INTERACTION OF HERBICIDES, DISEASE, AND CULTURAL PRACTICES ON GROWTH AND DEVELOPMENT OF NAVY BEAN (PHASEOLUS VULGARIS L.)

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ABSTRACT

INTERACTION OF HERBICIDES, DISEASE, AND CULTURAL PRACTICES ON GROWTH AND DEVELOPMENT OF NAVY BEAN (PHASEOLUS VULGARIS L.)

By

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Injury to navy bean (Phaseolus vulgaris L.) from EPTC (S-ethyl dipropylthiocarbamate) was investigated as a function of soil moisture, planting depth, soil compaction, temperature, available nitrogen and seed quality. Greenhouse and growth chamber experiments showed that both high and low moisture stress increased navy bean injury from EPTC. Soil compaction in combination with EPTC greatly reduced navy bean growth. Ambient temperatures of 20 to 30 C did not alter the susceptibility of navy bean to EPTC. Stimulation of navy bean growth by nitrogen as measured by fresh weight was inhibited by EPTC. Field and greenhouse studies showed that EPTC injury was greater to mechanically damaged seed than to non-damaged seed.

The interaction of EPTC and atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine] on navy bean was studied in field and laboratory experiments. Navy beans grown in soil

treated with the combination of EPTC and atrazine developed extensive leaf chlorosis, greater than the atrazine treatment alone. Navy bean yields in field studies in 1972 were reduced 39% when grown in the presence of 0.55 kg/ha of atrazine and 3.36 kg/ha of EPTC, whereas, the single application of each herbicide reduced yields 0 and 14%, respectively. In 1973, atrazine at 0.28 kg/ha did not significantly reduce yields, but in combination with 2.24, 3.36 and 4.48 kg/ha of EPTC yields of navy bean were reduced 40, 30, and 62%, respectively. The combination of atrazine and EPTC also reduced plant weight and delayed flower and pod development. EPTC was found to increase wind blast injury under conditions of low relative humidity, high winds, and limited soil moisture. EPTC at 2.24, 3.36 and 4.48 kg/ha reduced the chloroform-soluble leaf extract from the surface of navy bean leaves. Scanning electron micrographs confirmed that EPTC at 10^{-6} M altered leaf surface waxes. EPTC at 10^{-5} and 10^{-6} M increased the transpiration rate. Increased uptake of ^{14}C-atrazine from solution was associated with the increased transpiration rate.

Applications of EPTC at 3.36 and 4.48 kg/ha increased root rot severity and reduced yields in artificially infested soil. EPTC at 3.36 kg/ha and chloramben (3-amino-2,5-dichlorobenzoic acid) at 3.36 kg/ha in field experiments caused the greatest increase in root rot severity and yield reductions. Dinoseb (2-sec-butyl-4,6-dinitrophenol) and fluorodifen (p-nitrophenyl α,α,α-trifluro-2-nitro-p-tolyl ether) at 5.04 kg/ha

caused the least root rot development of any herbicide treatment. In growth chamber experiments at 24 C, EPTC at 3.36 kg/ha and alachlor [2-chloro-2',6'-diethyl-N-(methoxymethyl) acetanilide] at 2.80 kg/ha applied to soil infested with Fusarium solani reduced navy bean root and shoot growth. Root rot severity was greatest in plants grown on EPTC treated soil at the 20 C ambient temperature. The growth of navy bean was reduced more as the level of Fusarium solani chlamydospore inoculum level increased in the presence of EPTC, than without EPTC. Growth of a moderately root rot resistant navy bean line 'RRR041' was not reduced when grown in soil infested with Fusarium solani and treated with EPTC. None of the herbicides tested increased virulence of Fusarium solani when grown on potato dextrose agar supplemented with herbicides. EPTC-treated navy bean seedlings were more susceptible to Fusarium solani than untreated seedlings. Scanning electron micrographs indicated that navy bean plants grown in EPTC had altered hypocotyl surfaces. EPTC and dinoseb increased the exudation of electrolytes, amino acids and sugars from root and hypocotyl tissue. None of the herbicides tested increased Fusarium solani hyphal development in soil or liquid culture. Dinoseb reduced hyphal development by the greatest amount of any herbicide tested in soil and liquid culture.

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By

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To Bev, Dawn and Ryan

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TABLE OF CONTENTS

	Page
LIST OF TABLES	viii
LIST OF FIGURES	Xi
INTRODUCTION	1
CHAPTER 1: LITERATURE REVIEW	4
EPTC Volatility and Incorporation	4
EPTC Symptoms	6
EPTC Metabolism	7
Temperature	8
EPTC Placement, Uptake, and Planting Depth	9
Soil Compaction	11
Damaged Seed	13
Nutrient Level - Herbicide Interaction	14
Herbicide-Herbicide Interactions	15
Navy Bean Root Rot Development	18
Herbicide - Disease Interactions	20
CHAPTER 2: FACTORS AFFECTING EPTC INJURY TO NAVY BEANS	24
Abstract	24
Introduction	25
Materials and Methods	28

Page

Results and Discussion..... 31

Soil Moisture..... 31

Planting Depth..... 31

Mechanical Compaction..... 33

Temperature..... 33

Nitrogen..... 33

Damaged Seed..... 37

Literature Cited..... 41

CHAPTER 3: THE INTERACTION OF ATRAZINE AND EPTC ON NAVY BEAN..... 43

Abstract..... 43

Introduction..... 44

Materials and Methods..... 46

Field Experiments..... 46

Transpiration Study..... 47

Leaf Wax Determination..... 47

Atrazine Uptake Study..... 48

Results and Discussion..... 49

Field plot studies..... 49

Transpiration Studies..... 54

Leaf Wax Determinations..... 57

Uptake of ^{14}C-Atrazine..... 62

Literature Cited..... 65

CHAPTER 4: HERBICIDE ROOT ROT INTERACTIONS IN NAVY BEANS..... 67

Abstract..... 67

Introduction..... 68

Page

Materials and Methods..... 71

Preparation and Infestation of Soil with *Fusarium solani*..... 73

Herbicide-*Fusarium solani* Interaction..... 75

Temperature..... 76

Inoculum Level..... 76

Genetic Resistance..... 76

Fusarium solani Virulence..... 76

EPTC-Induced Navy Bean Predisposition to Root Rot..... 77

Hypocotyl Surface..... 77

Exudate Studies..... 78

Fusarium solani Growth in Liquid Culture..... 79

Fusarium solani Chlamydospore Germination in Soil..... 79

Results..... 79

Growth Chamber Studies..... 82

Temperature..... 84

Inoculum Level..... 85

Genetic Resistance..... 88

Fusarium solani Virulence..... 88

EPTC-Induced Navy Bean Predisposition to Root Rot..... 88

Root and Hypocotyl Exudates..... 94

Effect of Herbicides on *Fusarium solani*..... 99

Chlamydospore Germination..... 99

Discussion..... 100

Literature Cited..... 106

Page

CHAPTER 5: SUMMARY AND CONCLUSIONS..... 109

APPENDICES..... 113

Appendix A..... 113

Appendix B..... 114

LIST OF REFERENCES..... 136

LIST OF TABLES

Page

CHAPTER 2

1. Effect of EPTC and moisture levels on the dry weight of 25-day-old navy bean plants..... 32

2. Effect of EPTC and planting depth on the dry weight of 25-day-old navy bean plants..... 34

3. Effect of EPTC and mechanical soil compaction on the dry weight of 25-day-old navy bean..... 34

4. The effect of temperature and EPTC on the dry weight of 25-day-old navy bean plants..... 35

5. Effect of EPTC and nitrogen on the fresh and dry weight per plant of 25-day-old navy bean..... 36

6. Effect of damaged seed and EPTC on plant height and yield of navy bean, East Lansing, Michigan, 1972, 1973..... 38

7. Effect of EPTC, damaged seed and planting depth, on the dry weight of 20-day old navy bean plants. 39

CHAPTER 3

1. Effects of EPTC and atrazine on fresh and dry weight of 35-day-old 'Sanilac' navy bean plants and final grain yield..... 50

2. Effects of EPTC and atrazine on bud, flower and pod formation of 'Sanilac' navy bean planted June 15, 1972..... 52

3. Effects of EPTC and atrazine on bud, flower and pod formation on navy bean planted June 15, 1973. 53

4. The effect of EPTC and temperature pretreatment on the quantity of water loss from 15-day-old navy bean plants at 30 C for 48 hr..... 58

Page

5. Effect of EPTC on the chloroform-soluble fraction from the surface of navy bean leaves..... 61

6. The effect of EPTC pretreatment for 16 days on the uptake of ^{14}C-atrazine by navy bean plants after 15 hours at 30 C..... 63

CHAPTER 4

1. Disease ratings and yields of navy bean grown in Fusarium solani natural and artificially infested soil..... 80

2. Navy bean growth in soil infested with Fusarium solani and treated with several herbicides..... 83

3. Growth of navy bean in soil infested or non-infested with Fusarium solani and treated with EPTC at three temperatures..... 86

4. Effect of Fusarium solani inoculum level and EPTC on navy bean growth..... 87

5. Growth of 'Sanilac' (susceptible) and 'RRR041' (moderately susceptible) navy bean in soils treated with EPTC and infested with Fusarium solani..... 89

6. The virulence on navy bean of Fusarium solani chlamydospores grown on potato dextrose agar containing several herbicides..... 90

7. The effect of navy bean pretreatment with several herbicides for 10 days on the subsequent root rot development after 30 days..... 91

8. Total carbohydrates exuded from navy bean seeds treated with several herbicides at 10^{-6} M..... 97

9. Efflux of amino acids, total carbohydrates and electrolytes from 6-day-old navy bean roots and hypocotyls grown in the dark at 26 C then placed in herbicide solutions for 5 hours..... 98

10. Fusarium solani growth in potato dextrose broth containing several herbicides..... 100

11. Percent chlamydospore germination in soil treated with several herbicides..... 103

Page

APPENDIX B.

B-1. Total protein content of navy bean seeds treated with EPTC and atrazine - 1972..... 114

B-2. Efflux of electrolytes from navy bean root and hypocotyl tissue treated with several herbicides. 115

B-3. Efflux of electrolytes from navy bean root and hypocotyl tissue pretreated with several herbicides..... 116

B-4. Efflux of amino acids and total carbohydrates from navy bean roots and hypocotyl tissue treated with several herbicides..... 117

B-5. Effect of EPTC on Fusarium solani chlamydospore germination in the spermosphere of navy bean after 48 hours in soil at 50% moisture..... 118

B-6. The effect of EPTC and Zn levels on navy bean yields..... 119

LIST OF FIGURES

Page

CHAPTER 3

1. Navy bean plants with wind blast injury. Untreated plants (A) and plants treated with 3.36 kg/ha of EPTC (B)..... 56

2. Electron micrographs of navy bean leaves at 1000 X. Leaf (A) is from a plant grown in Hoagland's solution. Leaf (B) is from a plant grown in Hoagland's solution with EPTC at 10^{-6} M..... 60

CHAPTER 4

1. Root rot rating system - 1972-1973..... 74

2. Predisposition of navy bean hypocotyl tissue to Fusarium solani. Plant (A) received no herbicide treatment prior to exposure to Fusarium solani chlamydospores. Plants (B) and (C) were pretreated with EPTC at 10^{-5} and 10^{-6} M, respectively. The hypocotyls of plants (D) were treated with acetone prior to exposure to the chlamydospores..... 93

3. EPTC affect on navy bean hypocotyl surfaces. Plant (A) was treated with 10^{-6} M EPTC for 12 days and plant (B) 12-days-old without a herbicide treatment. Plant (C) was treated with EPTC at 10^{-6} M for 20 days and (D) received no herbicide treatment..... 96

4. Fusarium solani hyphal development in potato dextrose broth containing herbicides. Hyphal development in check (A), EPTC 10^{-5} M (B), chloramben 10^{-5} M (C), and dinoseb 10^{-5} M (D) at 480 X..... 102

Page

APPENDIX B

B-1. EPTC-atrazine interaction at East Lansing, Michigan, 1972. Treatment (A) EPTC at 3.36 kg/ha (B) atrazine 0.56 kg/ha and (C) EPTC and atrazine at 3.36 + 0.56 kg/ha..... 121

B-2. EPTC-root rot interaction, 1972. Plants (A) were grown in soil with a natural root rot population. Plants (B) were grown in soil with a natural root rot population and treated with EPTC at 3.36 kg/ha..... 123

B-3. EPTC-root rot interaction, 1972. Plants (A) grown in soil with a natural root rot population. Plants (B) were grown in soil with a natural root rot population and treated with 3.36 kg/ha of EPTC..... 125

B-4. EPTC-Fusarium solani interaction in a growth chamber study at 20, 25 and 30 C. At each temperature A, C, E and G plants were grown in non-infested soil with EPTC at 0, 2.24, 3.36 and 4.48 kg/ha. Plants B, D, F and H were grown in soil infested with Fusarium solani and treated with 0, 2.24, 3.36 and 4.48 kg/ha of EPTC, respectively..... 127

B-5. Navy bean growth in non-infested soil or soil infested with Fusarium solani and treated with several herbicides. Plants were treated with (A) EPTC at 3.36 kg/ha (B) alachlor at 2.80 kg/ha (C) trifluralin at 0.84 kg/ha and (D) dinoseb at 5.04 kg/ha..... 129

B-6. Navy bean growth in non-infested soil or soil infested with Fusarium solani and treated with two herbicides. Plants were treated with (A) EPTC + trifluralin and (B) chloramben..... 131

B-7. Electron micrographs of 18-day-old navy bean hypocotyls treated with (A) chloramben at 10^{-5} M and trifluralin at 10^{-5} M..... 133

B-8. Electron micrograph of 30-day-old navy bean hypocotyls at 5000 X. Plant (A) was grown in EPTC at 10^{-6} M while plant (B) received no herbicide treatment..... 135

INTRODUCTION

Navy bean production in Michigan is not a recent phenomenon. Farmers in the Michigan territory sold navy beans to Commodore Perry on Lake Erie in 1812 (120, 142). The Thumb district and Saginaw Valley of Michigan, have in the past, and even today, produce nearly all the white-pea-bean (navy bean) crop. This area is characterized by fertile Brookston soils. These are loams and clay loams with a dark-colored plow layer underlain by wet, mottled, gritty clay to a depth of several feet. They have a high pH and are high in organic matter and retention of moisture. Climatically these areas tend to provide a cool and humid growing season.

During the last 10 years, average yields of navy bean have decreased. In 1963, the projected state yield for 1980 was set at 1800 lb/a (2016 kg/ha). The 4-year average navy bean yield between 1960 to 1963 was 1320 lb/a (1478 kg/ha), however, the 1970 to 1973 4-year average was 1050 lb/a (1176 kg/ha) (103).

Navy bean is reported sensitive to many environmental factors and cultural practices, which result in production problems, such as, insects, diseases, herbicide injury, physiological and mechanical disorders and nutrient deficiency (33). All of these problems have contributed to the declining

navy bean yields. It is of utmost importance that each of the contributing environmental and cultural factors be studied and evaluated in terms of a systems approach, wherein the interaction of production components are studied. Until now this has been ignored. Thus, the possible interactions between component factors of navy bean and dry bean production have not been identified or studied.

One of the major underlying causes of poor navy bean yields may be poor soil tilth, resulting from the navy bean-sugar beet rotation. This rotation returns very little plant material to the soil. Poor rotations, extensive seed bed preparation and subsequent between the row tillage may all have contributed to the poor growing conditions. Tradition and herbicide incorporation has dictated repeated tillage of the land prior to planting. This extensive tillage of the heavy soil with high moisture retention may result in compacted soil with poor structure. Soil compaction may contribute to many other problems, such as increased disease development.

In 1962, less than 1% of the one-half million acres (.2 million ha) of navy bean grown in Michigan were treated with herbicides (61). However, in 1970, more than 90% of the navy bean acreage was treated with at least one herbicide (101). The major herbicide used for weed control in navy bean during the last 10 years has been EPTC. Along with the alterations in soil structure, tillage equipment, and cultural practices, a new factor, herbicides, has been added to navy bean production.

The objective of this study was to determine the possible interactions of herbicides with a number of management factors in navy bean production. This thesis studied the effect of temperature, soil compaction, seed quality, fertility, soil moisture, planting depth, herbicide residue and disease, on navy bean tolerance to EPTC.

CHAPTER 1

LITERATURE REVIEW

In 1962, when only 1% of the one-half million acres of navy beans grown in Michigan were treated with herbicides, several herbicides were reported for use in Michigan (61). Up to this time none of the herbicides used were entirely satisfactory. Injury to the crop, poor weed control, high cost, and lack of federal clearance for use on navy bean was a major problem. Navy bean was shown to be tolerant to EPTC in Michigan (61) and other reports showed it useful on other types of beans in other areas (39, 97). Further work in Michigan and other locations showed the need for incorporation of EPTC to limit volatilization losses from the soil for complete weed control (8, 86, 101, 112, 125).

EPTC volatility and incorporation. EPTC has a high vapor pressure, and is readily volatilized from the soil and must be incorporated to accomplish weed control. Kanke et. al. (86) found that incorporation of EPTC to a depth of 2.54 to 5.08 cm gave greater weed control. They also showed that under dry conditions incorporation was necessary to obtain weed control. However, if the application of EPTC was followed by rainfall, there was no need for incorporation.

Fang et. al. (56) found the loss of ^{35}S-EPTC from drying of a moist soil was related to the amount of water being vaporized and also the soil properties. The higher the organic matter and clay content, the smaller the EPTC loss due to evaporation of soil water. Gray and Weierich (66) showed that the deeper EPTC was incorporated into moist soil the greater the EPTC retention. Incorporation to 1.27 cm was better than surface treatments in plots moistened by rain. Much less EPTC loss occurred when the soil dried and stayed dry, than if kept moist by rain. They showed that sprinkling with one inch of water in 3 days caused large losses of EPTC except at incorporation depths of 7.6 cm. Gary and Weierich (66) showed that EPTC loss was greatest in all cases during the first 15 minutes. In wet soil they found considerable loss of EPTC occurring in incorporated treatments. When not incorporated, about 90% of the applied EPTC was lost in 24 hours from sand and 75% from clay. Gray and Weierich (67) reported that in a loamy sand after application of 8 inches of water, the EPTC was completely leached out of the top 7.6 cm of soil. The depth of leaching decreased as the amount of clay content and organic matter increased. Koren et. al. (87) along with Mortland and Meggitt (106) showed that adsorption of EPTC is related to the chemical properties of the herbicide. The nature of the functional groups played a very important role in the adsorption of EPTC. Ashton and Sheets (9) found an inverse relationship between injury to oats Avena sativa L. and soil adsorption of EPTC in various soil types.

Fang et. al. (56) found that at 0 to 3 C radioactive EPTC in the soil remained unchanged from the original activity, concluding that little vaporization occurred at low temperatures. At 25 to 30 C they found that the radioactivity level dropped slightly in the first few days and continued to decrease with time. There was some upward movement of ^{35}S-EPTC from the lower soil layer, resulting in some accumulation of ^{35}S-EPTC at the soil surface. Gray and Weierich (66) reported that increasing the temperature from 9 to 15 C caused an increase in the vapor loss of EPTC from moist soil, but not from dry soil.

EPTC symptoms. Dawson (38) showed that EPTC causes kinking of the first internode of barnyardgrass (Echinochloa crusgalli (L.) Beauv.) resulting in a zig-zag growth pattern. The leaves remained rolled. He concluded that the major effect of EPTC was on leaf development. At high levels of EPTC treatment, no leaf development was observed beyond the coleoptile. Tissue studies by Dawson showed that barnyardgrass seedlings treated with EPTC had altered less mesophyl with reduced intercellular spaces.

Gentner (65) found that EPTC inhibited the deposition of epicuticular wax on cabbage Brassica oleraces L. This inhibition was correlated directly with the rate of EPTC application and only the leaves in the bud at the time of application were affected. He also found that EPTC in the gaseous phase was effective in reducing foliage wax formation. There was a correlation between the transpiration rate of the affected

leaves and EPTC treatment. As the EPTC rate increased, the transpiration rate of the affected leaves increased. Still et. al. (139) has found that EPTC treatment caused a significant reduction of wax formation on both leaf surfaces of pea (Pisum sativum L.) leaves. Wilkinson and Hardcastle (152) reported that petiole cuticle thickness of sicklepod was decreased 25% by EPTC at 4.48 kg/ha.

EPTC metabolism. Nalawaja et. al. (108) reported that ^{14}C-EPTC was degraded to CO_2 in plants and was then incorporated into a number of naturally occurring constituents such as fructose, glucose and amino acids, via photosynthesis.

Ashton (7) found that EPTC did not affect photosynthesis but did increase the respiration of excised embryos of maize (Zea mays L.) and mung bean when data was expressed on fresh weight basis, but when calculated on a per embryo basis, the stimulation was not evident.

Mann et. al. (95) reported that EPTC inhibited ^{14}C-l-leucine incorporation into protein by segments of barley (Hordeum vulgare L.) coleoptile. Moreland et. al. (104) showed that in soybean [Glycine max (L.) Merr.] hypocotyl sections, EPTC at 6×10^{-4} M inhibited protein synthesis by 24%.

Moreland et. al. (104) reported that the gibberellic acid-induced de novo α-amylase synthesis in barley half-seeds was inhibited 39% by 6×10^{-4} M EPTC.

Beste and Schreiber (14) showed that EPTC inhibited growth and RNA synthesis in soybean tissue. The combination of 2,4-D with EPTC was antagonistic and caused an increase in total

RNA synthesis.

Temperature. Fang et. al. (56) showed that temperature played a role in the amount of EPTC that remained in the soil after treatment. Little vaporization occurred at lower temperatures leaving a higher concentration in the soil. Applications to a cool soil maintained a larger quantity of EPTC. Waldrep and Freeman (149) related the length of exposure to EPTC to injury since at lower temperatures, plants were exposed to the herbicide for a longer period of time. Curless (37) working with butylate (S-ethyldiisobutylthiocarbamate) showed that there was more injury to corn at higher temperatures when 3.36, 5.04 and 6.72 kg/ha were applied. The number of injured plants increased as the temperature and herbicide rate increased. At lower temperatures germination was delayed allowing for dilution of the herbicide by adsorption to organic matter and soil clay colloids. At the higher temperatures germination was more rapid and plant emergence took place while the herbicide concentration in soil was still high.

Wright and Rieck (153) have reported that certain corn hybrids may be injured more with butylate at higher temperatures while others show more injury at lower temperatures.

The phytotoxicity of alachlor [2-chloro-2',6'-diethyl-N-(methoxymethyl)acetanilide] to corn has been reported to be greater at higher 32/20 C day-night temperatures (44). However, alachlor is more toxic to navy bean at 20 C than at 30 C (114).

The phytotoxicity of atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine] and linuron [3-(3,4-dichlorophenyl)-1-methoxy-1-methylurea] is greater within the range of 25 to 30 C. However, other reports indicate that corn may accumulate larger quantities of atrazine under cold, wet conditions, which resulted in injury (143). Trifluralin [α,α,α-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine] has been shown to accumulate at lower temperatures resulting in increased crop injury (74).

EPTC placement, uptake, and planting depth. Yamaguchi (154) reported that EPTC applied to roots was absorbed and distributed apoplastically throughout the plant. Prendville et. al. (117) and Oliver et. al. (111) showed that EPTC entered both roots and shoots and movement was both basipetal and acropetal.

Parker (112) showed that the characteristic toxic effects of the thiocarbamates were dependent upon shoot uptake prior to emergence from the soil. Dawson (38) reported that exposing the seeds and primary root of barnyardgrass to soil treated with EPTC did not result in injury, while only exposing the shoot caused severe injury. Appleby et. al. (5) found that EPTC was very toxic to oat through coleoptile uptake, but exposing the roots or seed caused little or no injury. Several other researchers have shown that shoot exposure was important as the site of EPTC uptake (86, 117).

Oliver et. al. (111) reported that the roots of barley are the major sites of EPTC uptake. In another report (117)

EPTC moved into the roots of corn and inhibited root growth while the shoots did not show reduced growth. The work by Prendville et. al. (118) and Oliver et. al. (111) are somewhat contradictory in that the former found wheat Triticum aestivum L. and barley severely injured when treated at the coleoptile internode, while the latter found the application of EPTC to the root zone caused the greater injury. Prendville et. al. (118) explained that the growth responses of species to EPTC applied to the shoots was dependent on the stage of plant development at which treatment occurred. Oliver et. al. (111) suggested that differential responses to EPTC by several species was the location of the intercalary growth. It occurred at the first internode in oat, at the second in corn and at the third in wheat and barley.

