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SEASONAL FECUNDITY DYNAMICS OF THE PINK BOLLWORM PECTINOPHORA GOSSYPIELLA (SAUNDERS)

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SEASONAL FECUNDITY DYNAMICS OF THE PINK BOLLWORM *PECTINOPHORA GOSSYPIELLA* (SAUNDERS)

by

Nathan Mark Schiff

A Thesis Submitted to the Faculty of the

DEPARTMENT OF ENTOMOLOGY

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ABSTRACT

Pink bollworm larvae from short and long staple cotton blooms and bolls were collected in the early and late season in Arizona. The emergent moths from these populations were paired and their egg production determined in the laboratory. The number of viable eggs produced by females producing viable eggs was similar for each population: 137.6 and 123.1 eggs for long and short staple bloom-reared populations, and 121.5 and 116 eggs for the long and short staple boll-reared populations. However, the percentage of females in each population that laid viable eggs varied with larval food source and cotton type: 48.7% and 59.4% for the short and long staple bloom-reared populations, and 52.1% and 19% for the short and long staple boll-reared populations.

INTRODUCTION

An important aspect of a pest management program is the ability to predict the population growth of the pest. To do this there must be a method of sampling that reflects the current population size, coupled with knowledge of the reproductive capacity of the pest population. In Arizona 2 types of cotton are grown, long staple (*Gossypium barbadense* L.) and short staple (*G. hirsutum* L.). A Major pest of both is the pink bollworm (PBW).

There have been a number of studies relating pink bollworm fecundity to a variety of parameters. Adkisson (1961) examined the effects of various larval diets on fecundity, Philipp and Watson (1971) and Fye and McAda (1972) looked at the effects of temperature. Raina and Bell (1978) showed the importance of adult feeding. However, no comparative study has been conducted between long staple and short staple field reared populations at different times during the growing season. The specific objectives of this research were to determine the fecundities of emergent pink bollworm females from larvae reared on cotton squares and cotton bolls of long staple and short staple cotton.

LITERATURE REVIEW

The Pink Bollworm

In the United States the pink bollworm *Pectinophora gossypiella* (saunders) is a serious pest of cotton grown in the southwest (Vail, Henneberry, Bariola, Wilson, Wilson, Kittock and Arle 1978). It was first discovered in the U. S. A. in Hearne, Texas in 1917 (Noble 1969), and in 1926 it was found in the eastern counties of Arizona (Wene, Carruth and Telford 1965; Noble 1969). Currently, in Arizona, pink bollworm (PBW) is a pest in all the cotton producing counties (Spears 1967; Vail et al. 1978).

Description of Life Stages

The most comprehensive description of the PBW was by Busk (1917). The following, except where otherwise indicated, is derived from that work.

The PBW egg is elongate, oval, flattened; about 1 mm long and 0.5 mm broad; the shell is pearly white, with a finely wrinkled surface. When newly laid, the egg has a slightly greenish tint. At maturity it turns reddish.

PBW larvae range in size from 1 mm when newly hatched to 11-13 mm in the last instar. They are cylindrical in shape with a reddish brown head and blackish brown mandibles. The early instars are white, with the dorsal side gradually becoming suffused with pink as the larvae mature.

The pupa is obtect, 8-10 mm long, reddish brown with the posterior end pointed and terminating in a short, stout, upwardly turned hook-like cremaster.

The adult is a small grayish brown moth with a wingspan of 15 to 20 mm (Noble 1969). he hind wings are acutely pointed and heavily fringed posteriorly (Loftin, McKinney and Hanson 1921).

Oviposition Behavior and Egg Development

Early in the season PBW eggs are laid on the squares, stems and terminal buds of the cotton plant. Later in the season when bolls are available, the eggs are laid singly above the calyx or in clusters at the base of the bolls (Brazzel and Martin 1957). In Texas eighty percent of egg laying occurs between 1600 Hr and 2400 Hr according to Lukefahr and Griffin (1957).

Incubation periods vary with temperature and moisture. Owen and Calhoun (1932) found that incubation was shortest (4.3 days) at a mean temperature of 29°C and longest (25.5 days) at 12°C when kept in small stender dishes with dampened blotters to provide moisture. Philipp and Watson (1971) in their study of the effects of temperature on population growth, observed that the incubation period, at a relative humidity of at least 50%, varied from

8.5 to 3.5 days at constant temperatures of 24°C and 30°C, respectively. Adkisson (1959) examined the effect of various humidity levels on hatchability of pink bollworm eggs and found that at 26.7°C constant temperature the percent hatch varied form 67.5% at 0° relative humidity to 90.4% hatch at 75% relative humidity.

Larval Development in Squares

In early season before bolls are present on the plants, larvae burrow into the squares to feed. During this period larvae need 10 day old squares to survive, since younger squares that are attacked are shed by the plant. During development, larvae produce webs which seal the tips of the flower petals together forming a rosetted bloom (Noble and Robertson 1964). In short staple cotton, rosetting is a sign of PBW infestation, but in long staple cotton where blooms rosette naturally, flower dissection must be made to determine whether a PBW has caused the rosette. Owen and Calhoun (1932) reared PBW larvae in short staple squares covered with cellophane bags in the field, and determined the larval duration to be 10.1 days with a range of 6-18 days. Butler and Henneberry (1976) using their own observations and the equation of Butler and Hamilton (1976) estimated an 8-9 day larval stage for short staple, square-reared larvae. There is no report in the literature of larval development time for PBW larvae reared in squares of long staple cotton. Economic damage to cotton by PBW larvae reared in squares is insignificant, except that it permits early season population establishment which is critical for the survival of PBW in annually planted cotton.

Larval Development in Bolls

In short staple cotton, bolls are only susceptible to PBW attack during a specific period. Bolls less than 14-days old are too wet for larval survival, bolls more than 21-days old are too hard and dry. When susceptible bolls are available (14-21 days old), first instar larvae burrow through the carpel wall to feed on the lint and seeds (Van Steenwyck, Ballmer and Reynolds 1976). The entrance hole is so small as to be undetected from the outside of the boll, but appears as a pimple-like growth about 2 mm wide and 1.5 mm high on the inner carpel wall (Khalifa 1966). Larvae are antagonistic to each other and will attack any other larvae they come into contact with in the boll (Brazzel and Martin 1955). The PBW commonly has 4 larval instars (Fife 1937) although Watson and Johnson (1974) in their laboratory study reported five instars in some cases. Larvae develop faster in older bolls but do more damage in the lint and seeds in younger bolls (Lukefahr and Griffin 1962). According to

Fry, Kittock and Henneberry (1978) the infestation susceptibility, as determined by the relationship between PBW per gram seed cotton and boll dry weight, was the same for both long staple and short staple varieties of cotton but the percentage of seed damaged was greater in the long staple cotton.

Owen and Calhoun (1932) reported that newly hatched larvae placed on bolls, in a short staple cotton field, and then enclosed in cellophane bags, fed for an average of 16.5 days with a range of 11-25 days. Noble (1969) stated that the larvae enter fruiting forms where they feed for 10-15 days. There is again no literature available for larval developmental time in long staple cotton bolls.

