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EGG HATCHING PATTERNS FOR INDIVIDUAL

WESTERN CORN ROOTWORM FEMALES

BY

ROBERT SHELTON HOOTEN

A thesis submitted in partial fulfillment of the requirements for the degree Master of Science, Major in Entomology, South Dakota State University 1979

EGG HATCHING PATTERNS FOR INDIVIDUAL WESTERN CORN ROOTWORM FEMALES

This thesis is approved as a creditable and independent investigation by a candidate for the degree, Master of Science, and is acceptable for meeting the thesis requirements for this degree. Acceptance of this thesis does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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Date

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TABLE OF CONTENTS

.

																							Page
INTRODUC	TION .		•••	•			•		•		•		•		•	•	0	•		•	•		1
LITERATU	JRE REV	IEW	• •		•		•	• •	۰	•	•	•		•	•	e				e	•		5
PROCEDUR	RE		• •		•	•	•				•			e .	•	•	e	•	•	۰	•	•	25
RESULTS	AND DI	SCUS	SION	•	•	•	•	• •	•			•		•	•	•	•	•		•		•	29
CONCLUSI	IONS .	• •	• •	۰													•	۰	•	•	0		48
REFERENC	CES CIT	ED.															•	•	•	۰	•		49
APPENDIC	CES		• •	•		•			°.	•	•			•				•	•	۰	•		55
Α.	Compa early				-				-											•	•		56
В.	Total locat										-						-		•		•		57
С.	Furac Beres	lan Ll sford	D ₅₀ wes	va] tei	lue rn	s co	fo: rn	r F ro			i f	ie ar	eld nd	l c WC	col CRN	.1e ID	ect 1a	ied	l vae	9			58

.

.

LIST OF TABLES

Table		Page
1.	Average weekly soil temperatures at 1" and 6" depths during the spring of 1975 in Brookings	30
2.	Average weekly soil temperatures at 1" and 6" depths during the spring of 1976 in Brookings	31
3.	Longevity of each Beresford female after the date of capture	33
4.	Longevity of each Elkton female after the date of capture	34
5.	Egg totals for individual field collected Elkton female WCR	36
6.	Larval numbers from individual field collected Beresford female WCR	37
7.	Larval numbers from individual field collected Elkton female WCR	38
8.	The period from oviposition to peak hatch, for the eggs from individual Beresford and Elkton female WCR	39
9.	The period from oviposition to peak hatch for the eggs from individual Beresford female WCR 1-10	40
10.	The number of days from oviposition until initial, 50%, and final hatch for eggs from Beresford females 1-10	42
11.	The number of days from oviposition to initial, 50%, and final hatch for eggs from Beresford females 11-25	43
12.	The number of days from oviposition to initial, 50%, and final hatch for eggs from Elkton females 16-30	44
13.	The number of days from oviposition to initial, 50%, and final hatch for eggs from WCRND females	45
14.	Larval length from 3 different populations	46

INTRODUCTION

The western corn rootworm, <u>Diabrotica virgifera</u> LeConte is a major economic pest of corn in the north-central region of the United States. In 1973, yield reduction and control costs credited to the rootworm complex for a seven state area was estimated at 82 million dollars (Gerloff 1976). In 1975, South Dakota corn producers suffered a combined yield reduction and control cost of 27.5 million dollars (D. Walgenbach, South Dakota State University, personal communication). The majority of the damage to corn by the rootworm stems from larval feeding leads to an increase in the lodging of corn plants and a decrease in ear size. The adult beetles cause damage, to a lesser extent than the larva, by feeding on the pollen, silks, and ear tips (Chiang 1973).

Gillette (1912) reported injury to corn in Colorado by the western corn rootworm in 1909. By 1948, Hixson et al. (1949) had found the western corn rootworm in eastern Nebraska only 70 miles from the Missouri River. A 1953 survey of Kansas indicated an eastward migration across Kansas at a rate of 30-35 miles per year (Burkhardt and Bryson 1955). The western corn rootworm's range by 1960 included parts of Nebraska, Colorado, Kansas and South Dakota and a small portion of several other states. The migration eastward of the western corn rootworm increased during the 1960's and early 1970's until by 1975 its eastern boundary was in lower Michigan and eastern Ohio (Gerloff 1976).

The more common methods of corn rootworm control are with soil insecticides applied at planting and crop rotation. The development of insecticide resistance was shown with aldrin and heptachlor used in the 1950's and 1960's for rootworm control. Howe et al. (1963) and Kantack (1965) reported that by 1962 an aldrin and heptachlor resistant population of western corn rootworms had been established in southeastern South Dakota corn fields. Soil applied organophosphate insecticides were recommended in 1964 for rootworm control in South Dakota and over 600,000 acres were treated (Kantack 1965). The development of resistance to aldrin and heptachlor shown by the western corn rootworm coincided with the migration of the adults. This led to a continued changing of recommendations from the hydrocarbon insecticides to the carbamate and organophosphate insecticides for rootworm control. The organophosphate and carbamate insecticides have controlled the western corn rootworm for the past 10 years; however, in the mid 1970's, field failures of some of these insecticides occurred. Metakamate was the first carbamate insecticide to be removed from the market because of its poor performance in the field. Diazinon, an organophosphate, and carbofuran, a carbamate, have also been experiencing field failures during the 1970's (Walgenbach and Sutter 1977). The knowledge of whether another resistant population of rootworms is developing is essential. Possibly the rootworm population could be manipulated in some way to prevent or slow the development of resistance.

Hamilton (1966) noted that adult resistance to aldrin and heptachlor decreased later in the season. He suggested that perhaps the more resistant larva, through a genetic linkage with resistance, tended to hatch earlier; thus the first adults to emerge might have come from these more resistant larva. Walgenbach and Sutter (1977) performed a

series of LD_{50} and LD_{90} tests on western corn rootworm larva from various geographical locations with organophosphate and carbamate insecticides during the 1970's. Their results indicated that the early hatching larva were more resistant than later hatching larva with some of the insecticides. The larva used by Walgenbach and Sutter (1977) were obtained from several hundred females caged together eliminating any determination of whether a single female's progeny hatched early or late. Musick and Fairchild (1971) determined the rate of hatch of western corn rootworm eggs in Missouri from 1965 to 1968. They conducted this field study to help explain the erratic performance of soil insecticides in controlling the western corn rootworm. The results indicated that the initial date of hatch for all four years was similar being on June 1 for 1965, 1966 and 1968 and being on June 4 for 1967. The rate of hatch varied from an average rate of 2.91% per day for 1965, 1966 and 1968 to a rate of 1.77% per day for 1967. The completion of hatch occurred about July 26 during 1967 giving a 52 day duration, while during the other 3 years the completion of hatch was about July 5, giving a 34 day duration. The hatching of the 1967 eggs late into July could account for possible field failures in fields treated with soil insecticides. The larva could possibly have avoided the insecticide because of the short residual activity of the carbamates and organophosphates. The reason for the variation of rate and duration of hatch in 1967 was not determined. Branson (1976B) exposed western corn rootworm eggs to simulated environmental conditions in the laboratory. He reported that after a long period of chill, greater than 16 weeks, the mean post chill time to hatch was 19 days; peak hatch occurred a mean

of 5 days after the initiation of hatch; and the mean duration was 35 days. Genetic variation in egg hatch seems apparent when comparing the 34 day duration reported by Musick and Fairchild (1971) to the 35 day duration reported by Branson (1976B). Branson's laboratory conditions in probability could not have duplicated the field conditions which existed for Musick's eggs; therefore, since the two durations are similar, genetics must be involved in the timing of egg hatch. Krysan and Branson (1977) tested inheritance of diapause intensity among three different populations of Western Corn Rootworm. They concluded that diapause intensity is intermediate between the parental lines with a larger contribution to intensity coming from the female.

Resistance to the carbamates may become widespread if the eggs of single female western corn rootworms hatch early. This would allow for the deveopment of a totally resistant population.