Gray and Weierich (68) using a charcoal barrier to allow separate exposure of seed, roots and shoots, found that in most species, root exposure caused more injury than shoot exposure, although, the shoot was the uptake site for some grasses. Corn showed almost equal injury from shoot and root exposure, whereas, wheat and barley were more severely injured by root exposure.

Planting depth can place a stress on germinating seedlings. Seedling vigor depended upon the quantity of reserve food supply and length of time for emergence (145). Field and growth chamber studies have indicated that the potential for butylate injury increased with increasing planting depth regardless of corn hybrid (153). However, in another test

shallow planting of corn in soil treated with alachlor resulted in greater injury (44).

Soil compaction. The crop rotations in the navy bean producing region of Michigan have changed from the use of legume plants in the rotation to a consistent sugar beet-navy bean rotation. This rotation adds very little organic matter to the soil, resulting in poor soil structure (25).

The average weight of wheel-type tractors tested in 1948 was less than 6,000 pounds (3.05 metric ton). However, in 1968, the average tractor weight was over 10,000 pounds (5.08 metric ton) (99), which has added to the soil compaction problem.

A compacted soil retains water longer (145) and has poor aeration (53). Navy bean germinates very poorly in cold, damp soil, and these seedlings are then subject to disease infection (120, 142).

Cook (32) grew navy bean in a 3-year rotation of oats, wheat and navy bean. During the first 3 years of the experiment, the beans averaged 19.8 cwt. per acre. In the second 3-year period the yields averaged significantly less, only 15.8 cwt. per acre (1770 kg/ha). During the third year period, the yields averaged only 12.9 cwt. per acre. He suggested that this rotation resulted in a deterioration of soil structure with time causing the observed yield reductions.

Erickson (52) reported an oxygen deficiency on all plots in an experiment evaluating various tillage systems in heavy soil at Saginaw, Michigan, 1973. This indicated the importance

of good soil tilth and its influence on the soil-root environment. Tillage practices which resulted in the best soil aeration and the least soil compaction gave the greatest yields. Plots compacted with a tractor and dragged to 3 inches deep, produced 8.4 cwt. per acre (940 kg/ha). The greatest yield was obtained following tillage with a spring chiseling 4 to 6 inches deep and a spring-tooth drag, yielding 18.6 cwt. per acre (2084 kg/ha) (26).

Robertson (124) showed that the highest yields of sugar beet were obtained on plots where legumes had helped to maintain and build soil structure. In compacted soil (72) there are four possible factors to which the change can be attributed, 1) a compression of the solid particles, 2) a compression of the liquid and gas within the spore spaces, 3) a change in the liquid and gas contents and 4) rearrangement of soil particles.

Another area closely related to soil compaction is the development of high-strength crusts. McIntyre (98) described a likely sequence of events for soil crust formation under field conditions as (a) breakdown of soil aggregates, (b) movement of fine particles into the upper few centimeters of soil and (c) compaction of the soil surface to form a thin film which restricts water entry. Under drying, rigidity of the rearranged particle depended, among other things, upon the area of contact between soil particles and upon the forces between the particles.

Crusted soil conditions that develop due to rainfall or mechanical compaction are a serious hazard to seedling emergence

and the establishment of crop stand for navy bean and sugar beet (2, 50, 55, 63, 122). Carns (20) showed that cotton Gossypium hirsutum L. emergence was reduced as crust strength was increased.

Pollack et. al. (115) showed that the formation of a soil crust tended to diminish the injury that fluorodifen [p-nitrophenyl α,α,α-trifluoro-2-nitro-p-toly ether] causes to navy bean seedlings. As the navy bean emerged, the preemergence herbicide was pushed aside by breaking the crust of the soil surface.

Damaged seed. Herter (73) and Whitney (151) in 1930, reported injury to navy bean resulting from the mechanical threshing operation. They reported the most severe injury resulted under conditions of low humidity.

Damage to seed is classified into two major groups (79) (a) external or visible damage and (b) internal damage that becomes evident only after imbibition and germination of the seed. The external damage is characterized by a cracked seed coat or splitting of the seed. Two important types of abnormalities resulting from internal damage are 'baldhead' and 'snakehead' seedlings (11, 12). A baldhead seedling lacks the epicotyl or apical growing point and forms no primary leaves. Snakehead plants retain the growing point but lack primary leaves.

Plants which develop from 'baldhead' and 'snakehead' seedlings are spindly, slower growing, later maturing and lower yielding (11, 36, 73, 110).

Barriga-Solorio (12) reported a possible vigor reduction in normal navy bean seedlings that he attributed to "invisible injury...that may have had some effect in the physiological processes of the seed".

Reviews of seedling vigor have been made by Delouche and Caldwell (43) and Isely (78, 79). Seed vigor concerns the performance under unfavorable environmental conditions of temperature, moisture and pathogens; or vigor per se reflected in germination and growth rates.

Schweitzer (133) showed reduction in vigor and seedling elongation of a normal appearing seedling produced from damaged navy bean seeds.

Nutrient level - herbicide interaction. There has been only limited work done on the influence of nutrients on herbicide uptake and phytotoxicity. Crafts in 1939 (34) studied the effects of several levels of Hoagland's nutrient solutions on the toxic effects of borax, arsenate, and chlorate. Chlorate was less injurious to oat as the strength of the nutrient solution increased while borax and arsenate phytotoxicity were not affected.

Bingham and Upchurch (15) observed a highly significant interaction between phosphorus and diuron [3-(3,4-dichlorophenyl)-1,1-dimethylurea]. Diuron at 2 ppm reduced the fresh weight of cotton from 84% to 3% as the phosphorus level of a sandy loam soil increased. Soil nitrogen, potassium and pH levels had little influence on the effect of diuron on plant growth. In a subsequent study he found that amitrole

was apparently more phytotoxic at high phosphorus levels.

McReynolds and Tweedy (100) reported that twice as much ^{14}C-simazine [2-chloro-4,6-bis(ethylamino)s-triazine] was translocated to the shoots of corn, rye (Secale cereale L.) and soybean grown in nitrate than in ammonium nitrogen.

Doll (49) showed that the toxicity of chloramben (3-amino-2,5-dichlorobenzoic acid), atrazine, and linuron increased as the NO_3^- level of the nutrient solution increased. However, the increased toxicity of chloramben, atrazine and linuron at higher NO_3^- rates was not due to increased chloramben uptake.

Herbicide-herbicide interactions. The present trend in weed control is to use herbicide combinations to increase the spectrum of weed control. Therefore, more than one compound is applied to a crop the same year, either as a tank mix or a preemergence followed by a postemergence treatment. Herbicide residue in the soil from the previous year also creates the potential for the interaction of this residue with the herbicide applied in the current year. These conditions then provide the potential for an interaction between chemicals, resulting in responses not predictable from each chemical alone (111).

Tammes (140) used the following terms to describe responses obtained with combinations of compounds:

Synergism - Cooperative action of two components of a mixture such that the total effect is greater or more prolonged than the sum of the effects of the two taken

independently.

Addition - Cooperative action, such that the total effect is equal to the sum of the effects of the components taken independently.

Independent effect - The total effect is equal to the effect of the most active component alone.

Antagonism - The total effect is smaller than the effect of the most active component alone.

There are several mathematical methods used to calculate expected responses of pesticide combinations (35, 57, 69, 91). The Colby (31) method is extensively used by herbicide researchers as a method for predicting responses to herbicide combinations. The Colby formula, $E_1 = \frac{X_1 \ Y_1}{100}$ where E_1 represents the expected response as a percent of control, and X_1 and Y_1 are the responses as percent of control obtained from applications of chemicals A + B singularly. If the observed value is less than the expected value, synergism is indicated, if they are equal, the response is additive and when the observed value exceeds the expected value, antagonism is indicated.

Many of the reports of synergistic herbicide responses involve the postemergence application of two or more compounds (18, 40, 88, 105, 136). Moore and Cheetham (105) found that 2,4-D [2,4-dichlorophenoxy acetic acid] and chloropropham (isopropyl m-chlorocarbanilate) acted in a synergistic manner on wild garlic. A pretreatment of wild garlic with chloropropham increased 2,4-D toxicity.

Binning et. al. (16) showed that ethephon (2-chlorethyl phosphonic acid) as a pretreatment increased the transport of dicamba (3,6-dichloro-o-anisic acid) to the basal bulbs of wild garlic. Devlin and Yaklich (45) showed that bean plants pretreated with gibberellin (GA) will absorb more naptalam (N-1-naphthylphthalamic acid) than untreated plants.

Dickerson and Sweet (48) reported synergism with the addition of 34.5 ml of 2,4-D to 1.12 kg of atrazine and 9.35 l of crop oil per ha on nutsedge. Lynch (92) also found that low rates of 2,4-D and atrazine on bean leaves resulted in a synergistic response.

Herbicides have been shown to predispose plants to increased toxicity from postemergence treatments applied at a later time. Gentner (65) showed that the application of dinoseb (2-sec-butyl-4,6-dinitrophenol) on cabbage leaves previously treated with EPTC increased toxicity. Dewey et. al. (46) and Preiffer et. al. (116) reported that TCA (trichloroacetic acid), dalapon (2,2-dichloropropionic acid), and dinoseb increased the toxicity of other herbicides applied to peas. Juniper (83), concluded that changes in susceptibility of crops after application of these herbicides was due to a reduction in external leaf waxes. Davis and Dusbabek (42) showed that exposing pea plants to diallate [S-(2,3-dichloroallyl)diisopropylthiocarbamate] vapors increased the subsequent foliar uptake of ^{14}C-2,4-D, atrazine, TCA and diquat [6,7-dihydrodipyrido(1,2-α:2',1'-c)pyrazinediium ion], which they attributed to a reduction of epicuticular waxes on

the leaves of the treated plants.

Dexter (47) showed increased phytotoxicity of phenmedipham (methyl m-hydroxycarbanilate m-methylcarbanilate) or pyrazon (5-amino-4-chloro-2-phenyl-2(2H)-pyridazinone) plus dalapon when applied postemergence after preplant EPTC or preemergence TCA treatments.

Duke et. al. (51) found increased injury to alfalfa in a greenhouse study following application to soils containing low levels of atrazine residues. The injury resembled an additional dose of atrazine and they concluded that it may have been the result of decreased atrazine detoxification.

Johnson (80, 81, 82), showed that sequential applications of chloroxuron (3-[p-(p-chlorophenoxy)phenyl]-1,1-dimethylurea) applied as a semidirected postemergence treatment following preplant treatments with nitralin, [4-(methylsulfonyl)-2,6-dinitro-N,N-dipropylaniline] resulted in synergism reducing soybean yields. Postemergence treatments of prometryne or chloroxuron following preplant applications of vernolate (S-propyl dipropylthiocarbamate) were also found to be more toxic to soybean.

Navy bean root rot development. Pathogens responsible for the development of root rot have been reported in all areas wherever navy beans are grown. Fusarium solani (Mart.) Appel and Wr. f. phaseoli (Burk.) Snyd. and Hans. is a primary pathogen in the "bean root rot complex". Rhizoctonia solani Kuhn and Thielaviopsis basicola Zoph. are of secondary importance. Fusarium, Rhizoctonia, and Thielaviopsis are capable of direct

penetration or entrance through natural openings or wounds (25, 27, 28, 29). These organisms are indigenous to the soil, and survive as soil saprophytes in the absence of a suitable host.

Root rot caused by Fusarium solani f. phaseoli appears as a reddish discoloration on the hypocotyl and root, extending to the soil line (19, 155). The plants are stunted, the leaves may turn yellow and the lower parts of the stem become pithy and dry. Often the secondary roots that develop from the taproot are killed. In that case a cluster of roots may develop above the lesion, a little below the soil line and if weather conditions are favorable an almost normal crop yield may be produced. Maloy and Buckholder (93) found that wheat grown immediately preceding navy bean significantly reduced root rot severity, however, relatively low levels of F. solani may result in high incidence of infection. Huber (77) showed in a Michigan study that corn preceding navy bean in the rotation resulted in the greatest reduction of root rot and in a significant increase in yield. The most severe root rot infection was observed when navy bean followed barley in the rotation. Maloy (94) showed that the mycelial form rather than the conidial form of Fusarium solani f. phaseoli predominated in the soil, in the prolonged absence of the host plant. Schroth and Hendrix (132) suggested that the survival of F. solani f. phaseoli in agricultural land is enhanced by diffusates from nonsusceptible plants and crop residues which enable the fungus to vegetate briefly and form new

chlamydospores. Maloy et. al. suggested that there could be a reduction in disease severity without an alteration in the pathogen population. This could be accomplished by a change to a less virulent mycelial type from a more virulent conidial type (70). Nash et. al. (109) reported that in natural soil Fusarium solani f. phaseoli exists chiefly in the form of chlamydospores. Macroconidia placed in soil either germinated into a short germ-tube which eventually produced chlamydospores or were converted directly into chlamydospores. Schroth and Snyder (131) showed that germination of chlamydospores were stimulated by "root exudates". Taussoun et. al. (144) showed that glucose was essential for the penetration stage of infection of navy bean hypocotyl by macroconidia of Fusarium solani f. phaseoli but glucose without a nitrogen source delayed host penetration and the onset of pathogenesis. Nitrogenous compounds favor early penetration and pathogenesis with the organic forms being more effective than the inorganic forms.

Maier (96) showed that Fusarium solani resulted in the most rapid disease development at 24 C with the most rapid development during the first week and the greatest severity occurring 3 weeks after emergence.

Herbicide - disease interactions. From an extensive review by Katan and Eshel (84) of the interactions between herbicide and plant pathogens, it is apparent that a number of herbicides interact with various pathogens to either increase, decrease or have no effect on disease development.

There are many reports of damping-off diseases, such as, Rhizoctonia solani interacting with herbicides to cause increased disease severity. Trifluralin has been shown to alter the tolerance of cotton to Rhizoctonia solani (4). The increase in Rhizoctonia disease in cotton has been shown to be erratic and environment dependent (4, 138). The growth of cotton was reduced by the combination of trifluralin or prometryne [2,4-bis(isopropylamino)-6-(methylthio)-s-triazine] and Rhizoctonia solani (21). Fusarium oxysporum has been shown to interact with several herbicides to increase disease severity on a number of plant species (22, 107, 148).

Studies on the direct effect of herbicides on pathogen growth and development show that herbicides may either increase or decrease hyphal production, or have no effect on the pathogen. The growth or reproduction of Fusarium sp. (23, 107, 113, 128, 135) Rhizoctonia solani (3, 13) and Sclerotium rolfsii (113, 128, 129, 130) have been shown to increase when grown in herbicide cultures or sterilized soil. Many of the reports show the growth to be concentration dependent. With increasing concentrations most herbicides inhibited growth or reproduction of the pathogens, while at low concentrations, growth was stimulated (113, 121, 126, 137). Tang (141) has shown that trifluralin increased germination and chlamydospore production in soil of Fusarium oxysporum f. sp. vasinfection. EPTC has been shown to increase mycelial production of Fusarium oxysporum and Sclerotium rolfsii (113, 129).

There are many reports of herbicides decreasing *in vitro* growth of pathogens (123, 127, 129) and reducing plant diseases. Dinoseb decreased disease incidence caused by *Fusarium oxysporum*, (123) *Cercospora arachidicola* (22), *Puccinia menthae* (75), and *Sclerotinia rolfsii* (22, 64) on several crops. The phenoxy herbicides are generally only slightly toxic to pathogens (30, 54, 90, 134). 2,4-D has been shown to reduce *Fusarium* wilt of tomato due to alterations in metabolism which regulate the growth of the parasite or formation of toxins (41).

Herbicides may also influence pathogen virulence. Weinhold *et. al.* (150) suggested that even though a herbicide did not alter growth there could be an alteration in metabolism which could increase pathogen aggressiveness. Hsia and Christensen (76) showed that 2,4-D increased *Helminthosporum sativum* pathogenicity.

Herbicides may also have a direct effect upon the host plant, which makes it more susceptible to disease organisms. The increase in American gooseberry mildew of black currant treated with simazine has been attributed to a rise in nitrogen content of the host due to simazine (147). Altman (3) showed that in sugar beet the amount of glucose exudate at the soil-plant interface was greater when plants were grown in herbicide treated soil, and concluded that this was the reason for their increased susceptibility to *Rhizoctonia*. Lai and Semeniuk (89) suggested that picloram (4-amino-3,5,6-trichloropicolinic acid) enhanced carbohydrate exudation which resulted in increased root rot of corn.

There are many reports indicating a natural biological balance between pathogens and saprophytic organisms (28, 62). There are several possible mechanisms of antagonism that may exist in soil, (1, 85) competition for limited quantities of nutrients, oxygen, space (4), the release of toxic products which inhibits pathogen growth, and (138) direct parasitism or predation. Many reports show that the application of herbicides at field rates do not greatly alter gross numbers of soil microflora (10, 17, 59, 60). However, a total number count does not reveal changes in specific species of organisms which are required for biological control of plant diseases (58). Soil pathogens invade roots through a rhizosphere in which there is a pronounced microbial activity. Foliar and soil applications of herbicides which lead to changes in microbial composition of the rhizosphere may well affect the inoculum level of the pathogen and consequently, disease incidence (89).

CHAPTER 2

FACTORS AFFECTING EPTC INJURY TO NAVY BEANS

Abstract

Injury to navy bean (*Phaseolus vulgaris* L.) from EPTC (*S*-ethyl dipropylthiocarbamate) was investigated as a function of soil moisture, planting depth, soil compaction, temperature, available nitrogen and seed quality. Greenhouse and growth chamber experiments showed that both high and low moisture stress increased navy bean injury. Deep planting increased the injury of navy bean from high rates of EPTC. Soil compaction in combination with EPTC greatly reduced navy bean growth. Ambient temperature did not alter the susceptibility of navy beans to EPTC. Stimulation of navy bean growth by nitrogen as measured by increased fresh weight and inhibited by EPTC. Field and greenhouse studies showed that EPTC injury was greater to mechanically damaged seed than non-damaged seed.

Introduction

Navy bean is very sensitive to environmental and cultural stress conditions, such as soil compaction, soil moisture stress and high or low temperatures. Each of these factors can greatly reduce navy bean vigor and ability to survive through the growing season. Plant tolerance to herbicides may be altered when grown under stress conditions resulting in reduced vigor (3).

The influence of simulated rainfall and soil moisture on herbicide action has received attention primarily under non-agricultural conditions (4, 11). The toxicity of diuron [3-(3,4-dichlorophenyl)-1,1-dimethylurea] has been shown to be greater under moist soil conditions than under dry soil conditions (22). Soil moisture plays a large role in the effectiveness of EPTC. Under high moisture conditions there is extensive vapor loss resulting in poor weed control (10, 13, 14).

Planting depth can place stress on germinating seedlings as seeds planted too deep may be less vigorous at emergence. Field and growth chamber studies have indicated that the potential for butylate (S-ethyl diisobutylthiocarbamate) injury increased with increasing planting depth regardless of corn (Zea mays L.) hybrid (24). However, in soil treated with alachlor [2-chloro-2',6'-diethyl-N-(methoxymethyl)acetanilide] shallow planting of corn resulted in the greatest amount of injury[1].

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[1]Devisetty, B. N. and R. G. Harvey. 1974. Depth of planting as a factor influencing corn injury from alachlor. Weed Sci. Soc. Amer. Abstr. No. 17.

Crusted soil conditions that develop due to rainfall or mechanical compaction are a serious hazard to seedling emergence and the establishment of a stand of many crops such as navy bean and sugar beet (Beta vulgaris L.) (2, 7, 9, 12, 19). Carns (5) showed that cotton (Gossypium hirsutum L.) emergence was reduced as crust strength was increased. Pollack[2] showed that the formation of a soil crust tended to diminish the injury that fluorodifen (p-nitrophenyl α,α,α-trifluoro-2-nitro-p-tolyl ether) caused to navy bean seedlings. As navy beans emerged the preemergence herbicide was pushed aside by breaking the crust of the soil surface.

Temperature has been shown to alter the phytotoxicity of several herbicides. The phytotoxicity of alachlor to corn has been reported to be greatest at higher 32 C/20 C day-night temperatures. Alachlor has also been shown to be more injurious to navy bean at 20 C than at 30 C (17). The phytotoxicity of atrazine [(2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine] and linuron [(3-(3,4-dichlorophenyl-1-methoxy-1-methylurea)] is greatest within the range of 25 to 30 C (15, 18). Under cold, wet conditions atrazine has been shown to accumulate and injure corn (20). Increased trifluralin (α,α,α-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine) injury at low temperature has been attributed to increased accumulation at the lower temperatures[3]. Certain corn hybrids may be injured more at higher temperatures,

[2]Pollack, T.,W. R. Furtick and G. Crabtree. 1973. Response of beans to fluorodifen under certain soil and light conditions. Weed Sci. Soc. Amer. Abstr. No. 39.

[3]Hawxby, K., E. Basler and P. W. Santelmann. 1971. Temperature effects on absorption and translocation of trifluralin in peanut seedlings. Weed Sci. Soc. Amer. Abstr. No. 48.

while others show more injury at lower temperatures to the same herbicide (24).

Soil fertility levels have also been shown to alter plant response to herbicides. Upchurch (21) studied 12 herbicides and found statistically significant herbicide-phosphate interactions with diuron, amitrole (3-amino-s-triazole), CDAA (N,N-diallyl-2-chloroacetamide), and simazine [2-chloro-4,6-bis (ethylamino)-s-triazine]. Adams (1) studied the effects of phosphorus and atrazine on the mineral composition of soybean [Glycine max (L.) Merr.]. He concluded that phosphorus appeared to increase the sensitivity of soybeans to atrazine. Doll[4] showed that the toxicity of chloramben (3-amino-2,5-dichlorobenzoic acid), atrazine and linuron to oat (Avena sativa L.) increased as the NO_3^- level was increased in nutrient solution.

Several[5] researchers have studied the performance of navy bean seedlings containing various degrees of cotyledon cracks and breaks (23). They found that damaged seed and loss of cotyledonary tissue resulted in smaller seedlings, fewer pods and lower yields per plant, and that the reductions were proportional to the amount of cotyledon lost.

The objectives of this study were to determine the effects of soil moisture, planting depth, mechanical soil compaction,

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[4]Doll, J. D. 1969. The influence of nutrient level and combination on herbicide uptake and phytotoxicity. Ph.D. Thesis. Mich. State Univ., E. Lansing. 83 p.

[5]Schweitzer, L. R. 1972. Reduction in seedling vigor and changes in metabolism during germination related to mechanical abuse of bean (Phaseolus vulgaris L.) seed. Ph.D. Thesis. Mich. State Univ., E. Lansing. 88 p.

temperature, nitrogen level, and seed quality, on navy bean tolerance to EPTC.

Materials and Methods

In greenhouse and growth chamber studies 'Sanilac' navy bean was grown in either 50 by 35 by 10 cm wooden flats or 946 ml cups placed in larger diameter containers that served as the water reservoir for subirrigation of the cups. The upper 5.0 cm of soil in either case was treated with EPTC in a separate flat or 946 ml container and thoroughly mixed. EPTC was applied to the soil at 935 l/ha and 2.11 kg/cm^2 pressure. The treated soil was placed on top of the untreated soil and seeds planted at 2.5 cm. A Conover sandy loam soil with 2.5% organic matter was used in all experiments. Experiments involving wooden flats had 75 seeds per replication while, experiments in 946 ml containers had 8 to 10 seeds. Greenhouse temperatures were maintained between 20 and 30 C. Natural illumination was supplemented with fluorescent light.

In a factorial greenhouse experiment to study the possible interaction of EPTC and soil moisture, EPTC at 2.24, 3.36 and 4.48 kg/ha was applied to a Conover sandy loam soil containing 15% moisture. Subsequent watering rates were 0.32, 0.64, 1.3 and 2.5 cm at planting and every 4 days following for a total of 16 days. After 16 days, water was applied equally by subirrigation to each treatment. After 25 days, the plants were harvested and dry weight recorded.