Since insects are poikilothermic, a more accurate means of describing developmental time than calender days would be in terms of heat accumulation (degree days, heat units), or number of days at a particular temperature. Under laboratory conditions, larval developmental time was shown to vary with temperature. Philipp and Watson (1971) reared larvae at both constant and fluctuating temperatures that correspond to temperatures in the field. The developmental times ranged from 27 days at constant 24°C to 21 days at temperatures fluctuating between 28°C and 37°C. This agrees with the work of Fye and McAda (1972) who found larval developmental times to vary between 34.6 days at 20°C constant temperature and 20.5 days at 30°C constant temperature.

Heat accumulation models have been developed in Arizona (Huber, Moore and Hoffman 1979) to predict spring emergence and generation times. It takes 750 $\pm$ 50 fahrenheit heat units for the first PBW generation to develop in the early season in cotton squares, while in mid and late season one generation reared on bolls takes 800 $\pm$ 50 fahrenheit heat units (Huber 1981).

Pupal Stage

Just prior to pupation the larvae cut-out of the bolls making a round exit hole approximately 2 mm in diameter. Ninety-eight percent of these larvae leave between 1000-1300 Hrs according to Butler and Henneberry (1976). The larvae drop to the ground where they prefer to pupate in damp loose, slightly lumpy soil, in which some trash has accumulated (Fye and Brewer 1975). Unless they find a pupation site quickly, high soil temperatures can cause mortality of the larvae and/or reduce the fecundity of the ensuing adult (Fye 1971). High soil temperatures are most important early in the season, before the plants are large enough to shade the soil (Fye 1971; Clayton and Henneberry (1979).

Temperature, as in larval development is the major factor in pupal duration. Guerra and Ouye (1968) found that artificial diet reared PBWs had a pupal development time of 8.9 days at 26.7°C and 14:10 light dark cycle, while Philipp and Watson (1971) found that the pupal stage varied from 15 days at a fluctuating 19.5°C to 37°C temperature regime to 6 days at a constant 32.5°C. This agrees with Fye and McAda (1972) who reported pupal development times of 15.5 days at 20°C ranging to 6.9 days at 33°C.

At ambient field temperatures Loftin, McKinney and Henson (1921) found that pupae from diapause larvae took 10.5 days for males and 10.0 days for females to develop as compared to 9.3 days for pupae from non-diapause larvae. Owen and Calhoun (1932) showed that under field conditions pupae from square reared larvae took an average of 8.1 days to develop with a range of 6-24 days, compared to 8.6 days with a range of 7-14 days for pupae from boll reared larvae.

Adult Stage

Lukefahr and Griffin (1962) found that most adults emerge between 0600 and 0900 Hrs. The adults are very seclusive during the day becoming active at sundown (Loftin et al. 1921; Noble 1969).

Mating

Squire (1937) and Lukefahr and Griffin (1957) found that mating normally occurs between 0200 and 0500 Hrs, after a 7 hour period of darkness with less than 3 foot candles of light. Under continuous light conditions mating occurred throughout the day (Ouye, Graham, Richard and Martin 1964). Henneberry and Leal (1979) also observed mating under continuous light conditions but at a reduced rate.

In field studies female PBW's collected at light traps were shown to have mated an average of 1.1 times determined by dissection of the bursa copulatrix and counting of the spermatophores (Graham, Glick, Ouye and Martin 1965). According to Ouye et al. (1964) one spermatophore was equal to one mating.

In the laboratory much higher mating frequencies were found of 2.3 times for females and 4.2 times for males (Ouye, Garcia, Graham and Martin 1965). In the laboratory at 30°C 41% of matings occurred within the first 6 days and the last mating on the 16th day after pairing. Of 94 pairs individually caged, 74.5% mated (Ouye et al. 1964). Raina and Bell (1978) report increased mating from 1-3 times at 25°C and 60% relative humidity, when adults were fed 10% sucrose or 10% honey solution.

Longevity

There are differing reports on the longevity of PBW adults depnding on larval diet. Temperature, humidity and availability of food to the adult are also important.

Natural Diet

Loftin et al. (1921) found that adults from boll-reared larvae and held in the laboratory lived for 14.7 days with ample water and 7.6 days under dry conditions. They noted that males and females lived approximately the same amount of time. In field cages in Texas, Owen and Calhoun (1932) reported similar findings. Female moths that emerged before cotton was available lived 3.1 days and males 2.4 days under dry conditions. When the soil was moist females lived 8.8 days and males 6.7 days. They went on to show that when cotton fruiting began, the average longevity of females increased to 11.6 days and that of males to 9.9 days. When cotton was in full fruit the longevity increased still further to 15.1 days for females and 12.0 days for males.

Adkisson (1961) reared PBW larvae on short staple cotton squares and bolls with ample sucrose solution for the adults, at 26.7°C constant temperature in the laboratory. He found that females reared on squares lived 15.7 days and on bolls lived 15.5 days. However, this data included only females that laid eggs.

Hussien, Shazli, Sawaf and Zaazou (1962) found that cotton reared diapause females lived 21.1 days and males 20.5 days at 18°C and 70% relative humidity. When the temperature was increased to 22°C longevity decreased to 11.4 days and 9.7 days for females and males, respectively. Henneberry and Leal (1979) using a cotton-reared diapause strain of PBW found longevity varied from 12 to 42 days for males and 16 to 50 days for females at constant temperatures of 32.2°C and 15.6°C respectively.

In a study on humidity, Hussien et al. (1962) discovered that changing from 70% relative humidity to 90% relative humidity increased longevity significantly from 9.7 days for females and 14.1 days for males, to 15.2 days and 15.7 days respectively.

Abd-El-Fatah-Khalifa El-Shaarawy, Salem and El-Serwiy (1981) compared the longevity of PBW adults when the larvae were fed on 3 different natural food plants, okra pods, cotton bolls and kenaf fruits. Females lived significantly longer than males in all cases. Those fed on okra pods lived longest - 22.0 and 19.0 days for females and males. Those on kenaf fruits lived the shortest amount of time 19.6 and 15.2 days. The moths from the larvae fed on cotton bolls lived 20.6 and 17.8 days. The experiment was carried out in April and May, at 23°C to 30°C and 65% to 85% relative humidity.

Artificial Diet

Philipp and Watson (1971) found that the longevity of PBW's reared in the laboratory on lima bean diet and at a relative humidity of at least 50% varied with temperature. The longevities ranged from 14.2 days to 26.3 days at fluctuating temperatures of 19.5°C to 37°C and 14°C to 33°C for the females. The male longevities ranged from 15 days to 25.3 days at 32.5°C constant temperature and fluctuating 14°C to 33°C. Fye and McAda (1972) also reared artificial diet fed PBW's at different temperatures. They found that adult longevity was greatest, 22.1 days for males and 27.8 days for females, at a daily mean temperature of 20°C. They found longevity was shortest, 10.3 days for males and 11.7 days for females, at a mean daily temperature of 25°C. Henneberry and Leal (1979) also did a study which involved the effects of temperature on longevity of artificial diet reared PBW's. They found maximum longevities, for males of 43 days at 21.1°C and females of 44 days at 35°C Corresponding minima were of 18 and 17 days respectively at a constant 32.2°C.

