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THE EFFECTS OF ONION AGRO-ECOSYSTEM STRUCTURE ON THE POPULATION DYNAMICS OF $\underline{\text{Delia}}$ antiqua (Meigen)

presented by

Brian R. Chambers

has been accepted towards fulfillment of the requirements for

Master of Science degree in Entomology

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Dean L. Haynes

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THE EFFECTS OF ONION AGRO-ECOSYSTEM STRUCTURE ON THE POPULATION DYNAMICS OF <u>Delia antiqua</u> (MEIGEN)

Ву

Brian R. Chambers

A THESIS

Submitted to
Michigan State University
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ABSTRACT

THE EFFECTS OF ONION AGRO-ECOSYSTEM STRUCTURE ON THE POPULATION DYNAMICS OF Delia antiqua (MEIGEN)

By

Brian R. Chambers

Population dynamics of <u>Delia antiqua</u> (Meigen) were assessed by the effects of planting time, soil treatment and harvest of onions; density and distribution of spring volunteer onions; post-harvest cover-crop planting time; and autumn larval freeze resistance. Earlier onion plantings received larger larval populations than later plantings regardless of the use of a soil insecticide. Onion stand reductions were not proportional to larval populations. Earlier plantings received more larvae per damaged plant. Volunteer onions that occured within seeded rows of onions caused higher stand loss to the surrounding rows than volunteers between onion rows. Post harvest field <u>D. antiqua</u> adult activity was affected by both the time of day and duration from harvest time. The time of soil conditioning for a post-harvest cover-crop affected <u>D. antiqua</u> more in relation to peak female fly field acticity than time of harvest. Autumn larvae were freeze resistant as a function of larval length and exposure to fluctuating environments.

DEDICATION

To Lauren with whom I share my strongest joy, fear, aspiration and dedication.

And to all other people who postpone immediate pleasures in the hope of achieving an unknown and distant goal.

ACKNOWLEDGMENTS

I wish to express my sincere appreciation to the entire faculty and staff of the Department of Entomology for their patience and understanding of me as I developed professionally and personally in my Masters degree program.

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To Susan Battenfield, Kelly Barden, Margaret McLeod and Maurine Tillmon, thank you for putting this thesis to rest.

Oh investigator, do not flatter yourself that you know the things

nature performs for herself, but rejoice in knowing the purpose of

those things designed by your own mind.

Leonardo Da Vinci

Madrid Codices

Clearly the problem of man and nature is not one of providing a

decorative background for the human play, or even ameliorating the

grim city; it is the necessity of sustaining nature as source of life,

milieu, teacher, sanctum, challenge and, most of all, of rediscovering

nature's corollary of the unknown in the self, the source of meaning.

Ian L. McHarg

Design With Nature

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INTRODUCTION

In 1976 a project was initiated at Michigan State University to study the onion agroecosystem from a systems perspective across discipline boundaries to affect alternative working hypotheses concerning its management. Research cooperators have included economists Mr. Alex Russell, Ms. Sharon Hart and Dr. Thomas Edens; plant physiologists, Mr. Nick Bolgiano and Dr. Gene Safir; nematologists, Dr. An MacGuidwin and Dr. George Bird; system scientists, Ms. Julia Pet and Dr. Lal Tummala; and entomologists, Mr. Francis Drummond, Mr. Thomas Ellis, Ms. Eleanor Groden, Ms. Marion Harris, Fred Warner, this author, Dr. Ray Carruthers, Dr. Dean Haynes, Dr. James Miller, Dr. Ed Grafius and Dr. Gary Whitfield. The overall objective of the project, conceived and originated by Dr. Dean Haynes, has been to conduct an ecosystem analysis to define and investigate the biotic and abiotic components of the onion system as they affect management of the pest populations.

Ecosystem components investigated prior to this thesis include; foliar insecticides, volunteers as initiators of spring damage, diurnal and spatial distributions of the 1st and 2nd generation of <u>Delia antiqua</u> (Meigen), the onion maggot (OM), OM overwintering mortality (Whitfield 1981), the biology and distribution of <u>Entomopthora muscae</u> (C.), a fungal pathogen of the OM (Carruthers 1981), onion mycorrhizae (Bolgiano 1982), onion nematode (Meloidogyne hapla (C.)) pathogenicity and ecology (MacGuidwin 1983), OM parasite biology and phenology, planting date effects of onions and radishes on OM damage and parasite phenology (Groden 1982), 3rd generation distribution and phenology of the OM, and post-harvest cover crop effects on OM larval

occurrence (Drummond 1982). Simulation models were developed for the onion plant, onion maggot, <u>E. muscae</u>, <u>Aphaereta pallipes</u> (Say), the braconid parasite of the OM, and economic components (Whitfield 1981; Drummond 1982; Carruthers 1981; Bolgiano 1982; Whitfield et al. 1982). Some aspects of this thesis were follow up studies while others employed techniques already developed to quantify previously unexplored relationships.

The objective of this thesis was to investigate the relationships of agroecosystem structure with D. antiqua through studies of:

- 1) planting time, soil treatment and harvest time of the onions;
- the density and location of volunteers in the onion field on the distribution of plant damage;
- 3) distribution of D. antiqua adults following harvest;
- 4) post-harvest cover-crop planting dates; and
- 5) cold hardiness in the third larval brood.

BACKGROUND

Onion (Alium cepa) production

Current onion production in the U.S. is over 100,000 acres per annum and may fluctuate 10 - 30 percent between years within a region (Fedewa 1982). Onions are grown commercially in all fifty states, 87 percent of the total occurring in seven states: California, Texas, Oregon, New York, Idaho, Colorado and Michigan. California and Texas together account for approximately 44 percent of the total U.S. production; however, they primarily produce non-

storage, spring and summer onions and, in the case of California, a large portion of the crop goes to processors. Michigan's onions are principally grown for storage (90%) and compete mostly with other states that produce storage varieties, namely: New York, Colorado, Idaho and Oregon (McLaughlin and Pierson 1981).

The single most important factor influencing Michigan onion prices is the quantity of onions produced in New york during a given marketing year, even more important than Michigan's own production. This situation results not only from the similarities of onion varieties and seasons, but also from the proximity of New York and Michigan to the same geographic marketing areas, especially the Middle and South Atlantic states. When New York producers have a large crop year they have more onions to ship and this typically depresses prices in many of the markets where Michigan shippers normally compete evenly with New York. Hence, Michigan shippers in some instances are compelled to accept market prices which are lower as a result of New York shippers' selling patterns. Higher Michigan prices occur when New York has a short crop year (McLaughlin and Pierson 1981).

Onion seedling emergence occurs between 57.5 and 163 degree days from planting with the form of a logistic growth curve (base 5.6°C)(Bolgiano 1982). Excessive nitrogen and high plant densities both delay maturity (Magruder et al. 1941). Water stress hastens maturity (Drinkwater and James 1955). Relative growth rates are maximal between 28 and 33 °C in Michigan (Bolgiano 1982). Onion bulbing is a function of both temperature and photoperiod with varieties of A. cepa available for both short and long day-length growing seasons (Robinson 1973 and Kedar et al. 1975).

In Michigan, onion field seeding is initiated as close to April 1 as weather permits. Major risks include late snow, frost and heavy spring rains. One or two fields in each region are harvested mid to late August for the early, fresh market, usually the first planted. The remainder of the onions are harvested at the initiation of plant leaf senescence, usually through September. Growers, anxious to harvest before the fall rains of late September will either roll or chop the tops of the onions and harvest before plant senescence. October would be considered an extremely late harvest. The onset of autumn temperature reductions and fluctuating moisture conditions encourages the spread of <u>Botrytis squamosa</u> (W), a leaf blight of onions. Market preferences for untarnished bulbs compel growers to harvest as soon as possible to lessen the discoloration of the onion bulb due to B. squamosa (Swanton 1977).

Delia antiqua (M.) biology

<u>D. antiqua</u> has been the major insect pest in many onion growing regions in the United States since it was introduced to North America in the mid-19th century (Fitch 1867). Doane (1953), Tozloski (1954), Workman (1958), Ellington (1963), Loosjes (1976), and Hucket (1924) have reviewed much of the biology known of <u>D. antiqua</u> prior to the M.S.U. agroecosystem project. Carruthers (1979 and 1981), Whitfield (1981), Groden (1982), Drummond (1982), and Harris (1983) have since contributed to the knowledge of <u>D. antiqua</u>'s basic biology. These studies and others will be appropriately referenced as they pertain to the background literature of this thesis.

In Michigan there are typically three overlapping generations per year of D. antiqua. Three adult flights occur with concomitant larval broods, one each in

the spring, summer and fall. Population densities differ between generations due to environmental conditions as well as cultural practices. Quebec, Canada regions have reported seasons when the second larval brood is the largest (Perron 1972). Michigan records the first and third broods to be the dominate populations (Whitfield 1981; Drummond 1982). A related onion maggot species in Egypt, D. alliaria has its largest numbers in the second and third larval broods (Abul-Nasar 1974).

Other closely related species of <u>Delia</u> include <u>D. platura</u> (R.), the seed corn maggot and <u>D. brassicae</u> (B.), the cabbage maggot. <u>D. platura</u> is a general seed and seedling herbivore, feeding on many types of seeds and germinating plants including onions, beans, corn, and other vegetables. <u>D. brassicae</u> is slightly more limited in its host range infesting cabbage and other brassicas, turnips and radishes (Groden 1982; Whitfield 1981). <u>D. antiqua</u> is host specific to <u>Allium</u> species with preference for <u>A. cepa</u> (Ellis and Eckenroade 1979). Other <u>Allium</u> hosts include garlic (<u>A. sativum</u>), leek (<u>A. ampeloprasum</u>), rakkyo (<u>A. chinese</u>), japenese bunching or welsh onion (<u>A. fistulosum</u>), chives (<u>A. schoenoprasum</u>), and chinese chives (A. tuberosum).

Adult

<u>D. antiqua</u> overwinters as diapaused pupae and emerges in mid spring. A degree day base of 4.4°C has been found to explain 50 percent of the variability in the emergence of the first flight (Whitfield 1981). Other factors believed to be affecting emergence include the pupae's overwintering soil depth and soil moisture. Fifty percent emergence of the first flight was found to occur by 400 degree days accumulation in Michigan (Whitfield 1982). Summer adult activity

and dispersal was 2.5 times greater outside onion fields than within a field in Holland (Loosjes 1976). Adult flight occured almost entirely below 120 cm within a field while only 75 percent occured below that level in field borders (Whitfield 1981). Ninety percent of the summer, second flight adults were found in field borders and adjacent carrot fields apparently seeking protection from high onion field temperatures and low moisture conditions (Carruthers 1981; Whitfield 1981). Summer adult field activity levels were found greatest between 5:30 - 9:30 a.m. and 5:30 - 8:30 p.m. (Whitfield 1981; Loosjes 1976).

<u>D. antiqua</u> adults have been cited as actively feeding on flowers or weed pollen following initial emergence (Baker 1928; Maan 1945; Rygg 1960; Carruthers 1979). Females are anatogenous, requiring a protein rich food source before oviposition (Miller and Haarer 1981; Missonier 1967). A preovipositional period of one to two weeks is dependent on temperature and on food availability (Theunissen 1976). Perron (1972) documented a decrease in oviposition when temperatures remained below 14 °C; a mean of 24.3 eggs per female in the first flight over a three year period; and a mean adult longevity of 66.6 days.

Ovipositing adults have been found to prefer plants wider than 2 mm in diameter at the base (Harris 1983; Harris and Miller 1982). Ovipositional preferences also occur for sprouting culls both of the spring and fall adult flights (Lovett 1923; Grey 1924; Whitfield 1981; Drummond 1982). Related Delia species differ in their ovipositional behavior. D. platura prefer newly emerged seedlings of lima beans and soybeans (Ibrahim and Hower 1979). D. floralis, a cabbage maggot, was found on swedes and turnips to be influenced not by the size of the plant but by its maturity (Rygg and Somme 1972). A similar phenomena

occurs with <u>D. brassicae</u> in radishes; ovipositional preferences occur at two seperate stages of plant development apart from size (Ellis et al. 1979). <u>D. alliaria</u>, the Egyptian onion maggot, prefers seeded onions over transplanted sets (Abul-Nasar and El-Sherif 1974).

Oviposition and its concomitant stand damage has been depicted as initially random in its field distribution and either initiated by <u>D. platura</u>, the seedcorn maggot or by <u>D. antiqua</u>'s attraction to volunteer onions in the field (Carruthers 1979; Whitfield 1981). Succeeding damage then spreads contagiously around the initial points of infestation. Flies are assumed to be ovipositing preferentially in previously damaged areas. Three chemicals, methyl propyl disulfide, cis-propenyl disulfide, and trans-propenyl disulfide are known to be attractive to <u>D. antiqua</u> for oviposition with higher concentrations occuring in field fresh onions over stored onions (Pierce et al. 1978).

Immatures

Eggs of D. antiqua are laid in the soil around the base of the onion plant. Normally, eggs are deposited in groups of three to six with ovipositon occuring repeatedly through the life of the female fly (Karuma et al. 1972). A lower developmental threshold has been estimated to occur at 3.88 °C with a required degree day accumulation of 50 degree days for eclosion (Carruthers 1979). Egg development to eclosion was shown to increase with a decrease in temperature from 32.2 °C to 10 °C (Ellington 1963). Zero egg survival occured at 37.7 °C and 100 percent relative humidity with 60 percent survival at 32.2 °C (100 percent RH) (Ellington 1963).

2

Only 23 percent soil moisture in muck soil was found to be needed for survival of one day old, first instar larvae before they reach the plant tissue of the onion (Workman 1958). Degree day requirements for completion of the three larval instars are reported to be 37, 89, and 161 degree days respectively (base 3.88 °C) (Carruthers 1979). First instar larvae have low survival in onion bulbs greater than or equal to 1 cm in diameter, the second and third instar's survival is independent of bulb size (Drummond 1982). A larva can consume up to 28 flagstage, seedling onions (Workman 1958). They can become freeze susceptible under early autumn field temperatures in Michigan (Drummond 1982). Larval survivorship and pupal size are both higher under alternating temperatures (10 - 20 °C) than a constant temperature of 23 °C (Robinson and Zurlini 1979).

Population Management History

The need for population management was first cited by Fitch in 1867. Lovett (1923) referred to over 70 management practices for <u>D</u>. <u>antiqua</u> and recommended the use of volunteers in flats as "traps" for eggs. Sweetened poison bait with chopped onion, sodium arsinate, molasses and water were being used at that time for direct row treatments (Lovett 1923). Mixtures of napthalene and furnace ash used after first plant damage obtained 80 percent population reduction (Grey 1924). The variety of agents used to control <u>D</u>. <u>antiqua</u> since then represents the evolution of pesticides in the U.S. (Carruthers 1981).

Substitution and replacement of insecticides occured in onions due to the loss of effectiveness of the chemicals to maintain <u>D</u>. antiqua field damage below economic levels. The reduced effectiveness has been reported the result of increasing pesticide resistance in D. antiqua populations (Brown 1958; Harris

1972; Harris et al. 1962; Howitt 1958; Guyer and Wells 1959). Harris et al. (1981) reported resistance levels of a Michigan strain, as compared to a susceptable laboratory strain, to various insecticides, in 1972 and 1978 following 20 generations of selection with parathion (Table 1). Resistance develops rapidly in this species due to its host specificity (Haynes et al. 1980). Once resistance occurs in a population, even if pesticide pressure is relaxed the resistance will only partially decrease (Keiding 1967).

Due to the limited nature of the onion crop (non-essential for subsistence) and current institutional constraints of pesticide registration, few compounds are being tested for onion production (Haynes et al. 1980). An average of 30 test compounds a year in the 1960's contrasts to only 3 per year in 1975 (Harris et al. 1981). Only two compounds remain effective in Canada by 1980 out of the 10 that had been used over the past 25 years (Harris et al. 1981).

Alternative management

Ecological data presented by Haynes et al. (1980), Whitfield (1981), Carruthers (1979 and 1981), Groden (1982), and Drummond (1982) suggests that non-chemical control alternatives will have to have some of the following characteristics:

- reducing pesticides which affect the efficacy of the bio-control agents
- 2) providing habitats appropriate for the agents' life histories (weedy crop borders, alternate hosts, etc.).

Mechanisms that interfere with <u>D</u>. <u>antiqua</u> population behavior mechanically as well as biologically also offer means of population management and are discussed below.

Table 1. Resistance levels of a Michigan onion maggot strain, as compared to a susceptable laboratory strain, in 1972 and in 1978 following 20 generations of selection with parathion (Harris et al. 1981).

| | Resista | nce level |
|-------------|---------|-----------|
| Insecticide | 1972 | 1978 |
| Parathion | 5.1 x | 24.4 x |
| Ethion | 3.0 x | 10.4 x |
| Fonofos | 5.1 x | 10.5 x |
| Carbofuran | 6.2 x | 10.1 x |

The first adult flight is the largest of the three in Michigan (Whitfield 1981). Economic damage is the most severe from the larvae of these adults and can cause over 65 percent of the total season's damage (Whitfield 1981). The survival of the larvae from the previous fall, the emergence and oviposition behavior characteristics of the spring adults, and the survival of their eggs could determine a major portion of the phenology and density characteristics of the first larval brood. Altering these three aspects of the <u>D</u>. antiqua populations could translate to also affecting a portion of the stand reduction in onions. It was for this reason that this thesis emphasizes study of the autumn and spring <u>D</u>. antiqua populations.

The use of delayed spring planting, early trap crops and fall cover crops have all been recommended to reduce the spring larval populations in the onion stands (Groden 1982; Drummond 1982). The only documented use of any of these measures in onions has been by Lovett (1923) and Grey (1924) with stand losses 10 to 15 percent less in fields using volunteers as traps than fields without them. Inter-row cover crops in oats and cabbage reduce oviposition by the frit fly, Oscinella frit (L), and the cabbage maggot, D. brassicae, respectively (Adesiyun 1979; Coaker 1980). In onions, the only documented occurrence of this is in the third adult flight. Fewer eggs were oviposited on sprouting culls in the presence of a cover broadcast planting following harvest (Drummond 1982). Evidence was shown by Adesiyun (1979) that the cover interfered with oviposition behavior by obstruction of preferred oviposition sites. In a cover-no-cover choice experiment Ryan el al. (1980) recorded an 81 percent reduction of oviposition of D. brassicae eggs.

Although no detailed management recommendations have been made for the use of traps or cover crops in onions, they do indicate a possible integration with other alternative management options. From a systems standpoint, manipulation of structural components in both the spring and fall generations to affect the rate of ovipostion and the distribution and survival of <u>D. antiqua</u> immatures represents an appropriate systems approach for applied research in onions. This thesis pursued this line of reasoning for assessing the effects of agro-ecosystem structure on <u>D. antiqua</u> population dynamics in onions.

INVESTIGATIONS

PLANTING AND HARVEST TIME OF ONIONS

Groden (1982) documented a decrease in <u>D. antiqua</u> onion damage as plantings were delayed at the Michigan State University Organic Soils Research Farm in Laingsburg, Michigan. Her plantings were made on May 7, May 14, May 28 and June 12, 1980 in replicated 6.75 m and 7.5 m plots with and without soil insecticide treatment. The decreasing damage trend was evident in both soil conditions. The evidence of higher levels of <u>D. antiqua</u> damage in early planted onions was also recorded by Grey in 1924 at the Lethbridge Entomological Laboratory in Alberta, Canada. Groden (1982) cited the concern that this phenomena had not yet been studied in a manner and scale appropriate to derive interpretations applicable to commercial production practices. A preliminary study was instituted in 1982 to investigate the effect of the range of commercial planting dates for that year on onion stand reduction, bulb size and yield as well as <u>D. antiqua</u> population phenology. Bulb size and yield calculations were made across the period of commercial harvest for the onion production region the study was conducted in.