A greenhouse experiment to study the possible interaction between EPTC at 2.24, 3.36 and 4.48 kg/ha and the planting depths of 2.5, 5.0 and 7.6 cm was initiated. Seeds planted at 2.5 cm were in direct contact with the treated soil layer, allowing for direct exposure of the hypocotyl, seed and roots. Seeds planted at 5.0 and 7.6 cm were below the EPTC-treated soil layer restricting exposure primarily to the emerging hypocotyl. The flats were watered to maintain similar and adequate soil moisture for growth. After 25 days the plants were harvested and dry weight recorded.

The possible interaction between EPTC and soil compaction was studied in a greenhouse experiment. Soil was treated with EPTC at 2.24 and 3.36 kg/ha and seeds planted as previously described. Soil compaction was accomplished by placing a steel plate over soil contained in 50 by 35 by 10 cm wooden containers and applying pressures of 5.6 and 11.2 kg/cm^2 to the plate with a hand screwpress. Soil moisture in the compressed soil was 15% and further watering was applied uniformly as needed. Plant weight measurements were taken after 25 days.

The possible interaction of EPTC and temperature was investigated in a growth chamber study. Plants were grown in 946 ml containers with subirrigation. The rates of EPTC used were 2.24, 3.36 and 4.48 kg/ha in combination with temperatures of 20, 25 and 30 C constant day and night temperatures. Fluorescent and incandescent lighting provided 20.5 klux in a 16-hour photoperiod. At the end of 25 days the plant dry weights were recorded.

The possible interaction of nitrogen and EPTC was studied in a factorial greenhouse experiment. EPTC at 3.36 and 4.48 kg/ha was applied to soil containing 56.0, 112.1 and 224.2 kg/ha nitrogen, applied as ammonium nitrate (NH_4NO_3). The plants were grown in 946 ml containers with subirrigation. After 25 days, the fresh and dry weight of the plants was recorded.

One-half of the seed for the damaged seed experiment was given a mechanical impact treatment by dropping the seeds 9.2 M onto a steel plate inclined 45^{o}. The seed was maintained at 11$\pm$ 0.5% moisture for the impact treatment. The impacted seeds were carefully examined and all broken seeds and seeds with visual cracked seed coats were discarded. Only physically intact seeds, showing no external evidence of damage were used.

The effect of damaged seed on the response of navy bean to EPTC was studied in a field experiment at East Lansing, Michigan in 1972 and 1973. Normal and damaged 'Sanilac' navy bean seeds were planted 2.5 cm deep in soil treated with 3.36 and 4.48 kg/ha of EPTC applied to 3.0 m four row plots in 0.76 m row widths with a tractor mounted sprayer. EPTC was incorporated twice to a depth of 7.6 cm with a spring tooth harrow. Plant height and grain yields were recorded in both years.

The effect of damaged seed and planting depth on navy bean tolerance to EPTC was studied in the greenhouse. EPTC was applied at 3.36 kg/ha in combination with 2.5, 5.0 and 7.6 cm planting depths with damaged or normal seed.

All data presented with the exception of Table 6, are the means of two experiments with three replications each.

Results and Discussion

Soil Moisture. The soil moisture level greatly influenced navy bean dry weight development. The moisture levels of 1.3 and 2.5 cm per 4 days resulted in the best growing conditions and the 0.32 cm moisture level produced the least favorable growing conditions (Table 1). The least reduction in dry weight by EPTC from that of the no herbicide treatment, resulted at the 1.3 cm moisture level. At this level, which provided for optimum growth, most of the herbicide was available to the navy bean plants. The plants grown under this optimum moisture condition were more vigorous and less sensitive to EPTC. From the data, it was evident that varying degrees of moisture stress played a role in navy bean tolerance to EPTC.

Planting depth. Growth of 25-day-old navy bean plants was reduced significantly at the 5.1 and 7.6 cm planting depths. Increasing the planting depth of navy bean from 2.5 cm to 7.6 cm in soil treated with 0, 2.24 and 3.36 kg/ha of EPTC reduced plant growth 50%. EPTC at 4.48 kg/ha reduced plant growth 71% at the 7.6 cm planting depth (Table 2). The increased injury with deeper planting may have been due to reduced seedling vigor and increased emergence time. This allowed the emerging hypocotyl tissue additional time to absorb larger quantities of EPTC. The 3.36 kg/ha rate of EPTC,

Table 1. Effect of EPTC and moisture levels on the dry weight of 25-day-old navy bean plants.

EPTC rate (kg/ha)	Moisture level (cm)[b]			
	0.32	0.64	1.3	2.5
	(mg/plant)[a]			
0	59 b	76 d	97 g	98 gh
2.24	61 b	70 c	104 h	91 f
3.36	47 a	57 b	87 ef	71 cd

[a]Means followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

[b]Each level of water was applied at planting and every 4 days following for 16 days, whereafter moisture was applied equally.

the recommended field rate of application for navy bean, caused the greatest reduction in dry weight from that of the corresponding no herbicide treatment, at the 2.5 cm planting depth.

Mechanical compaction. Soil compaction as a single factor resulted in a significant reduction in plant growth at all levels of herbicide application (Table 3). The 3.36 kg/ha rate of EPTC did not significantly further reduce plant growth at any level of soil compaction. EPTC at 4.48 kg/ha significantly reduced plant growth equally from that of the no herbicide treatment at all levels of compaction.

Temperature. The most optimum temperature for navy bean growth was at 25 C (Table 4). Temperature did not interact with EPTC to alter the growth of navy beans grown in soil treated with 2.24, 3.36 and 4.48 kg/ha of EPTC. The percent reduction in plant growth due to EPTC treatment at each temperature was not significantly different, supporting previous reports (17).

Nitrogen. Applications of 56.1, 112.1 and 224.2 kg/ha of nitrogen, resulted in a significant increase in fresh weight, but not dry weight, over that of the no nitrogen treatment (Table 5). There was no increase in fresh weight above that of the 56.1 kg/ha nitrogen level. Applications of EPTC at 3.36 and 4.48 kg/ha nullified the increased fresh weight observed with nitrogen applications alone. The application of 4.48 kg/ha of EPTC to plants grown with 224.2 kg/ha of nitrogen resulted in a significant reduction in fresh weight from that of the 4.48 kg/ha treatment with no nitrogen. There are many reports in the literature which show that tissue hydration

Table 2. Effect of EPTC and planting depth on the dry weight of 25-day-old navy bean plants.

Planting depth (cm)	Herbicide rate (kg/ha)			
	None	2.24	3.36	4.48
	(mg/plant)[a]			
2.5	106 j	97 i	82 fg	84 gh
5.1	88 gh	91 hi	74 e	68 d
7.6	54 c	56 c	41 b	24 a

[a]Means followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 3. Effect of EPTC and mechanical soil compaction on the dry weight of 25-day-old navy bean.

EPTC rate (kg/ha)	Soil compaction level (kg/cm^2)		
	None	5.6	11.2
	(% of control)[a]		
0	100 e	65 cd	62 bc
3.36	95 e	64 cd	61 bc
4.48	69 d	57 b	48 a

[a]Means followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 4. The effect of temperature and EPTC on the dry weight of 25-day-old navy bean plants.[a]

EPTC rate (kg/ha)	Temperature		
	20 C	25 C	30 C
		(mg/plant)[b]	
0	139 b	152 b	139 b
2.24	121 ab	157 b	139 b
3.36	91 a	132 ab	112 ab
4.48	83 a	105 a	87 a

[a]Temperature X EPTC interaction was nonsignificant.

[b]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 5. Effect of EPTC and nitrogen on the fresh and dry weight per plant of 25-day-old navy bean.

EPTC rate (kg/ha)	Nitrogen (kg/ha)[b] 0 fresh wt (g/plant)[a]	56.1 fresh wt (g/plant)	112.1 fresh wt (g/plant)	224.2 fresh wt (g/plant)	0 dry wt (mg/plant)	56.1 dry wt (mg/plant)	112.1 dry wt (mg/plant)	224.2 dry wt (mg/plant)
0	1.9 bc	2.4 d	2.4 d	2.4 d	261 c	271 c	246 c	275 c
3.36	1.8 bc	2.1 cd	1.7 b	1.5 ab	186 ab	195 b	182 ab	180 ab
4.48	1.8 bc	1.5 ab	1.5 ab	1.3 a	164 ab	151 ab	173 ab	139 a

[a]Means followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test. Comparisons are only valid within either fresh or dry weight.

[b]Nitrogen applied as ammonium nitrate (NH_4NO_3).

can be directly correlated with nitrogen levels (6, 16). From the data it was apparent that without EPTC, navy bean showed an increase in fresh weight and tissue hydration in response to nitrogen. The failure of navy bean treated with EPTC to show increased fresh weight development when treated with nitrogen, may have been the result of increased water loss due to EPTC treatment. As previously reported, EPTC has been shown to increase water loss from navy bean plants[6]. The potential increase in tissue hydration due to nitrogen could be nullified by increased water loss resulting from the EPTC treatment.

Damaged seed. Data from 1972 and 1973 field trials showed that damaged seed alone did not result in reduced plant height or yield. Both plant height and yield of plants grown from damaged navy bean seed were significantly reduced when treated with 3.36 and 4.48 kg/ha of EPTC (Table 6). These data indicated that damaged seed is more sensitive to EPTC than normal navy bean seed. In greenhouse experiments, EPTC at 3.36 kg/ha did not reduce the growth of plants from normal seed at the 2.5 and 5.1 cm planting depths. However, EPTC did significantly reduce plant growth of normal seeds at the 7.6 cm planting depth (Table 7). EPTC at 3.36 kg/ha reduced plant growth of navy bean plants developed from damaged seed at all planting depths. These data supported the field and greenhouse

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[6]Wyse, D., W. F. Meggitt and D. Penner. 1971. Factors influencing the response of navy beans to EPTC. Proc. N. Cent. Weed Contr. Conf. 26:52.

Table 6. Effect of damaged seed and EPTC on plant height and yield of navy bean, East Lansing, Michigan, 1972, 1973.[b]

EPTC rate (kg/ha)	1972				1973			
	Normal seed	Damaged seed	Normal seed	Damaged seed	Normal seed	Damaged seed	Normal seed	Damaged seed
	Plant height (cm/plant)[a]	Plant height (cm/plant)	Yield (kg/ha)	Yield (kg/ha)	Plant height (cm/plant)[a]	Plant height (cm/plant)	Yield (kg/ha)	Yield (kg/ha)
0	22.1 c	21.1 c	1680 c	1620 bc	19.1 ab	20.3 a	1872 ab	1764 bc
3.36	21.6 c	18.3 b	1668 c	1326 a	20.0 ab	16.8 c	1944 a	1398 d
4.48	17.8 b	12.4 a	1518 b	1266 a	18.5 b	13.7 d	1620 c	1236 e

[a]Means followed by corresponding letters are not significantly different at the 5% level by Duncan's Multiple Range Test. Comparisons are only valid within either 1972 or 1973.

[b]In 1972 the plots were sprayed and planted June 20 and plant height measured 35 days later. In 1973 the plots were planted and sprayed June 22 and plant height measured 30 days later.

Table 7. Effect of EPTC, damaged seed and planting depth, on the dry weight of 20-day-old navy bean plants.

Planting depth (cm)	Normal seed		Damaged seed	
	EPTC kg/ha		EPTC kg/ha	
	None	3.36	None	3.36
	(mg/plant)[a]			
2.54	172 g	174 g	170 g	132 d
5.1	150 f	144 ef	135 de	109 bc
7.6	118 c	104 b	109 bc	88 a

[a]Means followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

data that damaged seed and planting depth increased navy bean susceptibility to EPTC.

Navy bean is susceptible to many stress conditions. Any one of the factors studied in these experiments has the potential of greatly altering the growth of navy bean. The application of EPTC to plants under moisture, deep planting and seed quality stress can accentuate the stress condition, the interaction resulting in reduced plant growth.

Literature Cited

1. Adams, R. S., Jr. and W. G. Espinoza. 1969. Effect of phosphorus and atrazine on mineral composition of soybeans. J. Agr. Food Chem. 17:818-822.

2. Allison, L. E. 1952. Effect of synthetic polyelectrolytes on the structure of saline and alkali soils. Soil Sci. 73:443.

3. Audus, L. J. 1964. The physiology and biochemistry of herbicides. Academic Press Inc., New York, New York. 555 p.

4. Burnside, O. C. and W. G. Lipke. 1962. The effect of applied water on preemergence applications of amiben. Weeds. 10:100-103.

5. Carns, A. 1934. Soil crust. Agricultural Engineering. 15:167-169.

6. Carroll, J. C. 1943. Effects of drought, temperature, and nitrogen on turf grasses. Plant Physiol. 18: 19-36.

7. Drew, L. O. and W. F. Buckele. 1963. Emergence force of plants. Proc. Am. Soc. Agr. Engr. Trans. Paper No. 62:641.

8. Drew, L. O., T. H. Garner and D. G. Dickson. 1967. Seedling thrust vs. soil strength. Proc. Am. Soc. Agr. Eng. Trans. Paper No. 67-163.

9. Edwards, F. E. 1966. Cotton seedling emergence. Mississippi Farm Res. 29(11):4-5.

10. Fang, S. C., P. Theisen and V. H. Freed. 1961. Effects of water evaporation, temperature and rates of application on the retention of ethyl-N,N-di-n-propylthiocarbamate on various soils. Weeds. 9:569-574.

11. Gantz, R. L. and F. L. Slife. 1960. Persistence and movement of CDAA and CDEC in soil and the tolerance of corn seedlings to these herbicides. Weeds. 8: 599-606.

12. Garner, T. H. and H. D. Bowen. 1966. Plant mechanics in seedling emergence. Soc. Agr. Engr. Trans. 9:650-633.

13. Gray, R. A. 1965. A vapor trapping apparatus for determining the loss of EPTC and other herbicides from soils. Weeds. 13:138-141.

14. Gray, R. A. and A. J. Weierich. 1965. Factors affecting the vapor loss of EPTC from soils. Weeds. 13:141-147.

15. Koslowski, T. T., S. Sasaki and J. H. Torrie. 1967. Influence of temperature on phytotoxicity of triazine herbicides to pine seedlings. Amer. J. Bot. 54:790-796.

16. Pellett, H. M. and E. C. Roberts. 1963. Effect of mineral nutrition on high temperature induced growth retardation of Kentucky bluegrass. Agron. J. 55:473-476.

17. Penner, D. and D. Graves. 1972. Temperature influence on herbicide injury to navy beans. Agron. J. 64:30.

18. Penner, D. 1971. Effect of temperature on phytotoxicity and root uptake of several herbicides. Weed Sci. 19: 571-576.

19. Richards, L. A. 1953. Modulus of rupture as an index of crusting soil. Soil Sci. Soc. Amer. Proc. 17:321-323.

20. Thompson, L., Jr., F. W. Slife and H. S. Butler. 1970. Environmental influence on the tolerance of corn to atrazine. Weed Sci. 18:509-514.

21. Upchurch, R. P., G. R. Ledbetter and F. L. Solman. 1963. The interaction of phosphorus with the phytotoxicity of soil applied herbicides. Weeds. 11:36-41.

22. Upchurch, R. P. 1957. The influence of soil-moisture content on the response of cotton to herbicides. Weeds. 5:112-120.

23. Waters, E. C., Jr. and J. D. Atkins. 1959. Performance of snapbeans (Phaseolus vulgaris) seedlings having transversely broken cotyledons. Proc. Amer. Soc. Hort. Sci. 74:591-596.

24. Wright, T. H. and C. E. Rieck. 1974. Factors affecting butylate injury to corn. Weed Sci. 22:83-85.

CHAPTER 3

THE INTERACTION OF ATRAZINE AND EPTC ON NAVY BEAN

Abstract

The interaction of EPTC (S-ethyl dipropylthiocarbamate) and atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine] on navy bean was studied in field and laboratory experiments. Navy bean (Phaseolus vulgaris L.) treated with the combination of EPTC and atrazine developed extensive leaf chlorosis greater than the atrazine treatment alone. Navy bean yields in field studies in 1972, were reduced 39% when grown in the presence of 0.55 kg/ha of atrazine and 3.36 kg/ha of EPTC, whereas, the single application of either herbicide reduced yields 0 and 14%, respectively. In 1973, atrazine at 0.28 kg/ha did not significantly reduce yields, but in combination with 2.24, 3.36 and 4.48 kg/ha of EPTC, yields of navy bean were reduced 40, 30 and 62%, respectively. The combination of atrazine and EPTC also reduced plant weight and delayed flower and pod development. EPTC was found to increase wind blast under conditions of low relative humidity, high winds and limited soil moisture. EPTC at 2.26, 3.36 and 4.48 kg/ha reduced the chloroform-soluble leaf extract from the surface of navy bean leaves and scanning electron

micrographs confirmed that EPTC at 10^{-6} M altered leaf surface waxes. EPTC at 10^{-5} and 10^{-6} M increased the transpiration rate of navy bean plants. Increased uptake of ^{14}C-atrazine from solution was associated with the increased transpiration rate.

Introduction

Atrazine is widely used for selective weed control in corn (*Zea mays* L.) and EPTC is used extensively for weed control in navy beans. In recent years the trend in Michigan has been to incorporate corn into the sugar beet-navy bean rotation to add more organic matter for improved soil structure.

Atrazine, under certain conditions, presents a carry over problem recognized for a long period of time. The atrazine application rate and climate play an important role in atrazine residue levels the following year. Several reports show the atrazine residue problem to be more severe in arid regions and under prolonged cold and dry periods (2, 3, 7) with less persistence in warm and humid climates (5, 11, 14, 16). Year to year variations in rainfall and temperature also play an important role in determining atrazine residue (1, 10). Talbert and Fletchall (16) indicated that 0.16 kg/ha of atrazine and 0.31 kg/ha of simazine [2-chloro-4,6-bis(ethylamino)-*s*-triazine] were present 1 year after application of 2.24 kg/ha of each.

Meggitt[1] observed no effect on oat, soybean and navy bean 1 year after application of 2.24 kg/ha of atrazine in Michigan, however, 1 year after the second or third annual application of 2.24 kg/ha the sensitive crops were severely injured.

Atrazine residue in the soil from the previous year creates the potential for interaction with the herbicide applied for weed control in the current year. Duke (6) showed increased injury to alfalfa following EPTC application to soils containing low levels of atrazine residues. The injury resembled an additional dose of atrazine and could be the result of reduced atrazine detoxification or increased atrazine uptake due to EPTC treatment.

TCA (15) and EPTC (8, 15) have been shown, along with other thiocarbamates, to inhibit the deposition of foliar waxes on cabbage and pea. Gentner (8) showed that the transpiration rate of the leaves treated with EPTC increased. He hypothesized that this increase in transpiration may influence the uptake of other compounds which move freely in the transpiration stream. There is considerable evidence showing s-triazine herbicide absorption by roots and translocation via the apoplastic system to plant shoots, the rate of translocation being proportional to the rate of transpiration or water loss (13, 17). Therefore, the increase in transpiration resulting from EPTC treatment could allow for increased atrazine uptake and translocation.

[1]Meggitt, W. F. 1969. Persistence of herbicide residues in soils. Michigan State University, East Lansing, unpublished report.

This investigation studied (a) the interaction effects of the combination of EPTC and low levels of atrazine in the field and (b) the effect of EPTC on wax deposition on navy bean leaves and its effect on transpiration and subsequent uptake of ^{14}C-atrazine.

Materials and Methods

Field experiments. Field experiments were conducted in 1972 and 1973 at East Lansing, Michigan to study the interaction between EPTC and atrazine on navy bean. The experiments were conducted on a Conover sandy loam with 2.5% organic matter. Treatments were replicated three times in a split-plot design. Plot size was 3.0 m x 15.2 m with four rows in 0.76 m row widths. Atrazine and EPTC were incorporated with a spring tooth harrow to a depth of 7.6 cm. In 1972, atrazine was applied at 0.069, 0.138, 0.276 and 0.552 kg/ha and incorporated on May 15. EPTC was applied at 3.36 and 4.48 kg/ha and incorporated June 15. In 1973, atrazine was applied at 0.07, 0.14, 0.28 and 0.55 kg/ha and incorporated on June 2 and EPTC at 2.24, 3.36 and 4.48 kg/ha was applied and incorporated on June 15. 'Sanilac' navy bean was planted June 15, in both 1972 and 1973. In 1972, 8.4 cm of rain fell from the time of atrazine application to planting and 0.84 cm of rain fell during the same period in 1973. All plots were cultivated twice and hand weeded as needed during the growing season. Fresh and dry weight of plants were recorded 35 and 30 days after planting in 1972 and 1973, respectively. Bud, flower and

pod counts were taken on three dates in 1972 and 1973 as a measure of plant development. At maturity plants were hand pulled and grain yields recorded.

Transpiration study. Five 'Sanilac' navy bean seeds were planted in washed sand in a 296 ml waxed cup. Solutions of EPTC at 10^{-5}, 10^{-6} and 10^{-7} were prepared in modified Hoagland's no. 1 solution at pH 6.8. The plants were grown at 20, 25 and 30 C for 15 days with a 16 hr day and 22.0 klux from fluorescent and incandescent lighting. The plants were then removed from sand culture and placed in 300 ml Erlenmeyer flasks containing Hoagland's no. 1 solution. Two plants were placed in each flask and stoppered with sponge plugs. The total weight of flask, solution, and plants was measured after 1 hr at 30 C. The transpiration rate was determined by the change in weight of the system after 24 hours at 30 C and 24.1 klux. The data are expressed as ml transpired per cm^2 leaf area and are the means of two experiments with four replications per experiment.

Leaf wax determination. The effect of EPTC on leaf wax production was determined by growing navy beans in soil treated with 2.24, 3.36 and 4.48 kg/ha of EPTC in the greenhouse at 20 to 30 C with natural illumination supplemented with fluorescent lighting. Two hundred leaves per replication were dipped in chloroform for 20 seconds, the extract was placed in weighing tins and dried to constant weight at 40 C. The chloroform-soluble wax extract was expressed as mg wax per dm^2 of leaf area. The data reported are the means of two

experiments with three replications per experiment. Unifoliate leaves from plants grown in 2.24, 3.36 and 4.48 kg/ha of EPTC were frozen in liquid nitrogen and freeze-dried for 12 hr. Sections were cut from the freeze-dried tissue for observation by scanning electron microscopy ('SEM'). The tissue was coated with a uniform coating of 500 Å of gold evaporated onto the surface. The samples were then examined with an advanced Metals Research 900 (AMR 900) SEM and representative sections photographed.

Atrazine uptake study. An experiment was initiated to determine if atrazine uptake by navy bean plants pretreated with EPTC was similar to that of plants with no EPTC pretreatment. Five 'Sanilac' navy bean seeds were planted in washed no. 7 graded quartz sand in a 295 ml paper cups with drainage holes in the bottom and placed in plastic 295 ml cups to allow for subirrigation by daily additions of the EPTC solutions. A solution of EPTC 10^{-6} M was made up in Hoagland's no. 1 solution at pH 6.8. After 16 days in a greenhouse maintained at 25 $\pm$ 5 C, the plants were removed from sand culture and placed in solutions of uniform ring labeled (18.3 mCi/mM) ^{14}C-atrazine. The ^{14}C-atrazine solutions contained 0.5 µci radioactivity /100 ml nutrient solution. The plants were allowed to take up the ^{14}C-atrazine for 15 hr at 30 C and 22.1 klux. The plants were then removed from the ^{14}C-solution and the roots washed five times with distilled water. The plants were sectioned into root, stem, unifoliate and trifoliate leaves. A portion of the oven dried tissue was

weighed and combusted by the Schoeninger combustion method of Wang and Willis (18).

Results and Discussion

Field plot studies. Navy bean yields in 1972 were reduced 39% with EPTC at 3.36 kg/ha in combination with 0.55 kg/ha of atrazine; however, these herbicides as single treatments reduced yield only 0 and 14%, respectively (Table 1). In 1973, the level of atrazine in the soil at planting was greater than in 1972 due to decreased time between atrazine application and planting, and less rainfall during the same period. This was evident by increased toxicity of the lower levels of atrazine to the navy beans (Table 1). In 1973, the 4.48 kg/ha rate of EPTC reduced yields 17%. Atrazine at 0.14 kg/ha did not reduce yields; however, in combination with EPTC at 4.48 kg/ha yields were reduced 40%. Atrazine at 0.28 kg/ha did not cause a significant reduction in yield but in combination with 2.24, 3.36 and 4.48 kg/ha of EPTC navy bean yields were reduced 40, 30 and 62%, respectively.