Butler and Foster (1979) also worked on longevity of PBW adults at constant and fluctuating temperatures. They obtained figures one-half as large again as Graham et al. (1967) and Philipp and Watson (1971) and Fye and McAda

(1972), but they kept individual adults separately without allowing them to mate.

Sidhu, Simwat and Dhawan (1980) studied the longevity of the pink bollworm at different combinations of temperature (20, 25, 30, 35 and 40°C) and relative humidity (40, 60 and 80%). They found that longevity increased with humidity at 20°C from 6.1 days at 40% relative humidity to 22.7 days at 80% relative humidity. At 25°C longevity was 16.2 days at 40%, it increased to 18.4 days at 60% and decreased to 15.0 days at 80% relative humidity. at 30°C longevity decreases with increase in humidity from 18.6 days at 40% to 11.2 days at 80%. Longevity was greatly reduced at all relative humidities when at or above 35°C.

Preoviposition Period

An important aspect of the pink bollworm reproductive behavior is the preoviposition period. Though it has been shown that the adults are reproductively mature and do not mate the first night after emergence, eggs are seldom laid at that time.

Owen and Calhoun (1932) in studies under field conditions indicated that females never laid eggs before the second night and that one female laid her first eggs on the tenth night. They agreed with Lukefahr and Griffin

(1957) however, when they determined that peak ovipositon occurred on the third night after emergence.

Adkisson (1961) found differences in average preoviposition according to the larval diets. The longest was 5.4 days for square reared PBW and the shortest was 3.8 days for those reared on a wheat germ diet. Philipp and Watson (1971) fed larvae on lima bean diet and paired the adults at different temperatures. They found that average preoviposition ranged from 3.2 days at 19.5°C to 37°C fluctuating temperature to 7.6 days at 14.5° to 33°C fluctuating temperature.

Fecundity

Fecundity studies of the PBW can also be divided into two groups by considering the larval diet. Most of the studies have been done on PBW populations reared on artificial diets. A few studies have been done where the PBW population was reared on cotton fruiting forms in the field.

Natural Diet

Willcocks (1916) and Taylor (1936) found that large females may produce 350 to 500 eggs while small moths may produce as few as 200 to 250 eggs. Correspondingly Wellso and Adkisson (1962) found approximately 550 eggs in various stages of development in the ovaries of recently emerged moths.

Owen and Calhoun (1932) found that under field conditions, short staple cotton reared, long cycle adults (from diapause larvae) laid 105.9 eggs and short cycle (non diapause larvae) moths laid an average of 180.7 eggs. Only females laying fertile eggs were considered. Owen and Calhoun (1932) also mention that the greatest average number of eggs laid in any 12 hour period were deposited on the first night of oviposition and there was a gradual falling off in the rate of egg deposition for each succeeding night until oviposition ceased.

Lukefahr and Griffin (1956) fed adult moths a simulated cotton nectar, honey and molasses, and a honey-molasses-protein hydrolyzate mixture, to see if adult fecundity was affected. All three combinations of sugars significantly increased fecundity when compared with a distilled water control. The average number of eggs laid varied from 146 per female for the simulated cotton nectar to 117, 107 and 68 for the control.

In all the following studies adults were fed some form of sugar solution. Adkisson (1961) reared PBW larvae on natural and synthetic diets at 26.7°C in a darkened room. He found that artificial-diet reared moths laid significantly more eggs than moths reared on short staple cotton bolls which were significantly more fecund than moths reared on short staple cotton squares. This agreed with the findings of Taylor (1936) and Fenton and Owen (1953). Of the five different larval diets the females from the wheat germ diet laid 393.3 eggs, those on a 1% oil added, cottonseed meal diet laid 376.9 eggs, those on a 5% oil added, cottonseed meal diet laid 323.5 eggs, those reared on cotton bolls laid 210.7 eggs and on squares 119 eggs. Since different percentages of females mated, Adkisson compared only individuals that produced 16 or more eggs. Females laying less than 16 eggs appeared to be virgin. Adkisson (1961) also found that the greatest number of eggs were produced on the seventh night following caging, which disagrees with Owen and Calhoun (1932) who did not publish their numbers for comparison.

Hussien et al. (1962) studied the effects of temperature humidity and population density on the fecundity of PBW from resting (diapause) larvae removed from infested double seeds of cotton. At 22°C and 18°C the moths laid averages of 33.4 and 14.4 eggs, respectively. These numbers are considerably lower than those of any other researcher and may be due to both temperatures being below optimal ranges of the PBW, and/or the effects of overwintering.

Humidity studies performed by the same authors at 30°C and relative humidities of 90% and 70% gave average egg productions of 29.4 and 58.4 eggs respectively. Again the numbers were very low and there were a large percentage of females that laid no eggs.

Hussien et al. also looked at the effects of population on fecundity and found that 2 males to 1 female per cage gave higher egg lay than a 1:1 male:female ratio, - 58.4 to 16.8 eggs. Unlike other researchers, Hussien et al. considered all females whether they laid eggs or not. Recalculating their data to include only females that laid 16 or more eggs gives averages of 70.2 eggs at 22°C, 31.6 eggs at 18°C, 73.5 eggs at 90% relative humidity and 133.7 eggs at 70% relative humidity. Unfortunately all the sample sizes are now less than 10 females.

Henneberry and Leal (1979) studied the effects of temperature on fecundity of PBW's on both overwintered diapause larvae from cotton seed and artificial diet reared PBW. Maximum egg lay occureed at 26.7°C and was 197 eggs per female and 147 eggs per female for the artificial diet and cotton seed reared populations, with 82% and 77% viability, respectively. Maximum egg lay of 16 eggs and 2 eggs per female was recorded at 15.6°C with 17% and 21% viabilities. They also pointed out that females mated on the third, fourth and fifth days after emergence laid 137, 172 and 131 eggs each, significantly more than moths mated on the first and second nights after emergence (28 and 88 eggs per female). Percent hatches for the delayed mating females was reduced to 55%, 57% and 51% (third, fourth and fifth nights) and still further to 8% and 36% for those mated on the first or second night. Correspondingly, longevity of these delayed mating females was significantly longer at 20.2 days, 27.7 days and 21.4 days than those females mated on the first and second nights of 17.1 and 17.9 days. Adkisson (1961) found that females that lived the longest laid the most eggs. Perhaps the delay in mating was significant in increasing the longevity of adult females and thus their fecundity.

Abd-El-Fatah-Khalifa et al. (1981) determined the fecundity of females developed from diapause larvae fed on okra pods, cotton bolls and kenaf fruits. They found that moths fed on okra pods laid the highest number of eggs (52.4 eggs) followed by those fed on cotton bolls (45.0 eggs) and then kenaf fruits (27.0 eggs). The moths were reared at 23°C to 30°C and at 65% to 85% relative humidity.