Study site

Experimental plots were established on approximately one acre of land on a commercial muck farm in Grant Township, Newaygo County, Michigan. Planting time plots measured 90 meters by 45 meters to a side across 16 double rows of onions (Figure 1). Each plot was subdivided into two subplots of eight double

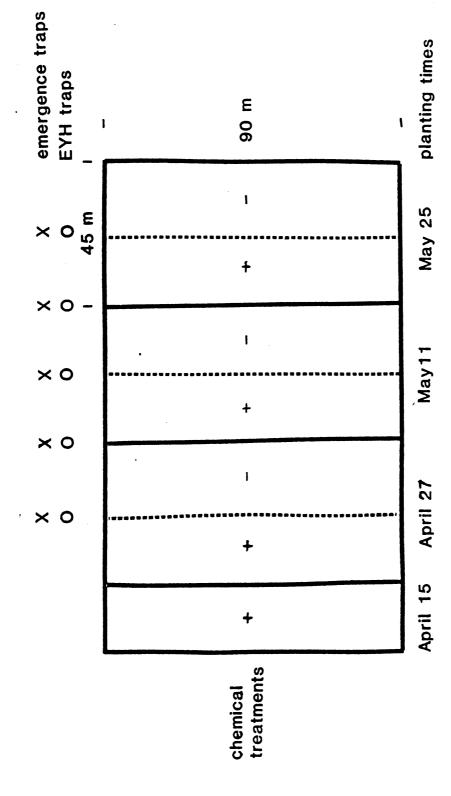


Figure 1. Plot design of planting and harvest time study in Grant, Michigan, 1982.

rows. The subplots consisted of either the presence or absence of the soil insecticide Dyfonate (O-ethyl S-phenyl ethylphosphonodithioate). The treatment combinations will be referred to as planting time subplots.

No herbicides, fungicides or foliar insecticides were applied to the study site. All volunteer onions were removed from the area at sprouting to prevent their effect on aggregate damage from being confounded with error variance (Whitfield 1981).

The study site was located adjacent to the earliest planted field in the region that year, April 15. No untreated subplot for April 15 planting was possible. The field was planted before the investigator had communicated adequately with the commercial grower. Sampling was done in the April 15 plantings, the eight double rows immediately adjacent to the experimental study site.

Sampling Methods

The procedures for sampling were extremely time consuming. Sample intervals, replication and precision were weighted against available resources. Methods for onion plant, <u>D. antiqua</u> adult, larval, and pupal population and Dyfonate residue sampling are discussed separately below.

Onion

Onion stand counts were initialized in each planting time subplot ten days after each seeding. Ten, randomly selected three meter lengths of onion row were assessed for total number of plants and damaged plants. All stand and damage counts after the initial one were made in one random 30 meter length of

row. These post-initial stand estimates were started on June 4 in each subplot and were made every seven to ten days thereafter until July 23. Final stand estimates were done in each subplot on August 20 with ten, random three meter samples.

First generation <u>D</u>. <u>antiqua</u> plant damage was indicated by flaccid, yellowing plant stems. By the end of July however, as the plants became larger and more sturdy maggot damage was no longer evident above ground. In the August 20 stand sample, stand damage was assessed by excavating the plants and inspecting them for entrance holes and larval specimens.

Onion bulb size and yield estimates were derived for each planting time subplot as well as the April 15 planted field for three sample harvest periods; August 20, September 13, and September 24. The August 20 sample coincided with the commercial harvest of the April 15 planted field. The September 13 sample represented a mid-season commercial harvest. The September 24 sample represented a late season harvest and coincided with the last commercial harvest in the Grant region for 1982.

The planting time subplots were assessed for average bulb size and relative yield from one, randomly selected 10 meter length of row. The sample was excavated, the bulbs were counted and recorded for size at geatest diameter and presence of root maggot damage.

D. antiqua

Adult

Two forms of adult monitoring were made for this experiment; spring emergence and female activity. Emergence was monitored with five emergence

traps placed adjacent to the study site (Figure 1). These traps were originally designed to measure emergence rates of cereal leaf beetle adults, <u>Oulema melanopus</u> (L) (Gage 1974). The traps consisted of a Lumite screen supported by an aluminum tripod structure such that emerging adult flies would enter a collection device at the top. A rubberized canvas material was sewn to the bottom of the screen and was buried at each location to a depth of six inches to enclose all flies covered by the trap. The trap covered an area of 0.84 m² (one square yard).

The traps were placed at the site April 15 and emptied every three to four days from May 4 to July 31. Since the experimental site was planted in onions the previous year the emergence of first flight <u>D</u>. antiqua in the vicinity of the experiment could be monitored by these placements.

Female D. antiqua field activity was monitored using baited traps developed by Miller and Haarer (1981) (Figure 2). Enzymatic yeast hydrolyzate (EYH) as a bait is believed by Miller and Haarer to attract female onion flies in search of an exogenous protein source just prior to oviposition. Laboratory cultured D. antiqua females aggregated on EYH dishes, peaking in this behavior eight days following emergence (Miller and Haarer 1981). Vitellogenisis cannot occur in the laboratory without an exogenous source of protien (McLeod 1964). If EYH causes gravid females to respond comparably in the field then a close approximation of oviposition activity can be made.

Large cone traps, somewhat similar to the design of Dindonis and Miller (1980) were constructed from 8 x 8 mesh screen. A 30 cm diameter (bottom) x 30 cm high cone with a 2.5 cm opening was topped with a 13 cm diameter x 15 cm high clear outer acetate cone (Figure 2). The outer acetate cone had an inner

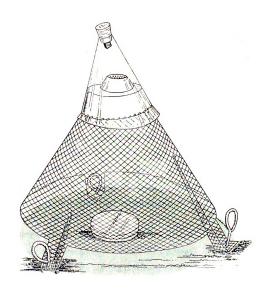


Figure 2. Design of baited fly traps using enzymatic yeast hydrolyzate.

acetate cone with a 13 cm diameter x 8 cm height and plaster of paris layered between the two cones for weight. Legs for the trap formed by the screen mesh held the cone 5 cm above the ground, while wire stakes secured the trap to the ground. EYH would be placed in a petri dish directly beneath the trap. Air would circulate beneath the trap and carry volatiles downwind and along the soil. Only walking insects or those flying close to the ground were able to enter the trap directly. A 1 cm³ volume of Vapona was tacked to a cork that sealed a 2 cm diameter opening at the top of the upper acetate cone. Flies entering the trap would attempt to leave by flying upward into the upper cone and die upon contacting the Vapona.

In anticipation of approximating oviposition frequency five EYH traps were placed adjacent to the emergence traps bordering the study site (Figure 1). The traps were placed in the field on May 25 and emptied every three to four days until July 31. Counts of both male and female <u>D. antiqua</u> were made to verify the effectiveness of the female monitoring. All fly specimens were returned to the laboratory for identification and recording.

Immature

Larvae were sampled to examine the phenology and relative density of D. antiqua immatures in the planting time subplots. Three sections of 0.6 meters of onion row were sampled twice a week at three and four day intervals. This sampling was conducted in each subplot from June 6 to July 31. The sections were randomly selected within areas were root maggot plant damage was evident (flaccid, yellowing leaves). Each onion in the sample was removed and disected in the field for root maggots. Once the onions were removed and larvae

retrieved, the soil beneath the sample length up to a 20 cm depth, 20 cm wide was sifted to collect any larvae or pupae in the soil (a 20 x 20 x 60 cm volume). All specimens were returned to the laboratory for identification and recording.

Dyfonate residue

Residue samples were made to account for any degradation that might have occurred through the season in each treated, planting time subplot. One, $162.56 \times 10.16 \times 12.70 \text{ cm}$ (64 x 4 x 5 in) volume of row was excavated immediately after each planting in the treated subplots. A sample was also made at that time from the untreated subplot as a check.

The soil volume was tumbled for 15 minutes in a cement mixer to homogenize the soil with the Dyfonate residue. A 50 ml subsample was removed from the center portion of soil in the mixer and sent to the Michigan State University Analytical Laboratory for Dyfonate residue analysis. This sampling was repeated after seeding twice a week in each treated subplot until July 31.

Results and Analysis

Onion

Stand damage .

Stand and damage averages are listed in Tables 2, 3 and 4 for each planting time subplot. Damage was higher in the untreated subplots in contrast to the treated for all planting times.

For the sample periods from June 4 to July 23 differences of stand and damage rates are not statistical since only one 30 meter sample was made from

Table 2. Average plants per meter for stand and damage density estimate of the April 27 planting for the treated and untreated subplots across the season.

| | | | | Sample period | period | | | |
|-----------|--------------------|---------|---------|---------------|--------|---------|---------|----------------------|
| Subplot | (initial) May 6 | June 4 | June 14 | June 30 | July 7 | July 16 | July 23 | (final) August 20 |
| Treated | | | | | | | | |
| stand | 53.64 | 52.85 | 47.90 | 46.88 | 45.80 | 44.58 | 43.93 | 48.90 |
| damage | | 0.78 | 4.95 | 1.02 | 1.05 | 1.90 | 0.65 | 1.15 |
| % damage | (1.5)* | 3.60 | 10.34 | 2.10 | 2.30 | 4.30 | 1.40 | 2.30 |
| Untreated | | | | | | | | |
| stand | 48.34 | 17.88 | 5.12 | 3.87 | 3.67 | 3.44 | 3.00 | 1.24 |
| damage | | 11.25 | 3.64 | 1.24 | 0.18 | 0.23 | 00.00 | 90.0 |
| % damage | (63.0)* | ٠ 69.92 | 71.00 | 24.00 | 5.00 | 6.00 | 0.00 | 5.00 |

* estimated

Table 3. Average plants per meter for stand and damage density estimates of the May 11 planting for the treated and untreated subplots across the season.

| | | | | Sample period | eriod | | | |
|-----------|---------------------|---------|---------|---------------|--------|----------------|---------|----------------------|
| Subplot | (initial) May 21 | June 4 | June 14 | June 30 | July 7 | 16 לוטל 7 לוטל | July 23 | (final) August 20 |
| Treated | | | | | | | | |
| stand | 52.79 | 50.95 | 49.51 | 48.72 | 47.80 | 47.28 | 46.26 | 49.83 |
| damage | | 1.84 | 1.44 | 0.78 | 0.91 | 0.52 | 1.02 | 0.82 |
| % damage | (3.5) | * 3.60 | 2.91 | 1.61 | 1.92 | 2.00 | 2.19 | 1.65 |
| Untreated | | | | | | | | |
| stand | 51.30 | 30.31 | 25.80 | 8.79 | 6.82 | 92.9 | 95.9 | 5.25 |
| damage | | 20.99 | 17.10 | 1.98 | 0.05 | 0.13 | 0.19 | 0.24 |
| % damage | (41.0) | * 69.00 | 99.00 | 22.50 | 0.78 | 2.00 | 3.00 | 4.60 |

* estimated

Table 4. Average plants per meter for stand and damage density estimates of the May 25 planting for the treated and untreated subplots across the season.

| | | | Ň | Sample period | þ | | |
|-----------|---------------------|---------|---------|---------------|---------|---------|----------------------|
| Subplot | (initial) June 4 | June 14 | June 30 | July 7 | July 16 | July 23 | (final) August 20 |
| Treated | | | | | | | |
| stand | 53.40 | 57.80 | 57.64 | 57.18 | 57.08 | 57.05 | 56.97 |
| damage | 0.16 | 0.13 | 0.13 | 0.16 | 0.46 | 0.10 | 0.13 |
| % damage | 0.30 | 0.23 | 0.22 | 0.28 | 0.80 | 0.17 | 0.23 |
| Untreated | | | | | | | |
| stand | 49.47 | 45.54 | 39.86 | 36.81 | 34.97 | 34.19 | 32.44 |
| damage | 10.50 | 8.63 | 2.67 | 3.65 | 1.84 | 0.79 | 1.74 |
| % damage | 21.00 | 18.90 | 14.20 | 8.20 | 5.25 | 2.30 | 5.30 |
| | • | | | | | | |

each subplot. Relative trends can however be discerned. The April 27 planting's untreated subplot had an estimated 63 percent reduction through the month of May in contrast to the estimated 40 percent reduction in the May 11 planting's untreated plot. By June 4, when the May 25 planting was initially assessed, the earlier plantings were yet receiving proportionately higher rates of stand loss in both the treated and untreated subplots. The relative trends indicate that the earlier two plantings were prone to stand loss early in the season and continued to be more so than the last planting regardless of soil treatment.

After relatively high stand damage early in the season a marked reduction occurred in each of the subplots after June 30. This trend of high early season damage and a marked mid-season reduction was comparable to that found by Whitfield (1981) and Groden (1982).

Statistical tests for differences can be made on the initial and final stand count data since replicate sampling was performed. Table 5 lists the means of the initial and final stand estimates for each planting time subplot. Scheffe multiple range comparisons among the initial stand estimates revealed insignificantly different means for all subplots. This implies that by the tenth day after planting each subplot's stand was at a comparable level of plant emergence. The pooled mean of the initial stands was 51.5 plants per meter.

Paired difference tests between the initial and final stand estimates were significant for only the untreated subplots. Stand density was significantly lowered over the season in all untreated plantings (Table 5). The largest reductions occurred in the April 27 and May 11 untreated subplots. Final densities were 1.2 and 5.2 plants per meter respectively. In contrast, the May 25 planting's untreated subplot was moderate in its stand loss with a final stand

Table 5. Scheffe multiple range comparisons of mean stand densities for initial and final estimates by planting time and soil treatment. Means are in plants per meter.

| | | Treated | | | Untreated | |
|---------------|---------|---------|---------|---------|-------------------|-------------|
| Planting time | initial | final | % chng. | initial | final | % chng. |
| April 15 | 51.2cd | 44.3c | - 13.5 | 1 1 | \$ 8 9 9 | ! ! ! |
| April 27 | 53.6cd | 48.9cd | - 8.7 | 48.3cd | 1.2a | - 97.5 |
| May 11 | 52.7cd | 49.8cd | - 5.5 | 51.3cd | 5.2a | 8.68 - |
| May 25 | 53.4cd | p6.95 | + 6.5 | 49.4cd | 32.4b | - 34.4 |
| | | | | | | |

* numbers followed by a common letter are insignificantly different (p = .05)

density of 32.4 plant per meter. This subplot's stand density was significantly different from all other treated and untreated subplots.

The means of the initial and final stand counts were not statistically different within any of the treated subplots. However, relative differences did occur within them. The relative percent change from the initial to final stand means, shown in Table 5 highlights a trend in the treated plantings. The April 15 treated subplot was reduced 13.5 percent less in its mean stand density by the end of the season. The April 27 planting's treated stand was reduced 8.7 percent and the May 11 planting's treated stand 5.5 percent. However, the May 25 treated stand increased a relative 6.5 percent in mean plant density by the end of the season.

Discussion

The three earlier treated planting's relative stand reduction trend can be explained by the higher rates of <u>D</u>. antiqua damage that were found in the untreated subplots. Groden's 1980 experiments also had the trend of decreasing damage by delaying planting evident in both respective soil treatments. The relative stand increase in the last planting can only be explained by onion seed germination occurring after the June 4 initial stand estimate. Comparing the June 4 mean stand estimate with the June 14 for the May 25 planting's treated subplot in Table 4 shows a relative stand increase from 53.4 to 57.8 plants per meter. Since all treated subplots' initial mean densities were insignificantly different from the May 25 planting's initial, it is likely that the earlier plantings also experienced germination after their initial stand determinations. Because of late germination the stand reduction estimates within a treated subplot were likely biased downward.

Bulb diameter

Two forms of statistical analyses were performed on the bulb diameter data recorded for each planting time subplot across the three sample harvest periods. First, Scheffe multiple range tests were made to determine where and when significant differences occurred between the average bulb diameters of the subplots. Second, a multiple regression analysis was performed on selected independent variables which data was available for that were believed to best explain the differences found. The extent that the selected independent variables actually characterized the mean bulb diameter differences detected by the multiple range test were assessed by the multiple regession.

Mean bulb diameter data for each harvest sample period by planting time and soil treatment are listed in Table 6 with the results of the Scheffe multiple range test. The largest average onion bulbs of 8.4 and 6.8 cm for the three sample periods occurred across the untreated subplots harvest samples of the April 27 and May 11 plantings, respectively. These subplots had the lowest final stand densities of 1.2 and 5.2 plants per meter. It was not unexpected for them to also average the largest onions.

Although the treated subplots were all insignificantly different from each others average final stand density their mean bulb diameters were detectably different at the August 20 harvest sample. A statistical trend of decreasing bulb diameters from 5.3 cm to 3.6 cm, at this sample period for the treated subplots was evident as the plantings were delayed from April 27 to May 25. The April 15 planting's treated subplot was an anomoly in relation to this trend. Although the April 27 and May 11 treated subplot's August 20 mean bulb diameters were significantly different from each other, neither were significantly different from the April 15 planting's 4.9 cm mean.

Table 6. Scheffe multiple range comparisons of mean bulb diameters for each planting time subplot by harvest sample period. Mean are in centimeters. (n=5625)

| | ber 24 | untreated | | 8.49k | 7.87j | 6.04gh |
|-----------------------|--------------|---------------|----------|----------|---------|---------|
| | September 24 | treated | 5.80fgh | 5.37def | 5.63def | 5.43def |
| Harvest sample period | September 13 | untreated | | 8.28k | 5.72efg | 4.84b |
| Harvest sa | Septem | treated | 6.16h | 5.38e | 5.45def | 4.62b |
| | August 20 | untreated | | 8.44k | 6.891 | 4.09a |
| | Augu | treated | 4.94bc | 5.26cd | 4.65b | 3.85a |
| | | Planting time | April 15 | April 27 | May 11 | May 25 |

 \star numbers followed by a common letter are not significantly different (p = .10)

Interpretation of the April 15 planting's bulb diameter data is confounded by cultural practices. These onions were rolled August 10 in preparation for the early, fresh market. Preparing onions for market in this manner appears to curtail bulb development. However, the April 15 planting did recover from the mechanically induced stress by the later harvest samples with bulb diameters of 6.1 and 5.8 cm's.

The significant trend of decreasing bulb diameters also occurred in the untreated subplots for the August 20 harvest samples. The average bulb diameters decreased from 8.4 to 6.9 to 4.1 cm's as the subplots' planting times were delayed from April 27, May 11 and May 25 repsectively. This trend remained significant for the untreated subplots in both the September 13 and September 24 harvest samples.

The bulbs were relatively unconstrained by crowding in the untreated subplots due to the considerable stand reductions. The average diameter does appear to be limited by the planting time. The untreated subplots of the April 27 and May 11 plantings remained significantly different (8.5 - vs - 7.9 cm) in the September 24 harvest sample even though their final densities of 1.2 and 5.2 plants per meter were not significantly different.

The treated plantings did not maintain the significant trend of decreasing mean bulb diameters as planting times were delayed. By the September 13 harvest the April 27 and May 11 planting's treated subplots were insignificantly different. The trend had become totally indistinct by the September 24 harvest sample. None of the treated subplots by this time differed significantly in mean bulb diameter. This included the May 25 planting which had the highest relative stand density of 56.9 plants per meter. The upper limit of bulb development for

the range of final stand densities (44.3 to 56.9 plants/meter) exhibited in the treated subplots appears to be between 5.4 and 5.8 cm's. The treated densities appeared to curtail bulb size development prior to any limit imposed by the time of planting as observed in the untreated subplots.

Contrasting the May 25 planting's treated and untreated subplots mean bulb sizes across the sample harvest periods also depicts an inter-relationship between the time of planting, final stand density and bulb size. The untreated subplot was 43 percent less in average final stand density from its treated counterpart. Even with this disparity in stand densities mean bulb size differences were not significant until the last harvest sample. The period for growth necessary for bulb sizes to be detectably constrained by stand density in the May 25 planting did not occur until September 24.

Five variables possibly explain the differences in mean bulb diameters detected by the Scheffe multiple range test. They are

- the time of harvest sample;
- 2) the length of time allowed for onion growth;
- 3) the time of planting;
- 4) the soil treatment (presence or absence of Dyfonate); and
- 5) the density of the stand at the end of the season

A multiple regression was performed on these variables to determine which were most inter-correlated and which independently characterized the differences in the bulb diameters detected. Since treatment combinations were not replicated, error variance terms were not available for treatment combinations. The regression model, in this case is used to only characterize the data set.

The multiple regression was first made using a forward, step-wise regression on the five previously stated variables expressed in the following seven ways

- 1) Julian date of planting (PD);
- 2) Julian date of harvest sample (HD);
- 3) degree day accumulation above 5.5°C (the developmental base of onion) at planting from April 1 (PDD);
- 4) degree day accumulation at harvest sample (base 5.5°C) from April 1 (HDD);
- 5) accumulated degree days for growth (the difference between harvest and planting degree days) (DDGR);
- 6) soil treatment (absence = 1, presence = 2) (ST); and
- 7) harvest stand count (HST).