The purpose of this study was to determine the hatching pattern of WCR eggs from early and late emerging adults.

LITERATURE REVIEW

Temperature, moisture, cultural practices and soil type all have an influence on the hatch of western corn rootworm eggs in the spring. Wilde (1971) found that a threshold temperature of $52^{\circ}F$ and a thermal constant of 380 degree days must be obtained before Minnesota collected western corn rootworm eggs would hatch. This is consistent with the 52°F threshold temperature and 400 degree day thermal constant established for the northern corn rootworm, Diabrotica longicornis Say, by Chiang and Sisson (1968). Various tillage operations are known to cause a down ward movement of the eggs in the soil profile. Chiang (1965) reported that if 83% of the eggs were in the top 2", after harvest, disking, plowing, and raking, 14% would be at 10" and 1% even deeper and only 4% would remain at the 2" depth. Some northern corn rootworm eggs buried too deeply in the soil to receive enough heat units to hatch the first year have been observed hatching the second year (Chiang 1965). Chiang (1965) observed that percentage of hatch and egg survival increased as the egg depth in the soil increased. Calkins and Kirk (1969) reported that with a lack of winter precipitation a higher mortality rate occurred among those eggs nearer the soil surface. The percentage of egg hatch was significantly reduced in the laboratory when the eggs were allowed to dry shortly after oviposition (George and Ortman 1965). The highest rate of hatch occurred when rootworm eggs were continually exposed to contact moisture during a laboratory test conducted by Mihm et al. (1974).

Musick and Fairchild (1971) reported that western corn rootworm egg hatch began during the first of June and continued into July for the Missouri area. Newly hatched larva migrate to the corn roots and begin feeding primarily on the root hairs. Food uptake increases as the larva mature causing them to burrow into the cortical parenchyma (Chiang 1973). Root feeding is reduced once the cortex of the root acquires a tough exodermis and the larva will progressively move toward new areas of root growth (Apple and Patel 1963). Corn roots are the major host of the western corn rootworm larva; however, Branson and Ortman (1967A) reported that there were alternate hosts on which the larva could survive. Branson and Ortman (1967A) found nine grass species which supported the western corn rootworm larva for at least 10 days and in a later study they increased this number to 18 grass species (Branson and Ortman 1970). On 13 of 18 potential hosts the western corn rootworm was able to complete its life cycle (Branson and Ortman 1970). The adults from larva reared on green and yellow foxtail, Minter and Selkirk wheat, Omugi barley, Oahe intermediate wheat grass and corn laid viable eggs and appeared to have no reduction in fertility (Branson and Ortman 1967B).

The survival rate of the larva in the soil is dependent upon the stress situations placed on the larva by soil type and distance from the host. Short and Lufdtke (1970) determined that western corn rootworm larvae could migrate up to 32 inches through the soil to obtain food. As the distance from the host plant to the larva increased however; a decrease in adult emergence and root damage occurred. Short (1970) and Sechriest (1969) showed that the greatest concentration of larva was around the corn roots. A minimum or no-till tillage practice would keep the greatest concentration of larva around the roots of the

previous corn crop. Chiang et al. (1971) found that in a minimum tillage field a reduction in larval numbers occurred when the new corn rows were placed midway between the rows of the preceding year. Turpin and Peters (1971) investigated larval survival in relation to scil texture in the laboratory and found that rootworm larva moved from sandy to clay soils but not from clay to sandy soils. Turpin and Peters (1971) determined that as the percentage in the clay content of soils increased survival rate of the western corn rootworm increased. A tendency for dessication caused by abrasion of the larval cuticle by sandy soils was considered the mechanism involved.

The larva remain in the soil and form earthern cells in which to pupate. Sechriest (1969) reported that in Illinois, 90% of the larva pupated in the top two inches of soil. Minnesota pupae have been observed as far as 25 inches from the main corn roots and 9 inches deep (Chiang 1973). These facts indicate that larval movement toward the soil surface does not necessarily occur and that the adults may have to move considerable distances in the soil upon emergence. Sampling for pupae is difficult in that they are fragile and the pupal stage is short (Chiang 1973).

The largest percentage of rootworm beetles emerge between 8 and 10 inches from the base of the corn plant (Short 1970). Ball (1957) reported western corn rootworm adults first appeared around Lincoln, Nebraska in early July. Lockwood (1978) trapped western corn rootworm males with pheromone traps during the first week in July in South Dakota. Male western corn rootworm adults emerged 3 to 5 days prior to the females (Short and Hill 1972). The western corn rootworm females in Nebraska

emerged over a six week period starting in the middle of July and continued until the end of August (Short and Hill 1972). Adult western corn rootworms have been observed feeding upon every growing part of the corn plant above ground (Bryson et al. 1953). Partial or incomplete ear filling occurs when rootworm adults feed upon the silks and tassels interfering with pollination and fertilization (Sifuentes and Painter 1964). Adult feeding upon the ear tip kernels causes an increased potential for disease development within the ear.

Ball (1957) observed that mating of the western corn rootworm occurred from July to August in Nebraska. The female does not appear to mate more than once, whereas the male is capable of and may have repeated matings (Branson et al. 1977). Branson indicated that in a few cases a second mating took place before oviposition, but after oviposition no mating took place. Female beetles prefer moist soil of large particle size for oviposition (Kirk et al. 1968). Kirk et al. (1968) observed that the rootworm females also selected cracks, insect holes or animal burrows for ovipositional sites. Howe et al. (1963) postulated that corn fields maturing later due to planting date or varietal differences would attract ovipositing females, thus increasing the population potentials in the following growing season. Branson and Johnson (1973), in the laboratory, established a mean preoviposition period of 14.3 days for South Dakota beetles. Short and Hill (1972) determined that a small percentage of Nebraska western corn rootworm females started oviposition at an age of 12 to 14 days. By the age of 18 and 21 days over 50% of the females started oviposition and all females were ovipositing by the 31st day (Short and Hill 1972). Hill (1975) reported a pre-

oviposition period of from 9 to 22 days with a mean of 12.2 for Nebraska western corn rootworm females. In general, it can be assumed oviposition occurs from 2 to 3 weeks after the emergence of the female. Hill (1975) determined that Nebraska western corn rootworm females laid 24% of their total eggs by August 25, 57% by September 10, 83% by September 30, and 90% by October 10. This would mean that the majority of eggs are laid before the first killing frost, thereby insuring a large potential population of rootworms the next spring. Ball (1957) established that 80% of the western corn rootworm eggs were oviposited in the upper 6" of soil. Oviposition will occur from August until a killing frost, the eggs will then overwinter in a diapause state (Ball 1957).

Laboratory rearing methods of the western corn rootworm were investigated in the 1960's to aid in understanding the biology of this pest. Howe and George in 1966 published one of the first reports on the mass rearing of the wastern corn rootworm. They fed the adult beetles with squash, lettuce, seedling corn, silks and immature ears. Hamilton (1972) established that a diet of pollen substitute plus plant tissue increased the egg production in the southern corn rootworm, <u>Diabrotica undecimpunctata howardi</u> Barber, over a straight diet of plant tissue. Guss et al. (1976) altered a dry diet previously formulated for the southern corn rootworm. The alterations consisted of ball milling the diet into a fine powder then forming it into a pellet suitable for the western corn rootworm. Western corn rootworm females fed straight dry diet laid 71% of the eggs compared to beetles fed on fresh corn leaves. Oviposition in the western corn rootworm is affected not only by diet, but also by ovipositional sites, temperature,

and photoperiod. Guss et al. (1976) determined that under laboratory conditions female western corn rootworms show a preference for ovipositional sites of white gauze or sifted soil that are covered over uncovered or black gauze sites. Kirk et al. (1968) tested soil aggregate size in relation to the ovipositional preference of the western corn rootworm and found that as the soil aggregate size increased, ovipositional preference increased. Kirk et al. (1968) tested the ovipositional preference of the western corn rootworm to moist and dry soil dishes and found that females laid 9.3 times better in the moist egg dishes. Mihm et al. (1974) determined that optimum temperatures in the laboratory for oviposition is a constant 20° C or a fluctuating $25^{\circ}/15^{\circ}$ C. Mihm et al. (1974) also noted that the longevity of the female beetles was greater at cooler temperatures. Female western corn rootworms reared by Hill (1975) had a mean longevity of 78.2 days and a mean fecundity of 1087 eggs laid in 13.5 clutches. Egg laying commenced at a high level, after a mean preoviposition period of 12.2 days, with one of the first four clutches being the largest. Branson and Johnson (1973) reported a mean preoviposition period of 14.3 days, a mean longevity of 94.8 days, and a mean fecundity of 1023 eggs. The high level of oviposition at the beginning and the peak about 10 to 15 days later indicated by Branson and Johnson (1973) agrees with what Hill (1975) reported. Ball (1971) reported that maximum oviposition in western corn rootworms occurred in the early morning hours, yet oviposition did occur all day. Chiang (1973) reported that maximum egg deposition occurs when female beetles are subjected to an 8 to 12 hour photophase.