Artificial Diet

Graham, et al. (1967) attempted to find the optimum temperature for mass rearing the adults of PBW, after the mass rearing techniques of Richmond and Ignoffo (1964) were developed. They achieved a maximum egg production of 280 eggs per female at 26.7°C with only 4% of the females not producing. Fecundity dropped off on either side of 26.7°C to 22.0 eggs at 16°C and less than 1 egg per female at 41°C. They did not present any viability data for the eggs produced.

Philipp and Watson (1971) and Fye and McAda (1972) also measured the fecundities of female PBW's at different temperatures. Philipp and Watson (1971) found that maximal egg lay occurred at 30°C constant temperature (159 eggs per female) and at fluctuating temperatures between 28°C and 37°C (163 eggs per female). Fye and McAda (1972) however determined maximum egg lay to occur at a mean daily temperature of 20°C (169 eggs/female). Minimum egg lay occurred at 33°C of 21 eggs/female and 16.3 eggs per female in the 1971 and 1972 studies respectively.

Because extremely high temperatures occur in cotton fields in the summer months in Arizona, several researchers have studied the effects of high temperatures for various amounts of time on various stages of the PBW. Fye and Poole (1971) experimented by exposing PBW larvae to high temperatures of 35°C and 40°C for different time intervals. They found that exposure to 35°C for 16 hours daily reduced fecundity and fertility about 50%.

Henneberry, Flint and Bariola (1977) showed that constant temperatures between 32-35°C or higher and fluctuating temperatures with a mean of 32°C or higher generally resulted in reproductive failure of PBW. They determined that males exposed to these high temperatures did not mate and/or transfer sperm effectively. They also showed that females exposed to high temperatures had reduced egg lay and egg viability.

In a third study Henneberry and Clayton (1982) showed reduced reproduction in moths surviving brief larval exposure to high soil surface temperatures. They mated females exposed to 49°C with unexposed males and vice-versa. In all cases fecundity was reduced over unexposed females mated with unexposed males.

Henneberry and Clayton (1980) in studies of mating, reproduction and longevity of a laboratory reared, and a native strain of PBWs, found that the native strain of boll reared PBW from Phoenix laid 44 eggs per female. The yield from an artificial diet reared population treated similarly was 123 eggs per female. The percent hatches for the two populations were 74% and 57% respectively. The moths for this study were kept, 5 pairs to a cage, at 26.7°C constant temperature and 14:10 light dark cycle. These results agree with Adkisson (1961) and Henneberry and Leal (1979) who both found that artificial diet reared PBW laid more eggs than natural diet reared populations, but the actual number of eggs laid was much lower than those found by the other researchers.

When the literature cited above had been considered it was determined that the optimal conditions for maximal survival, longevity and fecundity of the pink bollworm adults were a constant temperature of 26.7°C, at least 50% relative humidity, and a sucrose solution available as a food source for the adults. Consequently these were the conditions used to maintain the adult population throughout the course of these studies. A 14:10 light dark cycle was also used to both simulate daylight under field conditions and to prevent diapause.

Diapause

Diapause is a delay in development which, although its effect is usually to facilitate survival during unfavorable periods, is not immediately referable to the adverse environmental conditions (Chapman 1971). Diapause can occur at any stage in the life cycle, but in any one species occurs at a specific stage. In the PBW it occurs at the last larval instar. Diapause may be obligatory, or as in the PBW in temperature cotton growing regions of the world, faculative (Adkisson 1961a). Diapause PBW larvae are heavier, have slower rate of heart beat and consume oxygen at about one-sixth the rate of non diapause individuals. In addition the diapause condition is accompanied by an atrophy of the male gonads and an increase in fat content of both sexes (Adkisson, Bell and Wellso 1963). Most of the resting (diapause) larvae pass the winter in the cotton bolls in which they developed. Some of them, however, on completion of feeding cut their way out of the bolls and spin cocoons in the soil or in the debris on the soil surface (Chapman and Hughes 1941).

Bull and Adkisson (1962) concluded that several factors; photoperiod, temperature and oil content of the bolls, all combined to induce diapause. Diapause occurs in late September and October because that is when the critical photoperiod is reached, the temperatures cool down and the bolls dry out increasing their oil content. Adkisson et al. (1963) and Lukefahr, Noble and Martin (1964) showed that photoperiod was the most important factor, and that the critical photoperiod was 13 hours.

After the winter, photoperiod temperature and moisture are the most important factors terminating diapause. Wellso and Adkisson (1964) showed that laboratory reared diapause pink bollworms, terminated diapause more rapidly under 14 or 16 hour photoperiods than larvae under 8 or 12 hour photoperiods. They also showed that larvae terminated diapause and pupated more rapidly at 100% relative humidity than those held at 65% relative humidity. Watson

Lindsey and Slosser (1973) simulated field conditions occurring at Mesa, Arizona for the midmonth periods of March, April and May in the laboratory. They found that conditions occurring in mid-April were most favorable for pupation and survival.

MATERIALS AND METHODS

Origin and Collection of the Pink Bollworm Populations

Six populations of PBWs were compared in this study. Five populations were field collected from cotton at differing stages of physiological development. The sixth population was laboratory-reared, on an artificial wheat germ diet at the USDA Western Cotton Research Laboratory in Phoenix, Maricopa County, Arizona. The field populations were collected for two different types of cotton: American Upland (*Gossypium hirsutum* L.) a short staple cotton, and American Egyptian (Pima S-5) (*G. barbadense* L.) a long staple cotton. Populations were collected at two different times of the year in order to obtain PBW which were reared in squares and bolls. Early season larvae were collected from cotton blooms; late season larvae were collected from susceptible cotton bolls.

The first population was collected from short staple blooms in San Luis, Yuma Co., Arizona, on June 9, 1982. The second population was collected from long staple blooms in Safford, Graham County, Arizona, on July 27, 1982. The third population was collected from short staple cotton at the University of Arizona Experimental Station in Yuma, Yuma

Co., Arizona, on September 2, 1982. The fourth population was collected from susceptible bolls in a long staple cotton field in Safford, Graham Co., Arizona on September 17, 1982. Due to low infestations and disease, supplementary collections of Safford long staple bolls were made on September 24, 1982 and October 2, 1982. The fifth population was collected from susceptible bolls in a long staple cotton field in Coolidge, Pinal Co., Arizona, on September 13, 1982. All the blooms and susceptible bolls were immediately transported, either in insulated ice chests or air-conditioned vehicles to the laboratory on the University of Arizona Campus, Tucson, Pima Co., Arizona.