Temperature data was available from the Grant, Michigan State Agricultural Experiment Station starting from April 1, 1982.

Degree days for growth (DDGR) was most correlated with bulb diameter in this first analysis (r = .36). Julian date of planting and harvest sample were both highly correlated with their degree day counterparts (r = .98). Only degree days at planting and harvest were retained for use in the next regression analysis. Soil treatment and the density of the stand at harvest were only moderately correlated (r = .70) so both terms were saved for the second analysis.

A second regression analysis was performed to determine which interaction terms of the remaining variables from the previous analysis were appropriate for inclusion. The interaction terms were entered based on their level of correlation

with other variables as well as on the results of the F-tests performed for significant coefficients (Gill 1978).

After an initial run of this analysis it was decided to drop degree days at planting (PDD). It was determined that the interaction of degree days at planting with degree days at harvest (PDD·HDD) was highly correlated with degree days at planting (PDD) and obtained a higher R². Because of this it appeared that PDD·HDD would substitute well for PDD.

Table 7 lists the regression statistics for the final set of independent variables that explained the greatest amount of variance in the data, were least correlated with each other and each passed the F-tests for inclusion in the regression. In the final analysis the accumulated degree days for growth (DDGR) most characterized the differences in bulb diameters. The density of the stand at the end of the season (HST) had the second greatest ability to explain the differences in the data. Since stand differences varied in direct relation to the use of dyfonate it was not surprizing that soil treatment (ST) was the fourth most significant term in the explanation of the variance. The signs of the correlation coefficients were negative for both ST and HST signifying that as the insecticide was used and the stand densities became higher the bulb size decreased.

The third most significant and explanatory term was the interaction of the degree days at planting and the degree days at harvest (PDD·HDD). The importance of this interaction term is that it highlights the non-parallel relationship between the two parameters in the characterization of bulb size. It was not just the physiological time of harvest (HDD) under which a subplot was sampled but the subplot's simultaneous identification with a particular planting

Table 7. Multiple regression statistics for the onion bulb diameter under field conditions characterized by degree days for growth (DDGR), the degree day accumulation atplanting (PDD), the density of the onion stand at harvest (HST), the degree day accumulation at harvest (HDD), and presence or absence of a soil insecticide treatment (ST).

| Source | DF | Sum of Squares | Mean Square | F |
|------------|------|----------------|-------------|-----------|
| Regression | 7 | 2369.25 | 338.46 | 215.03*** |
| Residual | 5617 | 8841.24 | 1.57 | |

^{***} (p = .001)

| Variable | F to enter | R^2 | В |
|-----------|------------|-------|------------------------|
| DDGR | 3.26* | .130 | 1.26×10 ⁻² |
| HST | 218.92*** | .047 | -2.23×10^{-2} |
| HDD | 96.20*** | .003 | -1.09×10^{-2} |
| ST | 82.81*** | .012 | 1.96 |
| HST•PDD | 8.05** | .001 | 1.09x10 ⁻⁵ |
| ST · DDGR | 36.63*** | .005 | -4.66×10^{-4} |
| HDD•PDD | 88.25*** | .012 | 3.03x10 ⁻⁵ |
| constant | | | 2.33 |

^{*} (p = .10) ** (p = .005) ***(p = .001)

 $R^2 = .2113$

time (PDD) which characterized the differences in bulb diameters. The unique combination of degree days at planting and degree days at harvest (PDD·HDD) explained, in part, a significant trend in the data set.

Similar explanations can be made of the succeeding significant variables in the regression. What is crucial to this analysis is the ability of the available independent variable data, in toto, to explain the variance of the dependent variable data. The coefficient of multiple determination (R²) is often used as a statistical measure of a regession's explanatory value of causal (or functional) relationships (Gill 1978). In this regression the R² was 0.216. Approximately 22 percent of the variation in bulb diameter was explained by the selected regression variables and their derived coefficients.

Discussion

The resultant regression variables were able to characterize and verify an influence on the bulb diameter but they did not explain or predict the differences well. Replication of the treatment combinations would have permitted a formal analysis of variance to be made to determine which design variables actually could explain the variance in the data. Larger sample sizes for normally distributed data often reduce variance to mean ratios (Steele and Torrie 1980). A regression from treatment combination replicates would therfore likely be more explanatory (Cochran and Cox 1957).

The regression analysis results of Table 7 do corroborate the Scheffe multiple range tests on the mean bulb diameter data (Table 6). Bulb diameter differences were greatest between the significantly different final stand densities (HST) as listed in Table 5. The subplots in which more time had been

allowed for growth had significantly larger bulb sizes, especially in the untreated subplots (DDGR and ST). The early, insignificantly different mean bulb diameters of the May 25 planting's subplots likely placed soil treatment (ST) as the fourth term in the regression even though overall, treated - vs - untreated subplots were markedly different in mean bulb diameters. Generally as the harvest times were further into the autumn, bulb sizes were significantly larger in relation to their planting times (PDD·HDD).

Yield

Commercial onion yield is derived by the density and size distribution of onions per unit area. Typically, onion yield in the United States calculated in bushels per acre of U.S. #1 onions. A bushel of #1 onions is defined as 50 pounds of onion bulbs between one and a half and three inches in diameter with 80 percent between two and three inches.

Relative yield estimates were determined from the subplots for each of the harvest samples by the use of a computer algorithm. This algorithm incorporated the diameters of the undamaged bulbs measured at each harvest sample, a regression between bulb volume and weight and a sorting routine which determined the maximum number of appropriate 50 pound units possible from each data set.

Bulb weights were not determined in the field so a regression between weight and bulb diameter was calculated from a commercial bag of #1 onions. Each individual bulb in the 50 pounds was weighed and sized at its greatest diameter. The regression on weight was developed from a spherical estimate of bulb volume $(4/3 \text{ pi } (1/2 \text{ diameter})^3)$.

Table 8. Regression statistics for the wieght of an onion bulb in grams characterized by $4/3*pi*(1/2(DIA)^3$.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|----|----------------------|----------------------|--------------------------|
| Regression | 1 | 2.22×10 ⁵ | 2.22×10 ⁵ | 1.38×10 ³ *** |
| Residual | 99 | 1.60×10 ⁴ | 1.62×10 ² | |

^{*** (}p = .001)

$$Y = 14.14 + 3.218 (4/3*pi*(1/2(DIA)^3)$$

 $R^2 = .9329$

The regression of estimated bulb volume on its weight explained 93 percent of the variation in bulb weight for the measured 50 pounds of onions (Table 8). This regression was then used in the computer algorithm which selected the necessary range of bulb sizes to derive the maximum number of 50 pound units that could be attained from each subplot's harvest sample. The results were then extrapolated to acre level determinations. Table 9 lists the results of the estimates of relative yield for each of the harvest samples.

Discussion

The bushels per acre levels were each determined from a single, 10 meter length of row sampled at each harvest period. Because of this, differences in yield between the planting time subplots and sample periods are only relative. The relative yield differences do however corroborate the interpretations made of the bulb diameter analysis in conjunction with the estimates of the final stand densities. The analysis portrays the trade-offs between the density of the onion stand and the size distribution of the bulbs in the determination of a commercial yield.

The significantly larger bulbs of the untreated subplots were assumed to occur because of the extremely low density of the stands. This apparently also translated into lower relative yield estimates in the untreated subplots. The April 27 and May 11 plantings untreated subplots received the greatest stand reductions with final stand densities between 1.2 and 5.2 plants per meter. These plots also consistently had the lowest relative yield estimates for all the harvest periods.

Table 9. Harvest date yield in bushels per acre.

| | | | Harvest date | date | | |
|---------------|---------|-----------|--------------|-----------|---------|--------------|
| | Augr | August 20 | September 13 | er 13 | Septem | September 24 |
| Planting date | treated | untreated | treated | untreated | treated | untreated |
| April 15 | 884 | | 661 | | 1,413 | |
| April 27 | 1,115 | 21 | 1,378 | 22 | 1,197 | 124 |
| May 11 | 797 | 134 | 1,076 | 131 | 1,030 | 66 |
| May 25 | 212 | 135 | 581 | 384 | 985 | 619 |
| | | | | | | |

The mechanical stress induced on the April 15 planted onions for the early harvest was likely responsible in part for the relatively low yield of 884 bushels per acre for the August 20 harvest sample. The final density of this stand of 44.3 plants per meter was also relatively lower than the other treated plots (Table 5). This lower density also could have contributed to its depressed yield.

The treated subplots that had comparable stand densities at the end of the season followed the same relative trend in yield estimates as they did bulb diameters. They were markedly different in the estimates in the first harvest sample. As with bulb diameter, the April 27 planting had the largest relative yield for this period of 1,115 bushels per acre followed by the April 15 and May 11 plantings of 884 and 767 bushels per acre, respectively. The May 25 planting had the lowest relative yield of 212 bushels at the August 24 harvest period. At this time neither the May 11 or May 25 treated plantings appeared ready for harvest.

The May 11 treated planting had an average bulb size comparable to the April 27 planting's at the September 13 harvest sample. The relative yield for the two plantings were 1,378 and 1,176 bushels per acre for the April 27 and May 11 plantings, respectively. These yields appear more comparable and hence the May 11 would likely have been ready for harvest in a commercial setting by September 13.

The May 25 planting at 212 bushels per acre for the August 20 harvest was apparently still at an immature level of bulb development. As the bulb diameters increased for each of the harvest samples so did the relative yield estimate for both the treated and untreated subplots in the May 25 planting. The September 24 harvest sample for this planting had an estimate of 985 and 619

bushels per acre for the treated and untreated plots. The treated sample indicated that possibly it was at a level of development appropriate for a commercial harvest. The untreated subplot, even though it had significantly larger bulbs on the average, had a relative yield indicative of its lower stand density.

D. antiqua

Adult

Emergence rates of the spring adults from the five emergence traps are shown in Figures 4 and 5 for Julian day and degree day accumulation (base 4.4°C for emergence) time scales, respectively. The average emergence trap catches were averaged over the time units from the previous sample and plotted at the midpoint of the sample interval. Relative trend differences in the two time scales occur due to the variable temperature fluctuations from day to day.

To estimate the cummulative percent emergence at a given degree day the average trap catch from each sample interval was distributed evenly, in whole units across the degree day interval in which the catch was made. The cummulative proportion of the total was then determined at the degree day times of the evenly distributed, interval catch data (Figure 6).

Fifty percent of the monitored adult population had emerged by degree day 900 (Julian day 162; calendar day June 11). In relation to the stand germination counts ten days after seeding, cummulative emergence can be used to contrast the proportion of the total to which that each planting was exposed. One percent of the adults had emerged by the April 15 planting's, ten day stand count. Two percent had emerged by the April 27 planting's initial stand count.

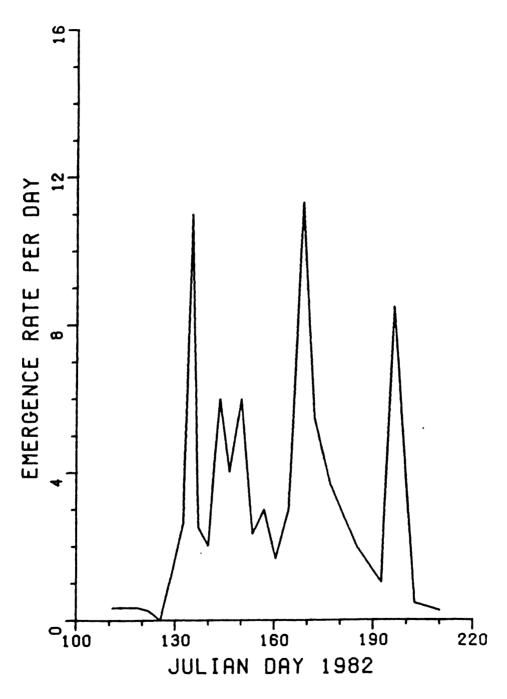


Figure 3. Emergence rates of spring adult $\underline{\textbf{D}}$. $\underline{\textbf{antiqua}}$ by Julian days.

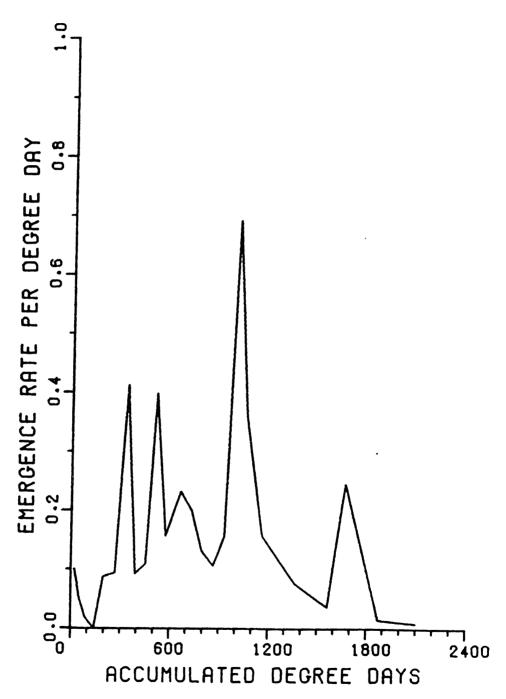


Figure 4. Emergence rates of spring \underline{D} . $\underline{antiqua}$ by accumulated degree days.

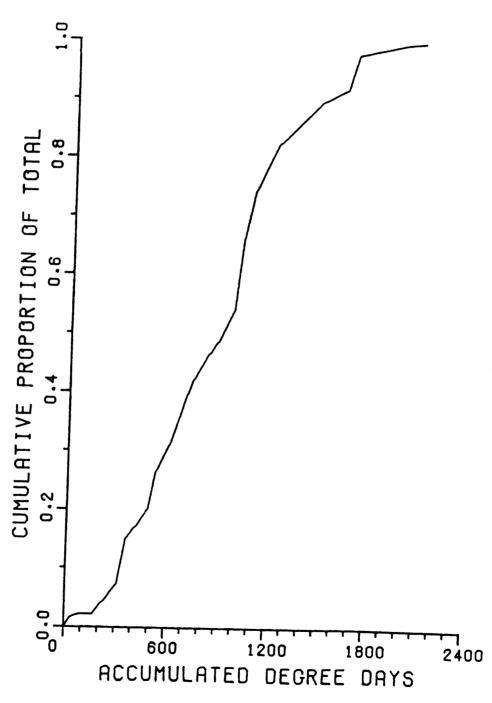


Figure 5 . Cummulative proportion of total $\underline{\textbf{D}}$. $\underline{\textbf{antiqua}}$ spring emergence.

Twenty percent emergence had occurred by the stand initialization of the May 11 planting and 42 percent had emerged by the May 25 planting's June 4 stand count. Less than half of the monitored adults had emerged by the time all of the planting's had plant emergence to 51.5 plants per meter.

The catch results of the EYH baited attraction traps for both the females and male onion flies are shown in Figure 7. As expected the ratio of females to males was high, ranging from 2:1 to 5:1. Peak female activity at these traps occurred at 1050 degree days (Julian day 171; calendar day June 20). This peak, in light of a pre-ovipositional period of one to two weeks (Theunissen 1976; Biscoe and Kitching 1974), correlates well with the time of 50 percent emergence at 900 degree days, 9 days previous.

Discussion

Contrasting the analysis of the plant sampling with D. antiqua adult monitoring indicates plausable interactions of the fly population with the onion planting times. Assessment of female fly activity by EYH traps indicates possible peak oviposition rates around June 20. This is well after all the stands were established to at least 51.5 plants/meter (Table 5). Since the relative rate of stand damage did not increase during this time (Tables 2, 3 and 4) ovipostion must have been occurring on previously damaged plants. This phenomena was documented by Whitfield (1981) as damage initially occurring in a random pattern in the field with succeeding damage occurring in previously infested areas contagiously.

In relation to the emergence data the majority of stand damage appears to have occurred prior to even 50 percent of the total adult emergence from the

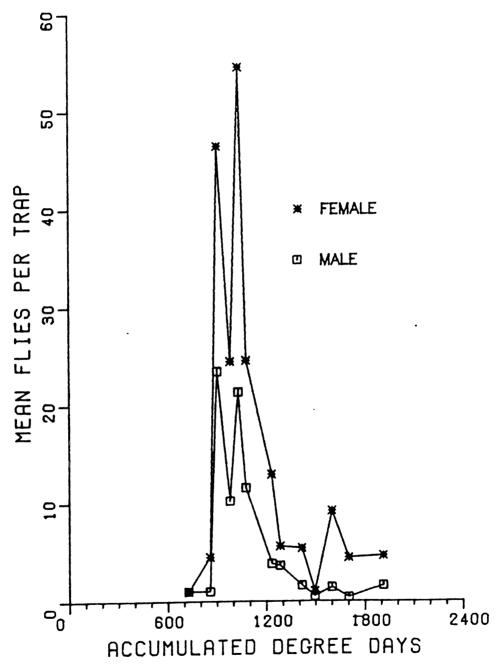


Figure 6. Mean \underline{D} . antiqua adults per EYH trap for both males and females.

surrounding area. It would therefore be the early emerging flies that oviposited the eggs for the most damaging immatures of the population. The oviposition by the mid and later emerging adults did not proportionately increase the rate of stand loss. It probably only increased the density of larvae per damaged plant. This also corroborates well with Whitfield's 1981 findings of early stand loss being greatest with a contagious distribution of plant damage following initial damage.

Egg dessication from high summer soil surface temperatures is another possible factor that could explain the lack of stand loss when female flies appeared to be most active. Summer soil surface temperatures have been recorded in black muck soils as high as 60°C (Warner 1984). Ellington's 1963 report of zero egg survival at 37.7°C (100% RH) indicates the strong possibility of abiotic mortality being very significant. Although not recorded, muck soil surface temperatures at this study site could have easily exceeded this threshold of 37.7°C by late June, 1982.

Immature

Drummond (1982) developed indices of <u>D. antiqua</u> larval length for use in field determination of instar. His estimates, 2.9 mm for separating first and second instars and 5.1 mm for distinguishing between second and third instars, were used for rapid and inexpensive instar determinations of larval specimens in this experiment.

Three sets of analyses were performed on the larval instar data. The first, an estimate of the immature population's overall maturity or development used an index of the weighted mean immature life stage (WMI) (Fulton 1975). The

instar data for the second and third analyses were used in an index of field instar density. The second analysis used the instar density index as a function of planting time and sample period for each soil treatment. In the third analysis the index was used to derive the total sample season's production of each life stage per acre for each subplot.

The analysis of the WMI could indicate whether the planting time subplots differed in the maturity of the immature populations at any point in time. The effect of the differing planting times might be that the earlier planted have a more mature larval population than the later planted onions. This would imply that the earlier plantings not only received more eggs, but that they received them substantially early in the season.

The analysis of the instar density indexes as a function of the design variables of planting time and sample period separately for each soil treatment would allow a determination to be made of the extent that each variable influenced the instar density of each treatment subplot. The effect of the soil treatment was obvious in the stand, bulb diameter and yield data. It would not further any understanding to include this parameter in a single regression analysis.

The total production derivation estimates the average number of individuals in a sample season that move through a unit area in a field. The analysis of the density index used to calculate the total production of each instar in each planting time subplot will give a relative comparison of each subplot's ability to foster and sustain <u>D</u>. antiqua immatures. For this experiment it will be a relative measure since treatment combinations were not replicated. These three analyses will be described in detail with results and discussions in the following sections.

Weighted mean immature (WMI)

Fulton (1975) developed an index for determining the maturity of a sampled population by weighting the age classes and adjusting for the proportion of time spent in each age class relative to the entire life span of all classes:

$$WMI = \sum_{i} P_{i} \cdot (i) \cdot N_{i} / \sum_{i} P_{i} \cdot N_{i}$$
 (1)

where WMI = weighted mean immature

P = proportion of the (i)th life stage duration relative to total life stages in degree days;

N; = number of individuals in the (i)th life stage in sample;

· (i) = life stage number.

Carruthers (1979) estimated mean degree day requirements for the life stages of <u>D</u>. antiqua from the literature. These estimates were used to derive P_i values for equation (1) (Table 9). WMI indexes were determined for each sample period replicate within each planting time subplot.