The use of a trap crop, adult suppression with insecticides, and crop rotation are three control methods which are used presently or have been investigated for control of the western corn rootworm. Hill and Mayo (1974) determined that corn planted at a later date than the main crop would attract western corn rootworm adults. The females would oviposit in the trap crop lessening the potential damage in the area surrounding the trap crop the following year. Pruess et al. (1974) attempted population suppression by adult control with ultra low volume malathion. The 16 square miles treated had no economic infestations the following year; however, western corn rootworm adult migration into the treated area was sufficient enough to warrant a second treatment the following year. Pruess et al. (1974) stated that since treatments were needed each year area suppression did not appear feasible, but adult control in individual fields might be an acceptable alternative. Crop rotation was a method commonly used before the advent of insecticides. Hill et al. (1948) reported effective control in Nebraska of both the northern and western species during the 1940's.

Control of corn rootworm larva with soil insecticides is the most common control measure today used against the western corn rootworm. Cox and Lilly (1953) applied aldrin, gamma BHC, chlordane, and dieldrin as soil insecticides and showed a reduction in rootworms and lodging. Severe lodging was reduced by 90 to 100 percent and yield increases varied from -2.4 to +21.8 bushels per acre for the treated fields. Previous insecticide studies by Muma et al. (1949) showed benzene hexachloride as giving adequate control of rootworms. The data indicated that soil treatments of benezene hexachloride applied in the

spring of one year affected two generations of western corn rootworm. Peters (1964) reported that the planting of continuous corn and the rotation of corn were the most important factors influencing rootworm damage. He reported that soil insecticides were used for the control of corn rootworms based on previous cropping information rather than evidence of pest populations (Peters 1975). Peters (1975) conducted a ten year survey of Iowa corn fields to determine if the application of insecticides for rootworm control was of value or just being used as prophylactic treatments. He concluded that in continuous corn an aldrin or heptachlor soil insecticide treatment increased the yield by 8.0 bushels per acre.

In the late 1950's, western corn rootworm resistance to the chlorinated hydrocarbons was apparent. Ball and Weekman (1962) tested for resistance among two different geographical populations of rootworms in Nebraska. LD₅₀ ratings were determined for the adult western corn rootworms two hours after they were treated with aldrin, heptachlor, and diazinon. They found that adults collected from irrigated continuous corn fields were 100X more tolerant to aldrin and heptachlor than those adults collected from non-continuous corn areas. The LD₅₀ ratings for diazinon were the same for both populations, indicating no resistance. The following year Ball and Weekman (1963) reported that after testing 18 sites in Nebraska, the greatest resistance to aldrin and heptachlor was shown by western corn rootworm adults from around the Platte River. This area coincided with that part of the state were chlorinated hydrocarbons had been used the longest and where control problems had existed since 1959. Although Ball and Weekman (1963) tested only

adults, they mentioned that according to other data from other sources western corn rootworm larva most likely would show higher resistance than adults. Hamilton (1966) conducted LD₅₀ tests on western corn rootworm larva and found they were 2 to 4 times more resistant than the adults to chlorinated hydrocarbons. Hamilton (1965) reported resistance to aldrin but not diazinon was present in western corn rootworm adults collected from Iowa, Kansas, Missouri, Minnesota, and South Dakota.

Kantack (1965) estimated 496,000 acres of corn were severely damaged in South Dakota by resistant western corn rootworms in 1963. The 1963 adult survey revealed that the rootworm infestation was spreading northward in South Dakota at a rate of 50 to 75 miles per year. By 1964, organophosphate insecticides were being recommended for rootworm control in South Dakota because of the lack of control obtained from the chlorinated hydrocarbons. Since the western corn rootworm had developed resistance to the hydrocarbons Ball (1973) tested for possible resistance to diazinon and phorate. The test was conducted annually from 1963 through 1972 on western corn rootworm adults in Nebraska. Ball (1973) reported an increase in LD50 values that was statistically insignificant and what also appeared to be a reversion in susceptibility to diazinon. Rather than becoming resistant it appeared the western corn rootworm was becoming more susceptible to diazinon.

The carbamates and organophosphates have a shorter residual life in the soil than the chlorinated hydrocarbons. Research was done on the timing of application, method of application and emergency applications. Post planting treatments of soil applied insecticides for control of corn

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rootworms was first demonstrated in 1947 (Hill et al. 1948). In 1968 and 1969, Hills and Peters (1972) used Bux, Dyfonate, Thimet, Diazinon, Furadan and fensulfothion at planting time and three varied postplanting time treatments for rootworm control. Corn roots were dug, washed, and assigned an adjusted root damage rating (Hills and Peters 1971). Hills et al. (1972) concluded that only the last postplanting time treatment of Diazinon significantly controlled rootworm larva better than the Diazinon at planting treatment. All the other insecticides showed no significant difference between the timing of treatments for corn rootworm control. Hills and Peters (1972) investigated methods of application, insecticidal formulations, and rates of application for some carbamate and organophosphate insecticides. Their results indicated that by using half the recommended rate, a corn producer would sacrifice a significant degree of control. Hills and Peters (1972) established that in general, postplanting treatments applied at cultivation time gave the most effective control. They also concluded that liquid formulations gave better control with fertilizer combinations; while granular formulations gave better control as broadcast, band, or postplanting treatments. The possibility of an emergency treatment after planting for control was investigated by Mayo (1976). He concluded that emergency postplanting treatments may be economically beneficial, but major damage will have occurred reducing the yield from what could have been expected had a treatment at planting been used. The hatching of western corn rootworm eggs depends partially upon the intensity of their diapause (Krysan and Branson 1977). A closer examination of the dormant state of diapause will be discussed.

It is common for organisms inhabiting inconstant environments to possess a resting stage (Lees 1955). This dormant state is characterized by a temporary failure of growth and it enhances the organisms' resistance to adverse climatic conditions. Dormancy is well documented in literature but there is a variability in the nomenclature used. Wheeler (1893) introduced the term, diapause, to describe a stage in the embryogenesis of the grasshopper, Xiphidium ensiferum. The stage between anatrepsis and catatrepsis where the embryo remained poised with the head directed towards the posterior pole, he considered diapause. Henneguy (1904), going a step farther, considered diapause as not a stage but a condition of arrested growth. Henneguy (1904) believed diapause could take place at any growth stage, egg, larva, pupa, or adult. Shelford (1929) suggested that in those instances where development or activity was spontaneously arrested diapause occurred and an interruption of growth directly controlled by unfavorable conditions would be referred to as quiescence. Lees (1955) felt that since, in many cases, the onset of diapause can be traced to an environmental stimulus the following definitions would be better suited:

Quiescence - growth or development resume immediately when favorable conditions return:

Diapause - growth or development is slow and irregular when favorable conditions return:

Lees (1955), to better examine embryonic diapause, divided the periods of growth and differentiation in the embryo into three phases. The first phase started with the successive divisions of the cleavage nuclei and ended with the formation of the blastoderm. Early growth

of the embryo up to the beginning of the formation of the mesoderm was the second phase. This phase is one of growth but not differentiation and corresponds approximately to anatrepsis. The third phase, catatrepsis, has active growth and cellular differentiation. Lees (1955) stated that there are no known species where diapause supervened before the formation of the blastoderm. Rockstein (1978) wrote that true diapause is characterized by an extremely low metabolic rate and relative inactivity as well as certain biochemical specializations. He stated that egg diapause may occur at any of several stages of embryonic development. These stages range from just after germ band formation, around the time of blastokinesis, to the full grown larval stage just before hatching. Mansingh (1971) stated that Muller divided dormancy into four categories, oligopause, quiescence, parapause, and euidiapause. Dormancy caused as the direct result of the deviation of an environmental factor from its optimum, beginning and ending with retardation was quiescence. Oligopause begins and ends gradually and is caused by the same environmental factor of equal intensity. Parapause is similar to oligopause but the intensity of the environmental factor differs between the beginning and end of dormancy. He considered two different environmental factors initiating and terminating diapause as euidiapause (Mansingh 1971). Mansingh (1971) stated that one of the major shortcomings in Muller's proposition is that it attaches no significance to the extent and severity of adverse conditions in different geographical areas and the physiological and biochemical adjustments required to overcome them. Mansingh (1971) divides dormancy into three categories, then subdivides these into three subcategories. The categories are

divided by initiators of dormancy. If dormancy is initiated by temperatures lower than optimum it is hibernation, temperatures higher than optimum is aestivation, and factors other than temperature is athermopause. The subcategories are divided by extent and length of the adverse weather conditions. A slight and short term adversity is quiescense, mild and long term adversity is oligopause, and extreme and long term adversity is diapause. Mansingh (1971) further divides diapause into high intensity diapause (teleopause) and low intensity diapause ateleopause). Mansingh (1971) noted that usually half way through diapause, the dormant individuals become capable of initiating development when returned to adequate conditions. The conditions for reactivation may vary with species; however, they are always well below the threshold for morphogenesis. Mansingh (1971) suggests that after an organism is reactivated it enters a second diapause, which is less intense than the first. The organism remains in this state until favorable conditions return. In general, almost all overwintered insects show visible developmental activity within two weeks when returned to optimum conditions.

Hormonal basis for embryonic diapause is lacking; however, it is thought to be controlled by a diapause hormone in the mother. Hasegawa (1963) extracted from the commercial silkworm, <u>Bombyx mori</u>, the diapause hormone and injected it into pupa that were expected to produce nondiapause eggs. The effect of the injection on the young oocytes was to cause their development into diapausing eggs. Rockstein (1978) stated that in <u>Bombyx mori</u> the subesophagial ganglion of the female secretes a peptide hormone which acts on the ovary. Eggs maturing under the influence of this hormone will arrest at the germ band stage and

have an increased storage of an inactive form of a non specific esterase (esterase A). A prolonged chilling is required to activate esterase A. As the egg is warmed the esterase A causes lysis in yolk cells allowing for the continuation of embryonic development. <u>Bombyx mori</u> eggs must be exposed to a chill period for the activation of the esterase A, otherwise they will not hatch. The fact that <u>Diabrotica virgifera</u> eggs will hatch without exposure to chill temperatures rules out the esterase A cycle; however, a diapausing hormone may still control the diapause in western corn rootworm eggs.

Very little research has been done on the initiation and termination of diapause in western corn rootworm eggs. There are two conflicting theories, at the present time, on the termination of diapause. Ball (1957), Howe and George (1966), Wilde (1971), Wilde et al. (1972) and Chiang (1973) reported that diapause could artificially be broken or terminated by subjecting the eggs to a chill period. Krysan (1972), Branson et al. (1975), Krysan et al. (1977), and Branson (1976A) feel that chilling only synchronizes hatch and has no influence on diapause. Krysan (1972) categorized western corn rootworm eggs as follows: prediapause - eggs in which the embryonic rudiment is not yet separated from the serosa; diapause - the embryo is developed beyond the germinal disc stage. He reported, using this criteria, that in western corn rootworm eggs held continuously at 20°C for 105 days, 31% of the eggs were in post-diapause, while 69% were still in diapause. Egg subjected to a 14 day prechill period and 21 day chill period had 83% in diapause and 17% in prediapause. A post chill period was not allowed for the 21 day chilled eggs; therefore, possibly diapause could have been broken

with a post chill period. A post chill period may have allowed for the development of eggs to the post-diapause stage in a shorter time and in a greater percentage than was indicated by the 105 day no chill test. The variation in embryo development among eggs subjected to equal treatments reinforces the idea that a genetic variation in egg development or hatch occurs. George and Ortman (1965) exposed western corn rootworm eggs to different chill periods and recorded the period of time to initial hatch in each case. They discovered that as the length of the cold treatment increased the post chill period until initial hatch decreased. Eggs held at 4°C for 41 days had an initial hatch period of 32 days, while a chill period of 153 days decreased the time to initial hatch to 18 days. George and Ortman (1965) also reported that eggs held at 22°C with no chill period began hatching after 44 days and 63% hatched at a uniformily slow rate for 145 days. Palmer et al. (1977) reasoned that since western corn rootworm eggs hatch without exposure to cold, they could be in various stages of embryonic development The overwintering of the western corn rootworm eggs in in the fall. these stages could lead to a long hatch period in the spring. Palmer et al. (1977) examined this situation and reported that lower chill temperatures adversely affect the survival rate of the more advanced stages of the embryo. Eggs held at 20° without a chill period hatched asynchronously because all developmental stages hatch (Palmer et al. 1977). The chill decreased the variability in the eggs by eliminating the advanced stages; thus, a more synchronous hatch occurred among chilled eggs. Branson (1976A) manipulated the western corn rootworm and succeeded in developing a non-diapausing population. He used nine generations

of selective pressure by discarding the later hatching larvae in a no-chill situation. He reduced the period to initial hatch in eggs, not exposed to chill periods from 60 days to 24 days. Similarly, the duration of hatch was reduced from 60 days to 25 days.

Chiang et al. (1972) studied the effects of the prechill duration, chill temperature, and chill duration on the hatching of western corn rootworm eggs. He concluded that as chill durations increased or chill temperatures decreased the viability of the eggs decreased. The viability of western corn rootworm eggs did increase as prechill durations increased up to 8 weeks. Chiang et al. (1974) tested chill temperatures and concluded that a chill temperature of 7.5°C or 10.0° C resulted in lower egg mortality and was better suited for cold storage of western corn rootworm eggs than a 5°C or lower chill temperature. Chiang et al. (1974) also noted that a post chill temperature of 30°C resulted in a quick hatch, while a 20°C temperature resulted in a greater egg viability. Branson (1976B) reported that chilling from 3 to 60 weeks did not significantly affect the time to initial hatch. The duration of hatch was significantly reduced from approximately 60 days to 15 days after 12 weeks of chill, but continued chilling no longer had an effect. The chilling of western corn rootworm eggs beyond 16 weeks is detrimental and over a period of about a year egg viability is slowly reduced by 80% (Branson 1976B).

Wilde et al. (1972) reported a regional variation in the length of time to initial hatch for western corn rootworm eggs. They found that South Dakota and Minnesota eggs required fewer days for initial and 50% hatch than Iowa, Kansas, Missouri, or Nebraska eggs. Krysan et al. (1977) examined Mexican western corn rootworm eggs and found they showed a difference in diapause intensity and resistance to desiccation when compared to South Dakota eggs. Mihm et al. (1974) reported that contact moisture was necessary for egg hatch and as the relative humidity decreased, egg survival decreased. Western corn rootworm eggs must absorb water to complete embryogenesis and this water uptake is apparently a post-diapause event (Krysan 1978). Krysan et al. (1977) reported that the Mexican western corn rootworm eggs are more resistant to desiccation and have a more intense diapause than South Dakota western corn rootworm eggs. Krysan and Branson (1977) crossed non-diapause, South Dakota, and Mexican western corn rootworms to test inheritance of diapause intensity. Five females and five males of appropriate populations were placed in a cage and five cages were used for each cross. All of the crosses produced fertile eggs except South Dakota and non-diapause males mated with Mexican females. The time interval from oviposition to hatch was graphed and Krysan and Branson (1977) noted that the crosses were intermediate between that of the parental lines. They concluded that a larger contribution to diapause intensity came from the female parent. The variation in the period of egg hatch among various geographical western corn rootworm populations may occur at a smaller scale in various fields within the same geographic This variation, if genetically linked to resistance may cause area. an increase of a resistant population of rootworms to develop in a specific area.