Maintaining Pink Bollworm Populations in the Laboratory

As soon as a population was brought into the laboratory the blooms of susceptible bolls were placed, one deep, on metal racks in five temperature cabinets maintained at 26.7°C $\pm$ 1°C and constant relative humidity. The plant material was moistened daily with distilled water. Underneath each metal rack was a wire mesh container to catch the emerging pink bollworms. The mesh containers were lined with moistened paper towels to provide a substrate on which the larvae could pupate, or incorporate into their cocoons. Many of the larvae on the blooms did not drop onto the paper towels but pupated within the dying blooms. Blooms were therefore dissected and the fourth instar larvae and/or pupae were placed directly into petri dishes in the temperature cabinets. The cabinets were kept at a constant temperature of 26.7°C $\pm$ 1°C and a constant 14:10 light/dark cycle. Humidity was maintained at a constant level by the presence of free water in a glass container. The emergent larvae from the bolls were collected daily and placed in plastic petri dishes, fifteen larvae per dish. Three layers of moistened paper toweling were also placed in each petri dish to provide a substrate for the larvae to build their cocoons.

When the larvae in the petri dishes pupated the pupae were placed in individual plastic creamer cups (14.8 mls) with cardboard tops. Also placed in each cup were 3 squares (2 × 2 cms) of moistened paper toweling, to cushion the pupa and prevent it from drying out. The date of pupation was marked on the top of each container. The creamer cups were then placed sideways in large egg cartons to facilitate handling. The egg cartons were stored in the temperature cabinets and the creamer cups checked daily for adult emergence.

When adults emerged they were paired and placed in paper ice cream containers (no. 16SN3 Lily^{R} Nestite^{R}) with clear plastic lids and a wax coating on the inside to prevent oviposition on the container walls. The container had two opposing wire mesh windows (5 × 5 cm) which were covered with squares of black velour paper (4 × 4 cm) held in place by a rubber band. The velour paper squares served as oviposition sites and were changed daily. Placed in each paper container was a black vial cap (1 cm deep 2 cm in diameter). The cap was packed with a piece of cotton dowel which was soaked with a 1 molar sucrose solution. This provided a food source for the adults on which they could stand, and yet the cotton dowel prevented them from drowning in the solution. These vial caps were filled each day and the cotton replaced every 4 or 5 days to prevent buildup of a sticky residue which might trap the adults.

Adults were checked daily for mortality and fecundity. The black velour squares were removed and stored in the same cabinets for five days to determine viability. By the fifth day the eggs had either hatched or the black head capsule of the young larvae would be seen inside the egg, using a binocular microscope.

Sexing Pink Bollworm

When the larvae emerged from the blooms or susceptible bolls they were quickly sexed by observing the dark dorsal testes visible through the cuticle in the males (Leppla 1972; Bartlett and Lewis 1978). Since this method proved to be less than 100% accurate the pupae were also sexed (Butt and Cantu 1962). This method proved to be inaccurate as well, so adults were sexed by examining moths externally for claspers or ovipositor. Carbon dioxide was used to anaesthetise adults for the examination. Finally as each individual died, it was removed from the container and sexed by counting the frenulae (females triple, males single, Busk 1917).

Disease and Sterile Technique

Larvae brought into the laboratory from Yuma County short staple bolls and Graham County long staple bolls appeared to suffer from a disease. The disease attacked males and females equally, and was characterized by a general sluggish behavior of the larvae, a red and puffy appearance and finally by the larvae turning black and drying up. To curtail this disease, all the equipment and cabinets used in this study were sterilized with a solution of 46 ml of 5.25% sodium hypochlorite solution in one liter of distilled water (Vail, Henneberry, Kishaba and Arakawa 1968). This disease killed 330 larvae (50%) from the Yuma County collected bolls population and 200 larvae (30%) from the combined Graham County, boll reared population. Affected larvae were sent to the Western Cotten Research Laboratory in Phoenix, Arizona where researchers were unable to identify the disease.

Delayed Pairings

For each population of pink bollworms, individual males and females were paired for mating either on the day they emerged or the next day, except for the Graham County long staple, boll reared, population. Because more females emerged earlier than males, some females were left unmated for up to six days. To ascertain the effects of remaining unmated for six days, a population of pink bollworms reared on a wheat germ diet at the Western Cotton Research Laboratory in Phoenix, Maricopa County, Arizona, was tested. Forty-nine females were held in cages for six days and then paired with fresh males. Forty-nine pairs were also caged on the day of their emergence to act as a control.

Statistical Analysis

Analysis of the data from this study required several statistical tests. Chi square 2 × 2 contingency tables were computed by hand; the other tests were performed using computer programs from the Statistical Library file of the University of Arizona Computer Center. The computations were performed on a Control Data Corporation Cyber 175 computer. The following is an outline of the statistics used: SPSS subprogram breakdown was used to compute the sum, mean, standard deviation and variance for the parameters; pupal duration, longevity, total eggs laid and viable eggs laid; for each population, by sex. Analysis of variance was then carried out using SPSS Anova Option 9 for each parameter. Parameter means for each population were then separated with SPSS oneway Option 8 using the LSD test at the .05 level.

An analysis was made not only of the number of eggs laid, as described above, but also of how many females laid eggs in each population. This was important if egg production of the whole population was to be determined. Since both fruiting form and cotton variety, were considered as factors possibly affecting whether eggs were laid or not, chi-square 2 × 2 contingency tables that were only significant for 1 interaction at a time, were considered insufficient. Consequently a multiway frequency table was produced and analyzed by a log linear model using BMDP program F4. The advantage of the log linear model was that it considered the relationship of both factors as they affected whether eggs were laid or not. The log linear model was used to generate the 2-way interaction; eggs laid by fruiting form by cotton variety, the significance of which was then analyzed using the chi-square test.

When the data were analyzed it was observed that holding females for six days without pairing reduced fecundity, and that the fecundity of the populations of long staple, boll reared pink bollworms from Graham Co. paired at once, and the Pinal Co. population, were similar. It was decided that if the fecundity of the Pinal Co. and Graham Co. populations were the same, they should be combined and treated as one. To do this not only the number of eggs laid must be the same but also the frequency with which they are laid. The LSD test indicated that the numbers of eggs laid were the same. A linear regression model with first order auto-correlated errors would determine if the frequencies were the same (Neter and Wasserman 1967; Conover 1980). The Prais Winsten was rated the best of these models by Park and Mitchel (1980) and it was employed in the form of a Fortran 5 program written by Brian Maurer of the University of Arizona Statistical Support Unit.

RESULTS AND DISCUSSION

Definitions of Populations

Blooms and bolls of long staple and short staple cotton were collected on the dates indicated in Table 1. The numbers of mature larvae that either cut out naturally, or were dissected out as indicated above, and their sex ratios male/female, are also shown. None of the sex ratios for the individual populations were significantly different from 1:1. The sex ratio of all the populations together was 915/952. Monitoring pink bollworms using sex pheromone traps is a common practice in Arizona, and since the PBW sex pheromone only attracts male moths, an accurate knowledge of the sex ratio is essential to determine the corresponding number of females in the field, and thus the reproductive potential of the population.

Pupal Duration

Data on the pupal duration of the various populations of this study are presented in Table 2. The overall average pupal duration for the 723 cotton reared PBW pupae, was 9.6 days with a range of 5 to 19 days. This agrees with the findings of Hunter (1926) of 6 to 20 days, but is a little

Table 1. Summary of pink bollworm collection data and sex ratio.