Three way analysis of variance was conducted on the WMI determinations by planting time, soil treatment, sample period, two-way and three-way interactions (Table 10). Since treatment combinations were not replicated, statistical theory suggests that insignificnt interaction terms be pooled for error mean square to perform modified F-tests on the main variables (Cochran 1957; Yates 1935; Cornish 1936). All interaction terms proved insignificant in the analysis (Table 10). The terms were pooled for use as error mean square in modified F-tests in a second analysis of variance. Planting time and sample period were the only significant variables in this analysis (Table 11).

Table 10. Mean degree day requirements (base 3.8^{0} C) for the life stage of \underline{D} . antiqua and the proportion of total development time spent in each lifestage (P_{i}) .

| | egg | lst instar | 1st instar 2nd instar 3rd instar | 3rd instar | pupa | adult |
|-----------------|------|------------|----------------------------------|------------|-------|-------|
| | | | , | , | , | |
| ٦. | .067 | .050 | . 119 | .216 | .410 | .138 |
| *Mean DD | 90.0 | 67.0 | 160.0 | 290.0 | 550.0 | 185.0 |
| -S - | 8.5 | 3.1 | 4.4 | 24.0 | 19.2 | 25.2 |
| × | | | | | | |

* Carruthers 1979

Table 11. Three way analysis of variance for WMI evaluated by planting degree day (PDD), soil treatment (ST), sample degree day (SDD) and interaction terms.

| Source | SS | DF | MS | F |
|----------------|---------|-----|---------|-------------|
| Main effects | 1667.29 | 1 | 1667.29 | 11490.10*** |
| PDD | 2.25 | 2 | 1.12 | 7.76*** |
| ST | 0.26 | 1 | 0.26 | 1.83 |
| SDD | 12.54 | 11 | 1.14 | 7.86*** |
| interactions | 9.31 | 50 | 0.19 | 1.35 |
| PDD ·ST | 0.07 | 2 | 0.03 | 0.24 |
| PDD·SDD | 3.94 | 19 | 0.21 | 1.43 |
| ST · SDD | 1.66 | 11 | 0.15 | 1.04 |
| PDD·ST·SDD | 3.64 | 1.8 | 0.20 | 1.39 |
| residual error | 14.95 | 103 | 0.14 | |

Bartlett X^2 test for homogeneity of variance: p = .198

Table 12. Three way analysis of variance for WMI evaluated by planting degree day (PDD), soil treatment (ST) and sample degree days (SDD) with insignificant interaction terms pooled for modified F-tests.

| Source | SS | DF | MS | F |
|----------------|---------|-----|---------|------------|
| | | | | |
| Main effects | 1667.29 | 1 | 1667.29 | 8750.67*** |
| PDD | 2.25 | 2 | 1.12 | 6.05*** |
| ST | 0.26 | 1 | 0.26 | 1.42 |
| SDD | 12.54 | 11 | 1.14 | 6.12*** |
| interactions | 9.31 | 50 | 0.19 | |
| residual error | 14.95 | 103 | 0.14 | |

Scheffe multiple range tests were made separately on planting times and sample periods (Tables 12 and 13). Planting times were not detectably different from each other. This was likely due to the lack of strong homogeneous variance (p = .198). Sample periods did contain significant differences. The first, second, third and fourth larval samples taken from all subplots on June 9, 16, 19 and 23, respectively had significantly different WMI mean estimates from the samples taken on August 13. The samples taken on July 9 were also significantly different from those taken on June 16 and 19 in their estimates of the average WMI.

Discussion

The mean WMI does not apparently differ between planting times across the season. Differences might have been significant if each planting time treatment was contrasted with the others by each sample period. The analysis of variance did not indicate that this was necessary since the interaction of the times of planting and sample period was not significant (p = .22). If the interaction was strongly significant then the two parameters would be assumed to be non-parallel in their simultaneous effect on WMI and individual contrasts would be required.

It was not surprising that the presence or absence of Dyfonate was not a significant variable. The WMI's estimate of the immature population's overall maturity indicates that the population matures at a rate that is unaffected by the soil treatment. This is reasonable since it can be assumed to be the first instar larvae that are killed by the soil insecticide. The chemical decreases the number of larvae entering the onions. It does not appear to affect the surviving larvae's rate of growth.

Table 13. Scheffe multiple range comparisons of WMI by planting date.

| Planting date | sample size | mean* | standard error |
|---------------|-------------|-------|----------------|
| April 27 | 63 | 3.25d | 0.06 |
| May 11 | 59 | 3.08d | 0.06 |
| May 25 | 46 | 3.10d | 0.07 |

^{*} means followed by the same letter are insignificantly different (p = .05)

Table 14. Scheffe multiple range comparisons of WMI by sample period.

| Sample date | sample size | mean* | standard error |
|-------------|-------------|---------|----------------|
| June 9 | 10 | 2.83ab | 0.07 |
| June 16 | 12 | 2.75a | 0.17 |
| June 19 | 13 | 2.74a | 0.13 |
| June 23 | 15 | 2.81ab | 0.07 |
| June 29 | 17 | 3.10abc | 0.07 |
| July 2 | 17 | 3.16abc | 0.10 |
| July 6 | 16 | 3.41abc | 0.10 |
| July 9 | 17 | 3.50bc | 0.09 |
| July 13 | 15 | 3.34abc | 0.14 |
| July 16 | 15 | 3.30abc | 0.10 |
| July 23 | 16 | 3.22abc | 0.11 |
| August 13 | 5 | 3.71c | 0.16 |

^{*} means followed by the same letter are insignificantly different (p = .05)

The significant differences in the mean WMI denoted by the Scheffe test across the sample periods does not reveal a striking trend. This could be for a combination of reasons. First, the sample period differences were derived by pooling across the planting time treatments. This could have been inappropriate since the interaction of planting time and sample was only mildly not significant (p = .22). Second, the time span of June 9 to July 23 under which larval sampling was conducted likely overlapped two larval broods of D. antiqua (Whitfield 1981). This would have caused confounding of the later instar larvae of the first brood with the early instars of the second by the index. Third, the method of sampling employed was biased against the pupal life stage since samples were removed from row sections only where damaged plants were aggregated. If damaged plants were blown away or totally dessicated after the larvae pupated the area would not be sampled and the pupae would not be fully monitored. Because of these three concerns the WMI does not appear to be a useful index of the population maturity of D. antiqua for the experimental design and sampling scheme used in this study.

Instar density index

The number of individuals for each sampling replicate in each lifestage was used to formulate an instar density index estimate. The index is not a density estimate per se because of the bias of sampling only where damaged plants were aggregated. A field estimate of the number of damaged plant aggregates for each subplot at each sample period should have been made along with a count of the number of damaged plants within each sampled replicate to be able to adjust for the sampling bias. Assumptions were made therefore concerning the density and distribution of damage to calculate the index.

The index was derived by assuming that the number of plants in the sampled 0.6 meter length of aggregate damage was equal to the stand count and that all the plants were equally infested with larvae. The stand estimate for each planting time subplot that was temporally closest to each larval sample period was used for this (see Tables 2, 3 and 4). The number of each instar per plant in the 0.6 meters was calculated by dividing the larval instar count of each replicate by the estimate of the number of plants in the length (equation 2). This was then multiplied by the product of the damage count made at the subplot's closest stand estimate and the row-meters of onions per acre (equation 3). This resulted in an index of density per acre for each lifestage in each subplot's sample period replicate (equation 4):

$$\frac{X \text{ instar (i)}}{0.60 \text{ m}} * \frac{\text{row-meter}}{Z \text{ plants}} = \frac{X \text{ instar (i)} * 1.667}{Z \text{ plants}}$$
(2)

$$\frac{\text{Y damaged plants}}{\text{row meter}} * \frac{4.7 \times 10^3 \text{ row meter}}{\text{acre}} = \frac{\text{Y damaged plants} * 4.7 \times 10^3}{\text{acre}}$$
(3)

$$\frac{\text{X instar (i) * 1.667}}{\text{Z plants}} * \frac{\text{Y damaged plants * 4.7 x 10}^3}{\text{acre}} = \frac{\text{Y damaged plants * 4.7 x$$

The instar density index derived for each replicate in each sample period was used as a single dependent variable (each instar alone) in a multiple regession. Independent variables were degree day accumulation from April 1 at each planting for adult emergence (base 4.4°C) and degree day accumulation from planting at each sample period for larval development (base 3.8°C). Regessions were conducted separately for each soil treatment. Tables 14 to 21 record the multiple regression statistics for each life stage by soil treatment.

Planting degree day, sample degree day and their interaction terms were significant for all life stages except the pupal in both soil treatments. The pupal stage for both soil treatments (Tables 17 and 21) had only the degree day accumulation at planting as a significant term in the regression on its density index. This could have been due to the bias of sampling in only areas of aggregated plant damage.

Discussion

This analysis indicates two important phenomena of the <u>D</u>. <u>antiqua</u> immatures occurring in the subplots; the population was smaller for all instars in the treated subplots and the population did not respond in a parallel manner between planting times and sample periods. The y-intercept for the instar density index regressions of the untreated plots ranged from 4.0 to 9.1 (excluding pupae). The y-intercepts for the regressions in the treated subplots ranged from .29 to .60 (excluding pupae). These relative differences appear to signify that the immature density indexes were on the average higher in the untreated subplots than the treated ones. This correlates well with the stand density data of Table 5.

Table 15. Multiple regression statistics for the density index of first instar larvae under treated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|--------|
| Regression | 3 | 0.292 | 0.097 | 4.07** |
| Residual | 104 | 2.492 | 0.024 | |

^{**} (p = .01)

| Variable | F to enter | R^2 | В |
|--------------------|------------|-------|--------------------------|
| Planting DD | 6.41** | .05 | - 0.617×10 ⁻³ |
| Sample DD | 6.82** | .02 | -0.138×10^{-3} |
| Planting sample DD | 3.62* | .03 | 0.304×10^{-6} |
| Constant | | | 0.284 |

^{*} (p = .10) ** (p = .01) $R^2 = .1050$

Table 16. Multiple regression statistics for the density index of second instar larvae under treated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Suqare | F |
|------------|-----|----------------|-------------|---------|
| Regression | 3 | 1.210 | 0.405 | 4.63*** |
| Residual | 104 | 9.100 | 0.087 | |

^{***} (p = .005)

| Variable | F to enter | R ² | В |
|--------------------|------------|----------------|--------------------------|
| Planting DD | 6.95** | .06 | - 0.123×10 ⁻² |
| Sample DD | 7.26** | .02 | -0.273×10^{-3} |
| Planting·sample DD | 3.74* | .03 | 0.591×10 ⁻⁶ |
| Constant | | | 0.571 |

^{*} (p = .10) ** (p = .01)

 $R^2 = .1178$

Table 17. Multiple regression statistics for the density index of third instar larvae under treated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|----------|
| Regression | 3 | 1.54 | 0.515 | 16.70*** |
| Residual | 104 | 3.20 | 0.031 | |

^{***} (p = .001)

| Variable | F to enter | R^2 | В | |
|--------------------|------------|-------|--------------------------|--|
| Planting DD | 21.07*** | .22 | - 0.127×10 ⁻² | |
| Sample DD | 16.87*** | .04 | -0.247×10^{-3} | |
| Planting sample DD | 8.82** | .02 | 0.539×10 ⁻⁶ | |
| Constant | | | 0.599 | |

Table 18. Multiple regression statistics for the density index of pupae under treated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Suqare | F |
|------------|-----|----------------|-------------|----------|
| Regression | 3 | 0.578 | 0.193 | 13.99*** |
| Residual | 104 | 1.432 | 0.014 | |

^{***} (p = .001)

| Variable | F to enter | R^2 | В |
|--------------------|------------|-------|--------------------------|
| Planting DD | 6.47** | .28 | - 0.469×10 ⁻³ |
| Sample DD | 0.51 | .00 | - 0.286×10 ⁻⁴ |
| Planting sample DD | . 0.51 | .01 | 0.798×10 ⁻⁷ |
| Constant | | | 0.212 |

^{**} (p = .01)

 $R^2 = .2875$

Table 19. Multiple regression statistics for the density index of first instar larvae under untreated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|---------|
| Regression | 3 | 60.86 | 20.28 | 5.65*** |
| Residual | 103 | 387.31 | 3.58 | |

*** (p = .001)

| Variable | F to enter | R^2 | В |
|--------------------|------------|-------|---------------------------|
| Planting DD | 9.06** | .06 | - 0.8909x10 ⁻² |
| Sample DD | 10.36** | .03 | -0.2003×10^{-2} |
| Planting·sample DD | 5.42* | .04 | 0.4507×10 ⁻⁵ |
| Constant | | | 4.0502 |

^{* (}p = .10) ** (p = .01)

 $R^2 = .1358$

Table 20. Multiple regression statistics for the density index of second instar larvae under untreated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|----------|
| Regression | 3 | 294.66 | 98.22 | 11.25*** |
| Residual | 108 | 943.06 | 8.73 | |

^{***} (p = .001)

| Variable | F to enter | R ² | В |
|--------------------|------------|----------------|--------------------------|
| Planting DD | 18.29*** | .07 | - 0.197x10 ⁻¹ |
| Sample DD | 22.05*** | .08 | -0.456×10^{-2} |
| Planting sample DD | 11.43*** | .08 | 0.201×10^{-4} |
| Constant | | | 9.072 |

^{***} (p = .001)

 $R^2 = .2380$

Table 21. Multiple regression statistics for the density index of third instar larvae under untreated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|----------|
| Regression | 3 | 133.79 | 46.26 | 11.78*** |
| Residual | 108 | 423.91 | 3.92 | |

^{***} (p = .001)

| Variable | F to enter | R ² | В |
|--------------------|------------|----------------|--------------------------|
| Planting DD | 22.02*** | .12 | - 0.286×10 ⁻² |
| Sample DD | 19.32*** | .03 | -0.145×10^{-1} |
| Planting·sample DD | 13.26*** | .09 | 0.737x10 ⁻⁵ |
| Constant | | | 6.322 |

^{***} (p = .001)

 $R^2 = .2466$

Table 22. Multiple regression statistics for the density index of pupae under untreated soil field conditions characterized by planting degree day and sample degree day.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|-----|----------------|-------------|--------|
| Regression | 3 | 17.92 | 5.98 | 3.67** |
| Residual | 108 | 175.80 | 1.63 | |

^{**} (p = .01)

| Variable | F to enter | R^2 | В |
|--------------------|------------|-------|--------------------------|
| Planting DD | 2.76* | .22 | - 0.331x10 ⁻² |
| Sample DD | 0.35 | .07 | 0.248×10^{-3} |
| Planting·sample DD | 1.22 | .02 | 0.144×10^{-5} |
| Constant | | | 0.441 |

^{* (}p = .10)

 $R^2 = 0.3042$

The significance of the degree day accumulation at planting with the degree day accumulation at each sample period indicates a non-parallel relationship between the two parameters. The immature density index responded across the sample periods differently for each planting time treatment. This can be observed by pooling the immature density indexes for each subplot within each sample period and plotting the total immature density indexes across the season. Figures 8, 9 and 10 are the graphs for each planting time and soil treatment.

The density indexes for the total immatures depict both the marked difference between soil treatments as well as the differential response across the sample season between planting times. A relative trend of decreasing immature density indexes in both treated and untreated subplots as planting times are delayed is also apparent. The untreated density index for the immatures of the April 27 planting peaked at almost 125,000 per acre early in the season. The untreated immatures of the May 11 planting peaked around 38,000 per acre and the May 25 planting's peaked around 880 total immatures per acre. The treated immatures followed a similar trend as the plantings were delayed.

The lack of correspondence of the pupal density indexes with the larval instar's indicates an artifact of the sampling bias. Samples were chosen on the basis of evidence of larvae by aggregates of damaged plants. If the section of row had been damaged long enough for a number of pupae to develop it was likely that the plants had dried and were blown away. Areas of damaged row evidenced by missing plants were not sampled and hence a major portion of the pupae were not monitored. This could have caused the pupal density indexes to appear as an anomoly in regard to the instar indexes.

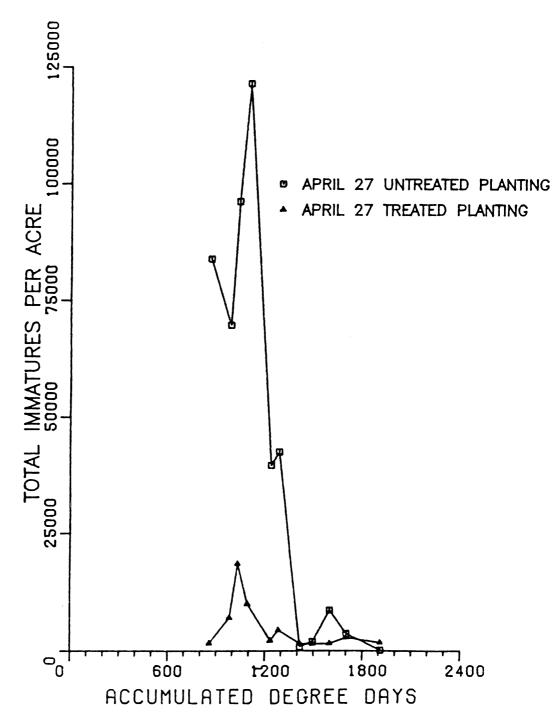


Figure 7. Pooled immature density index for the April 27 planting by soil treatment and sample period.

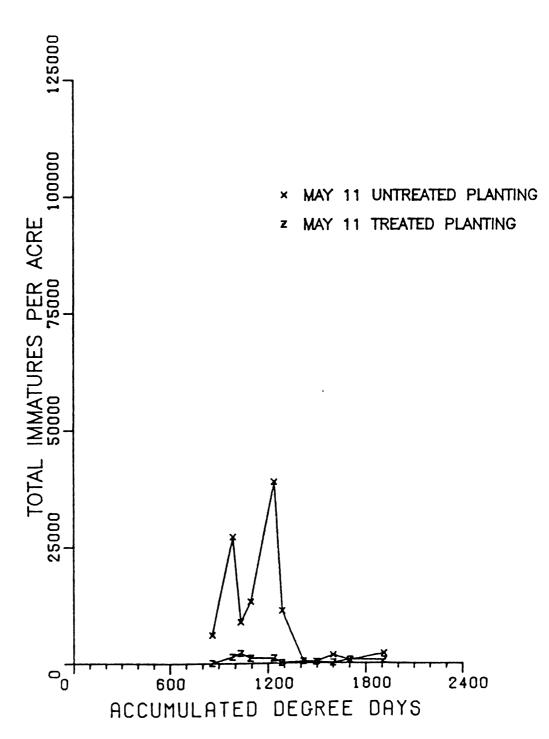


Figure 8. Pooled immature density index for the May 11 planting by soil treatment and sample period.

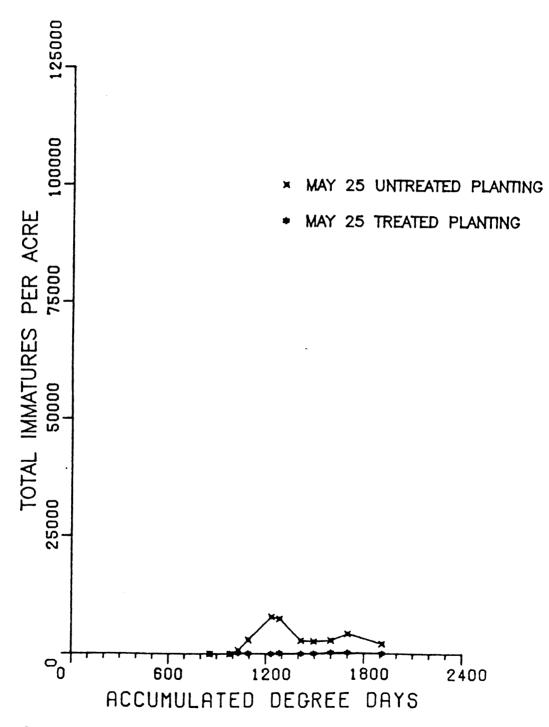


Figure 9. Pooled immature density index for the May 25 planting by soil treatment and sample period.