Walgenbach and Sutter (1977) in the mid and late 1970's were testing carbamates and organophosphates against various geographic western corn

rootworm larva. LD_{50} tests were performed in the laboratory using a microapplicator similar to the one described by Hamilton and Dahm (1960). A pattern began to develop with some of the insecticides when the field studies and LD_{50} tests were correlated. It appeared that early hatching larva were more resistant than later hatching larva.

Resistance by the western corn rootworm to the widely used carbamate insecticides was discussed by Walgenbach and Sutter (1977). During the early 1970's circumstantial field evidence pointed to resistance to metakamate as the insecticide failed or performed poorly in fields where it had been used consistently for 3 to 5 years. Carbofuran showed a similar field performance pattern in South Dakota (D. Walgenbach, South Dakota State University, personal communication). Kantack (1974) recommended the alternative use of carbamate and organophosphate insecticides for corn rootworm control. This procedure was widely followed in South Dakota and adjacent states and reduced field failures of corn rootworm insecticides. Kuhlman and Wedberg (1977) reported that poor performance of carbofuran occurred on 4 of 11 fields with a 2 or more consecutive year history of carbofuran use. He found that terbufos gave significantly better control on these fields than carbofuran. Fields having a history of organic phosphate insecticide use received rootworm control equal to or better than terbufos in 7 of 7 tests with carbofuran. He reported that on the basis of these tests it may be désirable to alternate carbamate and organic phosphate insecticides particularly if carbofuran had been used for several consecutive years in the same field. Kuhlman's (1978) research the following year indicated that a two year alternate insecticide program

was needed on carbofuran field failures before carbofuran was again used.

Ball and Weekman (1963) began monitoring rootworm resistance to insecticides in Nebraska during 1962. Topical evaluations were conducted on adults with mortality determined at the end of a 2-hour period. Results indicated that adults collected from areas where control problems existed and where aldrin had been used the longest were more resistant to aldrin than were adults from other areas. Since that time, published work has been limited to adult susceptibilities to the rootworm insecticides on a 2-hour mortality period. Correlations between adult topical data and field performance of the insecticides have not been established. Adult topical data taken 2 hours after have been shown to be misleading compared to 48 hour data and has no apparent correlation with larval susceptibility (D. Walgenbach, South Dakota State University, personal communication). Early developing larvae have shown greater tolerance for carbofuran than were late developing larvae. This situation complicates field sampling techniques and adds to the effect of environmental rootworm mortality on the performance of insecticides. The differential selective pressures from weather, cultural operations and planting dates may, therefore, influence rootworm susceptibility to insecticides in addition to the influence from insecticidal pressure (Walgenbach and Sutter 1977).

Branson (1976A) manipulated the western corn rootworm population and developed a strain of non-diapausing western corn rootworms (WCRND). Selection pressure was applied by selecting and rearing only larvae from eggs that hatched within 12 weeks of oviposition. Two generations of recombination without selection pressure followed the initial selection. The M4 generation had a selection pressure of 11 weeks applied. Thereafter, increasingly intense selection pressure was applied in each generation (reduction of 1-2 wk/generation) through M10 generation. The population underwent a total of 12 generations of selective pressure reducing the mean time to initial hatch from 58 days to 23 days thus eliminating diapause. Walgenbach (South Dakota State University, personal communication) tested various rootworm insecticides against early and late developing western corn rootworm non-diapausing larvae. The early larva were collected from eggs hatching during the initial 2 days of hatch and the late larva were collected from eggs hatching after 2 weeks from initial hatch. The 3rd instar larva were used for the test and probit analysis of the results (Appendix A) indicated that the early WCRND were 2 to 4 times more resistant to metakamate, carbofuran, and dyfonate.

The surveying and knowledge of what type of population a corn producer had could help prevent field failures of certain insecticides. This knowledge would give the corn producer the tool needed for choosing the best means of control for the western corn rootworm.

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PROCEDURE

Western corn rootworm beetles were collected from the Southeast Experiment Farm near Beresford, South Dakota and from the Tim Merrill farm near Elkton, South Dakota on July 23, 1977. The beetles were caged in the laboratory and sexed according to criteria published by Branson et al. (1975). Paired beetles, one female and one male were placed in cages, 25 cages for the Beresford population and 30 cages for the Elkton population. The cages were made from round, clear, plastic containers¹ 9 cm in diameter and 4 cm in height. Holes 2 cm in diameter were cut into the lids of the containers and fitted with nylon screen attached with duco cement to provide ventilation. Temperature and photoperiod were controlled artificially and there was equal exposure for all the adults. Temperature was maintained at 23 \pm 3°C by the internal heating and cooling system of the building. A 12 hour photoperiod was induced from 8:00 a.m. to 8:00 p.m. using a timer connected to a 100 watt incandescent light since Chiang (1973) reported that the maximum oviposition occurred when female western corn rootworms are exposed to an 8-12 hour photoperiod.

Methods for rearing the beetles were similar to those published by Branson et al. (1975). Egg dishes were placed in each cage on the date of capture and replaced every seven days. Disposable 5.5 cm polystyrene petri dishes² filled with 60 or 80 mesh sterilized

1_{No.} 300 plastic container, mfg. Peima Industries, Inc., P.O. Box 975, Henderson, Kentucky 42420

²Kimble, Owen & Ill, Inc., P.O. Box 1035, Toledo, Ohio 43666

soil served as the egg dishes. The supply of 60 mesh soil was the greatest, therefore, it was primarily used. The egg dishes from Elkton cages 1-15 were used for egg counts and 80 mesh soil was used as the oviposition medium. Western corn rootworm eggs are larger than 60 mesh; therefore, 80 mesh soil would pass through a 60 mesh screen while the rootworm eggs remain behind (Lawson and Weekman 1966). The egg dishes were moistened with distilled water and were kept damp with no standing water in them.

The beetles were fed 1/2 inch portions of immature ears of sweet corn, small bundles of 318 g - 416 g Pioneer corn seedlings and water every other day. The corn bundles were placed over the egg dishes to keep them moist and to increase the ovipositional preference of the females toward the egg dish. The water was administered through a vial fitted with a cotton stopper which served as a wick. Sweet corn ears were placed in cold storage to provide food during the period after it was not available from the field. A dry diet developed by Branson et al. (1975) was used after the first week in October.

The egg dishes were labeled with the field location of the female, cage number of the female and the week the egg dish was placed in the cage. Contents of dishes containing eggs from Elkton females in cages 1-15 were washed over a 60 mesh screen and the total number of eggs per egg dish was recorded and compared to past studies. Egg dishes from Elkton cages 16-30 were placed in cold storage at 6° C \pm 3°. The egg dishes from Elkton cages 16-22 were removed January 9 and the egg dishes from cages 23-30 were removed April 17, both sets were placed into a Hythermo, model 5520, incubator at $25^{\circ} \pm 1^{\circ}$.

Egg dishes from Beresford cages 1-10 were placed directly into incubation without a cold treatment. Beresford cages 11-25 had their egg dishes placed in cold storage at $6^{\circ}C \pm 3^{\circ}$. The egg dishes from Beresford cages 11-17 were removed January 9 and the egg dishes from Beresford cages 18-25 were removed April 17, both sets were incubated at 25°C \pm 1°.