Fresh weight measurements in 1972 taken 35 days after planting indicated that 3.36 kg/ha EPTC in combination with 0.14 and 0.28 kg/ha atrazine reduced growth significantly over either treatment alone (Table 1). EPTC at 4.48 kg/ha in combination with 0.07, 0.14 and 0.28 kg/ha of atrazine also reduced plant growth more than either treatment alone. In 1973, both the 3.36 and 4.48 kg/ha rate of EPTC in combination with 0.28 kg/ha of atrazine reduced fresh and dry

Table 1. Effects of EPTC and atrazine on fresh and dry weight of 35-day-old 'Sanilac' navy bean plants and final grain yield.

EPTC rate (kg/ha)	Atrazine rate (kg/ha)	1972 Yield (kg/ha)[a]	1972 Fresh weight (g/plant)	1972 Dry weight (g/plant)	1973 Yield (kg/ha)	1973 Fresh weight (g/plant)	1973 Dry weight (g/plant)
----	----	1478 cdef	23.4 def	5.0 bcd	1411 gh	16.6 efg	3.4 def
----	0.07	1640 f	34.3 g	7.8 e	1660 i	18.2 fg	3.4 def
----	0.14	1552 ef	25.5 ef	5.0 bcd	1566 ghi	19.2 g	4.2 ef
----	0.28	1539 ef	27.0 fg	5.8 cd	1136 fg	14.0 def	3.6 def
----	0.55	1270 cd	----	----	511 b	----	----
2.24	----	----	----	----	1331 gh	12.1 abcde	3.1 bcde
3.36	----	1499 def	23.3 def	6.4 de	1357 gh	18.6 fg	3.8 ef
4.48	----	1458 cdef	23.0 de	6.5 de	1116 efg	14.4 efg	3.5 def
2.24 +	0.07	----	----	----	1337 gh	12.7 cde	3.0 bcde
2.24 +	0.14	----	----	----	1297 g	13.2 cde	2.9 abcd
2.24 +	0.28	----	----	----	941 de	13.1 cde	3.2 cdef
2.24 +	0.55	----	----	----	242 a	----	----
3.36 +	0.07	1331 cde	21.0 cd	6.0 d	1331 gh	18.4 fg	4.2 f
3.36 +	0.14	1284 cd	17.4 bc	3.8 ab	1109 def	12.2 bcde	2.9 abcd
3.36 +	0.28	1290 cd	17.0 b	4.3 abc	820 d	10.0 abcd	2.2 ab
3.36 +	0.55	907 a	----	----	396 ab	----	----
4.48 +	0.07	1384 cde	18.7 bc	5.4 bcd	800 cd	9.4 abc	2.3 abc
4.48 +	0.14	1250 bc	15.3 ab	3.8 ab	806 d	7.6 a	2.3 abc
4.48 +	0.28	1290 cd	13.0 a	3.2 a	517 bc	7.7 a	2.0 a
4.48 +	0.55	1055 ab	----	----	183 a	----	----

[a]Means within a column with similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

weight significantly. EPTC at 4.48 in combination with 0.14 and 0.28 kg/ha of atrazine reduced fresh and dry weight more than any of these treatments alone.

Reduced formation of buds and flowers on 8/2/72 resulting from the combination of EPTC at 3.36 kg/ha and 0.07, 0.14 and 0.28 kg/ha of atrazine indicated a delay in plant development over that of any one of these treatments alone (Table 2). On 8/8/72, no significant interaction between atrazine and EPTC was observed. On 8/14/72, a significant reduction in flower and bud formation was observed from the combination of EPTC at 3.36 kg/ha and atrazine at 0.28 and 0.55 kg/ha. These same treatments showed a significant yield reduction at harvest. In 1973, on July 25, EPTC at 2.24 kg/ha in combination with 0.28 kg/ha atrazine and EPTC at 4.48 kg/ha in combination with 0.07, 0.14 and 0.28 kg/ha atrazine significantly reduced bud and flower formation (Table 3). The combination 3.36 kg/ha of EPTC and 0.28 kg/ha atrazine significantly reduced pod formation. On 8/10/73 treatments showing reduced yields and delayed development had more buds and flowers per plant, indicating a delay in plant development. On the last observation date in 1973, flowering was almost completed. This allowed for analysis of total pods per plant in each of the treatments. EPTC at 2.24 and 3.36 kg/ha showed no interaction with atrazine in terms of pod formation. EPTC at 4.48 kg/ha in combination with all levels of atrazine significantly reduced pod set to a greater extent than any one of the treatments alone.

Table 2. Effects of EPTC and atrazine on bud, flower and pod formation of 'Sanilac' navy bean planted June 15, 1972.

EPTC	Atrazine	Observation Date 8/2/72		8/8/72		8/14/72	
		Buds and flowers per plant	Pods per plant	Buds and flowers per plant	Pods per plant	Buds and flowers per plant	Pods per plant
kg/ha	kg/ha						
----	----	5.5 cd	4.7 fg	14.2 e	4.9 abcd	10.4 f	16.8 de
----	0.07	7.6 d	6.3 g	13.6 e	4.8 abcd	9.9 ef	18.2 e
----	0.14	5.8 cd	5.3 fg	12.3 de	4.5 abcd	9.4 def	17.5 de
----	0.28	5.6 cd	4.4 ef	11.5 cde	3.2 ab	7.5 cdef	15.1 cde
----	0.55	3.5 bc	3.0 de	9.5 bcde	2.5 a	6.4 abcd	12.1 abcd
3.36	----	5.4 cd	4.7 fg	13.0 de	6.8 bcd	7.4 cdef	14.9 cde
3.36 +	0.07	2.0 ab	2.0 bcd	9.5 bcde	7.3 d	7.3 cdef	15.1 cde
3.36 +	0.14	2.6 ab	2.4 cd	8.5 abcd	6.5 d	7.6 cdef	15.3 cde
3.36 +	0.28	1.9 ab	0.2 ab	7.6 abc	6.3 cd	3.5 a	8.8 ab
3.36 +	0.55	2.0 ab	0.1 a	6.4 abc	5.1 abcd	3.4 a	7.7 a
4.48 +	----	3.0 b	2.7 d	7.7 abcd	6.0 bcd	7.0 bcde	14.0 bcde
4.48 +	0.07	2.0 ab	1.7 abcd	8.2 abcd	6.4 d	5.7 abcd	12.7 abcd
4.48 +	0.14	2.3 ab	2.2 cd	6.9 abc	5.5 abcd	7.1 cde	13.7 bcd
4.48 +	0.28	2.0 ab	0.2 ab	6.0 ab	4.8 abcd	5.6 abc	12.7 abcd
4.48 +	0.55	1.2 a	0.9 abc	4.0 a	3.2 ab	3.9 abc	9.7 abc

[a]Means within columns followed with similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 3. Effects of EPTC and atrazine on bud, flower and pod formation on navy bean planted June 15, 1973.

EPTC	Atrazine	Observation Date					
		7/25/73		8/1/73		8/10/73	
kg/ha	kg/ha	Buds and flowers per plant	Pods per plant	Buds and flowers per plant	Pods per plant	Buds and flowers per plant	Pods per plant
----	----	27.7 ef	5.2 c	2.0 a	16.2 e	0.3 ab	21.1 efg
----	0.07	31.4 g	5.1 c	2.3 a	16.8 ef	0.9 bc	24.0 g
----	0.14	28.6 efg	1.5 ab	2.6 a	16.4 ef	0.6 abc	22.7 g
----	0.28	14.3 c	4.1 c	9.2 d	9.2 b	2.1 de	15.9 cde
2.24 +	----	27.6 ef	2.3 ab	1.8 a	16.7 ef	0.0 a	22.3 fg
2.24 +	0.07	27.7 ef	5.2 c	1.9 a	15.4 de	0.0 a	20.3 defg
2.24 +	0.14	27.8 ef	2.6 b	2.5 a	13.2 cd	0.0 a	20.6 efg
2.24 +	0.28	22.7 d	0.0 a	4.4 b	10.7 b	1.3 cd	14.8 bcd
3.36 +	----	29.3 efg	2.6 b	2.2 a	18.1 f	0.0 a	22.2 fg
3.36 +	0.07	30.4 fg	2.4 ab	2.0 a	16.5 ef	0.3 ab	21.0 efg
3.36 +	0.14	24.3 de	1.4 ab	4.2 b	11.3 bc	1.3 cd	16.8 cdef
3.36 +	0.28	14.2 c	0.0 a	7.9 d	5.6 a	3.1 f	12.1 bc
4.48 +	----	16.4 c	1.2 a	7.2 cd	10.6 b	2.1 de	16.3 cde
4.48 +	0.07	11.2 b	0.0 a	6.2 c	5.5 a	2.5 ef	10.5 ab
4.48 +	0.14	8.1 b	0.0 a	7.3 cd	4.4 a	2.7 ef	11.9 bc
4.48 +	0.28	5.8 a	0.0 a	6.4 c	3.5 a	2.5 ef	7.7 a

[a]Means within a column followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

The injury symptoms of navy bean resulting from the EPTC-atrazine interaction were similar to atrazine alone and similar to those reported by Duke (6) on alfalfa. The navy bean plants treated with 3.36 and 4.48 kg/ha of EPTC showed cupping and crinkling of the unifoliate leaf tips. The injury to navy bean treated with 0.55 kg/ha of atrazine showed leaf chlorosis. Navy bean treated with EPTC and atrazine at 3.36 and 0.55 kg/ha, respectively, developed extensive leaf chlorosis. Duke (6) has suggested that the increased toxicity of the combination may be due to EPTC preventing the formation of a detoxifying enzyme in alfalfa which prevented the metabolism of atrazine.

Another possible explanation for the interaction could be increased atrazine uptake induced by EPTC. Gentner (8) has shown that EPTC treatment increased the transpiration rate of cabbage leaves and suggested that the increased transpiration rate may influence the passive uptake of materials in the transpiration stream. Atrazine which is passively absorbed by roots could be swept up in larger quantities by the transpiration stream resulting in increased atrazine toxicity.

Transpiration studies. In 1971, severe wind blast injury or leaf desiccation was observed in navy bean treated with 3.36 kg/ha of EPTC, suggesting that EPTC altered the plants water economy (Figure 1). The extensive wind blast injury was observed under conditions of low humidity, high winds and limited soil moisture for 2 days. Plants treated with EPTC had more wind blast injury than plants in the hand weeded

Figure 1. Navy bean plants with wind blast injury. Untreated plants (A) and plants treated with 3.36 kg/ha of EPTC (B).


A


B

check. The wind blast injury was consistent throughout the four EPTC-treated replications. This observation supports Gentner's (7) earlier observation on cabbage leaves that EPTC can alter plant water economy.

Plants grown at 20, 25 or 30 C in controlled environmental chambers and treated with EPTC at 10^{-5} and 10^{-6} M had increased transpiration rates over that of the plants not treated with EPTC (Table 4). The greatest amount of water loss per unit area of leaf was observed at the pretreatment temperature of 20 C when water loss was measured at 30 C. The increased transpiration rate resulting from EPTC treatment, regardless of pretreatment temperature, supported the earlier observation of decreased water economy and increased wind blast damage due to EPTC application.

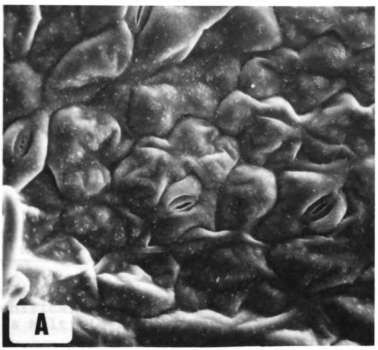
Leaf wax determinations. EPTC has previously been shown to reduce wax formation on leaf surfaces (8, 15). The amount of chloroform-soluble extract from navy bean leaves shows that soil-applied EPTC greatly reduced the amount of surface wax (Table 5). The 4.48 kg/ha rate of EPTC reduced wax deposition to 16% of the control. Scanning electron micrographs showed that EPTC altered the physical appearance of the navy bean leaf surface (Figure 2). The small flakes of epicuticular wax evident on the leaves of control plants do not appear on the leaves treated with EPTC at 10^{-6} M. The quantitative and qualitative data together suggest that EPTC altered the amount and structure of the surface waxes of navy bean leaves. This alteration in surface wax formation may account for the

Table 4. The effect of EPTC and temperature pretreatment on the quantity of water loss from 15-day-old navy bean plants at 30 C for 48 hr.

Herbicide treatment	Temperature pretreatment[a]		
	20 C	25 C	30 C
	(ml water lost/cm^2/48 hr)		
CHECK	0.312 d	0.238 b	0.184 a
EPTC 10^{-7} M	0.307 d	0.248 b	0.191 a
EPTC 10^{-6} M	0.405 e	0.302 cd	0.269 bc
EPTC 10^{-5} M	0.480 f	0.461 f	0.337 d

[a]Means having similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Figure 2. Electron micrographs of navy bean leaves at 1000 X. Leaf (A) is from a plant grown in Hoagland's solution. Leaf (B) is from a plant grown in Hoagland's solution with EPTC at 10^{-6} M.


A

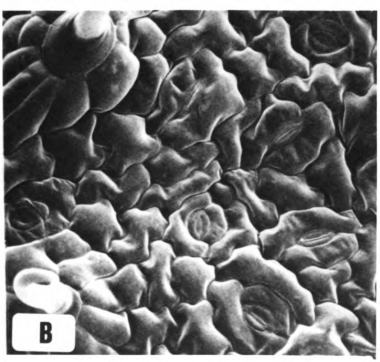

B

Table 5. Effect of EPTC on the chloroform-soluble fraction from the surface of navy bean leaves.

EPTC kg/ha	Chloroform-soluble extract (% of control)[a]
2.24	50 c
3.36	33 b
4.48	16 a

[a]Means within columns having identical letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

increased transpiration loss, and in turn account for the increased atrazine injury to navy bean when the two compounds are present in combination by increasing the passive uptake of atrazine and upward movement in the transpiration stream.

Uptake of ^{14}C-atrazine. In a study designed to measure the effect of EPTC on ^{14}C-atrazine uptake by 16-day-old navy bean plants, the water loss from these plants was found to be increased by EPTC similar to previous experiments (Table 6). When the quantity of ^{14}C-atrazine taken up by the navy bean plants was expressed as DPM per total plant dry weight, there was no apparent increase due to the EPTC treatment. When the radioactivity in the total plant was expressed per unit area of leaf, rather than on a leaf weight basis, there was a significant difference due to the EPTC treatment. This can be explained by the EPTC 10^{-6} M treatment increasing the density of the leaf tissue from 1.2 mg/cm^2 to 2.0 mg/cm^2. If the concentration of ^{14}C-atrazine increased in the leaf tissue with a higher density, the increase in ^{14}C-atrazine concentration when expressed in terms of leaf dry weight would be masked, due to the increased weight per unit area of leaf. Dawson (4) has shown that EPTC reduces intercellular spaces and increases cell compaction with a subsequent increase in tissue density. When the total radioactivity per plant was expressed per unit area of leaf, there was an increase of 51% over that of the untreated check. Plants pretreated with EPTC at 10^{-6} M increased water loss 33% over that of the untreated plants. When the radioactivity in the unifoliate

Table 6. The effect of EPTC pretreatment for 16 days on the uptake of ^{14}C-atrazine by navy bean plants after 15 hours at 30 C.

Herbicide pretreatment (M)	Transpiration rate (ml/cm^2)[a]	DPM/ Leaf area Total DPM (DPM/cm^2)	DPM/mg plant Dry weight Total DPM (DPM/mg)
Central	0.24 a	1304 a	552 a
EPTC 1.0×10^{-6}	0.36 b	2641 b	515 a

[a]Means within columns having similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

leaves was expressed per unit area of leaf, there was a 28% increase in atrazine uptake, approximately the same as the increase in transpiration.

These data support the hypothesis that increased water loss from navy bean treated with EPTC increases the uptake of atrazine and contributes to the increased atrazine toxicity observed in the field experiments from the combination of these two herbicides.

Literature Cited

1. Bucholtz, K. P. 1965. Factors influencing oat injury from triazine residues in soil. Weeds. 13:362.

2. Burnside, O. C., C. R. Fenster and G. A. Wicks. 1963. Dissipation and leaching of monuron, simazine, and atrazine in Nebraska soils. Weeds. 11:209-213.

3. Burnside, O. C., C. R. Fenster, G. A. Wicks and J. V. Drew. 1969. Effect of soil and climate on herbicide dissipation. Weed Sci. 17:241.

4. Dawson, J. H. 1963. Development of barnyardgrass seedlings and their response to EPTC. Weeds. 11:60-66.

5. Dowler, C. C., W. Forestier and F. H. Tschirley. 1968. Effects and persistence of herbicides applied to soil in Puerto Rican forests. Weed Sci. 16:45.

6. Duke, W. B., V. S. Rao and J. F. Hunt. 1972. EPTC-atrazine residue interaction effect on seedling alfalfa varieties. Proc. Northeast. Weed Sci. Soc. 26:258-262.

7. Fenster, C. R., O. C. Burnside and G. A. Wicks. 1965. Chemical fallow studies in winter wheat fallow rotations in western Nebraska. Agron. J. 57:469-470.

8. Gentner, W. A. 1966. The influence of EPTC on external wax disposition. Weeds. 14:27-31.

9. Grover, R. 1962. Uptake and distribution of ^{14}C-atrazine and ^{14}C-propazine in some tree seedlings. Plant Physiol. 37:12-13.

10. Harris, C. I. and T. J. Sheets. 1965. Persistence of several herbicides in the field. Proc. Northeast. Weed Contr. Conf. 19:359.

11. Harris, C. I., E. A. Woolson and B. E. Hummer. 1969. Dissipation of herbicides at three soil depths. Weed Sci. 17:27.

12. Sheets, T. J. 1961. Uptake and distribution of simazine by oat and cotton seedlings. Weeds. 9:1-3.

13. Sikka, H. C. and D. E. Davis. 1966. Dissipation of atrazine from soil by corn, sorghum, and johnsongrass. Weeds. 14:289-293.

14. Still, G. G., D. G. Davis and G. L. Zander. 1970. Plant epicuticular lipids: Alteration by herbicidal carbamates. Plant Physiol. 46:307-314.

15. Talbert, R. E. and O. H. Fletchall. 1964. Inactivation of simazine and atrazine in the field. Weeds. 12:33-37.

16. Vostral, H. J., K. P. Buchholtz and C. A. Kust. 1970. Effect of root temperature on absorption and translocation of atrazine in soybeans. Weed Sci. 18:115-117.

17. Wang, C. H. and D. L. Willis. 1965. Radiotracer Methodology in Biological Science. Prentice-Hall, Inc., N.J. 363 p.

CHAPTER 4

HERBICIDE ROOT ROT INTERACTIONS IN NAVY BEANS

Abstract

Applications of EPTC (S-ethyl dipropylthiocarbamate) at 3.36 and 4.48 kg/ha increased root rot severity and reduced yields in soil with an artificial root rot inoculum level. EPTC at 3.36 kg/ha and chloroamben (3-amino-2,5-dichlorobenzoic acid) 3.36 kg/ha in field experiments caused the greatest increase in root rot severity and yield reductions. Dinoseb (2-sec-butyl-4,6-dinitrophenol) and fluorodifen (p-nitrophenyl α,α,α-trifluoro-2-nitro-p-tolyl ether) at 5.04 kg/ha caused the least root rot development of any herbicide. In growth chamber experiments at 23 C, EPTC at 3.36 kg/ha and alachlor [2-chloro-2',6'-diethyl-N-(methoxymethyl)acetanilide] at 2.80 kg/ha applied to navy bean grown in soil infested with Fusarium solani reduced root and plant growth. Root rot severity was greatest in plants treated with EPTC at the 20 C ambient temperature. Navy bean growth was reduced more as the level of chlamydospore inoculum increased in the presence of EPTC than without EPTC. Growth of the navy bean line 'RRR041', which is moderately resistant to root rot, did not result in reduced plant growth when grown in soil infested with Fusarium solani and treated with EPTC. None of the

herbicides tested increased Fusarium solani chlamydospore virulence when grown on potato dextrose agar supplemented with herbicides. Navy bean seedlings treated with EPTC had increased susceptibility to Fusarium solani. Scanning electron micrographs indicated that navy bean plants grown in EPTC had altered hypocotyl surfaces. EPTC and dinoseb increased the exudates of electrolytes, amino acids and sugars from root and hypocotyl tissue. None of the herbicides tested increased Fusarium solani hyphal development in liquid culture or soil. Dinoseb reduced hyphal development by the greatest amount of any herbicide tested in liquid culture and soil.

Introduction

Navy bean yields in Michigan have declined during the last 10 years (19). Limited crop rotations and poor cultural practices have resulted in increased soil compaction and reduced soil aeration. Under these conditions which are ideal for root rot development, dry root rot has become a very serious problem in navy bean production.

Root rot caused by Fusarium solani (Mart.) Appel and Wr. f. phaseoli (Burk.) Snyd. and Hans. is one of the major diseases of navy bean (34). The significance of the root rot problem is often overlooked unless plant roots are actually analyzed for disease development or the environmental conditions accentuate the injury to above ground parts. Losses from root rot cannot be adequately determined because

satisfactory control measures are not available and no commercial variety of navy bean is resistant to root rot. In severe infestations the plant may die, but generally, only growth is retarded and the degree of plant growth reduction is weather dependent. Losses from this disease are generally greater under low moisture conditions, because few secondary roots are produced, however, a partial yield will generally be obtained (4, 5).

There are reports which indicate that herbicides may either increase or decrease the incidence of disease on various crops. Katan and Eshel (14) in a recent review listed 20 pathogens which showed increased incidence of plant disease, resulting from more than 16 herbicides. Trifluralin (α,α,α-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine) has been shown to increase damping off of cotton caused by Rhizoctonia solani (6, 23, 27). Standifer (27) indicated that temperature played a role in the interaction between Rhizoctonia solani and trifluralin. Early spring plantings of cotton had more damping off when treated with trifluralin, but later planting showed no increase in damping off due to trifluralin. Katan and Eshel[1] reported that diphenamid (N,N-dimethyl-2,2-diphenylacetamide) increased damping off in pepper caused by Rhizoctonia solani, but had no effect on Phythium sp. These reports indicated that an environmental-host-pathogen

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[1]Katan, J. and Y. Eshel. 1972. Increase in damping off incidence of pepper caused by diphenamid. Weed Sci. Soc. Amer. Abstr. 100 p.

interaction dictated the effect a herbicide will have upon disease development.

The direct effect of herbicides on the growth or reproduction of pathogens has not been studied extensively. Tang (29) reported that trifluralin increased germination and chlamydospore production in soil of Fusarium oxysporum f. sp. vasinfectum. Neubauer (21) has reported increased activity of Rhizoctonia solani in soil. In liquid culture, trifluralin either reduced or had no effect on mycelial production (11, 24). EPTC has been shown to increase mycelial production of Fusarium oxysporum at low concentrations and decrease it at higher concentrations (22). EPTC also increased mycelial development of Sclerotium rolfsii in sterilized soil (25).

Herbicides may alter the virulence of pathogens. Helminthosporium sativum grown on a medium supplemented with 2,4-D [(2,4-dichlorophenoxy)acetic acid] was shown to be more virulent than the control (13). In another experiment no increase in virulence of Rhizoctonia solani grown on diphenamid supplemented medium was found[1].

Herbicides may also have a direct effect upon the host plant which makes it more susceptible to disease organisms. The increase in American gooseberry mildew of black currant treated with simazine [2-chloro-4,6-bis(ethylamino)-s-triazine] has been attributed to a rise in the nitrogen content

[1]Katan, J. and Y. Eshel. 1972. Increase in damping off incidence of pepper caused by diphenamid. Weed Sci. Soc. Amer. Abstr. 100 p.

of the host due to simazine (31). In another report (32) it was suggested that s-triazine alteration of sugar production in plants may be important in a plants susceptibility to diseases.

It is generally accepted that the main cause of the stimulation of microorganisms in the rhizosphere is the excretion of organic substances -- mainly sugars from plant roots (8, 9, 26). Alterman (2) reported that in sugar beet, the amount of glucose exudate at the soil-plant interface was greater when plants were grown in soil treated with herbicide, and this was the reason suggested for their increased susceptibility to Rhizoctonia solani. Lai (16) related the incidence of root rot of corn in soil treated with picloram (4-amino-3,5,6-trichloropicolinic acid) with enhanced carbohydrate exudation.