Total production

Comparing densities between field subplots, sampled across time, can also be done by determining the total sample season production of a lifestage per unit area. Total seasonal production can be calculated in the following manner (Southwood 1966, 1976; Helgeson and Haynes 1969; Kiritana and Nakasuji 1967; Manly 1976; Lampert 1980)

$$TI_i = \Sigma (((DD_{ij} + D_{i,j+1})/2) * (DD_{j+1} - DD_j))$$
 (5)

where D_{ij} = density of the (i)th life stage on the jth sample date DD_{j} = accumulated degree days on the jth sample date

$$TP_{i} = TI/DD_{i}$$
 (6)

where DD_i = developmental time in degree days of the (i)th life stage

Total incidence (TI) provides an estimate of the total time (life stage degree days) a life stage is present in an area. Dividing the total incidence by the developmental time of the life stage (in degree days) adjusts for redundant observations. This adjustment results in total production (TP), the total number of individuals in a sample season estimated to move through a unit area in a field.

The <u>D</u>. antiqua instar density index data were transformed by this method to estimate the number of each instar that occurred per acre in each planting time subplot through the sample season. Density index replicates for each

treatment subplot were averaged within each sample period. The average density index for each sample period was used in the lifestage production algorithm for each lifestage. The analysis was performed with this data up to 1912 degree days (July 23). After this time the intervals between sample periods were too great for inclusion. Overlapping life stages require that the population be sampled frequently to estimate the number of individuals of a life stage produced per unit of habitat (Helgeson and Haynes 1969).

Table 22 lists the number of each life stage estimated to have passed through a unit acre. Since treatment combinations were not replicated no statistics are available other than means. The differences are only relative. The relative differences do corroborate well with the analysis of the stand reduction and density index data. Immature production totals show that each untreated subplot produced a greater relative density of immatures than its treated counterpart. This was also apparent for the individual lifestages.

The production trends for each lifestage, as the planting time is delayed also show consistent relative trends (Table 22). A greater number of each immature life stage is produced in the untreated subplots in each planting time treatment. As the planting time is delayed the contrast between treated and untreated remains. The delayed plantings did elicit a marked relative decline in the total immatures and the individual instars within a soil regime.

The total immature production in each planting time subplot, used as a percentage of total immature production for the whole experimental site keynotes significant trends for the experiment. Figure 11 shows the relationships. The trend of immatures produced per planting time subplot as the plantings were delayed is very similar to the total immature density index data

Table 23. Seasonal production per acre for each instar and total immatures by planting time and soil treatment.

| | | | Planting time | time | | | |
|----------------|-----------|----------|---------------|---------|-------------------|---------|--|
| | Apri | April 27 | May 11 | 11 | May 25 | 25 | |
| Lifestage | untreated | treated | untreated | treated | untreated treated | treated | |
| first instar | 156,368 | 8,314 | 20,135 | 601 | 3,445 | 169 | |
| second instar | 134,936 | 7,507 | 24,211 | 627 | 7,180 | 201 | |
| third instar | 58,978 | 5,791 | 14,449 | 1,390 | 7,252 | 210 | |
| pupa | 6,389 | 1,239 | 1,510 | 261 | 981 | 9/ | |
| immature total | 358,671 | 22,851 | 60,305 | 2,879 | 18,858 | 656 | |

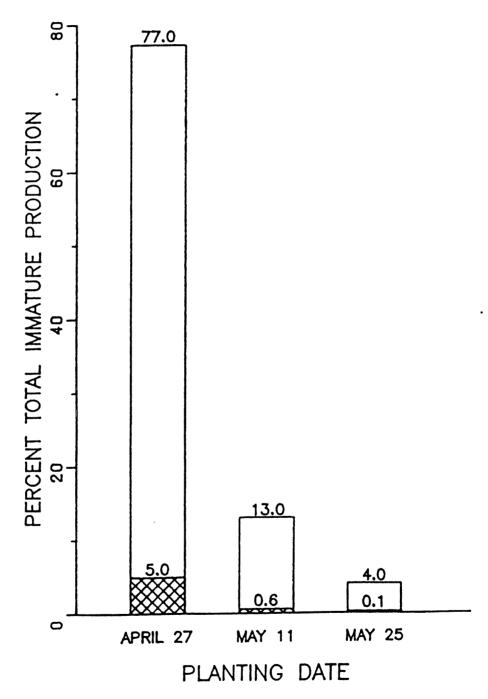


Figure 10. Total immature production by planting time and soil treatment as a percentage of total immature production for the whole experimental site.

plotted across time (Figures 8, 9 and 10). The untreated planting of April 27 produced 77 percent of the total <u>D</u>. antiqua immatures in the study site. The May 11 planting's untreated subplot produced 13 percent and the May 25 planting's produced 4 percent. The treated subplots show a similar decline in the percent of total immatures produced as the plantings were delayed.

The rate of change in the proportion of total immatures for the whole site as a function of degree day accumulation at planting can be contrasted between soil treatments to determine if a similar response occurred as the plantings were delayed. The slopes of the regressions of the pecent production of immatures as a function of planting times can be tested for parallel trends with a modified F-test (Neter and Wasserman 1974). Essentially, the difference between the mean square errors of the full model (two separate regressions) and the reduced model (data pooled into one regression) is tested for significance.

Table 23 contains the regression statistics for the comparisons of the regressions of degree day accumulation at planting on the percent production of <u>D. antiqua</u> immatures for the study site in each soil treatment. The slopes of the regression lines were determined not to be significantly different. This signifies that a similar functional response of immature <u>D. antiqua</u> was occurring across the planting times for both soil treatments.

Discussion

The analysis of the total production of the immatures was useful as a summary technique of the density index data as well as allowing for a statistical comparison to be made of the response of \underline{D} , antiqua to the two soil treatments. The total production data explains, in part the different levels of stand loss observed in the subplots.

Table 24. Regression statistics for testing slopes of the percent of total immature production as a function of planting degree days from the two soil treatments.

| Full model: | $Y_{ij} = B_{oj} + B_{ij}X_{ij} + E_{ij}$ | <pre>i = planting degree days j = soil treatment (Q or 1)</pre> |
|-------------|---|---|
|-------------|---|---|

Regression coefficients:

Treated

Untreated

$$b_{01} = 4.306$$
 $b_{11} = -0.8997 \times 10^{-2}$

 $b_{00} = 67.39$

 $b_{10} = -0.1357$

| source | SS | DF | source | SS | DF | |
|------------|--------|----|------------|--------|----|--|
| regression | 10.007 | 1 | regression | 2295.6 | 1 | |
| residual | 3.860 | 1 | residual | 895.0 | 1 | |
| total | 13.867 | 2 | total | 3190.6 | 2 | |

Sum of Squares for Full model:

$$SS_{E}(F) = SS_{E_{1}} + SS_{E_{2}} = 3.860 + 895.0 = 898.86$$

Reduced model:
$$Y_{ij} = B_0 + B_i X_{ij} + E_{ij}$$
 $i = 1,2,3$ $j = 1,2$

Regression coefficients:

Analysis of Variance:

$$b_0 = 35.81$$
 $b_1 = -0.07227$
 $F = SS_E(R) - SS_E(F) / \frac{SS_E(F)}{2}$
 $= 2.57$

$$F_{.05, 2, 2} = 19.00$$

The untreated subplots are most apparent in the depiction of overall trends. The April 27 and May 11 plantings' untreated subplots had the most extensive stand reductions (Table 5) as well as the largest percentage of D. antiqua immatures of 77 and 13 percent respectively (Figure 11). The two stands however were not significantly different in their final stand densities (1.2 and 5.2 plants/meter, respectively). This indicates that the larvae were likely at higher densities per damaged plant in the April 27 planting than the May 11 planting. In contrast, the May 25 planting's untreated subplot, with only an estimated four percent of the total immatures would have had a very low ratio of larvae to damaged plant to account for its 34 percent stand reduction.

The treated subplot data also shows a lack of correspondance between the reduction of the stands and the proportion of immatures that were estimated to occur in them. The April 27 treated planting with five percent of the estimated total population sustained only a relative eight percent stand reduction. A very high ratio of larvae to damaged plant in contrast to the May 25 planting's untreated data would have occurred in this treated stand.

The speculation made in regard to the adult female onion fly activity peak is reinforced by this analysis. The larval populations are augmented on areas of previous damage. These areas likely provided a major sink for the eggs being oviposited in late June. These comparisons would imply that the density of immatures is not directly proportional to the amount of stand reduction that occurred.

Dyfonate

Dyfonate residue levels in parts per million are listed in Table 24 by planting time and sample period. The data were best fit to a lognormal distribution with a mean of 3.46 and a variance of 20.20. The data were transformed by log(x) for further analysis.

Weather data were used from the Grant, Michigan State Agricultural Experiment Station to characterize the levels of Dyfonate residue across time in a multiple regression analysis. The degree day base used for Dyfonate degradation was 0° C. Independent regression variables chosen for the analysis were

- 1) Julian date of planting;
- 2) degree days accumulated at planting from April 1;
- 3) days from planting to sample period;
- 4) degree day accumulation from planting to sample period;
- 5) degree days accumulated at sample period from April 1;
- 6) accumulated precipitation from planting;
- 7) accumulated precipitation from previous sample

The two lowest residue data from the early samples (0.24 and 0.39 ppm) and the two highest from the later samples (7.3 and 6.6 ppm) were considered outliers and were excluded from the analysis. The correlation coefficient matrix for all variables is listed in Table 25. Four of the seven independent variables were approximately 30 percent correlated with the residue of Dyfonate; days from planting, degree days from planting, degree day of sample period and accumulated precipitation from planting. Degree days from planting was at

Table 25. Dyfonate residue in parts per million (ppm) by planting date and sample period.

| | | Plantin | g date | |
|----------------|----------|----------|--------|--------|
| Sample period | April 15 | April 27 | May 11 | May 25 |
| april 30 | 7.73 | 7.20 | | |
| lay 4 | 0.24 | 4.78 | | |
| May 7 | 1.95 | 0.39 | | |
| May 11 | 0.92 | 0.53 | 3.54 | |
| May 14 | 2.48 | 9.32 | 8.25 | |
| iay 18 | 6.31 | 1.24 | 9.98 | |
| May 21 | 1.44 | 8.20 | 8.46 | |
| fay 25 | 2.43 | 5.88 | 3.61 | 8.83 |
| lay 2 8 | 0.34 | 4.80 | 4.42 | 0.84 |
| Tune 1 | 0.43 | 4.21 | 8.77 | 1.67 |
| Tune 4 | 2.37 | 4.11 | 4.84 | 0.77 |
| Tune 8 | 0.99 | 1.04 | 0.89 | 0.94 |
| June 11 | 6.87 | 2.90 | 5.92 | 2.05 |
| Tune 19 | 1.76 | 1.17 | 6.95 | 0.65 |
| fune 23 | 0.40 | 0.91 | 0.70 | 5.95 |
| Tune 29 | 6.60 | 0.48 | 2.65 | 3.88 |
| July 2 | 2.52 | 0.50 | 1.61 | 4.08 |
| July 6 | 0.64 | 7.30 | 2.62 | 2.26 |
| July 9 | 0.11 | 1.82 | 4.30 | 0.21 |
| July 13 | 1.58 | 2.16 | 1.89 | 2.26 |
| July 16 | 2.24 | 0.51 | 2.48 | 2.89 |
| July 23 | | | 1.88 | 2.92 |
| July 31 | | | 2.33 | 2.37 |

Table 26. Correlation coefficients of selected independent variables in a regression analysis on Dyfonate residue in parts per million.

| | | | | | | 0.157 | (9) |
|------------------------------|------------------------------|-----------------------|---------------------------------|-----------------------------------|---|---|-----------------|
| | | | | | 0.864 | 0.081 | (2) |
| | | | | 0.875 | 0.975 | 0.101 | (4) |
| | | | 0.922 | 0.756 | 0.910 | 0.135 | (3) |
| | | - 0.291 | - 0.204 | 0.295 | - 0.176 | - 0.035 | (2) |
| | 0.991 | - 0.299 | - 0.208 | 0.287 | - 0.185 | - 0.036 | (1) |
| 0.159 | 0.124 | - 0.329 | - 0.363 | - 0.292 | - 0.363 | 0.081 | Dyfonate ppm |
| 1) Julian day of planting | 2) degree day of planting | days from planting | 4) degree days from planting | 5) degree day of sample period | <pre>6) precipitation from planting</pre> | 7) precipitation from previous sample | |
| 1) | 2) | 3) | 4) | 5) | (9 | 7) | |

least 90 percent correlated with both the accumulated precipitation from planting and days from planting and 80 percent correlated with degree days at sample period. Degree days from planting was therefore a good representation of the other variables that it was highly correlated with. Testing the remaining three independent variables with degree days from planting in a step-wise regression derived a significant regression coefficient for only the variable of degree days from planting. Table 26 contains the regression statistics for degree days from planting on the residue of Dyfonate in parts per million of the log transformed data.

Degree days accumulated from planting was not a controlled variable in this analysis. Each planting time subplot had independent intervals of accumulated degree days from planting at each sample period. To determine what interval of degree days from planting was neccessary for significant differences to occur the data was pooled at selected intervals.

Intervals of 600 degree days from planting were the smallest that expressed significant differences in a Scheffe multiple range test across the sample season (Table 27). The first two 600 degree day intervals averaged insignificantly different levels of Dyfonate residue. The last two (1200 DD to 1799 DD and 1800 DD to 2600 DD) intervals also averaged insignificantly different levels of residue as indicated by the transformed data. These two separate sets of intervals however, were significantly different from each other. The trend of lower Dyfonate residue counts was not significant unless the data of the first 1200 degree days from planting was contrasted with the remaing data for the whole experiment.

Table 27. Regression statistics for Dyfonate residue in parts per million transformed by $\log(x)$ under field conditions characterized by degree days from planting.

| Source | DF | Sum of Squares | Mean Square | F |
|------------|----|----------------|-------------|--------|
| Regression | 1 | 1.79 | 1.79 | 10.9** |
| Residual | 72 | 11.82 | 0.16 | |

^{**} (p = .001)

 $Y = 0.575 - 2.139 \times 10^{-4} (DD from planting) \pm 0.05$

 $R^2 = .131$

Table 28. Scheffe multiple range comparisons of means of log(x) transformed Dyfonate residue data at 600 degree day intervals from planting.

| degree days from planting | sample size | mean* | standard error |
|------------------------------|-------------|--------|----------------|
| 0 - 599 | 19 | 0.486a | 0.09 |
| 600 - 1199 | 20 | 0.496a | 0.08 |
| 1200 - 1799 | 20 | 0.145b | 0.08 |
| 1800 - 2600 | 15 | 0.130b | 0.10 |

^{*} numbers followed by a common letter are insignificantly different (p = .05)

Discussion

The four variables most correlated with the Dyfonate residue were all forms of time (days from planting, degree days from planting, degree day of sample period, and accumulated precipitation from planting). All were negatively correlated with the Dyfonate residue in ppm and highly correlated with each other. Each of these variables, increased in their value represented a decrease in the associated level of Dyfonate. The regression analysis however, had a very low R² value (0.13) for degree days from planting. Only general statements about an independent variable's affect on the residue level is possible.

The sampling methods employed do not allow for predictions of residue levels across the planting times or sample periods because of the large unexplained variance. This variance could have been accounted for in an analysis of variance if sample replicates were made from the field, the cement mixer as well as the laboratory analysis. The sampling intervals would need to remain relatively frequent (no greater than a week apart) in order to make comparisons with the larval and stand data. Replication of this nature was considered cost prohibitive in the study.

If the Scheffe multiple range test on the 600 degree day intervals did signify a trend actually characteristic of the Dyfonate degradation in the subplots then an inference can be made on the possible affect of the degradation of Dyfonate on stand reduction as planting times were delayed. The Dyfonate trend would appear to depict the residue sustaining itself for the first two 600 degree day periods and then drop more than 50 percent in its average concentration for the last two derived intervals. This would imply that the four plantings

of April 15, April 27, May 11 and May 25 had relatively high levels of Dyfonate at least until May 31, June 8, June 20 and July 5, respectively. After these dates of 1200 degree days from planting the Dyfonate would be expected to occur at appreciably lower levels in the subplots.

The times of Dyfonate residue thresholds contrasted with the stand reduction relative trend in the treated subplots portends a speculation on the influence that the level of Dyfonate might have had. Although the stand densities of the treated subplots were not significantly different from their respective initial stand counts (Table 5), a relative trend of decreasing stand reduction as plantings were delayed was apparent. The period of peak <u>D</u>. antiqua ovipostion was assumed to be occurring around June 20. This would have been after the 1200 degree day interval of Dyfonate degradation from planting for both the April 15 and 27 plantings. A decrease of the Dyfonate residue might possibly explain the relatively higher stand losses in the two early plantings.

The larval production trend however belies this possible effect of the level of Dyfonate residue. As discussed previously, both treated and untreated immature production rates as a function of planting times were parallel in nature. The relatively higher immature populations occurring in the earlier plantings of both the untreated and treated plots were believed to have arisen due to higher densities of larvae per damaged plant as opposed to more damaged plants per se. With the possibility of damage initially occurring randomly throughout a field and then expanding contagiously around the initial sites as cited by Whitfield (1981) it is difficult to partition the lower Dyfonate residue trend from the fly attraction to previously damaged areas in the variance of delayed planting data.

VOLUNTEER DENSITY AND ROW PLACEMENT

Whitfield (1981) determined the size of aggregated <u>D</u>. antiqua damage influenced by volunteer onion plants. Mean plant damage clump sizes surrounding single and aggregate volunteer densities were 32 cm and 35 cm in radius, respectively. Whitfield speculated that the area a volunteer affects <u>D</u>. antiqua damage may be geater than his study revealed. The population of <u>D</u>. antiqua damaged plants near sites of volunteers may consist of several distinct aggregates. An experiment investigating this possible phenomenon was conducted the spring and summer of 1982 at the Michigan State University Organic Soils Research Farm in Laingsburg, Michigan.

Methods

Two independent parameters were manipulated in this study; the size of the volunteer aggregate and the placement of the aggregate with respect to the onion row (Figure 12). Transplanted sprouting culls from a commercial field were used for the volunteer aggregates. The treatment combinations of size and placement were each transplanted into the center portion of three, five meter single rows of Spartan Banner onions. Three, five meter long guard rows separated each of the three treatment rows. Rows were 0.5 meters apart.

Aggregates were placed either within the center row of the three treatment rows or half of the way between two of the three for the placement variable. The size of the aggregates consisted of one, two and three volunteers. The single volunteer aggregates were placed at the 2.5 meter row-center of each treatment placement, either within the center row or half way between two of

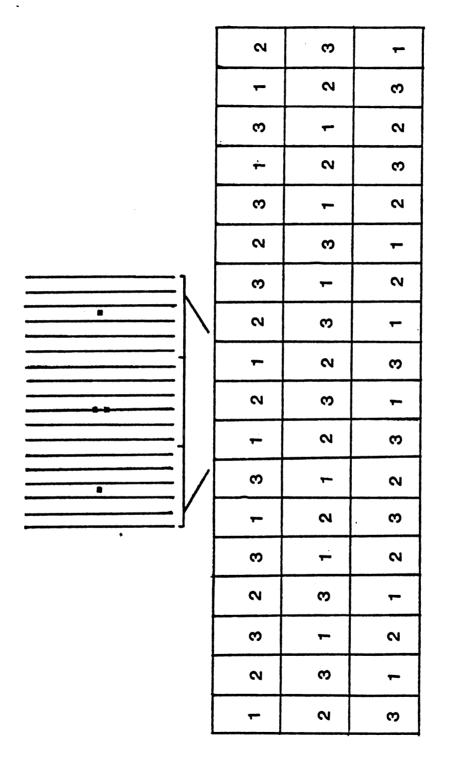


Figure 11. Plot design of one quarter acre of onions with aggregate volunteer treatments of one, two and three transplants alternated by between row and within row placement.

the three. The other aggregate sizes were spaced from the center of the treatments as defined by the locations of the single volunteer aggregates.

The transplants for the two volunteer aggregates were each spaced 15 cm from the previously defined centers along the length of the rows. The three volunteer aggregate transplants were placed with one at the center location and the other two each 15 cm distant. These occurred parallel with the onion row orientation for both the within row and between row placements of the aggregates.

The treatment combinations were completely randomized in 18 replicates across two, quarter acre plots. Plots were seeded May 18; volunteers were transplanted June 1 and plant sampling was conducted June 10, 1982. No pesticide applications were made in the plots.