While all the egg dishes were in cold storage, they were sealed in plastic sacks to prevent moisture loss. Hatch was checked every 1 to 3 days, depending on the time available, by placing a newly germinated kernel of B37-H84 Pioneer corn in the egg dish. Leaving the corn in the dish longer than 3 days caused problems in removal because of root growth. The inner surface of the lid of the egg dish was coated with stop cock grease to prevent larvae from escaping. The lid, surface of the soil, and kernel were all examined under 10X magnification for larvae. The egg dishes were discarded when no larva were recorded for a three week period after initial hatch. All larva, after being counted, were discarded except for two cases. Larva were collected during the first 3 examinations of the egg dishes from Beresford females numbers 11 through 17 and reared to third instar on germinated corn. The corn and larva were placed in round plastic containers lined with moist filter paper. The corn and filter paper were changed every few days to prevent mold and growth. Since the larva were collected during the first three examinations, they came from early hatching eggs. To determine if these larva showed similar LD50 ratings to carbofuran as other early hatching larva, an LD50 carbofuran test was performed on the 3rd instar larvae using a microapplicator

similar to Hamilton's (Hamilton and Dahm 1960). Probit analysis of the results from this test were recorded for insecticide susceptibility on a ug/g of insect tissue basis. Larva were also collected during the middle of hatch, May 4, 1978 from Elkton females numbers 23 through 30 and from Beresford females numbers 18 through 25 and reared as the previous larva. They were divided into three separate groups, Elkton, Beresford, and Beresford female 26 and placed in three growing containers. The larva were fed germinated corn and after 16 days larval lengths were recorded.

Early hatching larva were selected from the western corn rootworm non-diapause, WCRND, developed by Branson (1976A). These larva were selected by choosing only larva hatching during the first two days of hatch in a single WCRND egg dish obtained from Branson. The larva were reared through to the adult stage and designated as early nondiapausing. Similarly, larva hatching 2 weeks after initial hatch in the same egg dish were reared to the adult stage and designated as late non-diapause. Because of the few larva obtained from the egg dish and the mortality that occurred during rearing only 2 female and 2 male adults for each group survived. One female and one male of the same category were placed in a cage. The cages were then placed with the Beresford and Elkton cages and the adults were reared in the same manner. The egg dishes were not subjected to a cold treatment. The larva were counted using the same procedure as described. Only one female of each group laid fertile eggs. Longevity of all females in the study was recorded and the males were removed from the cages two weeks after their introduction.

The hatching pattern of the parental line must be established to determine whether the length of the period between oviposition and egg hatch is inherited. Individual female and male western corn rootworm beetles were collected from the field and their hatching pattern was determined. Beetle counts (Appendix B) from five pheromone check traps placed at the Southeast Experiment Farm near Beresford. South Dakota resulted in a daily 5 trap catch of 2 or less western corn rootworm beetles for July 6, 7 and 8. The 5 trap catch for July 11 was 15 indicating that the majority of the beetles collected July 23 were 2 weeks old or less. The relative early emergence of these beetles indicated they came from early developing eggs. The timing of adult emergence would not necessarily give an accurate account of the inherited hatching pattern for the adult. Temperature and moisture play a major role in the development and viability of western corn rootworm eggs. Soil temperatures vary each year and at different depths. Average weekly soil temperatures at Brookings, South Dakota varied between the 1" and 6" temperature readings as much as 23.7°F during 1975 (Table 1) and 4.5°F during 1976 (Table 2). Chiang (1965) reported that after farming operations eggs could be buried up to 12 inches deep in the soil. The variation in soil temperature and egg depth could cause early developing eggs to hatch later than late developing eggs depending on the location of these eggs in the soil. The temperature, moisture conditions and adult nutrition pattern during 1976 and 1977 are credited for causing a 90% to 95% reduction in the western corn rootworm population

Day of Year	Temp. at l" ^o F	Temp. at 6" °F	Difference
103	41.4	32.0	9.4
110	45.5	32.3	13.2
117	48.7	31.7	17.0
124	55.1	33.5	21.6
131	57.7	40.2	17.5
138	67.2	43.5	23.7
145	62.6	46.7	15.9
152	62.6	52.7	9.9
159	61.0	55.1	5.9
166	68.1	62.3	5.8

Table 1.--Average weekly soil temperatures at 1" and 6" depths during the spring of 1975 in Brookings.

Day of Year	Temp. at 1" °F	Temp. at 6" o _F	Difference
89	43.3	39.8	3.5
96	51.4	48.1	3.3
103	54.6	52.3	2.4
110	48.1	48.4	-0.3
117	46.8	47.3	-0.5
124	58.3	54.1	4.2
131	61.5	59.1	2.4
138	63.9	61.0	2.9
145	61.7	60.5	1.2
152	70.4	65.9	4.5
159	70.8	68.6	2.2

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Table 2.--Average weekly soil temperatures at 1" and 6" depths during the spring of 1976 in Brookings.

over eastern South Dakota during 1977. It could not be determined whether this reduction occurred among early or late developing eggs. Since environmental conditions and the reduction of the western corn rootworm population may have influenced the hatching pattern of the adults collected a second method of determining hatching patterns was used.

Susceptibility to carbofuran provides a means of differentiating between populations showing different developmental patterns (D. Walgenbach, South Dakota State University, personal communication). A carbofuran test (Appendix C) on larvae obtained from the second days eggs hatch from the ovipositional dishes of Beresford females numbers 11 through 17 indicated that these larva were closer in LD₅₀ values to late WCRND and 1976 Beresford larva. This indicates that the larva were susceptible to carbofuran and probably developed from a late developing parental line.

Longevity of the western corn rootworm females ranged from 14 to 112 days for the 25 Beresford females (Table 3) and from 14 to 110 days for the 30 Elkton females (Table 4). This agrees with the range of 19 to 126 days reported by Branson and Johnson (1973) and the 9 to 84 day range reported by Ball (1957). The 67 day mean longevity for the Beresford females and the 60.5 day mean longevity for the Elkton females falls between the 47 day mean reported by Ball (1957) and the 94.8 day mean reported by Branson and Johnson (1973). Diet plays a major role in the longevity of western corn rootworms (Singh and Howe 1971, Guss et al. 1976). Since different diets were used in Ball's, Branson's, and this study, the range in longevity is more important in determining

Female in Cage #	Longevity in days	Female in Cage #	Longevity in days
1	35	14	62
2	91	15	25
3	91	16	91
4	55	. 17	69
5	69	18	25
6	62	19	97
7	35	20	105
8	46	21	112
9	46	22	96
10	62	23	62
11	93	24	105
12	62	25	69
13	14		

Table 3.--Longevity of each Beresford female after the date of capture.

Average longevity 67 days.

65 C.			ILG AGINE IN
Female in Cage #	Longevity in days	Female in Cage #	Longevity in days
1	62	16	73
2	43	17	98
3	62	18	47
4	62	19	88
5	43	20	99
6	62	21	42
7	25	22	14
8	28	23	25
9	32	24	91
10	59	25	110
11	62	26	108
12	48	27	88
13	60	28	62
14	14	29	108
15	32	30	69

Table 4.--Longevity of each Elkton female after the date of capture.

Average longevity 60.5 days.

whether the longevity of the Elkton and Beresford females was acceptable rather than the mean longevity.

Egg totals per female for Elkton females numbers 1 through 15 (Table 5) ranged from 0 to 1109 with 60% of the females laying less than 100 eggs. The wide variation in egg totals presently can not be explained; however, the 1109 and 696 egg totals for two of the females indicates that the rearing procedure was adequate and probably did not cause the variation.

The larval totals of the Beresford population (Table 6) showed less variation than the Elkton population (Table 7). The larval total range, 0 to 914, for the Elkton population was similar to the range reported earlier for the egg totals from the same population and 53% of the females had less than 100 larva. There was no significant difference between the 335 day and 323 day means for the 162 day and 260 day cold treatments for the Beresford population; therefore, it is assumed the 2 different durations of $6^{\circ} \pm 3^{\circ}$ C chill had the same effect on egg viability.