Date	Food Source	Location	No. Larvae[1] ♀	O	Sex Ratio O/♀
6/9/82	Blooms	Yuma County	172	210	1.22
7/27/82	Blooms	Graham County	155	142	0.92
9/2/82	Bolls	Yuma County	272[2]	279[2]	1.02
9/13/82	Bolls	Pinal County	101	80	0.79
9/17/82-10/2/82	Bolls	Graham County	252[2]	204[2]	0.81

[1]Includes larvae that were damaged, didn't pupate, etc.

[2]Includes larvae that were diseased.

Table 2. Pupal durations of pink bollworm populations.

Population	Sex	Number Ob-served	Average Pupal Dura-tion (Days)	Standard Deviation	Range	LSD[a] Test (0.05 Level)
Yuma Co. short, staple, blooms	M	60	9.2	1.8	6-11	c
	F	15	8.9	1.7	6-12	cde
Graham Co. long staple blooms	M	106	8.5	0.9	7-12	cd
	F	106	8.3	0.1	6-11	c
Yuma Co. short, staple, bolls	M	92	9.7	1.3	7-12	cf
	F	92	9.1	1.2	5-12	c
Graham Co. long staple, bolls	M	84	10.7	2.7	5-19	g
	F	84	11.3	2.1	8-17	h
Graham Co. long staple, bolls delayed pairing	M	79	9.0	1.9	5-19	de
	F	27	10.3	1.4	8-17	fg
Graham Co. long staple, bolls paired at once	M	57	11.5	2.6	7-13	h
	F	57	11.8	2.3	8-14	h
Pinal Co. long staple, bolls	M	42	10.7	1.9	8-19	g
	F	42	9.9	2.5	5-13	f
Maricopa Co. wheat germ diet paired at once	M	49	5.9	1.0	3-7	a
	F	49	5.3	1.1	3-7	a
Maricopa Co. wheat germ diet delayed pairing	M	49	7.5	0.7	7-9	b
	F	49	4.9	0.3	4-5	a
Entire[b] population	-	723	9.6	----	5-19	---

[a]Populations followed by the same letter are not significantly different.

[b]Entire population refers to all cotton reared pupae.

shorter than the data of Busk (1917) who reported a range of 10 to 20 days.

The male pupae took longer to develop than the female pupae in all the populations except for the Graham Co. long staple boll reared population. This disagreed with Loftin et al. (1921) who noted that boll reared male pupae from diapause larvae developed more slowly, 10.5 days to only 10 days, than female pupae. Fye and McAda (1972) found that artifical diet reared male pupae also developed more slowly (8.1 days) than female pupae (7.7 days). The significance of females emerging one half day sooner than males is obscure in an insect population where emergence occurs over several days. Perhaps this allows females to feed before mating. The difference between male and female pupal durations was never more than one whole day except in the delayed pairing population.

The pupal durations of the Graham Co. long staple boll reared population and the Pinal Co. long staple boll reared population were longer than the pupal durations of any other populations and significantly longer, at the 0.05 level of the LSD test, than the Graham Co. long staple bloom reared population. There was however no significant difference between the Yuma Co. short staple, bloom and boll-reared populations. Except for the long staple boll reared populations there appears to be no differences in pupal duration between PBW reared on long and short staple cottons.

Owen and Calhoun (1932) indicated that pupae reared from short staple cotton bolls developed more slowly than pupae from short staple cotton squares. This is consistent with the data from the present study but the difference is not significant.

The pupal durations for the artificial diet reared populations in this study are shorter than those of the cotton reared populations. This is more likely an artifact of the experiment than a true reflection of the artificial diet. A large number of pupae were acquired for the delayed pairing studies, and the first ones to eclose were used. If the entire population had been averaged the pupal durations would probably have been comparable to those of the cotton-reared populations. Guerra and Ouye (1968), Philipp and Watson (1971), and Fye and McAda (1972) all achieved pupal durations from artificial diet reared larvae, similar to the pupal durations of the cotton reared populations of this study.

Adult Longevity

Data on the longevity of adult PBW moths of the various populations of this study are presented in Table 3. The average for the entire population of 894 cotton reared

Table 3. Longevities of pink bollworm populations.

Population	Sex	Number Ob- served	Average Long- evity (Days)	Standard Deviation	Range	LSD[a] Test (0.05 Level)
Yuma Co. short staple, blooms	M	123	16.0	4.6	2-27	abc
	F	123	15.9	3.8	8-29	abc
Graham Co. long staple blooms	M	106	15.8	3.3	7-23	abc
	F	106	14.9	3.3	1-25	a
Yuma Co. short staple, bolls	M	92	18.4	4.8	8-80	e
	F	92	18.7	4.7	9-30	e
Graham Co. long staple, bolls	M	84	16.9	5.6	6-40	cd
	F	84	17.9	5.5	7-30	de
Graham Co. long staple, bolls delayed pairing	M	27	17.6	7.2	6-82	cde
	F	27	18.1	5.6	7-30	de
Graham Co. long staple, bolls paired at once	M	57	16.6	4.7	7-40	bcd
	F	57	17.8	5.5	8-27	de
Pinal Co. long staple, bolls	M	42	18.9	7.2	6-39	e
	F	42	18.9	5.2	6-27	e
Maricopa Co. wheat germ diet paired at once	M	49	19.5	5.9	9-33	e
	F	49	18.4	4.1	9-29	de
Maricopa Co. wheat germ diet delayed pairing	M	49	14.9	5.7	5-26	ab
	F	49	18.9	3.9	11-26	e
Entire[b] Population	-	894	16.9	----	1-40	---

[a]Populations followed by the same letter are not significantly different.

[b]Entire population refers to cotton reared insects.

adults was 16.9 days ranging from 1 to 40 days. This is slightly longer than the 10 to 14 days reported by Noble (1969).

The longevities of moths reared from boll fed larvae are significantly longer at the 0.05 level of the LSD test, (18.4 and 16.9 days for the males and 18.7 and 17.9 days for the females), than the longevities of moths from bloom fed larvae (16.0 and 15.8 for the males and 15.9 and 14.9 days for the females). This was true of both the Yuma Co. short staple and the Graham Co. long staple reared populations. The population from Pinal Co. was boll-reared, and was not significantly different from the other boll-reared populations (18.9 days for the males and 18.9 days for the females). The average difference in longevities between short and long staple reared populations was 2.4 days. Owen and Calhoun (1932) stated that the life cycle, oviposition to adult, for square reared populations was 26.6 days, and for boll reared larvae 32.9 days. They did not indicate, however, whether this boll dependent increase in longevity also occurred in the adult. Contrary to the present study, Adkisson (1961) did not observe any differences in adult longevity for short staple, square-(15.5 days) or boll-(15.4 days) reared populations. He did find however that the adults that lived the longest laid the most eggs, and that short staple boll reared moths laid significantly more eggs than square reared adults.