Plant sampling

A block section of almost one third half of each quarter acre plot was sampled for plant damage on June 10, 1982. In one plot 13 treatment combinations and 14 sets of guard rows were entered. In the other plot 16 treatment combinations and 15 sets of guard rows were entered. The exact location of each damaged onion was recorded by its x,y co-ordinate in the plot and the treatment combination or guard row which it occurred in.

Results

Three analyses were made on the data set:

 the effect of the volunteer aggregate on the distribution of plant damage of the row that it was in or adjacent, at increasing interval distances away from the volunteers;

- 2) the effect of the volunteer aggregate's size on the rate of damage in the whole row of onions in which it resided or was adjacent; and
- 3) the effect of the treatment density of volunteers by their placement on the rate of plant damage within the surrounding treatment rows.

The effect a volunteer aggregate had within a row of onions and between onion rows on plant damage was assumed to be most detectable in the treatments of the three volunteer aggregates. Contrasting damage rates in interval distances (0 - 1 feet, 1 - 2 feet, 2 - 3 feet, etc.) away from the aggregates of the three volunteers allowed an assessment of any differences in the distribution of damage within a row that the aggregate occurred or was adjacent.

The x,y co-ordinate data was used to ascertain damage rates in eight, one foot intervals away from the volunteers for the onion rows that they were within or adjacent. The intervals that damage rates were determined for were; 0 - 1 feet, 1 - 2 feet, 2 - 3 feet, 3 - 4 feet, 4 - 5 feet, 5 - 6 feet, 6 - 7 feet, and 7 - 8 feet away from the center of the three volunteer aggregate. Figure 13 is a graph of the average damage rate for the one foot intervals away from the aggregates for each volunteer placement.

The damage for both placements appeared highest in the vicinity of the aggregates (0 - 1 feet) with a decline as the distance away from the volunteers increased. The trend of the damage appears to differ for the two placements in its apparent rate of decline. The between row volunteers show a steady, gradual decline in average damage as the distances increase. The aggregate of three volunteers within an onion row however, elicited a more rapid decline in damage away from them with a slight increase at the ends of the rows.

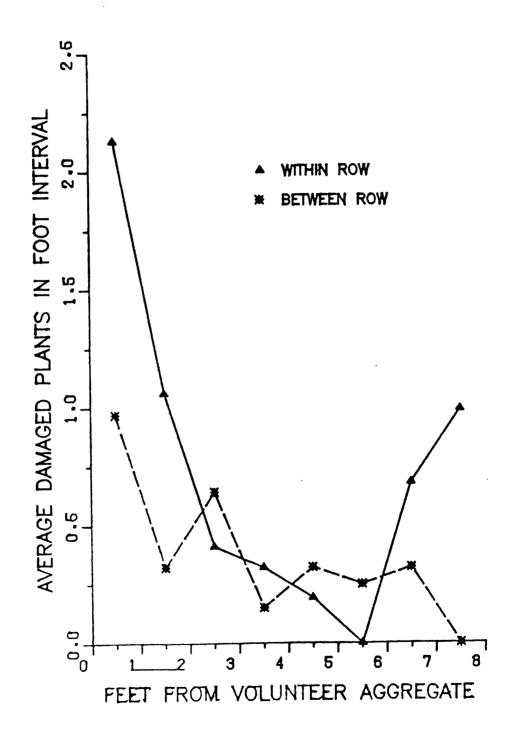


Figure 12. The average damage rate for one foot intervals away from a three volunteer aggregate at each treatment placement.

Tables 28 and 29 list the regression statistics for the average damage in a foot of onion row at one foot interval distances from the three volunteer aggregates for each of the treatment placements. These regressions were performed for data up to and including six feet from the aggregate. The within row aggregates' damage rate decreased at .38 damaged plants per interval foot while the between row aggregates decreased at .11 damaged plants per interval foot away from the aggregates. The regessions were made only up to six feet since the within row volunteer damage rates appeared to increase at the ends of the rows.

The average rates of damage between the two placements at each one foot interval were contrasted for differences. Table 30 records the results of a Scheffe multiple range contrast after the data was transformed by log(x + 1) to normalize the distributions. The three volunteers within a row had significantly more damage within a foot on either side of them than the between row volunteer aggregates. Damage was significantly different from zero for the within row aggregates from 0 to 2 feet away and from 6 to 8 feet away from them. The between row aggregates had damage significantly different from zero for 0 to 1 feet and 2 to 3 feet distances. Damage was on the average not significantly different from zero for 2 to 6 feet for the within row volunteers and from 3 to 8 feet for the between row volunteers. The within row aggregates of three volunteers drew more damage into their immediate vicinity than the between row volunteers with a greater decline in damage as the distance from them increased up to six feet.

The second analysis assessed the affect that the three sizes of volunteer aggregates and their placement had on the overall rate of damage within a row.

Table 29. Regression statistics for the average damage in a foot of onion row at one foot interval distances from three volunteer aggregates at within row placements to six feet.

| Source | DF | Sum of Squares | Mean Square | F | |
|------------|----|----------------|-------------|---------|--|
| Regression | 1 | 2.52 | 2.52 | 17.06** | |
| Residual | 4 | 0.59 | 0.15 | | |

^{**} (p = .01)

Y = 1.83 - 0.38(interval distance)

 $R^2 = .81$

Table 30. Regression statistics for the average damage in a foot of onion row at one foot interval distances from three volunteer aggregates at between row placements to six feet.

| Source | DF | Sum of Squares | Mean Square | F | |
|------------|----|----------------|-------------|------|--|
| Regression | 1 | 0.22 | 0.22 | 4.20 | |
| Residual | 4 | 0.21 | 0.05 | | |

^{* (}p = .10)

Y = 0.78 - 0.11(interval distance)

 $R^2 = .52$

Table 31. Scheffe multiple range comparison of one foot intervals of damage away from three volunteer aggregates by row placement (log(x + 1) data).

| interval distance | within row | | between | n row |
|----------------------|------------|------|---------|-------|
| | mean | s.e. | mean | s.e. |
| - 1 feet | 0.49c | 0.17 | 0.29b | 0.14 |
| l - 2 feet | 0.31bc | 0.11 | 0.12a | 0.07 |
| 2 - 3 feet | 0.15a | 0.15 | 0.21b | 0.09 |
| 3 - 4 feet | 0.12a | 0.12 | 0.06a | 0.06 |
| 4 - 5 feet | 0.07a | 0.07 | 0.12a | 0.07 |
| 5 - 6 feet | 0.00a | 0.00 | 0.10a | 0.09 |
| 6 - 7 feet | 0.22b | 0.07 | 0.12a | 0.12 |
| 7 - 8 feet | 0.30b | 0.12 | 0.00a | 0.00 |

^{*} numbers followed by a common letter are not significantly different (p = .10)

This analysis was done for the rows that the volunteers either resided in or were adjacent. The center guard rows were included as a zero density treatment to serve as controls for the volunteer placements that they were closest to.

Two-way analysis of variance for aggregate size and placement was conducted on the data after transformation by log(x + 1) to normalize its distribution (Table 31). Only the placement of the volunteers was significant with at least 90 percent confidence. In a Scheffe contrast of the two placements the rows that the volunteers resided in obtained a significantly higher average rate of damage than the rows that were adjacent to volunteers (Table 32).

To determine if the center guard rows were able to be addressed as controls they were contrasted with the densities and placement of the aggregates that they were closest to. Table 33 list the results of a Scheffe multiple range contrast across the treatment combinations and their associated guard rows. Significant differences did not occur in mean damage between any of the densities and center guard rows for either aggregate placement. The center guard rows could not be reasonably used as control rows.

The damage rates of the center guard rows however, appear to be proportional to the damage rates of the treatment combinations that they were closest to. The ones closest to within row volunteers had relatively higher damage rates than the ones closer to between row volunteers. This indicates that the increase in the damage that the within row volunteers elicited in the rows in which they resided could also have affected the damage rates of the rows surrounding them. A third analysis was conducted to determine the influence of volunteer aggregates on damage in the onion rows surrounding them.

Table 32. Two way analysis of variance for the damage rate per row that an aggregate of volunteers either resided or was adjacent evaluated by the volunteer density and placement.

| Source | SS | DF | MS | F |
|----------------|-------|----|-------|-----------|
| Main effects | 32.95 | 1 | 32.95 | 183.06*** |
| density | 0.06 | 3 | 0.02 | 0.11 |
| placement | 0.59 | 1 | 0.59 | 3.29* |
| interaction | 0.41 | 3 | 0.14 | 0.76 |
| residual error | 9.36 | 52 | 0.18 | |

^{***} (p = .001) * (p = .10)

Table 33. Scheffe comparison of average damage rate per row that a volunteer aggregate resided or was adjacent by treatment placement $(\log(x+1) \text{ data})$.

| placement | n | mean | standard error |
|--------------------------|----|-------|----------------|
| within row aggregate | 13 | 0.90a | 0.10 |
| between row aggregate | 16 | 0.63b | 0.09 |

^{*} numbers followed by a common letter are not significantly different (p = .10)

Table 34. Scheffe multiple range comparisons of volunteer density and placement for the average damage rate per rows that a volunteer aggregate resided or was adjacent $(\log(x+1))$ data).

| | | Volunteer placement | | | |
|---------|--------|---------------------|---------|------|--|
| | within | row | between | row | |
| density | mean | s.e. | mean | s.e. | |
| 0 | 0.81a | 0.14 | 0.67a | 0.09 | |
| 1 | 0.94a | 0.09 | 0.66a | 0.18 | |
| 2 | 0.76a | 0.25 | 0.77a | 0.20 | |
| 3 | 0.99a | 0.10 | 0.54a | 0.16 | |

^{*} numbers followed by a common letter in a column are not significantly different (p = .05)

In this analysis the surrounding rows of each aggregate up to and including the center guard row closest to it were pooled. The pooled data were transformed by log(x + 1) to normalize their distribution for statistical contrasts. To determine if the two placements of the aggregates affected damage in the rows surrounding them a Scheffe contrast was made between them (Table 34). The within row treatments still averaged higher rates of row damage over the between row volunteers. Pooling the onion rows surrounding the aggregates improved the confidence of the difference between the aggregate placements to 95 percent.

To determine if any trends existed across the densities of the treatment combinations a multiple contrast of them was done by each aggregate placement (Table 35). Although the contrasts were not significant within a placement across the density of the volunteers a relative trend was apparent. The average rate of damage for the rows surrounding a volunteer aggregate tended to increase as the size of the aggregate increased.

Discussion

This experiment and analysis portrays the differential influence volunteers have on onion damage due to their location with respect to the onion rows. For aggregates of three volunteers that occurred within an onion row the damage to the row was concentrated adjacent to the volunteers. Plant damage was also concentrated adjacent to the volunteers between onion rows but at a significantly lower rate than the within row.

The aggregates within the rows seemed to have a bimodel effect on damage that the between row volunteers did not. The rate of damage declined

Table 35. Scheffe comparison of average damaged plants per row by whole plot, volunteer placement $(\log(x + 1) \text{ data})$.

| placement | n | mean | standard error |
|-------------|----|-------|----------------|
| within row | 87 | 0.89a | 0.04 |
| between row | 52 | 0.65b | 0.05 |

^{*} numbers followed by a common letter are not significantly different (p = .05)

Table 36. Scheffe multiple comparisons of average damage per row by whole plot volunteer density and placement $(\log(x + 1))$ data.

| placement | density | n | mean | standard error |
|-------------|---------|----|---------|----------------|
| within row | 1 | 31 | 0.86abc | 0.06 |
| | 2 | 31 | 0.88bc | 0.07 |
| | 3 | 25 | 0.96c | 0.07 |
| between row | 1 | 21 | 0.62a | 0.08 |
| | 2 | 14 | 0.66ab | 0.13 |
| | 3 | 17 | 0.69ab | 0.07 |

^{*} numbers followed by a common letter are not significantly different (p = .05)

more readily away from the three volunteers within the rows than those between them. After six feet the damage rate increased for the within row volunteers possibly being influenced by the stakes that marked the ends of the five meter rows. The effect of the volunteers between the rows could be to allow a more even distribution of oviposition in the adjacent rows to occur. Volunteers within rows might be drawing flies more strongly to them than those between the rows when flies alight to initiate oviposition within six feet of them.

Damage was documented by Carruthers (1979), Loosjes (1976) and Whitfield (1981) as being contagious in field distribution. Whitfield documented damage first occurring randomly in a field and succeeding damage distributing itself around the initial points. If the initial sources of damage are already contagiously distributed around volunteers then any further damage would be expected to increase in the vicinity of the volunteers. This experiment showed that the volunteers that drew in more damage to them (those within the rows) had a greater propensity to attract damage to the surrounding rows of onions as well.

D. ANTIQUA ADULT POST HARVEST FIELD DISTRIBUTION

The distribution and movement of adult <u>D</u>. <u>antiqua</u> within a field following harvest could be important information for managment of the autumn population. In 1982 a 39 acre field, 872 meters by 183 meters was monitored following onion harvest in Grant Township, Newaygo County, Michigan for its onion fly density and distribution across time (Figure 14). An attempt was also made to determine if net adult movement into the field was occurring.

The use of an insecticide on the surface of one side of the field in conjunction with the adult fly monitoring was assumed to determine, in a relative manner the extent that adult movement occurred across the field's border in contrast to within its length. For this purpose the field was divided across its width with 402 meters of the field's total length occurring on the south side and 470 meters on the north side (Figure 14). The 183 meter width at this division constituted 14 percent and 16 percent of the total perimeters of the south and north sides', respectively. If, after the insecticide application, a decrease in the monitored adult field frequency occurred in only the sprayed side of the field, net movement could be assumed to be proportionately higher across the field's borders. If the net movement of adults was proportionately more within its length then the non-sprayed side's flies would readily redistribute into the sprayed side. This would be indicated by the frequency distribution of the flies not being different between the field sides following insecticide application. Evidence for either of these phenomena assumes that fly movement across the borders would be comparable for both the sprayed and non-sprayed sides.

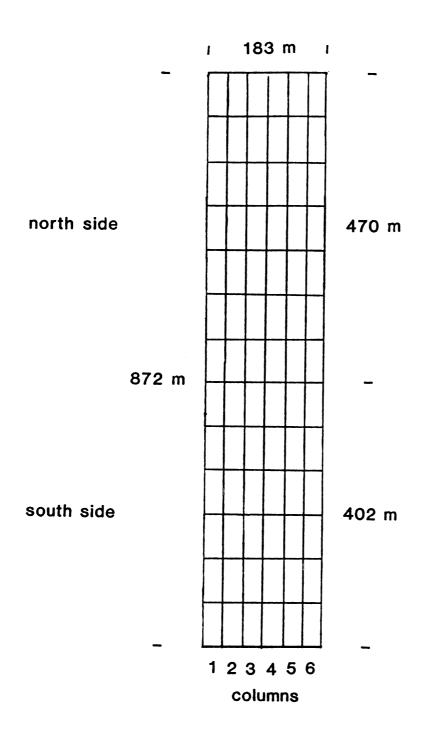


Figure 13. Quadrat design of research field for post-harvest adult \underline{D} . antiqua study.

Naled (Dibrom) was the insecticide used for this study. It was used for three reasons. First, it has a 2.45 times higher <u>D</u>. antiqua LD₅₀ than parathion, a commonly used foliar pesticide in onions (Niemczyk and Prins 1971). Second, it has a very short residual life and would only affect the resident fly population following its application. Third, its low mammal toxicity would allow for entry of the field by investigators for immediate post-spray monitoring.

Methods

As the field's harvest was being completed it was divided into 78 quadrats, 42 in the north side and 36 in the south side (Figure 14). Six quadrats occurred across the field's width with seven along the length of the north side and six along the length of the south side. Each quadrat was 30.5 meters by 66.75 meters to a side. The harvest was completed September 17, 1982, the middle of the harvest season for the Grant region that year.

A tractor mounted suction sampler, constructed after the design by Cobb and Ruppel (1976) was used for adult sampling in the quadrats (Carruthers 1981). The suction was created by a fan pulling the air at approximatley 180 miles per hour. Moving the vacuum head (10 cm by 213 cm) eight centimeters off of the ground at a slow rate would effectively sample the flies on the field surface. Sampling in this manner allowed for a consistent and rapid mass collection of the adult flies within each quadrat along the tractor's path.

At each sample period the tractor was driven down the middle of each quadrat along its length. Approximately seven percent of each monitored quadrat was sampled by this method. Sampling was conducted across contiguous quadrats along the whole field's length.

Immediately following completion of the field's September 17 harvest each quadrat was sampled. This sampling occurred between 10:00 a.m. and 2:00 p.m. It will be referred to as the post-harvest, mid-day sample.

The following morning, September 18, at 9:00 a.m. Naled was applied at 2 lbs active ingredient per acre by aerial application. This rate extrapolates linearly to 9.8 equivalents the registered rate of parathion in onions after adjusting for their LD₅₀'s. The chemical was applied on the north, downwind side of the field. A post-spray, mid-day monitoring of the adults in each quadrat was made betwenn 11:00 a.m. and 2:00 p.m. the same day. A post-spray, dusk sample was also made that evening between 5:00 p.m. and 8:00 p.m.

To account for any natural fluctuation of the adult quadrat frequency between sample periods a diurnal study of the field activity was made September 22, 1982. Quadrat monitoring was conducted at dawn, mid-day and dusk. The dawn sample period was between 6:00 a.m. and 9:00 a.m.; the mid-day was between 11:00 a.m. and 1:00 p.m.; and the dusk sample was between 5:00 p.m. and 7:30 p.m.

The maximum and minimum temperatures were comparable on all three of the sampling days. The maximum temperatures were 16.1, 17.7, and 16.1°C for September 17, 18 and 22, respectively. The minimum temperatures for the three days were 3.3, 7.2 and 3.8°C, respectively. A half inch of precipitation occurred on September 17 and 18.

Results

Three analyses were performed on the data. First an understanding of the field distribution of adults was derived by contrasting the differences in quadrat

frequencies of <u>D</u>. <u>antiqua</u> across the width of the field. This was compared to the frequency differences across the length of the field. Second, to determine if differences occurred between the field's sides a comparison of the fly frequencies between the north, sprayed side to the south, unsprayed side were performed within each sample period. Third, based on the results of the previous analyses contrasts were made between each of the sample periods to determine the effect of the spray in contrast to the pre-spray and diurnal data. These analyses determined the relative field distribution of adults and an estimate of the relative proportion of net movement across the borders in contrast to within its length.

The frequency distribution of each data set was fit best to a negative binomial. The data were transformed by log(x + 1) to normalize the distributions for statistical analysis.

The post-harvest, mid-day monitoring data were analysed for the field distribution of the onion flies. This was done by comparing field differences between its width and length. The frequency of <u>D</u>. antiqua adults were contrasted across the width of the field by pooling data from quadrat columns along the entire length of the field. Table 36 lists the results of a Scheffe multiple contrast of the pooled quadrats. The quadrat columns in the table refer to the contiguous quadrats along the length of the field as labelled in Figure 14.

Quadrat columns did differ significantly in onion fly frequencies across the width of the field. Column I was significantly higher in its estimate of mean adults per quadrat than each of the other columns of quadrats. Two and four had the lowest mean estimates of adults. The flies appear to be differentially distributed across the width of the field.

Table 37. Scheffe multiple comparisons of mean adult \underline{D} . antiqua per quadrat by field columns designated in Figure for the post-harvest, mid-day sample period (log(x + 1) data).

| quadrat column | n | mean | standard error |
|----------------|----|--------|----------------|
| 1 | 13 | 1.46c | 0.04 |
| 2 | 13 | 0.18a | 0.10 |
| 3 | 13 | 0.48ab | 0.09 |
| 4 | 13 | 0.27a | 0.09 |
| 5 | 13 | 0.73b | 0.12 |
| 6 | 13 | 0.37ab | 0.07 |

^{*} numbers followed by a common letter are not significantly different (p = .05)

To determine if the flies differentially distributed themselves across the length of the field each column of quadrats was pooled by the sides of the field they occurred in. Seven quadrats were pooled within columns on the north side of the field and six were pooled within columns on the south side. Table 37 lists the results of Scheffe contrasts of the sides of the field within each column of quadrats. No significant differences were derived between field sides within these contrasts. The lack of statistical differences when contrasting the two sides suggests that the flies did not differentially distribute across the length of the field.

Comparing the north and south sides of the field for differences in the frequency distribution of <u>D</u>. antiqua adults per quadrat was done for each sample period. The results of chi-square tests made on the untransformed data are recorded in Table 38. Each side of the field's adult frequency distribution was not statistically different from the other within each sample period.