Palmer et al. (1977) showed that chilling adversely affects western Corn rootworm eggs in more advanced stages of development causing a consolidation of egg hatch. The 8 day variation in the number of days to peak hatch for Beresford females 11-25 (Table 8) and the 76 day variation (Table 9) recorded for Beresford females 1-10 indicates that a consolidation of hatch did occur when a $6^{\circ} \pm 3^{\circ}$ C chill period was induced. The 105-181 day range reported for peak hatch for Beresford females 1-10 (Table 9) indicates that a 10 week variation occurs among the progeny of late developing adults.

emale in Cage #	Total Eggs
1	134
2	9
3	231
4	396
5	4
6	696
7	0
8	46
9	23
10	9
11	1109
12	189
13	7
14	1
15	2

Table 5.--Egg totals for individual field collected Elkton female WCR.

Mean: 190.4 Range: High 1109 Low 0

Beresford Female in Cage #	Total Larva	Beresford Female in Cage 4	Total Larva	
1	382	11	425	
2	234	12	540	
3	490	13	84	162 Day
4	530	14	515	Cold Treatment
5	406	15	78	
t 6	189	16	425	
7	219	17	278	
8	245	Mean	335	
9	319	10	220	
10	220			
Mean	323			
				260 Day
				Cold Treatment
		25	404	
		Mean	521	
	Female in Cage # 1 2 3 4 5 5 5 6 7 8 9 10	Female in Cage # Total Larva 1 382 2 234 3 490 4 530 5 406 6 189 7 219 8 245 9 319 10 220	Female in Cage # Total Larva Female in Cage # 1 382 11 2 234 12 3 490 13 4 530 14 5 406 15 6 189 16 7 219 17 8 245 Mean 9 319 18 10 220 19 Mean 323 20 21 22 23 24 22 23 25 24 25	Female in Cage # Total Larva Female in Cage # Total Larva 1 382 11 425 2 234 12 540 3 490 13 84 4 530 14 515 5 406 15 78 6 189 16 425 7 219 17 278 8 245 Mean 335 9 319 18 230 10 220 19 581 Mean 323 20 613 21 928 22 620 23 440 24 353 25 404 353 35

Table 6.--Larval numbers from individual field collected Beresford female WCR.

-		 Contraction of the Contraction of Co			
2 1	lkton fema in Cage #	Total Larva		Elkton female in Cage #	Total Larva
-	16	477	gene breasen an wegen agen an reform ^{an o} the entry wegen and beidde	23	
	17	347		24	2/
160 Dava	18	232		25	-
162 Day Cold Treatme	nt 19	13	260 Day Cold Treatment	26	914
	20	391	Cold Treatment	27	
	21	200		28	12
	22	400-400-600 400-400-600		29	868
	Mean	237		30	2
:				Mean	224

Table 7.--Larval numbers from individual field collected Elkton female WCR.

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-		an standar fan Merenige en gependelen af an Marine fan Denarde fan de		
	Reresford Female in Cage #	Number of Days to Peak Hatch	Elkton Female in Cage #	Number of Days to Peak Hatch
	11	25	16	25
	12	30	17	28
162 Day	13	28	18	25
Cold Treatmen	14	25	19	28
	15	25	20	25
	16	30	21	28
	17	28	22	
1	18	23	23	
	19	24	24	400-400
260 Day	20	24	- 25	
Cold Treatmen	1t 21	24	26	23
	22	27	27	-
	23	24	28	22
	24	22	29	24
	25	24	30	
-	High	30		28
	Low	22		22
	Mean	23.9		25.3

Table 8.—The period from oviposition to peak hatch, for the eggs from individual Beresford and Elkton female WCR.

				Contraction of the	Server any are
	Beresford Female in Cage #	Number of Days to Peak Hato	Femal	e in	Number of Days to Peak Hatch
	1	105	. 6		181
	2	142	7		144
No Cold Treatment	: 3	142	8		132
	4	181	9		154
	5	105	10		144
	High:	181	Low: 105	Mean:	143

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Table 9.-- The period from oviposition to peak hatch for the eggs from individual Beresford females WCR 1-10.

The hatching period was broken down into the time from oviposition, excluding cold treatment periods, to initial hatch, 50% hatch, and final hatch. A large variation occurred within the initial hatch, 50% hatch, and final hatch periods between Beresford females numbers 1 through 10 (Table 10). The 63-115 day initial hatch range recorded for Beresford females 1-10 indicates that adults with a similar hatching pattern have progeny which hatch at different rates. The duration of hatch for the eggs from Beresford females 1-10 ranged from 82 to 133 days. This indicates that the progeny of any individual female whose eggs were not subjected to a chill period hatched over a period of 3 to 4 months.

A chill period, as previously mentioned, consolidates egg hatch; therefore, the variation between the days to initial hatch for the eggs from Beresford females 11-25 (Table 11) and Elkton females 16-30 (Table 12) is only 8 days and is much shorter than the 52 day variation recorded for Beresford females 1-10. There was no significant difference between the Elkton and Beresford populations when the days to initial hatch, 50% hatch, and final hatch for the eggs exposed to a 60 ± 30 C chill period were compared. Since the chill duration was not recorded in the periods to hatch for Beresford females 11-25 and Elkton females 16-30 it was assumed the hatching patterns would be similar to a nondiapausing female. The mean of 22.7 days and 23.2 days reported for the Beresford and Elkton populations is similar to the 23 and 25 days to initial hatch reported for the early and late WCRND females (Table 13).

	Beresford Female in Cage #	Days to Initial Hatch	Days to 50% Hatch	Days to Final Hatch
	1	81	126	189
	2	86	145	207
	3	86	139	202
	4	91	170	202
No Cold Creatmer	nt 5	63	136	196
	6	91	184	208
	7	88	136	196
	8	109	145	191
	9	86	159	197
	10	115	173	207
	High	115	184	208
	Low	63	126	189
	Mean	89.6	151.3	199.5

Table 10.--The number of days from oviposition until initial, 50%, and final hatch for eggs from Beresford females 1-10.

Beresford Female in Cage #	Days to Initial Hatch	Days to 50% Hatch	Days to Final Hatch
11	23	25	44
12	23	29	44
13	28	34	44
14	23	25	39
15	23	25	33
16	23	29	44
17	23	28	39
18	22	26	29
19	22	27	31
20	22	28	42
21 ·	22	27	36
22	22	28	36
23	22	27	31
24	20	23	28
25	22	27	29
High	28	34	44
Low	20	23	28
Mean	22.7	27.3	36.6
	Pemale in Cage # 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 High Low	Pemale in Cage # Initial Hatch 11 23 12 23 13 28 14 23 15 23 16 23 17 23 18 22 20 22 21 22 22 22 23 22 24 20 25 22 Eligh 28 Low 20	Pemale in Cage # Initial Hatch 507 Hatch 11 23 25 12 23 29 13 28 34 14 23 25 15 23 25 16 23 29 17 23 28 18 22 26 19 22 27 20 22 28 21 22 27 20 22 28 21 22 27 22 22 28 23 22 27 24 20 23 25 22 27 High 28 34 Low 20 23 25 22 27 26 34 20 23 25 20 23 26 23 24 20 26 23 23

Table 11.--The number of days from oviposition **to initial**, 50%, and final hatch for eggs from **Beresford** females 11-25.

	Elkton Female in Cage #	Days to Initial Hatch	Days to 50% Hatch	Days to Final Hatch
	16	23	28	44
	17	23	28	39
	18	23	28	39
162 Day Cold Treatment	19	28	33	34
	20	23	25	39
	21	23	28	36
	22		42-425	-
	23	63-00		
	24		65-69	-
	25	105-002	93	
260 Day Cold Treatment	26	22	27	39
	27	-	-	
	28	22	23	26
	29	22	27	36
	30	Q148	6945	
2	Contraction of the Contract of			44
	High	28	33	
	Low	22	23	26
	Mean	23.2	27.4	36.9
	Carlange for surgical between the state	The ball and plates, the baseling high many contrast of possible of the states		

Table 12.--The number of days from oviposition to initial, 50%, and final hatch for eggs from Elkton females 16-30. Table 13.--The number of days from oviposition to initial, 50%, and final hatch for eggs from the WCRND females.