This investigation studied the effect of herbicides on the development of root rot on navy bean, in field, growth chamber and laboratory experiments. The influence of herbicides on Fusarium solani virulence and navy bean resistance to root rot development was also studied.

Materials and Methods

In 1972, a field experiment was conducted at Saginaw, Michigan to evaluate the effect of EPTC on root rot development on navy bean grown on a Charity clay loam soil with 3.8% organic matter. EPTC at 3.36 and 4.48 kg/ha was applied to 3.0 m four row plots in 0.76 m row widths, with a tractor

mounted sprayer and incorporated to a depth of 7.6 cm with a spring tooth harrow. The experimental design was a split plot with the main plot being inoculum level. The two inoculum levels were produced by using the natural root rot population, or by applying a soil drench containing macerated navy bean tissue infected with root rot organisms. In 1972, the root rot inoculum was prepared by growing navy bean plants in soil infested with the "root rot complex". The infected navy bean roots and hypocotyls were macerated in a Waring Blender. The diluted slurry was applied to the soil over previously planted navy bean prior to emergence. After 35 days, 50 plants per replication were dug, rated for disease development and the plant weight recorded. Grain yield was measured at maturity. In 1973, the primary field tests were repeated and expanded to include the major herbicides presently used or with potential use in the near future for weed control in navy bean. The number of locations were increased to four. At three of the locations only natural root rot inoculum levels were used, however, at Saginaw, Michigan both natural and artificial inoculum levels were studied. The inoculum in 1973 was prepared by growing Fusarium solani on autoclaved wheat grain for 30 days. The grain was maintained at 60% moisture and 27 C throughout the incubation period. The infected wheat seed was dried at 37 C for 106 hr. Sixty grams of the inoculum was applied in the row along with the navy bean seed by using a belt planter allowing placement of the inoculum in close association with the germinating seeds.

Thirty days after planting, 50 plants per replication were dug and rated for disease development. Grain yields were measured at maturity. Root rot development was studied at three other locations with natural inoculum potentials; East Lansing, on a Conover loam soil with 2.0% organic matter and Huron County I and II on a sandy loam soil with 2.5% organic matter. 'Sanilac' navy bean was used in the East Lansing and Saginaw studies and 'Seafarer' navy bean was used in the Huron County I and II experiments. Root rot development was measured in both 1972 and 1973 at all locations according to the rating system in Figure 1. Grain yields were measured at all locations except the Huron County II location. All plots were kept weed free throughout the growing season.

A standard method was used for application and incorporation of the herbicides in greenhouse and growth chamber tests. All herbicides were applied at 935 l/ha and 2.11 kg/cm^2 pressure. The preplant incorporated herbicides were mixed into the upper 5.0 cm of an unautoclaved Conover sandy loam soil with a low root rot inoculum level. 'Sanilac' navy bean was used in all experiments unless otherwise indicated. The navy bean seeds were planted 2.5 cm deep in 946 ml containers which were placed in larger diameter 475 ml containers that served as the water reservoir for subirrigation.

Preparation and infestation of soil with Fusarium solani. Chlamydospores of Fusarium solani f. phaseoli isolated from navy bean hypocotyl tissue from the Saginaw Valley of Michigan, were produced in soil extract and used for soil infestation.

Figure 1. Root rot rating system - 1972-1973.

Rating	Description
1	Very little or no infected tissue
2	Discolored tissue over 20 to 50% of the hypocotyl. Root system functional.
3	Discolored tissue over 50 to 75% of the hypocotyl tissue. With 20% or less of the area covered with lesions. With some loss of the primary root system.
4	Tissue completely discolored with 20% or more of the area covered with lesions. The primary root system only slightly functional.
5	Primary root completely non-functional. With or without secondary root development.

The cultures were maintained on potato dextrose agar slants. Mycelium was transferred to Petri plates with potato dextrose agar, and incubated in the dark for 10 days at 25 C. Macroconidia were removed from four plates and placed in 100 ml of potato dextrose broth for 36 hr until the macroconidia germinated. The germinated macroconidia were washed by centrifugation and placed in 100 ml of sterile soil extract. The soil extract was prepared by mixing one liter of water with 400 g of Conover sandy loam soil, then letting it stand for 7 days, filtering the supernatant through Whatman no. 5 filter paper and then autoclaving for 30 min. (1). The germinated macroconidia produced chlamydospores after 4 to 8 days. The chlamydospores were washed three times by centrifugation and the resulting plug was broken up by mixing for 1 hr on a Sorvall Omni-mixer. The concentration of chlamydospores in suspension was then adjusted and applied to soil in the required amounts. The Conover sandy loam soil was air dried and sieved prior to application of chlamydospores with an atomizer and thoroughly mixed in a cement mixer for 25 min. The soil moisture was equalized. In all cases the herbicides were applied following infestation of the soil with Fusarium solani chlamydospores.

Herbicide-Fusarium solani interaction. The interaction of Fusarium solani with the major herbicides used in navy bean production was studied in a growth chamber experiment. The chambers were maintained at 23 C with 21.2 klux produced by fluorescent and incandescent bulbs. Chlamydospores were

added to the soil to a level of 3.0×10^3 spores/gram air dry soil. After 30 days, plant and root weights were taken.

Temperature. EPTC was applied to soil at 2.24 and 3.36 kg/ha and chlamydospores at 3.0×10^3 spores/gram air dry soil were added. Eight seeds per replication were grown in growth chambers maintained at 20, 25 and 30 C ambient temperature and 22.1 klux. After 30 days, plant weight was measured.

Inoculum level. The influence of Fusarium solani on the interaction between root rot severity and EPTC treatment was studied at three inoculum levels. EPTC was applied at 2.24 and 3.36 kg/ha. The experiment was conducted at 24 C and 22.2 klux. Plants were harvested after 30 days, shoot and root weights were measured.

Genetic resistance. The navy bean line 'RRR041', which is moderately resistant to root rot was compared to the susceptible 'Sanilac' variety in its response to Fusarium solani and EPTC treatment. Soil was infested with Fusarium solani chlamydospores at 3.0×10^3 spores/gram dry soil and treated with 2.24 and 3.36 kg/ha of EPTC. The plants were grown for 25 days at 24 C and 20.0 klux.

Fusarium solani virulence. Experiments to determine the effect of herbicides on the virulence of Fusarium solani chlamydospores were initiated. The chlamydospores were produced as previously described except the macroconidia were produced on potato dextrose agar supplemented with several herbicides. The rest of the procedure was the same as previously described. The chlamydospores were used to inoculate

soil to 3.0×10^3 spores/gram. After 30 days at 24 C the percent of infected hypocotyl tissue was measured. The degree of infection was an indicator of virulence.

EPTC-induced navy bean predisposition to root rot. 'Sanilac' navy bean was grown in sand cultures containing solutions of several herbicides. The plants were grown for 10 days in quartz sand cultures with herbicide solutions and then transferred to soil containing 1.0×10^4 spores/gram dry soil. To determine if an alteration in the hypocotyl cuticle surface would alter the navy bean tolerance to Fusarium solani, one group of plant hypocotyls were washed with acetone and the plants transplanted in infested soil. Each treatment was replicated four times with 10 plants per replication and the experiment was repeated. The percent infection of the hypocotyl tissue was measured after 25 days at 24 C with 21.2 klux.

Hypocotyl surface. A scanning electron microscopy ('SEM') study was conducted to study the qualitative alterations of hypocotyl surfaces from plants grown in several herbicides. 'Sanilac' navy bean was grown in sand culture containing herbicides in Hoagland's no. 1 solution for 20 days. The hypocotyl tissue was washed in water several times prior to processing, and segments of the hypocotyl were cut and freeze-dried for 24 hr. Sections were then cut from the freeze-dried tissue for observation. The tissue was coated uniformly with 500 $\mathring{A}$ of gold evaporated onto the tissue surface. The samples were then observed in an Advanced Metals Research 900

(AMR 900) 'SEM'.

Exudate studies. 'Sanilac' navy bean seeds were washed in sodium hypochlorate for 30 min. Five seeds were placed in sterilized 25 x 200 mm test tubes containing acid washed, sterilized quartz sand and herbicide solutions. Seeds were incubated at 28 C for 48 hr. The quartz sand and seeds were washed with three 10 ml aliquots of water and suction filtered through Whatman no. 1 filter paper in a Buchner funnel. The concentration of total carbohydrates was determined by the anthrone method (30, 33).

The effect of herbicides on the efflux of electrolytes, total sugars and amino acids from navy bean hypocotyl and root tissue was studied. 'Sanilac' navy bean seeds were germinated in the dark on sterilized germination paper for 6 days at 26 C. The stems of 6-day-old seedlings were cut leaving 2 cm of the hypocotyl intact with the roots. The cut hypocotyl tip was dipped in liquid wax to stop leakage of cellular materials from the cut surface. Two grams fresh weight of hypocotyl and root tissue was enclosed in a cheese cloth bag. The bags were then placed in 50 ml Erlenmeyer flasks containing 30 ml of each herbicide solution. The efflux of electrolytes was measured directly with a conductance meter. The efflux of amino acids and total sugars were measured after suction filtering the solutions through Whatman no. 1 paper in a Buckner funnel. Quantitative determination of amino acids present were determined by ninhydrin method (18) and sugars by the anthrone method (30, 33).

Fusarium solani growth in liquid culture. Washed chlamydospores were placed in potato dextrose broth made up in a phosphate buffer at pH 6.5 containing herbicides. The herbicides were dissolved in 95% ethyl alcohol and made into stock solutions of phosphate buffered potato dextrose broth. Forty ml of the herbicide potato dextrose broth was added to a 300 ml flask and 0.5 ml of washed chlamydospores were added to the solution. After 6 days, the contents of each flask were filtered through pre-weighed Dacron gauze. The gauze and mycelium was washed and dried to constant weight at 75 C in a drying oven, and the dry weight recorded.

Fusarium solani chlamydospore germination in soil. Washed chlamydospores were added to non-autoclaved soil to a concentration of 3.0×10^5 spores/gram dry soil. One-half of the soil was amended with glucose at 5 mg/gram soil, the other half received no additional energy source. Technical or formulated herbicides were added to the soil at field rates. Germination of the chlamydospores was measured after 36 hours by direct microscopic examination of a soil smear (20).

Data reported are the means of two experiments with three or more replications each.

Results

Yield and disease ratings from 1972 and 1973 field studies are presented in Table 1. In 1972, the application of 3.36 and 4.48 kg/ha of EPTC resulted in increased root rot injury to navy bean grown in soil with a natural or artificial root

Table 1. Disease ratings and yields of navy bean grown in *Fusarium solani* natural and artificially infested soil.[a]

Herbicide treatment (kg/ha)		Field trial-1972				Field trial-1973							
		Saginaw, Michigan				Saginaw, Michigan				Huron Co. I		Huron Co. II	East Lansing, Michigan
		Natural Infestation		Artificial Infestation		Natural Infestation		Artificial Infestation		Natural Infestation		Natural Infestation	Natural Infestation
		Disease rating	Yield (kg/ha)	Disease rating	Yield (kg/ha)	Disease rating[a]	Yield[a] (kg/ha)	Disease rating	Yield (kg/ha)	Disease rating	Yield (kg/ha)	Disease ratings	Disease rating	Yield (kg/ha)
Check		1.6 a	1828 a	1.9 a	1761 c	2.3 a	1431 f	2.3 a	1142 c	1.9 b	1445 d	1.7 a	1.9 a	1351 b
DNBP	5.04	----	----	----	----	2.6 b	1371 f	2.6 ab	1230 c	----	----	1.8 a	----	----
EPTC	2.24	----	----	----	----	2.9 c	1129 cd	3.4 c	934 b	----	----	3.3 c	2.3 a	1317 b
EPTC	2.52	----	----	----	----	----	----	----	----	3.4 c	544 a	----	----	----
EPTC	3.36	2.8 b	1613 a	3.6 b	1371 b	3.7 d	732 b	3.8 d	618 a	----	----	----	2.5 a	1357 b
EPTC	4.48	3.3 c	1552 a	3.8 b	1109 a	----	----	----	----	----	----	----	2.6 a	1116 a
Alachlor	2.24	----	----	----	----	----	----	----	----	1.9 b	1136 c	2.1 ab	----	----
Alachlor	3.36	----	----	----	----	3.0 c	1089 c	2.9 b	1216 c	----	----	----	----	----
Fluorodifen	5.04	----	----	----	----	2.5 b	1284 ef	2.4 a	1142 c	1.6 a	1512 d	1.8 a	----	----
Dinitramine	0.56	----	----	----	----	----	----	----	----	2.0 b	1317 cd	2.2 b	----	----
Dinitramine	0.84	----	----	----	----	2.9 c	1236 de	2.8 b	1176 c	----	----	----	----	----
Trifluralin	0.56	----	----	----	----	----	----	----	----	1.8 ab	1304 cd	1.8 a	----	----
Trifluralin	0.84	----	----	----	----	2.9 c	1230 cde	2.8 b	1236 c	----	----	----	----	----
Trifluralin	1.12	----	----	----	----	2.9 c	1164 cd	2.7 b	1203 c	----	----	----	----	----
Chloramben	2.24	----	----	----	----	----	----	----	----	----	----	3.3 c	----	----
Chloramben	3.36	----	----	----	----	3.8 d	632 a	3.9 d	625 a	3.3 c	679 b	----	----	----

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

rot infestation. Yields were not reduced by increased root rot with natural soil infestation; however, where the root rot inoculum level was increased, grain yields were significantly reduced by both levels of EPTC. The recommended use rate of EPTC on navy beans is 3.36 kg/ha. The 1973 experiments at Saginaw, Michigan, showed that all herbicide treatments in the study, under natural soil root rot infestation, increased root rot infection of the navy beans compared to the control. EPTC at 3.36 kg/ha and chloramben at 3.36 kg/ha caused the greatest increase in root rot infection and corresponding reduction in navy bean yield. Treatments of dinoseb at 5.04 kg/ha and fluorodifen at 5.04 kg/ha resulted in the least root rot development with no reduction in grain yield. Under artificial root rot infestation, applications of EPTC at 2.24 and 3.36 kg/ha and chloramben at 3.36 kg/ha increased root rot development of navy bean by the greatest amount and decreased yield accordingly. None of the other herbicide treatments significantly reduced yield, although, some treatments did increase the severity of the root rot infection.

At the Huron County I location, EPTC at 2.52 kg/ha and chloramben at 3.36 kg/ha applied to soil with a natural root rot inoculum potential, resulted in the most severe root rot infection and the greatest yield reduction. Chloramben at 2.24 and EPTC at 2.24 kg/ha caused the greatest increase in disease infection at the Huron II location. Plants treated with EPTC at 2.24, 3.36 and 4.48 kg/ha at the East Lansing location, under conditions of good soil tilth and low root

rot infestation did not show a significant increase in disease infection at any level of EPTC treatment. No reduction in yield was observed except at the 4.48 kg/ha rate which is 1.12 kg/ha above the recommended rate of application.

The two years of data indicated that EPTC and chloramben may increase the severity of Fusarium solani root rot and affect the yield of navy bean, depending upon environmental and cultural conditions. The 1973 growing season was advantageous for development of severe root rot infection of navy bean and subsequent yield reductions. The soil was wet early in the growing season and later became extremely dry. Yield reductions were very evident in treatments which developed extensive root rot. The primary root system decayed and became non-functional, and the shallow secondary roots did not adequately supply moisture for plant growth. Root rot infection was severe in 1972, however, yield reductions were not as great as soil moisture levels remained adequate throughout the growing season. Captan seed treatments did not alter the mid-season development of root rot regardless of herbicide or inoculum treatment in 1973.

Growth chamber studies. Fusarium solani, the dominant navy bean root rot organism in Michigan, was used to further study the interaction of several herbicides and root rot in growth chamber experiments. Only the combination treatment of trifluralin + EPTC at 0.84 + 3.36 kg/ha reduced fresh weight and dry weight of shoots and dry weight of roots in non-infested soil (Table 2). The combination treatment of trifluralin +

Table 2. Navy bean growth in soil infested with Fusarium solani and treated with several herbicides.

Herbicide treatment (kg/ha)		Fresh weight plant		Dry weight shoot		Dry weight root	
		Non-infested Soil (g)[a]	Infested Soil (g)	Non-infested Soil (g)	Infested Soil (g)	Non-infested Soil (g)	Infested Soil (g)
Check		2.11 ef	1.54 cde	0.230 e	0.158 cd	0.228 ef	0.184 cde
EPTC	3.36	1.67 def	0.53 a	0.172 de	0.056 a	0.180 cde	0.049 a
Trifluralin	0.84	1.73 def	1.65 cde	0.182 de	0.166 cde	0.227 def	0.134 bc
Dinoseb	5.04	1.96 def	1.99 ef	0.204 de	0.207 de	0.288 fg	0.316 g
Chloramben	2.24	2.22 f	1.56 cde	0.205 de	0.163 cde	0.265 fg	0.159 cd
Alachlor	2.80	1.86 def	1.00 abc	0.177 de	0.106 abc	0.133 bc	0.052 a
Trifluralin + Dinoseb	0.84 5.04	1.66 def	1.64 cde	0.159 cd	0.163 cde	0.221 def	0.114 bc
Trifluralin + EPTC	0.84 3.36	1.36 bcd	0.967 ab	0.148 bcd	0.095 ab	0.145 bc	0.079 a

[a]Means within each weight parameter followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

dinoseb at 0.84 + 5.04 kg/ha significantly reduced shoot dry weight and alachlor at 2.80 kg/ha, significantly reduced root dry weight from that of the no herbicide check in non-infested soil. Plants grown in soil infested with Fusarium solani chlamydospores and treated with EPTC at 3.36 kg/ha, alachlor at 2.8 kg/ha and the combination of trifluralin + EPTC at 0.84 + 3.36 kg/ha showed significantly reduced plant and root weight from that of the infested soil check (Table 2). Fresh weight of plants grown in soil infested with Fusarium solani was reduced 27% from those grown in non-infested soil. EPTC treatments at 3.36 kg/ha, alachlor at 2.8 kg/ha and trifluralin + EPTC and 0.84 + 3.36 kg/ha and infested with Fusarium solani, reduced fresh weight 68, 46 and 29%, respectively from herbicide treatments in non-infested soil. These data supported field observations that EPTC interacted with the root rot organism to increase root rot severity. However, in the growth chamber studies, the interaction of chloramben with root rot was not observed. Alachlor interacted with root rot in the growth chamber studies, but not in field studies, indicating that the herbicide root rot interactions are dependent upon many factors and the conditions controlling the interaction may be different for each of the three herbicides.

Temperature. The effect of temperature on root rot infection of navy bean resulting from the interaction of EPTC and Fusarium solani was studied in growth chambers at 20, 25 and 30 C. The greatest growth of the navy bean plants was observed at 30 C

with no significant difference when grown in infested and non-infested soil in combination with any rate of EPTC (Table 3). At 25 C there was no reduction in plant growth when grown in Fusarium solani infested soil at recommended rates of EPTC application. However, 4.48 kg/ha of EPTC reduced root weight of the plants grown in infested soil compared to plants grown in non-infested soil. The temperature which was least favorable for plant growth resulted in the greatest growth reduction in the infested soil with or without the presence of herbicides. Soil infested with Fusarium solani and treated with EPTC at 2.24, 3.36 and 4.48 kg/ha reduced shoot and root growth significantly from that of each herbicide treatment in non-infested soil and from that of the infested soil check receiving no herbicide treatment (Table 3). The interaction of EPTC and Fusarium solani infection on navy bean was most severe at the temperature least ideal for plant growth. Plants growing under stress conditions are frequently more susceptible to disease infection.

Inoculum level. Navy bean fresh weight and dry weight were reduced more as the Fusarium solani inoculum level increased in the presence of EPTC. Shoot fresh weight was not reduced at 2.0×10^3 spores/gram, but at 6.0×10^3 and 1.0×10^4 spores/gram fresh weight was reduced 10 and 17%, respectively. The application of EPTC at 3.36 kg/ha to soil infested with 2.0×10^3, 6.0×10^3 and 1.0×10^4 spores/gram reduced navy bean fresh weight 22, 37 and 36%, respectively from that of the same rate of EPTC in non-infested soil (Table 4). These

Table 3. Growth of navy bean in soil infested or non-infested with Fusarium solani and treated with EPTC at three temperatures.

Inoculum	Herbicide treatment (kg/ha)	Temperature (C) 20 Fresh weight plant (g)[a]	20 Dry weight plant (g)	20 Dry weight root (g)	25 Fresh weight plant (g)	25 Dry weight plant (g)	25 Dry weight root (g)	30 Fresh weight plant (g)	30 Dry weight plant (g)	30 Dry weight root (g)
Non-infested	Check	1.64 c	0.208 c	0.177 c	1.96 a	0.281 a	0.251 a	3.57 a	0.329 a	0.260 a
Infested	Check	1.19 b	0.157 b	0.126 b	1.86 a	0.265 a	0.224 a	3.42 a	0.317 a	0.197 a
Non-infested	EPTC 2	1.66 c	0.184 bc	0.185 c	2.38 a	0.300 a	0.255 a	3.62 a	0.324 a	0.220 a
Infested	EPTC 2	0.70 a	0.085 a	0.085 a	2.07 a	0.260 a	0.230 a	3.69 a	0.334 a	0.180 a
Non-infested	EPTC 3	1.48 c	0.186 bc	0.138 b	2.26 a	0.265 a	0.249 a	3.63 a	0.336 a	0.215 a
Infested	EPTC 3	0.90 a	0.095 a	0.084 a	1.88 a	0.229 a	0.215 a	3.51 a	0.346 a	0.201 a
Non-infested	EPTC 4	1.43 bc	0.178 bc	0.168 c	2.31 a	0.233 a	0.224 a	3.32 a	0.325 a	0.185 a
Infested	EPTC 4	0.79 a	0.092 a	0.066 a	1.76 a	0.216 a	0.167 b	3.46 a	0.330 a	0.206 a

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 4. Effect of Fusarium solani inoculum level and EPTC on navy bean growth.

Herbicide treatment (kg/ha)	Fresh weight plant (g)[a]				Dry weight plant (g)				Dry weight root (g)			
	Inoculum level (spores/gram)				Inoculum level (spores/gram)				Inoculum level (spores/gram)			
	0	2.0 x 10^3	6.0 x 10^3	1.0 x 10^4	0	2.0 x 10^3	6.0 x 10^3	1.0 x 10^4	0	2.0 x 10^3	6.0 x 10^3	1.0 x 10^4
Check	4.18 j	3.94 hij	3.76 ghi	3.47 fg	0.557 f	0.519 ef	0.495 def	0.451 de	0.440 i	0.410 fg	0.421 gh	0.334 d
EPTC 2.28	4.02 ij	3.75 gh	2.64 d	2.63 d	0.561 f	0.536 ef	0.425 cde	0.402 cd	0.456 i	0.433 ghi	0.390 ef	0.330 cd
EPTC 3.36	3.35 f	2.62 d	2.12 bc	2.14 bc	0.527 ef	0.462 def	0.423 cde	0.314 abc	0.439 ghi	0.374 e	0.314 cd	0.283 b
EPTC 4.48	2.98 e	2.24 c	1.78 a	1.95 ab	0.449 de	0.384 bcd	0.270 a	0.283 ab	0.381 ef	0.309 bc	0.214 a	0.171 j

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

data indicate that the degree of growth reduction or disease severity from the EPTC Fusarium solani interaction was dependent upon the soil inoculum level.

Genetic resistance. A line of navy beans 'RRR041', moderately resistant to root rot, did not show reduced growth in soil infested with Fusarium solani and treated with EPTC at 2.24 and 3.31 kg/ha (Table 5). However, the 'Sanilac' variety, which is susceptible to root rot, showed reductions in both fresh and dry weight when grown in soil infested with Fusarium solani and treated with 3.36 kg/ha of EPTC. These data indicated that plants moderately tolerant to root rot development suffer less from the root rot-herbicide interaction.

Fusarium solani virulence. None of the herbicide treatments increased the virulence of Fusarium solani as shown in Table 6. Thus, the EPTC-Fusarium solani interaction could not be explained on this basis.