The data were pooled for the whole field within each monitoring period for statistical tests between monitorings. This was appropriate since the chi-square tests detected no significant differences of the frequency distributions within any of the periods. The result of a Scheffe multiple contrast of the sample periods is listed in Table 39. The post-spray, dusk sample had the highest estimated mean number of adults per quadrat. The lowest mean quadrat frequency of adults occurred in the diurnal dawn sample period. Moderate adult frequencies occurred in the post-harvest, mid-day sample; the post-spray, mid-day sample; and the diurnal dusk sample.

Table 38. Scheffe comparisons of mean adult D. antiqua per quadrat by field sides within columns for the post-harvest, mid-day sample period $(\log(x+1), \det x)$.

| field side 1 2 3 4 5 5 south 1.44b 0.13a 0.48a 0.24a 0.75ab | | | | Quadrat column | column | | |
|---|------------|-------|-------|----------------|--------|--------|-------|
| 1.44b 0.24a 0.49a 0.31a 1.48b 0.13a 0.48a 0.24a | field side | 1 | 2 | က | 4 | 5 | 9 |
| 1.48b 0.13a 0.48a 0.24a | south | 1.44b | 0.24a | 0.49a | 0.31a | 0.71ab | 0.33a |
| | north | 1.48b | 0.13a | 0.48a | 0.24a | 0.75ab | 0.40a |

 \star numbers followed by a common letter in columns are not significantly different (p = .05)

Table 39. Chi-square statistics for the comparisons of the frequency distribution of adult \underline{D} . antiqua per quadrat of the field sides for each sample period.

| sample period | field side | n | DF | χ ² | p * |
|---------------|------------|----|----|----------------|------------|
| September 17 | | | | | |
| mid-day | north | 42 | 24 | 20.79 | 0.65 |
| | south | 36 | | | |
| September 18 | | | | | |
| mid-day | north | 42 | 11 | 7.99 | 0.71 |
| | south | 36 | | | |
| dusk | north | 28 | 20 | 20.28 | 0.44 |
| | south | 24 | | | |
| September 22 | | | | | |
| dawn | north | 42 | 10 | 3.63 | 0.92 |
| | south | 36 | | | |
| mid-day | north | 32 | 16 | 5.97 | 0.98 |
| | south | 28 | | | |
| dusk | north | 28 | 14 | 11.82 | 0.62 |
| | south | 24 | | | |

^{*} significance level

Table 40. Scheffe multiple range comparison of mean adult \underline{D} . $\underline{antiqua}$ per quadrat by sample period for the whole field $(\log(x+1) \ \overline{data})$.

| sample period | n | mean | standard error |
|---------------|----|---------------|----------------|
| September 17 | | | |
| mid-day | 78 | 0.58b | 0.06 |
| September 18 | | | |
| mid-day | 78 | 0.5 7b | 0.04 |
| dusk | 52 | 0.85c | 0.06 |
| September 22 | | | |
| dawn | 78 | 0.36a | 0.04 |
| mid-day | 60 | 0.74c | 0.04 |
| dusk | 52 | 0.55b | 0.06 |

^{*} numbers followed by a common letter are not significantly different (p = .10)

Discussion

The contrasts of the adult quadrat frequencies being significant only across the width of the field suggests that the flies distributed themselves relatively evenly along the length of the field. The field had its onions planted and harvested along its length. The culls left in the field were likely more evenly distributed across its length as well. The flies could therefore be orienting to the length of the field for either feeding or ovipostion on the culls. The lack of statistical differences between the north and south sides of the field in the frequency of adults per quadrat also suggests a length orientation of fly activity. For the post-spray, mid-day and dusk samples however this also implies that if the Naled had an effect on the north side's adults the population immediately recovered. This could have occurred from migration across the field's borders in conjunction with a redistribution of the adults from the south side.

To compare the sample periods of September 17 (pre-spray) with September 18 (post-spray) an understanding of the change in the adult frequency across the day is necessary. The adult activity in the diurnal study of September 22 represents the flies as relatively infrequent in the early morning, at peak frequency during mid-day and at a moderate level of field activity by dusk. The post-spray, mid-day and dusk samples did not follow this diurnal trend. Peak quadrat frequencies of <u>D</u>. antiqua did not occur until the dusk sample the day the Naled was applied. Possibly the insecticide impeded the mid-day peak in adult field activity.

Contrasting the post-harvest, mid-day sample to the post-spray, mid-day implies that the insecticide had little effect on the fly activity in the field. If, however compensation is made for the diurnal rythm detected in the September

22 samples a severe knockdown of the field population is evident. If this knockdown did occur the flies would have had to redistribute from the south side to account for the comparable frequency distributions of the adults found in the post-spray samples. This redistribution would be proportionally higher than movement into the field across the borders for the mid-day sample. By dusk however, it appears that net movement across the borders into the whole field was occurring since the frequency of flies detected in the field was significantly higher than the noon estimate.

POST HARVEST COVER CROP PLANTING TIME

Drummond (1982) recorded third flight, autumn <u>D</u>. <u>antiqua</u> females ovipositing preferentially on sprouting cull onions. In a post harvest cover crop experiment he found lower egg numbers on sprouting cull onions that occurred in 12.2 meter wide rye cover strips than on those that occurred in 12.2 meter wide fallow strips. Drummond also reported larval freeze death occurring after October 13, 1980 in his study fields. Speculation was made that any post harvest cover crop management of <u>D</u>. <u>antiqua</u> would need to have its impact on the population at least 300 degree days previous to the onset of larval freeze dessication in order to limit the population more than abiotic motality would. An experiment was conducted in the autumn of 1982 to further investigate the phenomena of both reduced ovipostion in the presence of a post harvest cover crop and differential population success in delayed seedings.

Methods

Three planting times of 12.2 meter wide broadcast oat seedings were made following the August 18 harvest of an early market field. This field was planted in onions April 15 in Grant, Michigan adjacent to the spring planting time experiment of this thesis. Seeded strips alternated across the 10 acre field in three sets of treatment replicates, each set with a fallow 12.2 meter wide control strip (Figure 15). The seeding time replicates were made on August 22, August 27 and September 4, 1982. The treatment strips were disced just prior to broadcast seeding of oats. Following seeding each replicate was cultipacted to work the seeds into the soil.

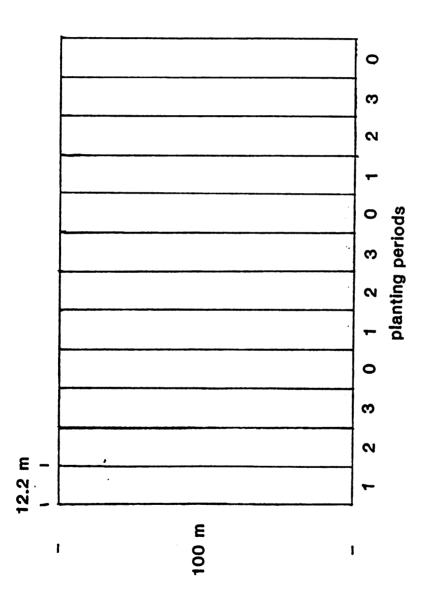


Figure 14. Field design for post-harvest cover crop study. Planting periods are August 22 (1), August 27 (2), and September 4 (3).

Sampling

<u>D</u>. <u>antiqua</u> was monitored for third flight adult field activity and egg frequency in the autumn and first flight adult emergence the following spring. These monitorings were conducted along parallel strip lengths of 100 meters within each treatment replicate (Figure 15).

Autumn field activity of <u>D. antiqua</u> was monitored with attraction traps baited with enzymatic yeast hydrolyzate (EYH) (see page for a discussion on the trap). Three EYH traps were placed across each seeding treatment replicate August 30; a total of 36 traps for the entire 10 acre field. All traps were emptied each day at dusk. Specimens were returned to the laboratory for identification and counting. This monitoring was continued until November 20 when no flies had been retrieved for a period of four days.

Egg sampling was conducted in each treatment replicate once each week following the August 20 harvest. This sampling was made in each treatment replicate before and after its seeding. To obtain an estimate of the egg population's frequency of occurrence cluster sampling was performed (Cochran 1977). A random length walk was made down each strip to establish a random point. The 15 nearest sprouting culls from the random point were then inspected for the number of eggs that were on and immediately adjacent to them. This was repeated three times in each treatment replicate. The egg sampling was conducted through the autumn until repeated sampling sessions showed that eggs were either to low in density or too widely dispersed to be effectively sampled and analyzed.

Autumn <u>D</u>. <u>antiqua</u> population success within each oat seeding and fallow strip treatment was estimated by monitoring first flight adult emergence within

each replicate the spring and summer of 1983. The frequency of spring emergence recorded in the treatments would allow for an estimate of the overall occurrence and survival of <u>D</u>. antiqua in them the previous autumn. Six emergence traps were used in each of the three seeding time and fallow strip replicates. Two trap sizes, covering 1 m² and 0.4095 m² in surface area were randomized within each replicate. Traps were emptied daily at dusk from April 1 to August 1, 1983. Trapping was stopped August 1, after less than one fly was removed from the whole field over a period of four days. The time of first and last emergence of the first generation adults in the field was assumed to be best estimated by monitoring across this time span.

Results

A total of 2,628 EYH attraction trap observations were made across the autumn monitoring season. Traps were devoid of specimens in 83 percent of the data. Because of this the <u>D. antiqua</u> adult activity data were deemed too sparse for statistical analysis.

Egg sampling was discontinued the second week of October. No eggs occurred on or adjacent to 92 percent of the 3,780 sprouting culls inspected by this time. Increasing the sample sizes would have been too costly so the egg monitoring was concluded. These data were also to sparse for analysis.

Adult emergence sampling was made to determine if significantly different frequencies of first flight adult emergence occurred in the cover-crop seeding treatments. Although emergence from each trap was monitored on a daily basis only total emergence by trap was necessary for the cover-crop treatment analysis. Emergence counts were therefore totalled for each trap across the

sampling season. The frequency distribution of the data for the trap totals fit a negative binomial with a mean adult total per trap equal to 11.07 and a "k" of 1.42. The trap totals were transformed by $\log_{10}(x+1)$ for statistical analysis.

Both trap sizes' frequency distributions fit the same negative binomial. This implied that both sizes estimated similar frequencies of emergence per trap. Since one surface area coverage was twice that of the other it can be assumed that the average patch size of emerging adults was less than or equal to the smaller trap (0.4095 m²). Also, for this similarity to appear in the data the average distance between adjacent patches of emerging adults would have to be greater than or equal to the difference between the coverage of the two traps. This interpretation of the data limits the ability to infer density per acre estimates of emerging adults in each treatment replicate.

The trap totals in this experiment were therefore used to analyze differences in the frequency of emergence per trap in each treatment replicate rather than the density of emergence. None of the design variables of seeding treatment, treatment replication or trap size were significant on analysis of variance. The data were pooled to inspect for relative trends in the seeding time treatments.

The means and standard errors of the transformed emergence totals for each treatment do show a relative trend (Table 40). The average total flies emerging per trap exhibited a relative increase as the plantings were delayed in the seeded treatments. The contol treatment with no seeding however, obtained the lowest relative estimate of emerging adults per trap.

Table 41. Means and standard errors of emergence totals for each seeding time treatment (log(x + 1) data).

| seeding time | n | mean | standard error |
|--------------|----|------|----------------|
| fallow | 18 | 0.80 | 0.10 |
| August 22 | 18 | 0.83 | 0.09 |
| August 27 | 18 | 0.92 | 0.09 |
| September 4 | 18 | 1.01 | 0.06 |

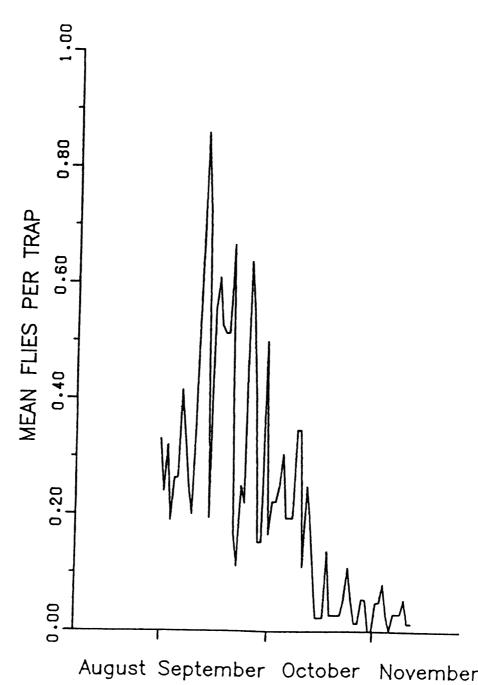
Discussion

The delayed seedings in this experiment were assumed to expose the cull onions to longer periods of oviposition before cover-crop seclusion. The work of Drummond (1982) anticipated the relative trends in the seeding treatments. He found lower egg frequencies on sprouting culls in 12.2 meter wide cover-crop treatments than in unseeded 12.2 meter wide strips. This would explain the relative increase in the frequency of emerging adult trap totals as the seeded treatments were delayed.

The control treatment with no seeding is an anomaly with repsect to this interpretation. It should have obtained the highest relative frequency of adult emergence trap totals, having never restricted the sites for oviposition. Increasing the monitoring of emergence within each treatment replicate would likely not have been enough to obtain interpretable significant differences in the delayed cover-crop treatments because of this anomaly.

Inspecting the EYH trap data for relative trends in female activity allows an alternative explanation to be proffered which includes the unseeded control strips. Figure 16 displays the female counts as daily means of the 36 traps. Relatively higher rates of female trap catches were occurring mid to late September. Moderate levels occurred early September and early October. Low catch rates occurred in late October and early November. The trap data, although consisting almost entirely of zero specimen counts does portray a relative trend of D. antiqua females in the autumn.

Miller and Haarer (1981) suggested that EYH as a bait was highly attractive to <u>D</u>. antiqua females just prior to vitillogenisis. If this differential attraction does occur in the field then it can be assumed that the higher rates of



August September October November Figure 15. Mean female <u>D</u>. <u>antiqua</u> per day of 36 EYH traps.

female EYH trap catches preceded high frequencies of oviposition. In this field study the female activity rates were relatively highest mid to late September. It appears that the cover-crop treatments of August 22, 27 and September 4 were made prior to the period of high female <u>D</u>. antiqua field activity and possibly high field rates of oviposition. In this respect, the relative trend of increasing frequencies of first flight adult emergence as the seedings were delayed can be reassessed.

Contrasting the seeding times and control (no discing, seeding or cultipacting) treatments with their temporal proximity to the higher female activity levels in the fall offers an alternative explanation of the relative spring emergence trend for all treatments. During the period of high female activity in the fall the last seeding of September 4 not only had less cover than the earlier seedings but the soil was also more recently disced and cultipacted. This soil treatment of the seeded strips likely increased the number of damaged and cut cull onions in them. Drummond (1982) recorded the damaged and cut culls to be 16 times more likely an oviposition site for <u>D. antiqua</u> than whole cull onions. The control strips, with neither discing nor cultipacting, could have had the lowest relative frequency of spring emergence because of the ovipostion preference to damaged cull onions in the seeded treatments. The trend across these treatments was likely a combination of both the oats' obstruction of oviposition sites as well as the seeding times' temporal proximity to the higher rates of adult female activity in mid to late September.

A proper experimental documentation of the validity of this interpretation would entail an added control treatment, larval sampling instead of egg sampling, cull type density and distribution estimates in the treatment replicates

across time, and increased sample sizes of spring emergence. At the time of each cover crop seeding an additional strip would have to be made which was disced and cultipacted but not seeded for each strip that was seeded. These control treatments could then be used to factor out the effect of the soil treatment on larval abundance rates and spring emergence from the effect of strictly the cover crop.

Egg sampling is inappropriate for intensive quantitative analysis because of the extremely low rates that were encountered in the field. Although Drummond (1982) was able to determine quantitative differences between monitored plots it does not appear that egg incidence is reliable enough to be routinely monitored. It would be more appropriate to sample for larvae. Relative immature population densities of <u>D</u>. antiqua could be ascertained if concomitant estimates of the density of cull types in each treatment replicate were made along with estimates of the larvae per cull type.

Increasing the sample size of emergence traps in each treatment replicate in the spring could also allow for discernable differences to be derived from the data. Monitoring for fly activity with the EYH traps, larval density in each treatment replicate, added control plots with discing and cultipacting as well as increased spring emergence monitoring could produce the data necessary for a fuller explanation to be made of the effects of delayed cover crop seedings on the population size and success of autumn <u>D. antiqua</u>.

LARVAL FREEZE RESISTANCE

Drummond (1981) recorded a high probability of <u>D</u>. antiqua larval survival up to the second week in October, 1980, citing freezing temperatures as the cause of reduced survivorship after that time. He speculated that a cover crop treatment designed to impact oviposition would need to be planted at a minimum, 300 degree days prior to this drop off in larval survival. This would be necessary to impact <u>D</u>. antiqua beyond levels of abiotic mortality already occurring in the environment. An investigation of cold hardiness in autumn larvae was conducted in the fall of 1982 to better understand this relationship.

<u>D. antiqua</u> pupae are believed to be freeze tolerant or resistant to abiotic stress such as that experienced in the winter (Whitfield 1981; Drummond 1982). Development of pre-winter, cold hardening could also occur in the autumn larvae (Horwath 1982). It is therefore likely that the larval mortality observed in the field due to freeze dessication occurred after an established level of cold hardiness had been exceeded by low temperatures.

Insect cold hardiness has been divided by Salt (1961) into three categories:

- cold acclimation preparation to avoid injury at temperatures below those at
 which continued growth is possible;
- 2) super-cooling or freeze resistance the ability to avoid freezing at temperatures below freezing; and
- 3) freeze tolerance the ability to withstand bodily freezing itself.

Freeze tolerant species are capable of surviving ice formulation in their extra cellular fluids while freeze susceptible species die if frozen. These latter species must generally lower their supercooling points by producing anti-freeze and/or ice nucleating agents within their extra cellular fluids. It is typically assumed that the critical parameter for freeze susceptible insects is not the melting point of their bodily fluids, but rather the temperature to which they will supercool before spontaneous nucleation (Duman et al. 1983; Knight 1967; Salt 1958). Temperature and time affect the nucleation and freezing of insects extrinsically, functioning as rate of cooling (Salt 1965). Intrinsically, proteins and polyols which produce a thermal hysteresis (a difference between freezing and melting points of several degrees) function as anti-freezes in a number of insects (Salt 1961; Asahina 1969; Danks 1978; Duman et al. 1982).

Physical and biological parameters of an organism can affect their supercooling points. A volume effect was cited by Salt (1965) as probable for insects based on a higher chance for nucleation in a larger volume of water than a smaller one that would have a lower number of nucleating agents overall. This is confounded with another physical factor, the surface area to volume ratio. Heat conductivity of the tissues and distance to the exterior, as well as temperature differential, determine heat loss to the environment and the probability of nucleation (Salt 1965). Thus, smaller larvae could have higher super cooling points due to their higher surface area to volume ratios. Age and the lifestage of an insect can also affect the supercooling point due to gut contents, past conditioning or physiological development (Salt 1961). Thermal hysterisis proteins are believed to be produced not only as anti-freeze agents but also to provide energy reserves for molting and metamorphosis (Duman et al. 1982; Duman and Horwath 1983).

The insects environment as well as biology is known to affect their freeze conditioning. Acclimatization, as defined by Hill (1976) appears as a change in an organism's environmental relationships after exposure to the environment's natural fluctuations. This is contrasted to acclimation which results from exposure to environments differing in only one or two well defined parameters. Temperature and photoperiod are believed to be the most important environmental cues used to trigger the production and loss of thermal hysterisis proteins and other cryo-protective agents. The environmental/physiological time processes which control anti-freeze levels involve the circadian system of the Pyrochoid beetle, Dendroides canadensis (Horwath and Duman 1982). Casagrande and Haynes (1976) elucidated the importance of seasonal change in cold hardiness from successive bouts of pre-conditioning and recovery with the temperature at which recovery occurred, the duration of the recovery period and the temperature during the recovery period all significantly affecting freeze mortality in adult cereal leaf beetles (Oulema melanopus L.). Duman (1979) and Duman and Horwath (1983) have shown fluctuations in cold hardiness while the Pyrchroid beetle, D. canadensis was still frozen and in winter diapause. One can deduce from these studies that freeze hardiness results from acclimatization in response to a series of environmental phenomena.