Female Type	Days to Initial Hatch	Days to 50% Hatch	Days to Final Hatch
WCRND Early	23	34	76
WCRND Late	25	33	53

		· · · · · · · · · · · · · · · · · · ·	
Date	Elkton Larva	Beresford Larva	Beresford Female 21 Larva
Мау 18	.3 ст	.4 cm	.3 cm
	.3 cm	.4 cm	.8 cm
	1.1 cm	.5 cm	
	1.2 cm	1.1 cm	
		1.2 cm	
May 20	.4 cm	.4 cm	.5 cm
	1.0 cm	.4 cm	.5 cm
	1.0 cm	.5 cm	.5 cm
	1.0 cm	.5 cm	.9 cm
	1.0 cm	.5 cm	.9 cm
	1.0 cm	1.0 cm	1.0 cm
	1.1 cm	1.1 cm	1.0 cm
	1.1 cm	1.1 cm	1.0 cm
	1.1 cm	1.2 cm	1.1 cm
	1.1 cm	1.3 cm	1.2 cm
	1.2 cm		1.2 cm

Table 14.--Larval length from 3 different populations.

First instar larvae were collected and placed on germinated corn on May 4. The Elkton larva were collected from eggs laid by eight females, while larvae from a single Beresford female, number 21, were reared leaving only seven females to contribute to the Beresford group. Due to the lack of time, larva were only moderately checked on May 18. The length of the larva was measured by placing them on a cm grid under 10X magnification. Only larva which appeared healthy and had good color were used. The larva were not stretched during measuring and they were measured only while they were at rest. The results (Table 14) indicate that growth rate is variable even among the progeny of a single female.

CONCLUSION

The egg hatching pattern was established for 10 western corn rootworm females whose eggs were not subjected to a chill period. The results indicate that a single female western corn rootworm's eggs will hatch over a 3-4 month period if the eggs are not exposed to a cold period. Since the interval of hatch occurred over a 12 to 16 week period, it appears that the progeny of a single female would be both late and early developing. The variation in larval length recorded for the Elkton, Beresford and Beresford female 21 larvae indicates that larval development does vary among females within a geographic area and among the progeny of individual females.

A chill period apparently consolidates hatch, since the egg hatching pattern for the chilled eggs from Beresford and Elkton females was only 24 days or less in length. The small variation between the Beresford and Elkton populations when the initial hatch periods, 50% hatch periods, and final hatch periods were compared indicates that hatching patterns for western corn rootworm females may be similar at geographic locations only 100 miles apart.

Further studies should be performed on adults from fields where carbofuran has a history of failure. The comparison of the hatching patterns from these adults would explain whether resistance could develop in an isolated geographic location from selective mortality of non-resistant adults.

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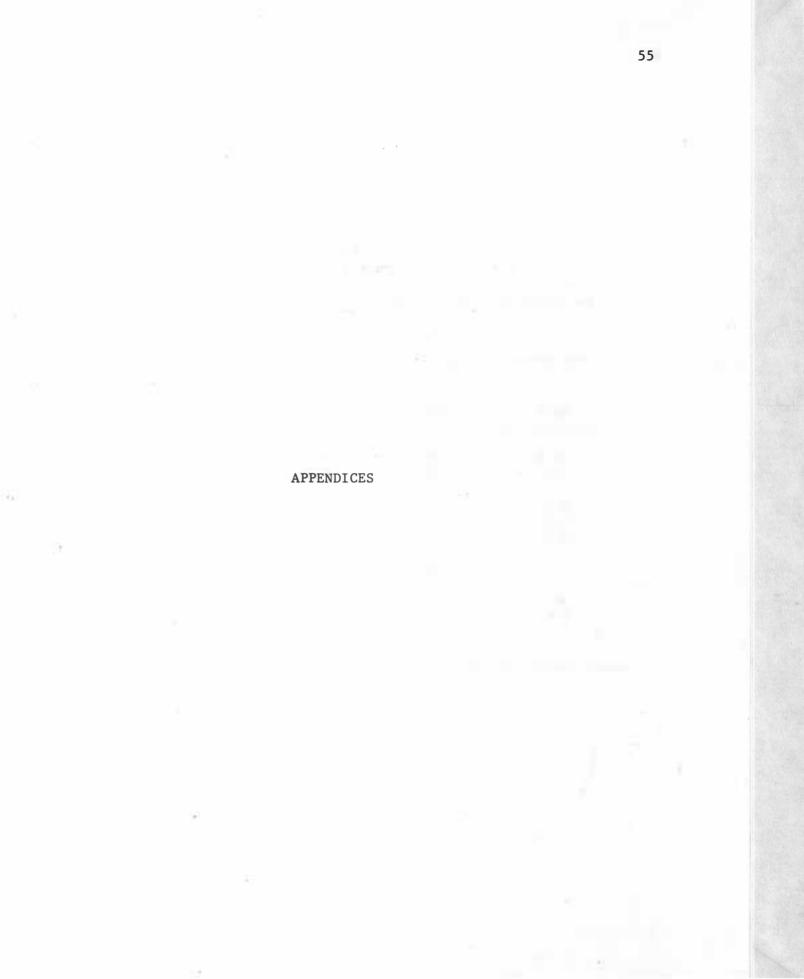
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APPENDIX A

Comparative susceptibility of 3 insecticides to early and late hatching WCRND larvae.^a

Ins ecticide	Larval Type	Date	LD ₅₀
Вих	Early WCRND	5-9-77	8.91
	Late WCRND	3-22-77	2.37
Furadan	Early WCRND Early WCRND Early WCRND Late WCRND	3-8-77 3-11-77 3-16-77 1-3-77 2-24-77	14.8 25.07 25.90 8.5
Dyfonate	Early WCRND	3-31-77	5.36
	Late WCRND	3-18-77	2.69

^a Unpublished data, D. Walgenbach, SDSU, Brookings, South Dakota

APPENDIX B

Total adult WCR counts from 5 pheromone traps located near Beresford, South Dakota.^a

	NCR Count 5 Traps
Date For All 7-6-77 7-7-77 7-8-77	5 Traps
7–7–77 7–8–77	1
7-8-77	T
	1
7-11-77	2
	15
7–13–77	25
7-15-77	53
7-23-77 Collect	ion Date

^a Michael Lockwood, unpublished data, South Dakota State University

APPENDIX C

Furadan LD_{50} values for F_1 and field collected Beresford western corn rootworm and WCRND larvae.^a

Larval Type	Location	Tre atment Date	LD ₅₀
1975 field ^b	Beresford	7-5-75	22.0
1975 fie ld	Beresford	7-10-75	12.9
1976 field	Beresford	7-14-76	9.2
1976 F ₁ lab ^c	Beresford	7-5-76	4.3
1977 F₁ lab	Beresford	3-31-77	15.5
1977 F₁ lab	Beresford	4-4-77	27.3
1978 F ₁ lab	Beresford	3-27-78	37.8
1978 F₁ lab	Beresford	4-13-78	54.6
Barly WCRNDd	Lab	3-8-77 3-11-77 3-16-77	14.8 25.1 25.9
Late WCRND ^e	Lab	1-3-77 2-24-77	8.5 9.8
WCRND (control)	Lab	2-13-78	13.8
1978 F₁ 1 ab (project)	Beresford	2-13-78	8.8

- Unpublished Data, D. Walgenbach, South Dakota State University.
- b Larva collected from the location indicated during the year indicated.
- **c** Larva reared from eggs laid by adults field collected the previous year.
- d Non-diapausing western corn rootworm larva hatching from egg dishes during the first 2 days of hatch.
- Non-diapausing western corn rootworm larva hatching from egg dishes 2 weeks after hatch has started.