In Table 4, the longevities of females that laid at least one egg and those that laid viable eggs are compared with the longevity of all females. In almost all cases the longevity of females that laid eggs was equal to, or longer than that of all females.

Unlike the pupal duration, there was no significant difference in adult longevity between long staple and short staple cotton reared populations. The Graham Co. boll reared population in fact had a shorter longevity than the Yuma Co. boll reared population. Conversely the Pinal Co. boll reared population longevity was longer.

The longevities of moths from artificial diet reared larvae were very similar, except for the males used in the delayed pairing experiment, which were significantly shorter lived, at the 0.05 level of the LSD test. Perhaps the older female damages the newly eclosed male during mating. Henneberry and Leal (1979) found that females paired with younger males had shorter longevities than females paired with older males. They did not include longevity data for males however. Except for the delayed pairing males, the longevities of the artificial diet reared moths in this study corresponded favorably with those obtained by Fye and

Table 4. Comparison of the longevity of female pink bollworm adults with longevity of females that laid eggs and longevity of females that laid viable eggs.

	Yuma Co. Short Staple Blooms	Graham Co. Long Staple Blooms	Yuma Co. Short Staple Bolls	Graham Co. Long Staple Bolls	Graham Co. Long Staple Bolls Delayed Pairing	Graham Co. Long Staple Bolls Paired at Once	Pinal Co. Long Staple Bolls	Maricopa Co. Wheat Germ Diet Paired at Once	Maricopa Co. Wheat Germ Diet Delayed Pairing
Female longevity[1]	15.9	14.9	18.7	17.9	18.1	17.8	18.9	18.4	18.9
Number sampled	123	106	92	84	27	57	42	49	49
Female longevity[1] females that laid at least 1 egg	16.5	14.8	19.4	18.2	16	18.8	19.3	18.4	19.5
Number sampled	75	76	55	31	7	24	23	47	45
Female longevity[1] females that laid at least 1 egg	16.3	14.8	19.2	18.4	24	17.8	16.7	17.9	19.0
Number sampled	60	63	48	10	1	9	10	42	37

[1]Days.

McAda (1972) at a mean daily temperature of 30°C. Philipp and Watson (1971), however, achieved a male longevity of 23.9 days at 30°C, nearly 4 days longer than the data presented here or the work of Fye and McAda (1972), and 6 days longer than the females in the same study. This large variability is not easily explained unless the conditions under which the moths had been reared in the laboratory had selected for these male longevities.

Preoviposition Period and Peak Egg Laying

Preoviposition times and peak egg laying periods are shown for both females that laid eggs, and females that laid viable eggs (Table 5). Preoviposition period was determined as the number of nights following emergence before eggs were laid. Peak egg laying was the day that most eggs were laid after the female eclosed.

Preoviposition periods were longer in the boll reared populations than in the bloom reared populations which in turn were longer than that of the artificial diet reared population paired at once. The absolute length of the preoviposition period for the boll reared populations was much longer than anything found by Adkisson (1961) or Philipp and Watson (1971). Recalculating the preoviposition periods using only the data from females that laid viable eggs,

Table 5. Preoviposition periods for pink bollworms, and day of peak egg lay.

Population	Days after Emergence to Peak Egg Lay	Number of Moths That Laid Eggs	Average Preoviposition Period (Days)	Range	Number of Moths That Laid Viable Eggs	Average Preoviposition Period (Days)	Range
Yuma Co. short staple, blooms	5	75	6.0	2-16	60	5.0	2-16
Graham Co. long staple, blooms	5	76	4.9	2-14	63	4.2	2-10
Yuma Co. short staple, bolls	7	55	7.3	2-20	48	6.8	2-20
Graham Co. long staple, bolls	11	31	9.2	1-21	10	6.5	1-14
Graham Co. long staple, bolls delayed pairing	15	7	9.7	5-14	1	14.0	----
Graham Co. long staple, bolls paired at once	11	24	9.0	1-21	9	5.6	1-10
Pinal Co. long staple, bolls	5	23	8.6	2-17	10	6.0	3-13

Table 5.--Continued

Population	Days after Emergence to Peak Egg Lay	Number of Moths That Laid Eggs	Average Preovi- position Period (Days)	Range	Number of Moths That Laid Viable Eggs	Average Preovi- position Period (Days)	Range
Maricopa Co. wheat germ diet paired at once	4	47	2.6	1-7	42	2.4	1-7
Maricopa Co. wheat germ diet delayed pairing	8	45	8.1	6-21	37	7.1	6-11

preoviposition periods of 5.6 days were found for the Graham Co. boll reared population, and 6 days for the Pinal Co. population. These numbers, though larger than those for diet reared populations, are closer to those reported by other researchers.

In the delayed paring study the preoviposition period of the population held 6 days, was almost exactly 6 days longer than that of the population paired at once. That the artificial diet reared PBW's had much shorter preoviposition times, and narrower ranges was not surprising. Reared for generations under constant conditions, PBW's that favor those conditions will be selected for, and gradually the natural variability of the population will be reduced.

Peak egg lay for both bloom populations was on the fifth day 2 days sooner than reported by Adkisson (1961). In the boll reared population it ranged from day 5 for Pinal Co. to day 11 for Graham Co. compared to day 6 in Adkisson's work.

In the artificial diet study, egg laying of the females paired at once peaked on day 4 whereas those held 6 days peaked on day 8, 4 days later. It is possible that the females held 6 days were "more ready" to lay their eggs than females paired normally. Further evidence of this "readiness" is that many of the females, held 6 days, laid some nonviable eggs only 1 day after pairing, and then a regular complement of viable eggs on the second night. Since lepidopterous species emerge as adults with their full complement of eggs already determined, and these females had 6 days to feed without laying any of their developing eggs, by the time they mated they were literally bursting with developed eggs, hence the early egg laying peak.

Mating

Since females were not dissected, nor the number of spermatophores in the bursa copulatrix counted, the only index of successful mating in this study was the number of females that laid viable eggs. The data for the different populations can be found in Table 6. Of the 447 cotton reared moths in these studies only 260 laid eggs and of those 223 or 49.8% of the total, laid viable eggs. The artificial diet reared population paired at once, however, had 85.7% of its members produce viable eggs.

The reason for this disparity in mating success could be due to the wider gene pool of the field population. In a field reared population at least some members have to be able to mate under the different conditions that sometimes occur, to ensure the populations survival. The artificial diet reared strain has been bred for many

Table 6. Viable egg production of pink bollworm populations.