This experiment tested whether or not cold hardening was occurring in the third larval brood of <u>D</u>. antiqua by making larval field collections in the late autumn and treating the specimens in three separate ways; initial field testing of supercooling points and holding for a week in two incubators, one at their optimal developmental temperature (Loosjes 1976) and the other at the previous week's average temperature. Comparing each of these treatments against the

other allowed determination to be made if cold hardening took place in the field in the form of freeze resistance and if constant warm and constant cold temperatures had a differential affect on its maintenance.

Methods

Larval collections were made November 9, 16, and 23, 1982 when daily average temperatures first approached <u>D</u>. <u>antiqua</u>'s developmental threshold of 3.8°C. The field collections were made from the field used for the cover crop experiment of this thesis. Collections were made randomly across all cover-crop treatment strips and pooled for laboratory analysis. Each field collection was divided into the three laboratory treatment groups.

The incubator group's instantaneous supercooling points were determined at the end of the week long incubation period. Their larval specimens were maintained on a diet of field fresh, partially rotten onions to provide a food resource that the majority of autumn field <u>D</u>. antiqua larvae normally consume.

An attempt was made to divide each field collection into groups comparable in their ratios of small, medium and large size larvae. Also, at the time of supercooling determination the larvae were tested in random ordering of the general larval sizes. This was done to minimize any confounding of the recording device's previous response with the size of the larvae. The freezing of the larvae resulted in an extension of their body length. Exact millimeter lengths of the larvae were recorded after the bodily extension upon freezing.

Supercooling points were determined with the use of a HONEYWELL Electronic 15° strip-chart recorder, a type-T thermal couple, an aluminum rod, and a liquid nitrogen bath. Each larval specimen was secured to the thermal

couple and inserted into the hollow end of the aluminum rod immersed in the liquid nitrogen. By supercooling the larvae very rapidly (1°C/sec) to extremely low temperatures (-60°C) nuclei were produced either inter or intra-cellularly (Danks 1978). The extensive supercooling resulted in the sudden freezing of the whole insect. At this point the heat of crystallization was released, detected by the thermal couple and recorded by the thermograph recorder.

Results

The data from this experiment were analyzed in three ways. First, it was determined if the length of the larvae being tested influenced the supercooling point determined for each specimen. The second analysis determined, within each sample period's treatment groups (initial field tested, cold incubator and 27 °C incubator) which one significantly differed from another. Third, paired difference tests were made between each incubator treatments' and initial field determinations' average supercooling points to determine if the incubators, as treatments were significant. The sample periods were used in this third analysis as experimental replications. These three analyses evaluated if the size of the larva influenced its supercooling point, if freeze resistance occurred in autumn D. antiqua larvae and if the resistance was differentially affected by constant warm or cold temperatures.

A regression of larval length on the supercooling point with the pooled data for all sample periods was significant (Table 41). A y-intercept of - 12.93°C was determined with a decreasing slope of 1.0°C for every millimeter larval length increased. Table 41 also records an analysis of covariance for larval length on the data by treatment and sample period. Both independent terms were

Table 42. Analysis of covariance by larval length for supercooling point of autumn \underline{D} . antiqua larvae evaluated by sample period and incubator treatment group.

| Source | DF | SS | MS | F |
|----------------|-----|--------|-------|---------|
| egression | 1 | 82.9 | 82.9 | 4.8*** |
| Main effects | 1 | 241.6 | 241.6 | 14.2*** |
| treatment | 2 | 474.2 | 237.1 | 13.9*** |
| sample period | 2 | 277.3 | 138.6 | 8.1*** |
| interaction | 4 | 162.8 | 40.7 | 2.4** |
| residual error | 136 | 2311.6 | 17.0 | |

^{**} (p = .05) *** (p = .025) **** (p = .001)

Regression of larval length on supercooling point:

Y intercept =
$$-12.93 \pm 4.66$$

slope = -1.01 ± 0.45
 $R^2 = 0.15$

significant as well as their interaction. A non-parallel relationship across treatments and sample periods was indicated by the significant interaction term in the analysis.

Adjusting the data by the larval length regession produces a common standard deviation for all treatment means (Gill 1978). This is necessary when a covariate is significant in order to compare treatment combinations. Table 42 lists the results of the paired contrasts on the adjusted treatment means within each sample period. In the first sample period of November 9 the cold incubator's adjusted mean was significantly 4.7°C higher than the initial field tested's. The warm incubator was not significantly different in this sample period from either the cold incubator or the initial field tested. In the November 16 collection both the incubators' means were significantly higher than the initial field tested, 4.65°C for the cold incubator and 5.14°C for the warm incubator. The two incubator groups were also not significantly different from each other in their mean estimates in this sample period. The last field collection of November 23 had the 27°C incubator's mean supercooling point significantly higher than the initial field tested, the cold incubator was not. Each initial field tested group did not significantly differ between any of the sample periods. The results of these contrasts are not consistent enough to determine if the incubators were affecting the supercooling points overall.

To determine if the incubators as treatments had an affect on the supercooling points of the larvae, paired difference tests were made. These were done between each incubator treatment's and respective initial field's adjusted mean supercooling points. The paired difference test was performed by a modified "t" statistic (Gill 1978). Table 43 contains the statistics for performing the tests using the sample periods as replicates.

Table 43. Adjusted treatment means of supercooling point determinations ($^{\circ}$ C) of treatment groups and paired contrasts within sample periods for autumn <u>D</u>. <u>antiqua</u> larave. Adjusted standard error for all means is 2.2.

sample period and previous week's average temperature November 9 November 16 November 23 treatment 7.5°C 3.9⁰C 3.7°C group initial - 15.87a - 14.69a - 15.84a 27°C incubator - 13.32ab - 8.75b - 10.41b cold incubator - 11.15b - 10.04b - 15.07a

^{*} numbers followed by a common letter within a column are not statistically different (p = .05)

Table 44. Paired difference test statistics for cold and 27°C incubator group's mean supercooling points from initial field determinations for autumn $\underline{\text{D}}$. $\underline{\text{antiqua}}$ larvae.

| cold in | cubator - vs | - initial field | |
|---|-----------------------|-----------------|------------------------------|
| H: $\overline{Y}_D = 0.0 - vs - \overline{Y}_D >$ | 0.0 | | |
| Ÿ _{initial} = - 15.501 | $S^2 = 0.496$ | cov = 1.324 | $\overline{Y}_D = 3.41$ |
| $\overline{Y}_{\text{cold}} = -12.087$ | $S^2 = 7.012$ | | $S^2(\overline{Y}_D) = 4.86$ |
| *t = (3.41 - 0.0)/(2.20/v | 3) = 2.68 | which exceeds | t.10,2 = 1.89 |

27°C incubator - vs - initial field

H: $\overline{Y}_D = 0.0 - vs - \overline{Y}_D > 0.0$ $\overline{Y}_{initial} = -15.501$ $S^2 = 0.496$ cov = 1.210 $\overline{Y}_D = 4.67$ $\overline{Y}_{27}^{\circ}{}_{C} = -10.827$ $S^2 = 5.346$ $s^2(\overline{Y}_D) = 3.42$ *t = $(4.67 - 0.0)/(1.85/\sqrt{3}) = 4.38$ which exceeds $t_{.05,2} = 2.92$

^{*} t = $(\overline{Y}_D - \mu)/(S(\overline{Y}_D)/\sqrt{r})$ is compared with $t_{\alpha,r-1}$ for the significance of the difference from μ

Overall, the differences between the initial field determinations and the two incubator treatments were significant. The cold incubator averaged a 3.4°C significant difference from the initial field determinations and the 27°C incubators averaged a 4.7°C significant difference. In this respect the ability to resist freezing was lower after a weeks confinement in either of the incubators. Thus, the autumn larvae had possibly lost the cold hardiness that they had accrued in the field.

The extent that larval specimens of one incubator treatment lost more freeze resistance than the other was addressed in a similar manner. Differences between each incubator group and respective initial field group were used in a paired difference of differences test (Table 44). The amount of supercooling ability lost was on the average 2.7°C more in the 27°C incubators than the cold incubators. This difference however was not significant.

Discussion

This analysis depicts well the complexity of freeze resistance in <u>D</u>. antiqua. The significant regression by larval length, showing a decrease in the supercooling point with increasing length implies that smaller, possibly earlier instar larvae are less cold hardy. This rules out the hypothesis of Salt's (1965) of higher supercooling ability in smaller volumed organisms. This experiment was unable to explain this phenomenon as either due to the differential surface area to volume ratios or biological factors of the larvae.

The autumn larvae had possibly developed a level of freeze resistance. This was evidenced by the significant supercooling differences between the initial field and incubator test groups. The larvae from the field did not increase in

Table 45. Paired difference of differences test between cold and 27° C incubators' differences from initial field supercooling point determinations for autumn <u>D. antiqua</u> larvae.

H:
$$\overline{Y}_{D_{cold}} = \overline{Y}_{D_{27}^{\circ}C} - vs - \overline{Y}_{D_{cold}} < \overline{Y}_{D_{27}^{\circ}C}$$
 $\overline{Y}_{D_{cold}} = 3.41^{\dagger}$
 $S^{2} = 4.86$
 $cov = 1.534$
 $(\overline{Y}_{D})_{D} = 2.70$
 $\overline{Y}_{D_{27}^{\circ}C} = 4.67^{\dagger}$
 $S^{2} = 3.42$
 $s^{2}(\overline{Y}_{D})_{D} = 0.99$

*t = $(1.26 - 0.0)/(2.14/\sqrt{3}) = 1.02$ does not exceed t.10,2 = 1.89

t mean difference from initial field determinations

^{*} t = $((\overline{Y}_D)_D - \mu)/(S(\overline{Y}_D)_D/\sqrt{3})$ is compared with $t_{\alpha,r-1}$ for the significance of the difference of the differences from μ

their supercooling ability after November 9. It was likely that the field sampling did not start early enough in the autumn of 1982 to document an increase in freeze resistance as measured by the instantaneous supercooling point.

Under laboratory conditions of constant temperatures both the cold and 27 $^{\circ}$ C incubators lost a significant amount of supercooling ability as compared to the initial field determinations. Although the 27 $^{\circ}$ C group appeared to lose more of its supercooling ability the difference of this from the cold incubator treatment was not significant. It is likely that the fluctuating temperatures and/or photoperiod exhibited in the field are necessary for the maintenance of cold hardiness for \underline{D} , antiqua as was found for the cereal leaf beetle and the Pyrochoid beetle (Casagrande and Haynes 1976; Horwath and Duman 1982).

To determine the extent that larval freeze dessication can have on a population of autumn <u>D</u>. <u>antiqua</u> information would be neccessary on what stages were susceptible and at what temperatures as well as how extremely low temperatures affected final development of the non-susceptible larvae to pupation. The greater susceptibility of the smaller larvae could greatly impact the age structure of the autumn population as well as its final density. However, it is probable that the larger more mature larvae, indicated to be more freeze resistant could never accrue the heat accumulation neccessary to complete their development to pupation. Elucidation of whether or not the non-susceptible larvae could continue development to the pupal and hence overwintering stage would therefore also be crucial to the understanding of the inherent population dynamics of the autumn D. antiqua.

SUMMARY AND CONCLUSIONS

Planting and Harvest Time of Onions

In this experiment stand reduction was only significant in the untreated subplots. It is believed that stand reduction estimates were confounded with late germination of the seeds contributing to an under estimate of crop loss. This under estimate could have been what caused only relative trends of stand reduction in the treated subplots to occur. Relative trends of percent stand loss however were consistent in both the treated and untreated subplots as the planting times were delayed. A trend of decreased reduction as onion planting was delayed occurred in both soil treatments.

Mean onion bulb diameters were largest in the early planted untreated subplots across all harvest periods. Significant differences of mean bulb diameters occurred within and between the soil treatments for the first harvest period of August 20. A significant trend of smaller mean diameters as the plantings were delayed was evident in both soil regimes. By the last harvest period of September 24, both the significant differences as well as the trend were absent in the treated subplots. The untreated ones however retained both the trend and the differences of mean bulb sizes at the last harvest period. The treated stand densities appeared to curtail bulb size development prior to any limit imposed by the time of planting. This was not evident in the untreated subplots.

The estimated yield from the harvest period of August 20 was relatively highest in the April 27 planting's treated subplot with 1,115 bu/acre. The April 15 planting with only 884 bu/acre likely had a lower yield due to the mechanical

damage imposed on the onions in preparation for commercaial harvest for the early fresh market. The higher rate of return for the early market would likely compensate a grower for this reduced yield. The May 11 treated subplot did not exceed 1,000 bu/acre until the September 13 harvest period. The May 25 treated subplot had an estimated yield of 985 bu/acre at the last harvest period of September 24. Only the September 24 harvest sample in the May 25 untreated planting obtained a sizable estimated yield of 619 bu/acre in the untreated subplots. The other untreated subplots had relative yields of 99 and 124 bu/acre by the last sample period for harvests.

<u>D. antiqua</u> population monitoring indicated that less than 50 percent of the spring adults had emerged by the time of the May 25 planting's initial stand count. By this time all stands were estimated to have had an initial density of 51.2 plants per meter. Peak female activity was estimated to have occurred in the EYH traps on June 20, nine days following 50 percent emergence. The majority of the stand reduction had occurred prior to both of these points in time for all subplots.

The immature sampling was found to be efficient for only the larval stages and not the pupal. Pupae were believed to be under estimated due to the sampling bias for currently damaged aggregates of plants. To better estimate pupal abundance it would have been necessary to sample in areas that had been damaged for a period of time long enough for the majority of the larvae to pupate. This likely would be after the plants had dried and blown away making it difficult for these areas to be found in homogeneous units.

Immature density indexes indicated that larval populations were at higher numbers per acre in the untreated plots than the treated plots. This was

anticipated by the stand reduction data. Greater densities of larvae occuured in the untreated subplots for all planting times. A significant trend of decreasing larval density indexes as the plantings were delayed was observed in both treated and untreated subplots. Relative peak larval densities occurred in the last week of June following the peak in adult female D. antiqua field activity of June 20.

A parallel trend of immature densities was indicated to exist between the two soil treatments. Seventy seven percent of the immature population developed in the April 27 planting's untreated subplot, 13 percent in the May 11 subplot and 4 percent in the May 25 planting's subplot. The respective treated subplots' similar trend consisted of 5, 0.6 and 0.1 percent of the total larval population for the experiment.

The untreated subplots of the April 27 and May 11 plantings, although not significantly different in final mean stand densities did appear to differ markedly in their density indexes of D. antiqua immatures. This could have been due to higher densities of larvae per damaged plant in the early planting of April 27. This phenomenon was also apparent by contrasting the comparable immature populations of four and five percent of the total in the treated subplot of the April 27 planting and the untreated subplot of the May 25 planting, respectively. The later planting subplot's final mean stand density was significantly 27 percent less than the former, earlier planted. The May 25 planting's untreated subplot likely had a much lower number of larvae per damaged plant than the April 27 planting's for this disparity in stand densities to occur. These observations reinforce the speculation that the eggs oviposited by the female flies which peaked in their field activity June 20 went into previously damaged plants. This would account for the lack of a proportional relationship between the larval

populations and stand losses. The density of immatures is therefore not necessarily proportional to the amount of stand reduction.

Dyfonate residue levels were found to be characterized by degree day accumulation from planting. The differences across time however were not great enough to explain the relative differences in final stand densities in the treated subplots. The trend of larval population densities as planting's were delayed was found to be not significantly different between the two soil treatments. Higher levels of larvae in the earlier plantings were believed to be due to higher numbers of larvae per damaged plant as opposed to more damaged plants per se. This is a more reasonable explanation for the trends in the treated plots than being due to differential levels of Dyfonate residue.

Volunteer Density and Row Placement

The within row aggregates of three volunteers drew significantly more damage into their immediate vicinity than the between row volunteers. The decline in plant damage as the distance down the row that the three volunteer aggregates were either in or adjacent was significant for both placements. The decline however was greater in the rows that the aggregates resided within.

The size of the aggregate was not a significant variable in this experiment on the rate of damage in the rows the aggregates were either residing in or adjacent. The placement of them however was a significant variable. The within row volunteers affected a higher rate of plant damage on the rows they occurred in than the aggregates placed between the rows affected on the rows adjacent to them.

In this experiment the onion rows surrounding each within row aggregate up to and including the center guard row closest to it had significantly more damage in them than those rows surrounding the between row aggregates. The size of the volunteer aggregates was again not significant, although a relative trend of increased row damage as the size increased was observed. As Whitfield (1981) suspected, there was an influence of the aggregates on plant damage further than his study was able to discern. This experiment showed that the volunteers that were able to draw more damage and hence ovipositing flies, to their immediate vicinity (those within the rows) also attracted more damage to the surrounding rows of onions.

D. antiqua Adult Post Harvest Field Distribution

The data from this experiment indicated the flies to be distributed along the length of the field. The flies appeared to be distributed along the same direction of the field as the onions were harvested. It was likely that the cull onions were also distributed more evenly along the length of the field. They could have caused the flies to be distributed along the field's length.

The north, sprayed side of the field was not significantly different in fly frequency from the south side for either of the post-spray sample periods. This implied that the flies were rapidly dispersing from the south side into the north side following the spray application of Dibrom. Corroboration was made with this by a comparison of the diurnal fly activity pattern detected. The deviation of the post-spray samples from the rythm detected in the diurnal study indicated the spray to have a depressing effect on the fly field frequency. The diurnal study suggested that peak field activity would occur during the middle of the day

and be significantly lower by dusk. The reverse was observed for the post-spray field estimates of adult <u>D</u>. <u>antiqua</u> activity. A significantly larger level of field activity occurred in the dusk, post-spray sample than in the dusk diurnal sample.

Adult <u>D</u>. antiqua field movement appears to be quite varied across the days following a field's harvest. The flies in this study were indicated to be moving rapidly along the length of the field, possibly orienting to the cull onions. Field frequency appears to increase through the duration of the day, possibly peaking around mid-day. This increase was also suggested by this study to be greater two days following the field's harvest than five days after harvest.

Post Harvest Cover Crop Planting Time

The frequency distribution of spring emergence was not significantly different between the two emergence trap sizes' coverage of 1.0 m² and 0.4095 m², respectively. It was inferred from this that the average patch size of emerging adult <u>D</u>. antiqua in this study was equal to or less than the smaller trap's surface coverage. The patches would also have to be distributed further apart than the difference of the two traps' longest sides (0.5905 m).

No significant differences were detected between any of the seeding time treatments of the oat cover or the fallow control. A relative trend however indicated that the oat cover seeding treatments made closest to the time of relative peak female <u>D</u>. antiqua fly activity had a higher frequency of adult emergence the following spring. It was inferred that this trend was due more to the discing and cultipacting of the treatment strips at seeding than the cover crops themselves. The fallow treatment yielded relatively the lowest frequency of emerging adults. This corroborated that it was the soil conditioning that

caused the relative trend. This explanation is probable in light of the preferrence of <u>D</u>. <u>antiqua</u> oviposition for damaged culls being 16 percent geater than for whole culls that Drummond (1982) observed.

Larval Freeze Resistance

A regression of larval length on supercooling point was statistically significant. Smaller larvae supercooled at higher temperatures than larger larvae at an average rate of 1°C per mm length. This study was unable to document whether this was due to the larger surface area to volume ratio of the smaller larvae or internal physiological factors specific to the lifestages involved.

Cold hardiness, in the form of freeze resistance was believed to have occurred in the autumn <u>D</u>. <u>antiqua</u> field larvae. This was evidenced by the significantly lower supercooling ability of the larvae that were held for a week in warm and cold incubators after being brought in from the field. The two incubators did not significantly differ from each other in their ability to depress the larvae's supercooling ability. Fluctuating cold temperatures or a photoperiod could likely have been necessary to maintain the supercooling ability the larvae had accrued in the field (Duman et al 1980; Casagrande and Haynes 1976).

The larvae tested directly from the field did not increase in their supercooling ability across the range of sample periods. It was likely that the sampling did not start early enough in the autumn to document an increase in freeze resistance. September, October and November sample periods might have been able to reflect field differences in supercooling point determinations of the larvae as a measure of the development of freeze resistance in autumn \underline{D} . antiqua.

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