EPTC-induced navy bean predisposition to root rot. Navy bean seedlings grown in sand culture containing several herbicides and transferred after 10 days to soil infested with Fusarium solani chlamydospores, developed extensive root rot only if treated with EPTC (Table 7). Plants receiving the other treatments had from 8 to 19% of the hypocotyl tissue infected by Fusarium solani, whereas, the EPTC treatments had 42 to 84% infected tissue (Figure 2). These data suggest that EPTC may predispose the navy bean hypocotyls to Fusarium solani infection by altering the hypocotyl surface wax formations in a manner similar to EPTC alteration of wax

Table 5. Growth of 'Sanilac' (susceptible) and 'RRR041' (moderately susceptible) navy bean in soils treated with EPTC and infested with Fusarium solani.

Navy bean variety	Herbicide treatment kg/ha	Fresh weight/plant (g)[a]	Dry weight/plant (g)[a]
Sanilac	-	2.40 b	0.371 b
Sanilac	2	1.32 ab	0.225 ab
Sanilac	3	0.89 a	0.166 a
RRR041	-	3.30 b	0.462 b
RRR041	2	2.78 b	0.404 ab
RRR041	3	2.37 b	0.375 b

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Table 6. The virulence on navy bean of Fusarium solani chlamydospores grown on potato dextrose agar containing several herbicides.

Herbicide treatment	(M)	Diseased hypocotyl tissue[ab] (%)
Check		20.4 a
EPTC	10^{-5}	14.8 a
	10^{-6}	17.8 a
Trifluralin	10^{-5}	21.6 a
	10^{-6}	23.3 a
Chloramben	10^{-5}	24.0 a
	10^{-6}	21.8 a
Alachlor	10^{-5}	24.0 a
	10^{-6}	21.9 a

[a]Means followed by similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

[b]Hypocotyl tissue was washed and percent of diseased tissue was determined visually.

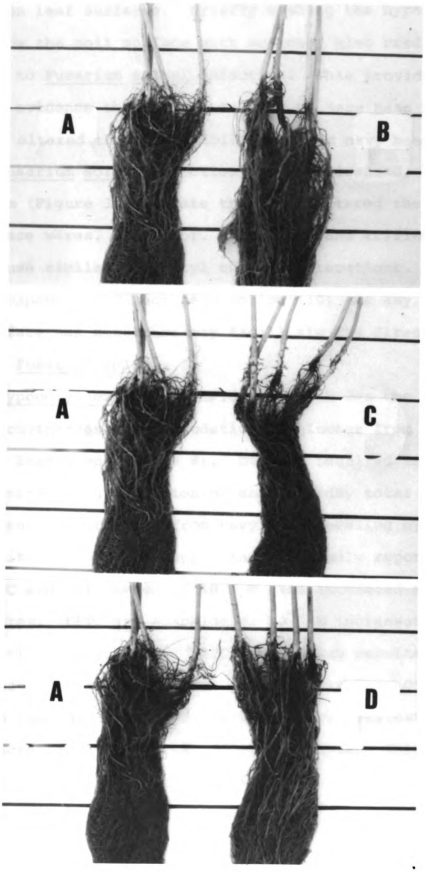
Table 7. The effect of navy bean pretreatment with several herbicides for 10 days on the subsequent root rot development after 30 days.

Treatment	(M)	Diseased hypocotyl tissue[ab] (%)
Check		8 a
Acetone		79 b
EPTC	5.0×10^{-5}	84 b
	1.0×10^{-5}	74 b
	1.0×10^{-6}	42 b
Dinoseb	1.0×10^{-5}	14 a
	1.0×10^{-6}	15 a
Alachlor	5.0×10^{-5}	19 a
	5.0×10^{-6}	12 a
Trifluralin	3.0×10^{-5}	19 a
	1.0×10^{-6}	15 a
Chloramben	5.0×10^{-5}	13 a
	1.0×10^{-5}	14 a

[a]Means followed by similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

[b]Hypocotyl tissue was washed and percent of diseased tissue was determined visually.

Figure 2. Predisposition of navy bean hypocotyl tissue to Fusarium solani. Plant (A) received no herbicide treatment prior to exposure to Fusarium solani chlamydospores. Plants (B) and (C) were pretreated with EPTC at 10^{-5} and 10^{-6} M, respectively. The hypocotyls of plants (D) were treated with acetone prior to exposure to the chlamydospores.


A
B
A
C
A
D

formation on leaf surfaces. Briefly washing the hypocotyl tissue below the soil surface with acetone, also predisposed the plants to Fusarium solani infection. This provides additional evidence that if the surface of navy bean hypocotyls are altered the susceptibility of the navy bean plant to Fusarium solani infection may be increased. 'SEM' micrographs (Figure 3) indicate that EPTC altered the hypocotyl surface waxes. Alachlor, chloramben and trifluralin did not cause similar hypocotyl surface alterations. Fusarium solani is capable of direct penetration (10) and any alteration in the surface wax structure may facilitate the direct penetration by Fusarium solani.

Root and hypocotyl exudates. EPTC at 10^{-6} M was the only herbicide which increased the exudation of glucose from germinating navy bean seeds (Table 8). Dinoseb resulted in the greatest increase in exudation of amino acids, total carbohydrates, and electrolytes from navy bean seedling hypocotyl and root tissue (Table 9), supporting previously reported data (28). EPTC and chloramben at 10^{-5} M also increased amino acid exudates. EPTC and alachlor at 10^{-5} M increased the exudation of carbohydrates. EPTC and dinoseb resulted in the greatest increase in efflux of electrolytes. Of the herbicides tested, EPTC and dinoseb have the greatest potential to increase the leakiness of plant tissue. This herbicide-induced increase in seed and root-hypocotyl exudation of cellular materials may increase chlamydospore germination and subsequent infection by Fusarium solani in the soil.

Figure 3. EPTC affect on navy bean hypocotyl surfaces. Plant (A) was treated with 10^{-6} M EPTC for 12 days and plant (B) 12-days-old without a herbicide treatment. Plant (C) was treated with EPTC at 10^{-6} M for 20 days and (D) received no herbicide treatment.

A
B
C
D

Table 8. Total carbohydrates exuded from navy bean seeds treated with several herbicides at 10^{-6} M.

Treatment	Total carbohydrates
	% of control[a]
Check	100 b
EPTC	139 c
Dinoseb	122 bc
Alachlor	116 bc
Fluorodifen	103 abc
Dinitramine	94 ab
Chloramben	94 ab
Trifluralin	68 a

[a]Means followed with similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

Table 9. Efflux of amino acids, total carbohydrates and electrolytes from 6-day-old navy bean roots and hypocotyls grown in the dark at 26 C then placed in herbicide solutions for 5 hours.

Herbicide treatment		Amino acids	Total carbohydrates	Electrolytes
		(% of control)[a]		
Dinoseb	1.0×10^{-5}	582 d	545 e	252 g
	1.0×10^{-6}	263 c	317 d	132 de
EPTC	1.0×10^{-5}	207 bc	169 c	176 f
	1.0×10^{-6}	114 a	118 ab	146 e
Chloramben	1.0×10^{-5}	227 bc	88 a	112 bc
	1.0×10^{-6}	107 a	101 ab	106 ab
Alachlor	1.0×10^{-5}	127 ab	172 c	110 bc
	1.0×10^{-6}	105 a	130 b	110 bc
Trifluralin	1.0×10^{-5}	86 a	85 a	125 cd
	1.0×10^{-6}	94 a	104 ab	94 a

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

Effect of herbicides on Fusarium solani. All herbicides tested decreased hyphal production in liquid culture, with dinoseb causing the greatest reduction in hyphal dry weight (Table 10). Examinations of hyphal structure indicated that no herbicides affected the hyphal development except dinoseb (Figure 4). Hyphae produced in the dinoseb treatment lysed and failed to form chlamydospores, which are the resting spores in the soil. Although dinoseb may indirectly increase the available energy source for chlamydospore germination, it inhibits normal development of the organism, partially explaining why the increased exudates resulting from the dinoseb treatment did not increase incidence in field and growth chamber experiments.

Chlamydospore germination. None of the treatments with either formulated or technical herbicides increased Fusarium solani chlamydospore germination in natural non-autoclaved soil with or without the addition of glucose as an exogenous energy source (Table 11). Both formulated and technical dinoseb, decreased chlamydospore germination.

Discussion

The field and growth chamber studies indicate that the herbicides EPTC, chloramben and alachlor can, depending upon the environmental-host-pathogen complex, increase root rot development on navy bean. In these studies root rot severity was increased by chloramben in the field tests, and by alachlor in growth chamber experiments, while EPTC increased root rot

Table 10. *Fusarium solani* growth in potato dextrose broth containing several herbicides.

Herbicide treatment (M)		Hyphal production (mg/flask)
Check		745 b
Trifluralin	5.0×10^{-6}	648 ab
Fluorodifen	5.0×10^{-6}	664 ab
EPTC	5.0×10^{-6}	654 ab
Chloramben	5.0×10^{-6}	662 ab
Alachlor	5.0×10^{-6}	633 ab
Dinitramine	5.0×10^{-6}	678 ab
Dinoseb	5.0×10^{-6}	523 a

[a]Means followed by similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

Figure 4. Fusarium solani hyphal development in potato dextrose broth containing herbicides. Hyphal development in check (A), EPTC 10^{-5} M (B), chloramben 10^{-5} M (C), and dinoseb 10^{-5} M (D) at 480 X.

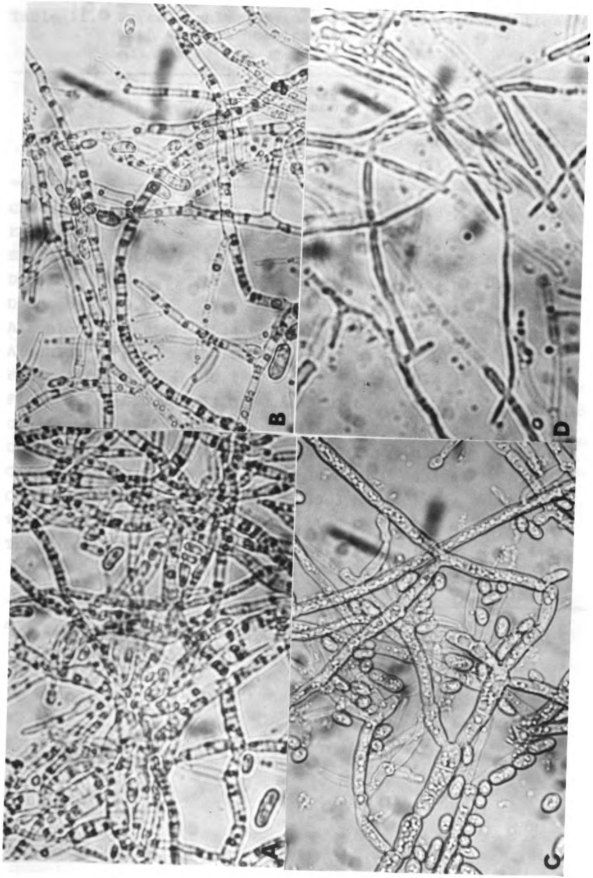

B
D
A
C

Table 11. Percent chlamydospore germination in soil treated with several herbicides.

Herbicide treatment (µg/g)		Soil amended with glucose		Natural soil	
		Technical (%)	Formulated (%)	Technical (%)	Formulated (%)
Check		36.5 b	35.4 b	2.3 a	2.4 a
EPTC	3.0	33.4 b	32.4 b	2.3 a	1.0 a
EPTC	6.0	34.3 b	33.2 b	0.8 a	1.5 a
Dinoseb	4.5	17.4 a	16.4 a	0.3 a	2.4 a
Dinoseb	9.0	11.9 a	10.0 a	0.3 a	1.5 a
Alachlor	3.0	35.4 b	34.2 b	1.4 a	2.3 a
Alachlor	6.0	34.8 b	34.8 b	1.9 a	1.5 a
Fluorodifen	3.0	36.4 b	34.8 b	2.4 a	1.9 a
Fluorodifen	6.0	37.0 b	34.7 b	1.8 a	2.5 a
Dinitramine	0.75	36.9 b	38.6 b	1.3 a	0.8 a
Dinitramine	1.5	37.2 b	32.0 b	0.9 a	0.9 a
Chloramben	3.0	33.4 b	34.6 b	0.5 a	0.6 a
Chloramben	6.0	34.7 b	33.3 b	1.3 a	1.5 a
Trifluralin	0.75	36.2 b	41.5 b	1.6 a	0.0 a
Trifluralin	1.5	37.7 b	40.0 b	1.4 a	1.4 a

[a]Means within columns followed by similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

severity in both field and growth chamber experiments. The degree of infection resulting from EPTC treatment was dependent upon temperature and inoculum level.

The EPTC effect on root rot severity was twofold: first, the increased leakage of cellular materials may increase chlamydospore germination, and secondly the alteration of hypocotyl tissue could increase the ease of hyphal penetration resulting in increased root rot severity. Fusarium solani chlamydospore (17) germination is nutrient dependent and penetration or infection is by direct penetration of hypocotyl tissue or through stomatal openings (10) and EPTC has been found to alter both of these relationships.

The observed dinoseb inhibition of Fusarium solani growth and development in soil and liquid culture may have some application to the field situation. Dinoseb is applied as a preemergence treatment. Barrons[2] (3) showed that roots of plants germinating near the soil surface appeared to be markedly affected by dinoseb, while deeper planting showed less injury. This indicates that the primary shoot apparently did not absorb toxic quantities as it pushed up through the soil. For dinoseb to increase the exudation of cellular materials, the roots must come in contact with it, due to the lack of translocation. Dinoseb inhibition of root rot development could occur if dinoseb was leached to the root zone or if it reached the root zone as a vapor. Under conditions of

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[2]Barrons, K. C. 1950. The relative toxicity of certain phenolic derivatives to the roots of major crop and weed plants. Ph.D. thesis, Mich. State Univ., E. Lansing. 99 p.

shallow planting, porous soil or heavy rainfall, dinoseb may play a role in root rot development due to its direct contact with the roots.

There are reports showing that dinoseb as a preemergence treatment can affect disease development and subsequent yields (12). Chappel and Miller (7) showed in laboratory experiments that the growth of Sclerotium rolfsii Sacc. was greatly retarded when inoculated on culture media containing field rate concentrations of dinoseb. They also recorded the incidence of stem rot on four farms where dinoseb was applied preemergence at 10 kg/ha concluded that dinoseb can influence disease development in peanuts (Arachis hypogaea L.).

Literature Cited

1. Alexander, J. V., J. A. Bourret, A. H. Gold and W. C. Snyder. 1966. Industion of chlamydospore formation by Fusarium solani in sterile soil extracts. Phytopathol. 56:353-354.

2. Altman, J. 1969. Predisposition of sugar beets to Rhizoctonia damping off with herbicides. Phytopathol. (Abstr). 59:1015.

3. Barrons, K. C. 1948. Preemergence weed control. Down to Earth. 4:(3)2-4.

4. Burkholder, W. H. 1920. The effect of two soil temperature on the yield and water relations of healthy and diseased bean plants. Ecology. 1:113-123.

5. Burkholder, W. H. 1924. The effect of varying soil moistures on healthy bean plants and on those infected by a root parasite. Ecology. 5:179-187.

6. Chandler, J. M. and P. W. Santelmann. 1968. Interactions of four herbicides with Rhizoctonia solani on seedling cotton. Weed Sci. 18:453-456.

7. Chappell, W. E. and L. I. Miller. 1956. The effects of certain herbicides on plant pathogens. Plant Dis. Rep. 40:52-57.

8. Cook, R. J. and W. C. Snyder. 1965. Influence of host exudates on growth and survival of germlings of Fusarium solani f. phaseoli in soil. Phytopathol. 55:1021-1025.

9. Cook, R. J. and N. T. Flentze. 1967. Chlamydospore germination and germling survival of Fusarium solani f. pisi in soil as affected by soil water and pea seed exudation. Phytopathol. 57:178-182.

10. Christou, T. and W. C. Snyder. 1962. Penetration and host-parasite relationships of Fusarium solani f. phaseoli in the bean plant. Phytopathol. 52:219-226.

11. Davis, D. and A. E. Dimond. 1953. Inducing disease resistance with plant growth regulators. Phytopathol. 43:137-140.

12. Garren, K. H. 1959. An evaluation of role of dinoseb in "non-dirting" control for peanut stem rot. Plant Dis. Rep. 43:665-667.

13. Hsia, Yu-Tien and J. J. Christensen. 1951. Effect of 2,4-D on seedling blight of wheat caused by Helminthosporium sativum. Phytopathol. 41:1011-1020.

14. Katan, J. and Y. Eshel. 1973. Interactions between herbicides and plant pathogens. Residue Reviews. 45:145-177.

15. Keyworth, W. C. and A. E. Dimond. 1952. Root injury as a factor in the assessment of chemotherapeutants. Phytopathol. 42:3-1-315.

16. Lai, M. T. and G. Semeniuk. 1970. Picloram induced increase of carbohydrate exudation from corn seedlings. Phytopathol. 60:563-564.

17. Lockwood, J. L. 1964. Soil fungistasis. Annual Review Phytopathol. 2:341-362.

18. Michigan Agricultural Statistics. 1960-1973. Michigan Crop Reporting Service.

19. Moore, S. and W. H. Stein. 1948. Photometric ninhydrin method for use in the chromatography of amino acids. J. Biol. Chem. 176:367-388.

20. Nash, S. M., T. C. Christou and W. C. Snyder. 1961. Existence of Fusarium solani f. phaseoli as chlamydospores in soil. Phytopathol. 51:302-312.

21. Neubauer, R. and Z. Avizohar-Hershenson. 1973. Effect of the herbicide, trifluralin, on Rhizoctonia disease in cotton. Phytopathol. 63:651-652.

22. Peeples, J. L. and E. A. Curl. 1969. Effect of paraquat, EPTC, and trifluralin on growth of Fusarium oxysporum, f. sp. vasinfectum in liquid culture. Phytopathol. (Abstr). 59:117.

23. Pinckard, J. A. and L. C. Standifer. 1966. An apparent interaction between cotton herbicide injury and seedling blight. Plant Dis. Rep. 50:172-174.

24. Rodriguez-Kabana, R., E. A. Curl and H. H. Funderburk, Jr. 1969. Effect of trifluralin on growth of Sclerotium rolfsii in liquid culture and soil. Phytopathol. 59:223-228.

25. Rodriguez-Kabana, R., E. A. Curl and J. L. Peeples. 1970. Growth response of Sclerotium rolfsii to the herbicide EPTC in liquid culture and soil. Phytopathol. 60:431-436.

26. Schroth, M. N. and W. C. Snyder. 1961. Effect of host exudates on chlamydospore germination of the bean root fungus, Fusarium solani f. phaseoli. Phytopathol. 51:389-393.

27. Standifer, L. C., Jr., D. R. Melville and S. A. Phillips. 1966. A possible interaction between herbicidal injury and the incidence of seedling disease in cotton plantings. Proc. S. Weed Conf. 19:126-127.

28. St. John, J. B. and J. L. Hilton. 1973. Lipid metabolism as a site of herbicide action. Weed Sci. 21:477-480.

29. Tang, A., E. A. Curl and R. Rodriguez-Kabana. 1970. Effect of trifluralin on inoculum density and spore germination of Fusarium oxysporum f. sp. vasinfectum in soil. Phytopathol. 60:1082-1086.

30. Trevelyan, W. E. and J. S. Harrison. 1952. Studies on yeast metabolism. I. Fractionation and microdetermination of cell carbohydrates. Biochem. J. 50:298-303.

31. Upstone, M. E. and J. C. Davis. 1967. The effect of simazine on the incidence of American gooseberry mildew on black currants. Plant Pathol. 16:68-69.

32. Van Der Zweep, W. 1970. Effects of herbicides on susceptibility of plants to pests and diseases. Proc. Brit. Weed Control Conf. 10:917-919.

33. Yemm, E. W. and A. J. Willis. 1954. The estimation of carbohydrates in plant extracts by anthrone. Biochem. J. 57:508-514.

34. Zaumeyer, W. J. and H. R. Thomas. 1957. A monographic study of bean diseases and methods for their control. U.S. Dept. Agr. Bul. No. 868.

CHAPTER 5

SUMMARY AND CONCLUSIONS

Field, greenhouse and laboratory studies on navy bean were initiated to determine the effect of disease, cultural, and environmental factors and their interaction with EPTC and several other herbicides.

Navy bean was found to be more sensitive to EPTC when grown under high or low moisture stress than plants grown under optimum soil moisture when plants are more vigorous and less sensitive to EPTC.

Navy bean grown in compacted soil had greatly reduced plant growth. EPTC at 4.48 kg/ha significantly reduced plant growth equally at all levels of compaction.

Increased planting depths resulted in decreased navy bean growth. Growth was reduced 71% when treated with 4.48 kg/ha of EPTC at the 7.6 cm planting depth. Reduced seedling vigor and increased emergence time may have allowed the emerging hypocotyl tissue additional time to absorb larger quantities of EPTC.

Ambient temperature variation between 20 and 30 C did not alter navy bean injury from EPTC.

Plants grown in soil supplemented with nitrogen produced more fresh weight. However, EPTC treatment at 3.36 and 4.48 kg/ha nullified this increase in fresh weight. The increased water loss from navy bean plants grown on soil treated with EPTC may have been the reason for the failure of nitrogen to increase navy bean fresh weight.

Navy bean plants grown from mechanically damaged seed were more sensitive to EPTC in greenhouse and field experiments. Both shoot height and grain yield of plants grown from damaged navy bean seed were significantly reduced by 3.36 and 4.48 kg/ha EPTC treatment. EPTC at 3.36 kg/ha reduced growth of navy bean grown from damaged seed at all planting depths. Plants which developed from undamaged navy bean seeds were only reduced at the 7.6 cm planting depth by EPTC at 3.36 kg/ha. Thus, seed quality and planting depth decrease the vigor of the navy bean aeedlings and reduce tolerance to EPTC.

The interaction of EPTC and atrazine on navy bean was studied in field and laboratory experiments. EPTC and low levels of atrazine interacted with EPTC to cause extensive leaf chlorosis, greater than that observed for atrazine alone. The synergistic interaction also resulted in dalayed maturity, decreased flower set, and yield reductions. The effects were all greater than an additive affect from each herbicide applied alone.

EPTC was shown to increase water loss from the navy bean plants. Plants treated with 3.36 kg/ha of EPTC showed extensive wind blast injury under hot and dry field conditions.

This increased water loss could be the result of alterations in leaf wax structure and quantity caused by EPTC treatment. The increased transpiration rate caused by EPTC treatment resulted in increased uptake of ^{14}C-atrazine, which is passively absorbed and moves in the transpiration stream. The increased uptake of atrazine could account for the increased atrazine injury in the presence of EPTC.

Root rot severity was increased and yields were reduced by EPTC at 3.36 and 4.48 kg/ha applied to soil with an added artificial root rot population. EPTC at 3.36 kg/ha and chloramben at 3.36 kg/ha caused the greatest increase in root rot severity and yield reduction in field experiments. EPTC at 3.36 kg/ha and alachlor at 2.80 kg/ha applied to soil infested with Fusarium solani also reduced root and shoot growth in growth chamber experiments at 24 C.

The largest increase in root rot severity resulting from applications of EPTC was observed at 20 C compared to 25 and 30 C. At the higher temperature, less EPTC may have been present in the soil, due to volatilization.

The EPTC-Fusarium solani interaction was increased as the chlamydospore inoculum level increased. A reduction in the inoculum level by crop rotations or other means could decrease the potential of the EPTC-Fusarium solani interaction in the field. A moderately root rot-resistant line of navy bean 'RRR041', did not show plant growth reductions when grown in soil infested with Fusarium solani and treated with EPTC.

None of the herbicides tested increased the virulence of Fusarium solani chlamydospores when grown on potato dextrose agar supplemented with herbicides.

Scanning electron micrographs of hypocotyl tissue indicated that EPTC was the only herbicide that altered the surface waxes. This alteration of hypocotyl tissue was related to increased susceptibility of navy bean to Fusarium solani. Plants grown in EPTC or plants with hypocotyls washed with acetone were found to be more susceptible to Fusarium solani.

