.0

of Thimet, at the particular concentration. With this hypothesis, the p.p.m. of Thimet were estimated in leaves for each treatment at particular stage of plant growth.

The data in some treatments were heterogeneous, so that the confidence limits included probabilities of high and low values. The 0.5, 1 and 2 pound in-row treatments in the first assay produced highly heterogeneous data. In general, higher amounts of toxicant were estimated to be present in the leaves 15 days after planting, and smaller amounts were found 25 days after planting in all except the 2 pound in-row and 1 pound broadcast treatments of Thimet. These differences may have been due to experimental error. There was a considerable decrease in quantities of toxicant 35 days after planting, with the exception of the 1 pound in-row treatment. It has been determined that Thimet following uptake in plants is converted into its isomers or metabolites. Some of these metabolites were found to be potentially more effective cholinesterase inhibitors than is pure Thimet (Metcalf _et al_, 1949). The amounts of toxicant estimated to be present in the leaves, then, could proportionally be in much lesser quantities. This experiment also indicated that the amount of toxicant estimated to be in the plants' tissues was not directly proportional to Thimet applied; however, the 2 and 3 pound treatments of Thimet did indicate relatively high amounts.

The method employed in preparing the leaf and pumpkin media was not completely satisfactory. A fairly high amount of moisture was required for a homogeneous mixture, which may have affected the outcome of the bioassays. Phytotoxicity symptoms were observed in the 2 pound in-row and 3 pound broadcast Thimet treatments, with slight stunting in growth.

SUMMARY

Soil applications of granular Thimet and Di-Syston were evaluated for (a) phytotoxicity and (b) control of foliage infesting insects and mites on bean plants. Experiments were conducted in the greenhouse with various rates and methods of insecticide applications, on 3 different soil types, with 3 soil moisture levels and 4 varieties of field beans. Bioassays were carried out with two-spotted spider mite, *Tetranychus telarius* (L.), bean aphid, *Aphis fabae* Scop., Mexican bean beetle, *Epilachna varivestis* Muls. and vinegar fly, *Drosophila melanogaster* Meig.

The results indicate:

1) A greenhouse mite culture of two-spotted spider mites was found to be resistant to soil applications of Thimet as high as 12 pounds per acre.

2) Band applications of Thimet at rates as low as 0.25 pounds per acre were effective in controlling a non-resistant strain of the two-spotted spider mite.

3) Band applications of 1, 2 and 4 pounds of Thimet, and 2 pounds of Di-Syston per acre were effective against Mexican bean beetle larvae and adults for 44 days after

planting. These rates did not control infestations of whiteflies.

4) Band applications were less effective for control of Mexican bean beetle larvae at 14 days than they were 20 days after seeding.

5) There was no apparent toxic effect on adult Mexican bean beetles fed on green bean pods from plants grown in muck, loam and loamy sand soils treated with 2 pounds of Thimet per acre.

6) Thimet residues from 3 (1 pound broadcast) to 5.8 (3 pounds broadcast) p.p.m. were present in bean leaves 15 days after seeding. Thirty-five days after seeding most residues had decreased to 1 p.p.m.

7) Comparable amounts of Thimet applied to beans in-row with seed, or mixed in the soil (broadcast) resulted in more phytotoxicity than when applied in bands 2 inches to the side and 1 inch below the seed.

8) Excessive moisture reduced the effectiveness of the systemic application and increased phytotoxicity symptoms.

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