EPTC was also found to increase the exudation of total carbohydrates from germinating navy bean seeds. EPTC and dinoseb caused increased exudation of electrolytes, amino acids and sugars from root and hypocotyl tissue. Increased plant exudates could result in increased germination of chlamydospores and cause increased disease infection.

Dinoseb was shown to reduce hyphal development in both soil and liquid culture. The dinoseb effect on exudate production may have been overcome by inhibition of Fusarium solani growth. However, EPTC stimulates the exudation of nutrients from tissue and does not inhibit Fusarium solani development. This could explain why EPTC increases root rot severity and dinoseb does not. None of the herbicides tested had a direct stimulatory affect on Fusarium solani chlamydospore germination or growth in soil or liquid culture.

APPENDICES

APPENDIX A

APPENDIX A

Modified No. 1 Hoagland's Solution

1.	1 M KH_2PO_4	2 ml/L
2.	1 M KNO_3	2 ml/L
3.	1 M $Ca(NO_3)_2 \cdot 4H_2O$	3 ml/L
4.	1 M $MgSO_4 \cdot 7H_2O$	2 ml/L
5.	1.5 g/L $MnCl_3 \cdot 4H_2O$	
	2.5 g/L H_3BO_4	
	0.1 g/L $ZnCl_2$	1 ml/L
	0.05 g/L $CuCl_2 \cdot 2H_2O$	
	0.05 g/L MoO_3	
6.	26.3 g/L Sequestrene^R	1 ml/L
	pH 6.5 to 6.8 with 1 M NaOH	

APPENDIX B

APPENDIX B

Table B-1. Total protein content of navy bean seeds treated with EPTC and atrazine - 1972.

Herbicide treatment (kg/ha)		Total protein/ gram seed (mg)
Check		260
EPTC	3.36	262
Atrazine	0.56	252
Atrazine	0.14	262
Atrazine	0.07	255
Atrazine + EPTC	0.56 + 3.36	263
Atrazine + EPTC	0.14 + 3.36	263

APPENDIX B

Table B-2. Efflux of electrolytes from navy bean root and hypocotyl tissue treated with several herbicides.

Herbicide treatment (M)	Conductance (μ mhos/g)[a] time - (hr)						
	1	2	3	4	5	6	7
Check	0.33 e	1.62 gh	1.78 c	1.84 hi	1.91 ig	2.11 g	2.61 de
DNBP 10^{-4}	1.42 a	5.49 a	8.59 a	12.59 a	13.51 a	24.52 a	42.42 a
DNBP 10^{-5}	0.65 cd	3.40 b	3.93 b	4.71 c	4.81 c	5.53 c	14.78 b
DNBP 10^{-6}	0.77 bc	1.88 efg	2.13 c	2.30 efg	2.52 ef	2.71 ef	2.89 de
EPTC 10^{-4}	0.76 bc	2.48 c	2.85 bc	2.50 e	2.74 e	3.31 de	5.26 c
EPTC 10^{-5}	0.69 cd	2.51 c	2.93 bc	3.02 d	3.37 d	3.51 d	3.41 cde
EPTC 10^{-6}	0.56 d	2.22 de	2.38 c	2.47 e	2.78 e	2.85 de	3.10 de
Chloramben 10^{-4}	0.86 b	2.26 d	5.52 b	5.75 b	5.93 b	6.08 b	6.26 c
Chloramben 10^{-5}	0.87 b	2.03 defg	2.10 c	2.10 fgh	2.14 ghi	2.17 fg	2.19 e
Chloramben 10^{-6}	0.86 b	1.76 ef	1.91 c	1.97 h	2.02 hij	2.08 g	2.43 de
Alachlor 10^{-4}	0.62 cd	2.04 defg	2.23 c	2.36 ef	2.36 fg	2.30 fg	3.55 cde
Alachlor 10^{-5}	0.67 cd	1.90 efg	2.07 c	2.08 fgh	2.10 ghi	1.95 g	2.45 de
Alachlor 10^{-6}	0.68 cd	1.94 efg	1.97 c	2.01 fgh	2.11 ghi	2.14 fg	2.45 de
Trifluralin 10^{-4}	0.74 bc	2.06 def	2.15 c	2.19 efgh	2.45 fgh	2.30 fg	2.69 de
Trifluralin 10^{-5}	0.74 bc	1.93 efg	2.10 c	2.19 efgh	2.39 fg	2.41 fg	2.63 de
Trifluralin 10^{-6}	0.67 cd	1.43 h	1.51 c	1.60 i	1.78 j	1.85 g	2.49 de

[a]Means within columns followed by similar letters are not significantly different at the 5% level of Duncan's Multiple Range Test.

APPENDIX B

Table B-3. Efflux of electrolytes from navy bean root and hypocotyl tissue pretreated with several herbicides.

Herbicide treatment (M)		Conductance (μ mhos/g)[a] time - (hr) 1	2	3	4	5	6
Check		1.00 cd	1.13 f	1.36 h	1.95 h	2.68 ef	5.22 gh
DNBP	10^{-4}	3.27 b	4.47 b	6.18 a	8.14 a	9.96 a	19.25 a
DNBP	10^{-5}	1.24 c	2.42 c	3.20 b	4.07 c	4.81 b	11.08 b
DNBP	10^{-6}	1.14 cd	1.68 de	2.48 cd	3.25 d	3.84 cd	7.62 de
EPTC	10^{-4}	1.01 cd	1.80 d	2.46 cd	3.18 de	3.80 cd	9.01 c
EPTC	10^{-5}	1.12 cd	1.67 de	2.71 bc	2.95 def	3.22 de	7.60 de
EPTC	10^{-6}	0.92 cd	1.40 def	1.97 defg	2.20 gh	2.55 ef	6.15 fg
Chloramben	10^{-4}	8.86 a	6.15 a	6.10 a	6.00 b	4.22 bc	8.13 cd
Chloramben	10^{-5}	0.86 d	1.52 def	2.18 def	2.42 a	2.95 ef	3.96 i
Chloramben	10^{-6}	1.13 cd	1.18 f	1.52 gh	1.97 h	2.86 ef	5.32 fgh
Alachlor	10^{-4}	1.01 cd	1.10 f	1.65 fgh	2.61 efg	2.92 ef	4.52 hi
Alachlor	10^{-5}	1.08 cd	1.51 def	1.81 efgh	2.13 gh	3.07 def	4.81 hi
Alachlor	10^{-6}	0.87 cd	1.47 def	1.86 efgh	2.36 gh	2.35 f	4.74 hi
Trifluralin	10^{-4}	1.05 cd	1.44 def	2.24 cde	2.55 fg	2.97 ef	6.42 ef
Trifluralin	10^{-5}	1.18 cd	1.25 ef	1.95 defg	2.54 fg	3.07 def	4.98 ghi
Trifluralin	10^{-6}	1.04 cd	1.15 f	1.69 fgh	2.25 gh	2.93 ef	5.24 fgh

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

APPENDIX B

Table B-4. Efflux of amino acids and total carbohydrates from navy bean roots and hypocotyl tissue treated with several herbicides.

Herbicide treatment (M)	Amino acids (mg/g)[a] time - (hr) 1	2	4	5	Total carbohydrates (mg/g) time - (hr) 1	2	4	5
Check	0.78 d	0.91 c	0.95 cd	1.10 e	1.08 d	2.23 h	2.40 h	3.60 ef
EPTC 10^{-4}	0.94 cd	1.20 c	1.98 cd	3.35 c	2.08 a	6.23 b	7.25 c	8.45 c
EPTC 10^{-5}	0.88 cd	0.99 c	1.45 cd	2.28 c	1.78 ab	3.35 ef	3.95 efg	6.08 d
EPTC 10^{-6}	0.81 cd	0.99 e	1.28 cd	1.25 e	1.23 cd	2.45 gh	3.20 fgh	4.28 ef
Trifluralin 10^{-4}	0.81 cd	0.92 c	0.93 d	0.98 e	1.58 bc	2.4 gh	2.35 h	3.43 ef
Trifluralin 10^{-5}	0.81 cd	0.82 c	0.87 d	0.95 e	2.10 ab	2.38 gh	2.58 gh	3.05 f
Trifluralin 10^{-6}	0.85 cd	0.87 c	0.93 d	1.03 e	1.83 ab	2.08 h	2.93 fgh	3.73 ef
DNBP 10^{-4}	8.00 a	23.90 a	36.00 a	51.30 a	1.70 abc	8.58 a	13.05 a	21.13 a
DNBP 10^{-5}	4.20 b	4.95 b	5.35 b	6.40 b	1.70 abc	8.33 a	12.15 ab	19.63 a
DNBP 10^{-6}	1.40 c	1.85 c	1.93 cd	2.60 c	1.75 abc	3.13 fg	8.83 bc	11.43 b
Chloramben 10^{-4}	0.82 cd	0.91 c	2.50 c	2.80 c	1.03 d	2.33 gh	2.75 gh	3.15 f
Chloramben 10^{-5}	0.84 cd	1.43 c	1.53 cd	2.50 cd	1.68 abc	2.48 fgh	2.48 h	3.15 f
Chloramben 10^{-6}	0.73 d	0.81 c	0.87 d	1.18 e	1.20 cd	1.83 h	2.55 h	3.65 ef
Alachlor 10^{-4}	0.75 d	0.94 c	2.13 cd	2.60 c	2.18 a	5.13 cd	5.23 cd	6.08 d
Alachlor 10^{-5}	0.76 d	0.91 c	0.98 d	1.40 de	2.03 ab	4.48 d	5.48 d	6.20 d
Alachlor 10^{-6}	0.83 cd	0.95 c	1.15 cd	1.16 e	2.10 ab	4.03 e	4.33 def	4.68 e

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

APPENDIX B

Table B-5. Effect of EPTC on Fusarium solani chlamydospore germination in the spermosphere of navy bean after 48 hours in soil at 50% moisture.[a]

Treatment (µg/g)	Distance from seed (mm)			
	0-1	2-3	4-5	6-7
	(percent germination)[b]			
Check	56.5	28.5	0	0
EPTC 3.0	58.9	37.0	9.5	0

[a]The method used to determine chlamydospore germination was developed by: Short, G. E. and M. L. Lacy. 1974. Germination of Fusarium solani f. sp. pisi chlamydospores in the spermosphere of pea. Phytopathol. 64:558-562.

[b]Means are the average of 6 replications of one experiment.

APPENDIX B

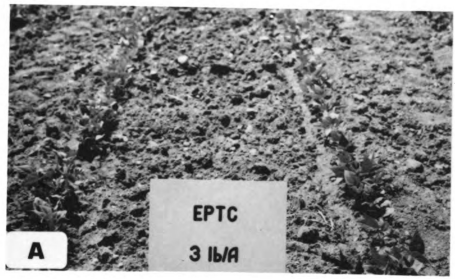
Table B-6. The effect of EPTC and Zn levels on navy bean yields.

EPTC treatment (kg/ha)	Zn level (kg/ha) 0 Yield (kg/ha)[a]	2.24 Yield (kg/ha)	4.48 Yield (kg/ha)
Check	474.9 a	2105.7 b	2173.0 b
EPTC 3.36	444.2 a	2231.4 b	2218.2 b
EPTC 4.48	560.7 a	2421.4 b	2047.3 b

[a]Means within columns followed by similar letters are not significantly different at the 5% level by Duncan's Multiple Range Test.

APPENDIX B

Figure B-1. EPTC-atrazine interaction at East Lansing, Michigan, 1972. Treatment (A) EPTC at 3.36 kg/ha (B) atrazine 0.56 kg/ha and (C) EPTC and atrazine at 3.36 + 0.56 kg/ha.


EPTC
3 lb/A
A

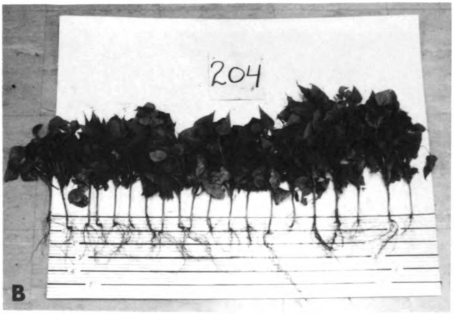

ATRAZINE
1/2 lb/A
B


ATRAZINE 1/2 lb/A
+
EPTC 3 lb/A
C

APPENDIX B

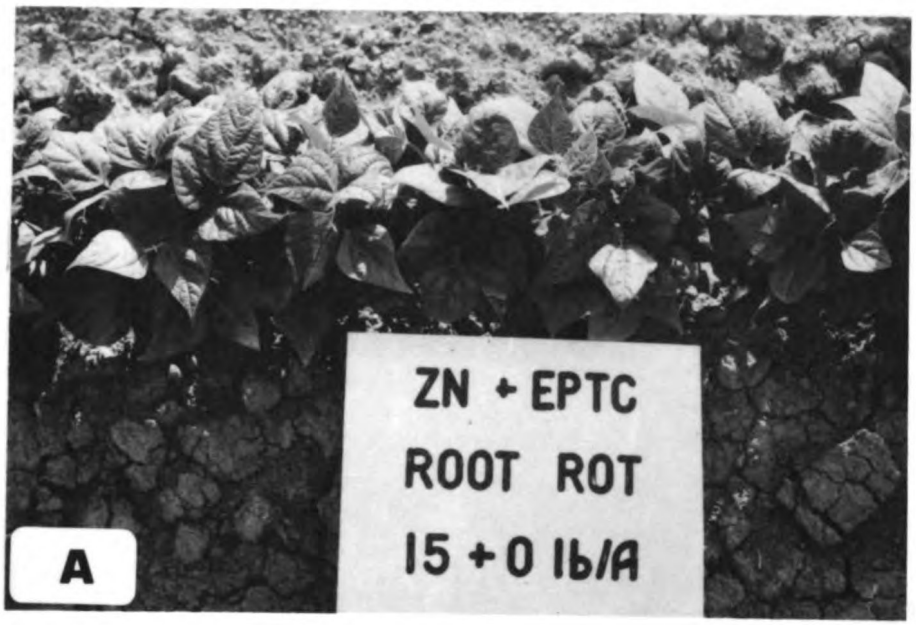
Figure B-2. EPTC-root rot interaction, 1972. Plants (A) were grown in soil with a natural root rot population. Plants (B) were grown in soil with a natural root rot population and treated with EPTC at 3.36 kg/ha.

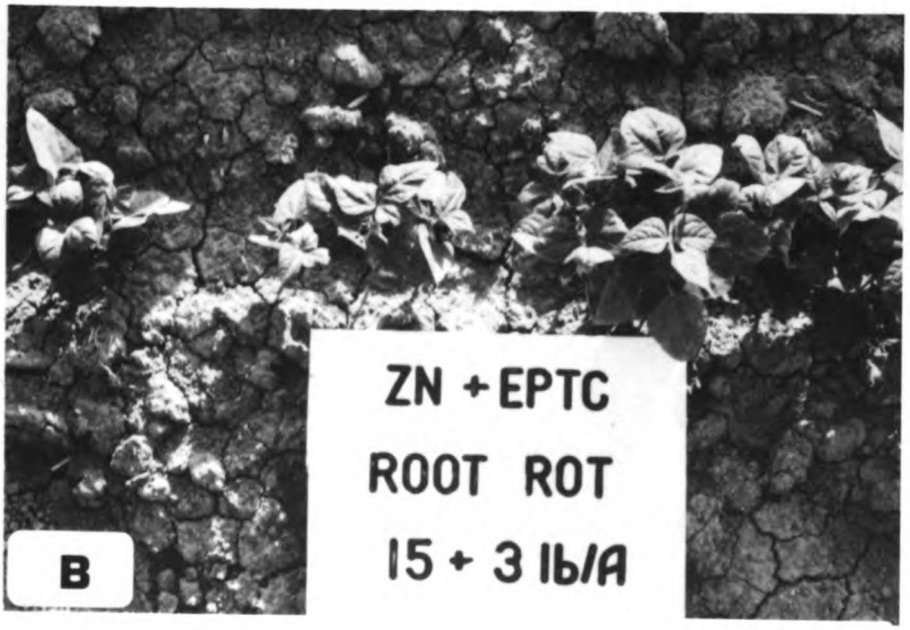
A



APPENDIX B

Figure B-3. EPTC-root rot interaction, 1972. Plants (A) grown in soil with a natural root rot population. Plants (B) were grown in soil with a natural root rot population and treated with 3.36 kg/ha of EPTC.


ZN + EPTC
ROOT ROT
15 + 0 lb/A
A

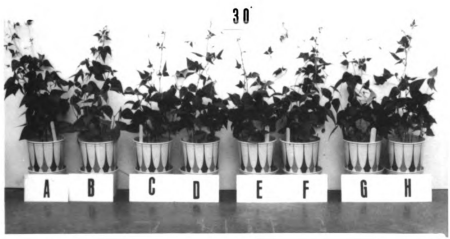

ZN + EPTC
ROOT ROT
15 + 3 lb/A
B

APPENDIX B

Figure B-4. EPTC-*Fusarium solani* interaction in a growth chamber study at 20, 25 and 30 C. At each temperature A, C, E and G plants were grown in non-infested soil with EPTC at 0, 2.24, 3.36 and 4.48 kg/ha. Plants B, D, F and H were grown in soil infested with *Fusarium solani* and treated with 0, 2.24, 3.36 and 4.48 kg/ha of EPTC, respectively.

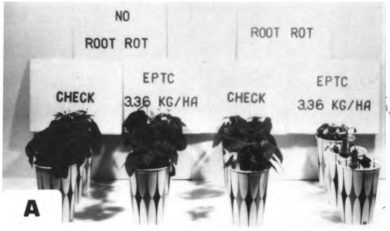

20°
A
B
C
D
E
F
G
H

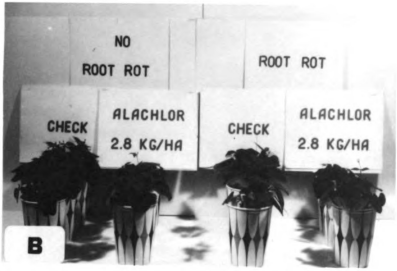

25°
A
B
C
D
E
F
G
H

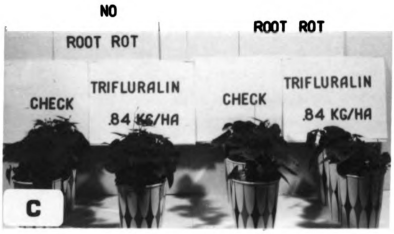

30°
A
B
C
D
E
F
G
H

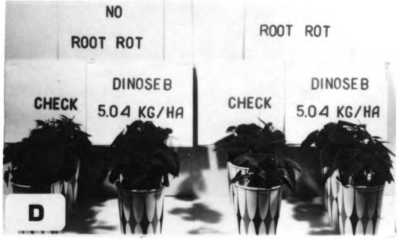
APPENDIX B

Figure B-5. Navy bean growth in non-infested soil or soil infested with Fusarium solani and treated with several herbicides. Plants were treated with (A) EPTC at 3.36 kg/ha (B) alachlor at 2.80 kg/ha (C) trifluralin at 0.84 kg/ha and (D) dinoseb at 5.04 kg/ha.


NO
ROOT ROT
ROOT ROT
CHECK
EPTC
3.36 KG/HA
CHECK
EPTC
3.36 KG/HA
A

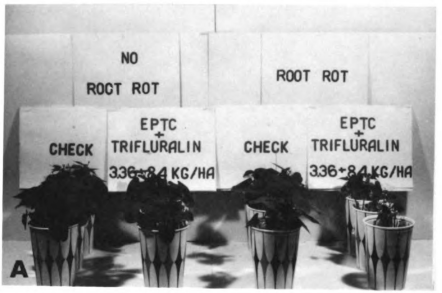

NO
ROOT ROT
ROOT ROT
CHECK
ALACHLOR
2.8 KG/HA
CHECK
ALACHLOR
2.8 KG/HA
B


NO
ROOT ROT
ROOT ROT
CHECK
TRIFLURALIN
.84 KG/HA
CHECK
TRIFLURALIN
.84 KG/HA
C


NO
ROOT ROT
ROOT ROT
CHECK
DINOSEB
5.04 KG/HA
CHECK
DINOSEB
5.04 KG/HA
D

APPENDIX B

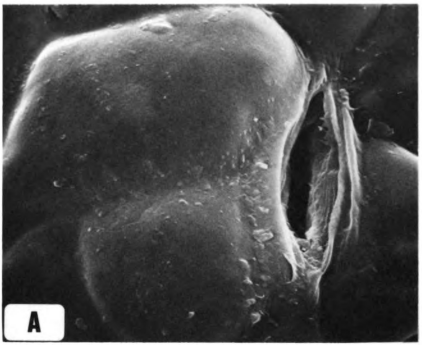
Figure B-6. Navy bean growth in non-infested soil or soil infested with Fusarium solani and treated with two herbicides. Plants were treated with (A) EPTC + trifluralin and (B) chloramben.

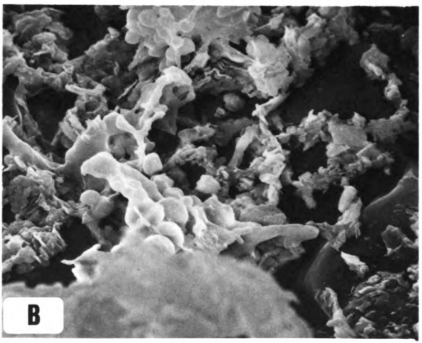

NO
ROOT ROT
ROOT ROT
CHECK
EPTC
+
TRIFLURALIN
3.36+8.4 KG/HA
CHECK
EPTC
+
TRIFLURALIN
3.36+8.4 KG/HA
A


NO
ROOT ROT
ROOT ROT
CHECK
CHLORAMBEN
2.24 KG/HA
CHECK
CHLORAMBEN
2.24 KG/HA
B

APPENDIX B

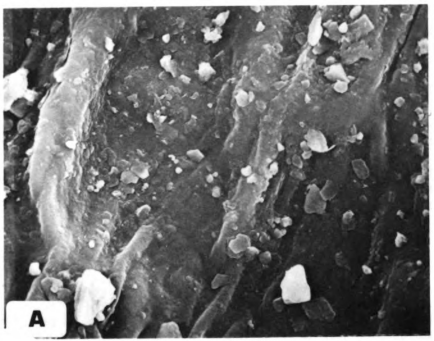
Figure B-7. Electron micrographs of 18-day-old navy bean hypocotyls treated with (A) chloramben at 10^{-5} M and trifluralin at 10^{-5} M.

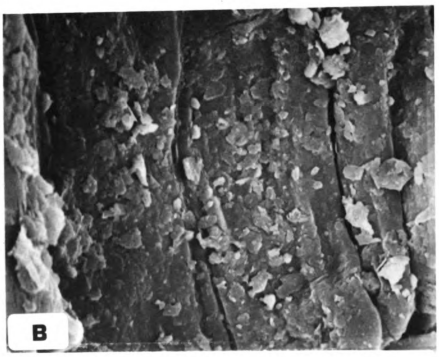

A


B

APPENDIX B

Figure B-8. Electron micrograph of 30-day-old navy bean hypocotyls at 5000 X. Plant (A) was grown in EPTC at 10^{-6} M while plant (B) received no herbicide treatment.


A


B

LIST OF REFERENCES

LIST OF REFERENCES

1. Alexander, M. 1961. Introduction to Soil Microbiology. New York. Wiley and Sons. 472 p.

2. Allison, L. E. 1952. Effect of synthetic polyelectrolytes on the structure of saline and alkali soils. Soil Sci. 73:443-454.

3. Altman, J. 1969. Predisposition of sugar beets to Rhizoctonia solani damping-off with herbicides. Phytopathol. (Abstr.) 59:1015.

4. Anderson, L. P. 1968. Histological and cytological responses of cotton to trifluralin and interactions with the damping-off pathogens. Ph.D. Thesis, Univ. of Georgia, Athens, Georgia. 92 p.

5. Appleby, A. P., W. R. Furtick and S. C. Fang. 1965. Soil placement studies with EPTC on Avena sativa. Weed Res. 5:115-122.

6. Asgrow Seed Company. 1949. A study of mechanical injury to seed beans. Asgrow monograph No. 1, Asgrow Seed Co., New Haven 2 Conn.

7. Ashton, F. M. 1963. Effect of EPTC on photosynthesis, respiration, and oxidative phosphorylation. Weeds. 11:295-297.

8. Ashton, F. M. and K. Dunster. 1961. The herbicidal effect of EPTC, CDEC, and CDAA on Echinochloa crusgalli with various depths of soil incorporation. Weeds. 9:312-317.

9. Ashton, F. M. and T. J. Sheets. 1959. The relationship of soil adsorption of EPTC to oats injury in various soil types. Weeds. 7:88-90.

10. Audus, L. J. 1964. Herbicides behavior in soil. II. Interaction with soil microorganisms. In L. J. Audus (ed.) The physiology and biochemistry of herbicides. New York Academic Press. 555 p.

11. Bainer, R. and H. A. Borthwick. 1934. Thresher and other mechanical injury to seed beans of the lima type. Calif. Agr. Exp. Sta. Bul. 580. 30 p.

12. Barriga-Solorio, C. 1959. Mechanical injury to pea bean seed treated at three moisture levels. M.S. Thesis, Mich. State Univ., E. Lansing. 98 p.

13. Beam, H. W. and E. A. Curl. 1971. Effect of fluomenturon and prometryne on Rhizoctonia solani in soil. Phytopathol. 61:884-885.

14. Beste, C. E. and M. M. Schreiber. 1972. RNA synthesis as the basis for EPTC and 2,4-D antagonism. Weed Sci. 20:8-11.

15. Bingham, S. W. and R. P. Upchurch. 1959. Some interactions between nutrient level (N, P, K, Ca) and diuron on the growth of cotton and Italian ryegrass. Weeds. 7:167-177.

16. Binning, L. K., D. Penner and W. F. Meggitt. 1971. The effect of 2-chloroethyl phosphonic acid on dicamba translocation in wild garlic. Weed Sci. 19:73-75.

17. Bollen, W. B. 1961. Interaction between herbicides and soil microorganisms. Ann. Rev. Microbiol. 15:69-89.

18. Bovey, R. W., F. S. Davis and H. L. Morton. 1968. Herbicide combinations for woody plant control. Weed Sci. 16:332-335.

19. Burkholder, W. H. 1919. The dry root-rot of the bean. Cornell Agr. Exp. Sta. Mem. 26:1003-1033.

20. Carns, A. 1934. Soil crust. Agri. Eng. 15:167-169.

21. Chandler, J. M. and P. W. Santelmann. 1968. Interactions of four herbicides with Rhizoctonia solani on seedling cotton. Weed Sci. 16:453-456.

22. Chappell, W. E. and L. T. Miller. 1957. The effect of certain herbicides on plant pathogens. Plant Dis. Rep. 40:52-56.

23. Chapra, B. K., E. A. Curl and R. Rodriguez-Kabana. 1970. Influence of prometryne in soil on growth-related activities of Fusarium oxysporum f. sp. vasinfectum. Phytopathol. 60:717-722.

24. Chatterju, P. 1958. The bean root rot complex in Idaho. Phytopathol. 48:197-200.

25. Christenson, D. 1974. Personal communication. Mich. State Univ.

26. Christenson, D. and A. E. Erickson. 1974. Mich. State Univ. Soil Sci. Rep. 2:6-7.

27. Christou, T. 1962. Penetration and host-parasite relationships of Thieleviopsis basicola in the bean plant. Phytopathol. 52:194-198.

28. Christou, T. 1962. Penetration and host-parasite relationships of Rhizoctonia solani in the bean plant. Phytopathol. 52:381-389.

29. Christou, T. and W. C. Snyder. 1962. Penetration and host-parasite relationships of Fusarium solani f. phaseoli in the bean plant. Phytopathol. 52:219-226.

30. Cohen, E., F. S. Lattar and R. Barkai-Golam. 1965. The effect of NAA, 2,4,5-T and 2,4-D on the germination in vitro of fungi pathogenic to fruits. Israel J. Agr. Research. 15:41-47.

31. Colby, S. R. 1968. Calculating synergistic and antagonistic responses of herbicide combinations. Weeds. 15:20-22.

32. Cook, R. L. 1938. Michigan State University unpublished data.

33. Cooperative Extension Service. 1968. Michigan dry edible bean problems. Michigan State University, Ext. Bul. No. E-629.

34. Crafts, A. S. 1939. The relation of nutrient to toxicity of arsenic, borate and chlorate in soils. J. Agr. Res. 58:637-671.

35. Crafts, A. S. and C. W. Cleary. 1936. Toxicity of arsenic, borate, chlorate, and their combinations in three California soils. Hilgardia. 10:401-412.

36. Crosier, W. 1942. Baldheads in beans, occurrence and influence on yields. Proc. Asn. Offic. Seed Anal. 34:118-123.

37. Curless, M. L. 1970. Response of corn hybrids and inbred lines to S-ethyl diisobutylthiocarbamate (butylate). M.S. Thesis, Mich. State Univ., E. Lansing. 61 p.

38. Dawson, J. H. 1963. Development of barnyardgrass seedlings and their response to EPTC. Weeds. 11:60-66.

39. Dawson, J. H. and V. F. Burns. 1959. Preemergence chemical control of annual weeds in field beans. Proc. Western Weed Contr. Conf. 39 p.

40. Davis, R. S., R. W. Bovey and M. G. Merkle. 1968. Effect of paraquat and 2,4,5-T on the uptake and transport of picloram in woody plants. Weed Sci. 16:336-339.

41. Davis, D. and A. E. Dimond. 1953. Inducing disease resistance with plant growth-regulators. Phytopathol. 43:137-140.

42. Davis, D. G. and K. E. Dusbabek. 1973. Effect of diallate on foliar uptake and translocation of herbicides in pea. Weed Sci. 21:16-18.

43. Delouche, J. C. and W. P. Caldwell. 1960. Seed vigor and vigor tests. Proc. Assn. Offic. Seed Anal. 50:124-129.

44. Devisetty, B. N. and R. G. Harvey. 1974. Depth of planting as a factor influencing corn injury from alachlor. Weed Sci. Soc. Amer. Abstr. No. 17.

45. Devlin, R. M. and R. W. Yaklich. 1972. Influence of two phenoxy growth regulators on the uptake and accumulation of naptalam by bean plants. Plant Physiol. 27: 317.

46. Dewey, O. R., P. Gregory and R. K. Pfieffer. 1956. Factors affecting the susceptibility of peas to selective dinitro-herbicides. Proc. Brit. Weed Control Conf. 1:313-314.

47. Dexter, A. G. 1971. Weed control and crop injury from herbicide combinations used in sugar beets. Proc. N. Cent. Weed Contr. Conf. 26:60.

48. Dickerson, C. T., Jr. and R. D. Sweet. 1968. Atrazine, oil and 2,4-D for postemergence weed control. Proc. Northeast. Weed Contr. Conf. 22:64.

49. Doll, J. D. 1969. The influence of nutrient level and combination on herbicide uptake and phytotoxicity. Ph.D. Thesis, Mich. State Univ., E. Lansing. 70 p.

50. Drew, L. O. and W. F. Buckele. 1964. Emergence force of plants. Proc. Am. Soc. Engr. Trans. Paper No. 62-641.

51. Duke, W. B., V. S. Rao and J. F. Hunt. 1972. EPTC-atrazine residue interaction effect on seedling alfalfa varieties. Proc. Northeast. Weed Sci. Soc. 26:258.

52. Erickson, A. E. 1974. Michigan State University unpublished data.

53. Erickson, A. E. and D. M. VanDoren. 1960. The relation of plant growth and yield to soil oxygen availability. Trans. 7th International Congress. Soil Sci. 3:428-434.

54. Erickson, L. C., T. A. DeWolf and B. L. Brannaman. 1958. Growth of some citrus fruit pathogens as affected by 2,4-D and 2,4,5-T. Bot. Gaz. 120:31-36.

55. Edwards, F. E. 1966. Cotton seedling emergence. Mississippi Farm Res. 29(11):4-5.

56. Fang, S. C., P. Theisen and V. H. Freed. 1961. Effects of water evaporation, temperature and rates of application on the retention of ethyl-N-N-di-n-propylthiocarbamate on various soils. Weeds. 9:569-574.

57. Finney, D. J. 1947. Probit analysis. 2 ed. Cambridge, Mass. Cambridge Univ. Press. 168 p.

58. Fink, R. J., O. H. Fletchall and O. H. Calvert. 1968. Relation of triazine residues to fungal and bacterial colonies. Weed Sci. 16:104-105.

59. Fletcher, W. W. 1960. The effect of herbicides on soil microorganisms. In, E. K. Woodland and G. R. Sagar (eds.), Herbicides and the soil. Oxford. Blackwell.

60. Fletcher, W. W. 1966. The effect of herbicides on soil microorganisms. Proc. Brit. Weed Contr. Conf. 3:896.

61. Foote, L. E. and B. R. Churchill. 1962. A study of chemical and cultural weed control treatments in navy, cranberry and kidney beans. Mich. Agr. Exp. Sta. Quar. Bul. 45:318-324.

62. Garett, S. D. 1970. Pathogenic root-infecting fungi. New York. Cambridge Univ. Press. 198 p.

63. Garner, T. H. and H. D. Bowen. 1966. Plant mechanics in seedling emergence. Soc. Agr. Engr. Trans. 9:605-633.

64. Garren, K. H. 1959. An evaluation of role of dinoseb in "non-dirting" control for peanut stem rot. Plant Dis. Rep. 43:665-667.

65. Gentner, W. A. 1966. The influence of EPTC on the external foliage wax deposition. Weed Sci. 14:27-30.

66. Gray, R. A. and A. J. Weierich. 1965. Factors affecting the vapor loss of EPTC from soils. Weeds. 13:141-147.

67. Gray, R. A. and A. J. Weierich. 1968. Leaching of five thiocarbamate herbicides in soils. Weed Sci. 16: 77-79.

68. Gray, R. A. and A. J. Weierich. 1969. Importance of root, shoot, and seed exposure on the herbicidal activity of EPTC. Weed Sci. 17:223-229.

69. Growing, D. P. 1959. A method of comparing herbicides and herbicide mixtures at the screening level. Weeds. 7:66-76.

70. Hanse, H. N. 1938. The dual phenomena in imperfect fungi. Mycologia. 30:442-445.

71. Hardenburg, E. V. 1972. Experiments with field beans. Cornell Univ. Agr. Exp. Sta. Bul. 776. 28 p.

72. Harris, W. L. 1971. The soil compaction process in compaction of agricultural soils. Amer. Soc. Agric. Eng. 471 p.

73. Herter, L. L. 1930. Thresher injury, a cause of baldhead in beans. J. Agr. Res. 40:371-384.

74. Hawxby, K., E. Basler and P. W. Santelmann. 1971. Temperature effects on absorption and translocation of trifluralin in peanut seedlings. Weed Sci. Soc. Amer. Abstr. No. 48.

75. Horner, C. E. 1965. Control of mint rust by propane gas flaming and contact herbicides. Plant Dis. Rep. 49:393-395.

76. Hsia, Yu-Tien and J. J. Christensen. 1951. Effect of 2,4-D on seedling blight of wheat caused by Heminthosporium sativum. Phytopathol. 41:1011-1020.

77. Huber, C. M. 1963. Investigations on root rot of beans caused by Fusarium solani f. phaseoli. Ph.D. Thesis, Mich. State Univ., E. Lansing. 153 p.

78. Isely, D. 1957. Vigor tests. Proc. Ass. Office. Seed Anal. 47:176-182.

79. Isely, D. 1958. Testing for vigor. Proc. Assn. Offic. Seed Anal. 48:136-138.

80. Johnson, B. J. 1970. Combinations of herbicides and other pesticides on soybeans. Weed Sci. 18:128-130.

81. Johnson, B. J. 1970. Effects of nitralin and chloroxuron combinations on weeds and soybeans. Weed Sci. 18: 616-618.

82. Johnson, B. J. 1971. Effects of sequential herbicide treatments on weeds and soybeans. Weed Sci. 19:695-700.

83. Juniper, B. E. 1957. The effect of preemergent treatment of peas with trichloroacetic acid on the submicroscopic structure of the leaf surface. New Phytol. 58:1.

84. Katan, J. and Y. Eshel. 1973. Interaction between herbicides and plant pathogens. Residue Reviews. 45:145-177.

85. Katan, J. and J. L. Lockwood. 1970. Effect of pentachloronitrobenzene on colonization of alfalfa residues by fungi and streptomycetes in soil. Phytopathol. 60:1578-1582.

86. Knake, E. L., A. P. Appleby and W. R. Furtick. 1967. Soil incorporation and site of uptake of preemergent herbicide. Weeds. 15:228-232.

87. Koren, E., C. L. Foy and F. M. Ashton. 1969. Adsorption, volatility, and migration of thiocarbamate herbicides in the soil. Weed Sci. 17:148-153.

88. Krawiec, S. and D. J. Moore. 1968. Interactions of Tordon herbicide applied in combinations. Down to Earth. 24:7-10.

89. Lai, M. T. and G. Semeniuk. 1970. Picloram-induced increase of carbohydrate exudation from corn seedlings. Phytopathol. 60:563-564.

90. Lewis, R. W. and C. L. Hammer. 1946. The effect of 2,4-D on some microorganisms. Mich. State Col. Agr. Exp. Sta. Quart. Bull. 29:112.

91. Limpel, L. E., P. H. Schuldt and D. Lamont. 1962. Weed control by dimethyltetrachloroterephthalate alone and in certain combinations. Proc. Northeast. Weed Contr. Conf. 24:30.

92. Lynch, M. R., R. D. Sweet and C. T. Dickerson. 1970. Synergistic responses of atrazine in combination with other herbicides - A preliminary report. Proc. Northeast. Weed Contr. Conf. 24:33.

93. Maloy, O. C., Jr. and W. H. Burkholder. 1959. Some effects of crop rotation on the Fusarium root rot of bean. Phytopathol. 50:583-587.

94. Maloy, O. C., Jr. 1960. Physiology of Fusarium solani f. phaseoli in relation to saprophytic survival in soil. Phytopathol. 50:56-61.

95. Mann, J. D., L. S. Jordan and B. E. Day. 1965. A survey of herbicides for their effect upon protein synthesis. Plant Physiol. 40:840-843.

96. Maier, C. R. 1961. Effects of soil temperature and selected crop residues on the development and severity of Fusarium root rot of bean. Plant Dis. Rep. 45: 960-964.

97. Marshall, E. R., G. Bayer and D. Robinson. 1956. Tests with new materials for preemergence weed control in red kidney beans. Proc. Northeast. Weed Contr. Conf. 10:143-146.

98. McIntyre, D. J. 1958. Soil splash and the formation of soil crusts by raindrop impact. Soil Sci. 85:261-265.

99. McKibben, E. C. 1971. Introduction in compaction of agricultural soils. Amer. Soc. Agric. Engineers. 471 p.

100. McReynolds, W. D., Jr. and J. A. Tweedy. 1969. Effect of nitrogen form on the uptake and herbicidal activity of simazine in corn, rye, and soybeans. Weed Sci. Soc. Amer. Abstr. No. 213.

101. Meggitt, W. F. 1969. Michigan State University. Unpublished data.

102. Menzies, F. D. 1963. Survival of microbial plant pathogens in soil. Bot. Rev. 29:79-122.

103. Michigan Agricultural Statistics. 1960-1974. Michigan Crop Reporting Service, SRS, USDA.

104. Moreland, D. E., S. S. Malhotra, R. D. Gruenhagen and E. H. Shokraii. 1969. Effects of herbicides on RNA and protein synthesis. Weed Sci. 17:556-563.

105. Morre, D. J. and R. D. Cheetham. 1970. Response of 2,4-D resistant monocot species to CIPC-2,4-D combinations. Proc. N. Centr. Weed Contr. Conf. 25:103.

106. Mortland, M. M. and W. F. Meggitt. 1966. Interaction of ethyl-N-N-di-n-propyl-thiocarbamate (EPTC) with montmorillonite. J. Agr. Food Chem. 14:126-129.

107. Nair, P. N. 1958. Effect of maleic hydrazide, thiourea, and 2,4-D-dinitrophenol on resistance to flax wilt. Phytopathol. 48:288-289.

108. Nalewaja, J. D., R. Behrens and A. R. Schmid. 1967. Uptake, translocation, and fate of ^{14}C-EPTC in alfalfa. Weeds. 12:269-272.

109. Nash, S. M., T. Christou and W. C. Snyder. 1961. Existence of Fusarium solani f. phaseoli chlamydospores in soil. Phytopathol. 51:308-312.

110. Nutile, G. E. 1946. Growth and yield of baldhead bean plants in the field. Proc. Assn. Offic. Seed Anal. 36:184-186.

111. Oliver, L. R., G. N. Prendeville and M. M. Schreiber. 1968. Species differences in site of root uptake and tolerance to EPTC. Weed Sci. 16:534-537.

112. Parker, C. 1966. The importance of shoot entry in the action of herbicides applied to the soil. Weeds. 14:117-121.

113. Peeples, J. L. and E. A. Curl. 1969. Effect of paraquat, EPTC and trifluralin on growth of Fusarium oxysporum, f. sp. vasinfectum in liquid culture. Phytopathol. Abstr. 59:117.

114. Penner, D. and D. Graves. 1972. Temperature influence on herbicide injury to navy beans. Agron. J. 64:30-31.

115. Pollack, T., T. Furtick and G. Crabtree. 1973. Response of beans to fluorodifen under certain soil and light conditions. Weed Sci. Soc. of Amer. Abstr. No. 39.

116. Preiffer, R. K., O. R. Dewey and R. T. Burnskill. 1957. Further inbestigations of the effect of pre-emergence treatment with trichloroacetic acid and

dichloropropionic acids on the subsequent reaction of plants to other herbicidal sprays. 4th Internat. Congr. Crop Prot. 1:523.

117. Prendeville, G. N., Y. Eshel, M. M. Schreiber and G. F. Warren. 1967. Site of uptake of soil-applied herbicides. Weed Res. 7:316-322.

118. Prendeville, G. N., L. R. Oliver and M. M. Schreiber. 1968. Species differences in site of shoot uptake and tolerance to EPTC. Weed Sci. 16:538-540.

119. Putnam, A. R. and D. Penner. 1974. Pesticide interactions in higher plants. Residue Reviews. 50:(In Press).

120. Rather, H. C. 1942. Field Crops. McGraw-Hill Book Company, Inc. 454 p.

121. Richards, R. R. 1949. Responses of representative fungi to certain growth regulating substances. Bot. Ga. 110:523-549.

122. Richards, L. A. 1953. Module of rupture as an index of crusting soil. Soil Sci. Soc. Amer. Proc. 17:321-323.

123. Richardson, L. T. 1959. Effect of insecticides and herbicides applied to soil on the development of plant disease. II Early blight and Fusarium wilt of tomato. Can. J. Plant Sci. 39:30-38.

124. Robertson, L. S. 1952. A study of the effects of seven systems of cropping upon yields and soil structure. Proc. Amer. Soc. Sugar Beet Tech. 255-264 pp.

125. Robinson, L. R. and C. R. Fenster. 1968. Residual effects of EPTC and trifluralin incorporated with different implements. Weed Sci. 16:415-416.

126. Rodriguez-Kabana, R. 1968. Effect of atrazine on growth of Sclerotium rolfsii and Trichoderma viride in soil. Can. J. Microbiol. 14:1283-1288.

127. Rodriguez-Kabana, R. 1969. Effect of trifluralin on growth of Sclerotium rolfsii in liquid culture and soil. Phytopathol. 59:228-232.

128. Rodriguez-Kabana, R. and E. A. Curl. 1970. Effect of atrazine on growth of Fusarium oxysporum f. sp. vasinfectum. Phytopathol. 60:65-69.

129. Rodriguez-Kabana, R. and H. H. Funderburk, Jr. 1967. Effect of paraquat on growth of Sclerotium rolfsii in liquid culture and soil. Phytopathol. 57:911-915.

130. Rodriguez-Kabana, R. and J. L. Peeples. 1970. Growth response of Sclerotium rolfsii to the herbicide EPTC in liquid culture and soil. Phytopathol. 60:431-436.

131. Schroth, M. N. and W. C. Snyder. 1961. Effect of host exudates on chlamydospore germination of the bean root rot fungus Fusarium solani f. phaseoli. Phytopathol. 51:389-393.

132. Schroth, M. N. and F. E. Hendrix, Jr. 1962. Influence of non-susceptible plants on the survival of Fusarium solani f. phaseoli in the soil. Phytopathol. 52:906-909.

133. Schweitzer, L. R. 1972. Reduction in seedling vigor and changes in metabolism during germination related to mechanical abuse of bean (Phaseolus vulgaris L.) seed. Ph.D. Thesis, Michigan State University.

134. Shennan, J. L. and W. W. Fletcher. 1965. The growth in in vitro microorganisms in the presence of substituted phenoxyacetic and phenoxybutyric acids. Weed Res. 5:266-274.

135. Sikka, H. C., R. W. Souch, D. E. Davis and H. H. Funderburk, Jr. 1965. Effect of atrazine on the growth and reporduction of soil fungi. Proc. S. Weed Conf. 18:616.

136. Skaptason, J. S. 1969. Synergism from phenoxy, propionic, and benzoic acid herbicide mixtures. Proc. N. Cent. Weed Contr. Conf. 24:58.

137. Smith, N. R. and J. L. Shennan. 1966. The effect of substituted phenoxyacetic and phenoxybutyric acids on the growth of Aspergillus niger. J. Gen. Microbiol. 43:293.

138. Standifer, L. C., Jr., D. R. Melville and S. A. Phillipps. 1966. A possible interaction between herbicidal injury and the incidence of seedling disease in cotton plantings. Proc. S. Weed Conf. 19:126.

139. Still, G. G., D. G. Davis and G. L. Zander. 1970. Plant epicuticular lipids: Alteration by herbicidal carbamates. Plant Physiol. 46:307-314.

140. Tammes, P. M. L. 1964. Isoboles, a graphic representation of synergism in pesticides. Neth. J. Plant Pathol. 73 p.

141. Tang, A., E. A. Curl and R. Rodriguez-Kabana. 1970. Effect of trifluralin on inoculum density and spore germination of Fusarium oxysporum f. sp. vasinfectum in soil. Phytopathol. 60:1082-1986.

142. Thompson, H. C. 1931. Vegetable Crops. McGraw-Hill Book Co., Inc., New York. 560 p.

143. Thompson, L., Jr., F. W. Slife and H. S. Butler. 1970. Environmental influence on the tolerance of corn to atrazine. Weed Sci. 18:509-514.

144. Taussoun, T. A., M. Nash and W. C. Snyder. 1960. The effect of nitrogen sources and glucose on the pathogenesis of Fusarium solani f. phaseoli. Phytopathol. 50:137-140.

145. Trouse, A. C., Jr. 1971. Soil conditions as they affect plant establishment, root development and yield. 241-252 pp. In compaction of Agricultural Soils. Amer. Soc. Agric. Engineers. 47 p.

146. Upchurch, R. P., G. R. Ledbetter and F. L. Selman. 1963. The interaction of phosphorus with the phytotoxicity of soil applied herbicides. Weeds. 11:36-41.

147. Upstone, M. E. and J. C. Daves. 1967. The effect of simazine on the incidence of American gooseberry mildew on black currants. Plant Pathol. 16:68-69.

148. Waggoner, D. E. and A. E. Damond. 1952. Effect of stunting agents, Fusarium lycopersici and maleic hydrazide, upon phosphorus distribution in tomato. Phytopathol. 42:22.

149. Waldrep, T. W. and J. F. Freeman. 1964. EPTC injury to corn as affected by depth of incorporation in the soil. Weeds. 12:315-317.

150. Weinhold, A. R., R. L. Dodman and T. Bowman. 1972. Influence of exogenous nutrition on virulence of Rhizoctonia solani. Phytopathol. 62:278-281.

151. Whitney, W. A. 1930. Mutilated seed, a contributing factor in defective stands of lima beans. Phytopathol. (Abstr). 20:134-135.

152. Wilkinson, R. E. and W. S. Hardcastle. 1969. EPTC effects on sicklepod petiolar fatty acids. Weed Sci. 17:335-338.

153. Wright, T. H. and C. E. Rieck. 1974. Factors affecting butylate injury to corn. Weed Sci. 22:83-85.

154. Yamaguchi, S. 1961. Absorption and distribution of ^{35}S-EPTC. Weeds. 9:374-380.

155. Zaumeyer, W. J. and H. R. Thomas. 1957. A monographic study of bean diseases and methods for their control. U.S. Dept. Agr. Bull. No. 868.