Population	Number of Females That Laid Viable Eggs	Average No. of Eggs of Females That Laid Viable Eggs	% Females That Laid Viable Eggs	Total Eggs	% Viable
Yuma Co. short staple, blooms	60	137.6	48.7	9,040	91.3
Graham Co. long staple, blooms	63	123.1	59.4	8,227	94.3
Yuma Co. short staple, bolls	48	121.3	52.1	6,262	92.9
Graham Co. long staple, bolls	10	122	11.9	1,924	63.4
Graham Co. long staple, bolls delayed pairng	1	175	37.7	435	40.2
Graham Co. long staple, bolls paired at once	9	116	15.7	1,489	70.2
Pinal Co. long staple, bolls	10	115.3	23.8	1,518	75.9

Table 6.--Continued

Population	Number of Females That Laid Viable Eggs	Average No. of Eggs of Females That Laid Viable Eggs	% Females That Laid Viable Eggs	Total Eggs	% Viable
Maricopa Co. wheat germ diet paired at once	42	260.1	85.7	12,562	86.9
Maricopa Co. wheat germ diet delayed pairing	37	168.1	75.5	7,104	87.5
Graham Co. long staple, bolls paired at once and Pinal Co. long staple, bolls	19	115.7	19.2	3,007	73
Entire Population[a]	223	108.5	49.8	26,971	89.7

[a]Refers to all insects fed on cotton.

generations under the exact same conditions in a laboratory. Because of this possible loss in genetic variability, interpretation of laboratory data needs to be considered cautiously when applying this information to field situations.

The long staple boll-reared populations have much lower percent mating success than the bloom reared or the Yuma Co. boll-reared population. This is possibly a reflection of the effects of breaking diapause too soon on the males of the long staple boll-reared populations. Adkisson et al. (1963) indicated that the gonads of the male PBW atrophy when they enter a state of diapause. Perhaps there was not enough time for the gonads to regrow in the shortened diapause period.

Log Linear Model

A log linear model, or multiway frequency table, was used to analyze the mating success, or egg laying response, of both total and viable reproduction. Since the sample sizes of both the Graham Co. boll-reared population paired at once and the Pinal Co. population were smaller than those of other populations, they were combined so that a relatively equal sample size comparison could be made. Before the populations were combined however, the egg laying frequencies were compared and found to not differ significantly at the 0.05 level of the students T-test, using the Prais Winsten auto-correlated linear regression model.

The populations analyzed were (1) the Yuma Co. short staple bloom-reared, (2) the Graham Co. long staple bloom-reared, (3) the Yuma Co. short staple boll-reared, (4) the combined Graham Co.-Pinal Co. long staple boll-reared population. The results and the corresponding statistics are shown in Table 7 for total egg production and Table 8 for viable egg production.

The results of the 2 way interaction were significant for the total egg laying, and highly significant for the viable egg laying analysis. Essentially this means that both fruiting form and cotton variety were responsible for the differences in whether eggs were laid or not. An ordinary chi-square analysis would have shown only the "eggs laid by fruiting forms" interaction to have been significant.

Fecundity

Data for the total egg production of female PBW's is presented in Table 9 and for viable egg production in Table 6. The average fecundity for all 447 cotton reared females is 60.3 eggs, 89% of which are viable. If only the 260 females (58.2%) that laid eggs are considered, they laid an average of 103.7 eggs each.

Table 7. Log linear model (multiway frequency table) of egg laying female pink bollworms analysis of three parameters.

		Short Staple	Long Staple	Total
Eggs Yes	Blooms	75	76	151
	Bolls	55	47	102
	Total	130	123	253
Eggs No	Blooms	48	30	78
	Bolls	42	53	95
	Total	90	83	173

Parameters: A = cotton variety 1 = short staple

2 = long staple

B = fruiting form 1 = blooms

2 = bolls

C = egg laying 1 = yes

2 = no

Selected Statistics:

Interaction	Chi Square	Probability
AB	0.90	0.3422
AC	0.07	0.7908
BC	8.87	0.0029**
ABC	4.72	0.0298*

*Significant at 0.05 level.

**Significant at 0.01 level.

Table 8. Log linear model (multiway frequency table) of viable egg laying female pink bollworms analysis of three parameters.

		Short Staple	Long Staple	Total
Eggs Yes	Blooms	60	63	123
	Bolls	48	19	67
	Total	108	82	190
Eggs No	Blooms	63	43	106
	Bolls	44	80	124
	Totals	107	123	230

Parameters: A = cotton variety 1 = short staple

2 = long staple

B = fruiting form 1 = blooms

2 = bolls

C = egg laying 1 = yes

2 = no

Selected Statistics:

Interaction	Chi Square	Probability
AB	0.57	0.4496
AC	3.73	0.0533
BC	14.01	0.0002**
ABC	22.20	0.0000**

**Significant at 0.01 level.

Table 9. Total fecundity of pink bollworm populations.

Population	Number Observed	Average No. Eggs/ Female Based on All Females	LSD Test 0.05 Level	Number of Females That Laid Eggs	Average No. Eggs/ Female of Females That Laid Eggs	% Females That Laid Eggs
Yuma Co. short staple, blooms	123	73.5	c	75	120.5	60.9
Graham Co. long staple, blooms	106	77.6	c	76	108.2	71.7
Yuma Co. short staple, bolls	92	68.1	bc	55	113.8	59.8
Graham Co. long staple, bolls	84	22.9	a	31	62.1	36.9
Graham Co. long staple, bolls delayed pairing	27	16.1	a	7	62.1	25.9
Graham Co. long staple, bolls paired at once	57	26.1	a	24	62.1	42
Pinal Co. long staple, bolls	42	36.1	ab	23	6	54.8

Table 9.--Continued

Population	Number Observed	Average No. Eggs/ Female Based on All Females	LSD Test 0.05 Level	Number of Females That Laid Eggs	Average No. Eggs/ Female of Females That Laid Eggs	% Females That Laid Eggs
Maricopa Co. wheat germ diet paired at once	49	256.4	d	47	267.3	95.9
Maricopa Co. wheat germ diet delayed pairing	49	145.	e	45	157.8	91.8
Entire Population[a]	447	60.3	----	260	103.7	58.2
Graham Co. long staple, bolls paired at once and Pinal Co. long staple, bolls	99	----	----	49	63.9	45.5

[a]Entire population refers to all cotton reared insects.

Comparing the individual populations there is no significant difference between the number of eggs laid by the short staple, bloom and boll reared moths, or the number of eggs laid by moths reared from blooms of either long or short staple cotton. All three populations, however, laid significantly more eggs than the Graham Co. long staple boll-reared populations. If the average number of viable eggs of females that laid viable eggs is considered, however, there was no difference in fecundity between moths reared on blooms or bolls of either long or short staple cotton. In other words viable egg production was the same for all females, and that only the number of females laying eggs in each population regulated the viable egg production of that population.

These results differ however from the findings of Taylor (1936), Fenton and Owen (1953) and Adkisson (1961) who all found that more eggs were laid by moths from boll-reared larvae than moths from square reared larvae.

The frequency of the populations reared on artificial diets was as expected significantly greater than the fecundity of the cotton reared populations. The fecundity of the population held for 6 days before pairing was significantly less at the 0.05 level than that of the population paired at once. The difference in the total number of eggs laid between the population held 6 days and the population paired at once was similar to the number of eggs laid in the first 6 days by the population paired at once. The differences were not observed in the Graham Co. boll reared population but this may have been due to the small sample size.

The numbers of eggs laid per day for each population is recorded in Table 10 and plotted versus days after emergence in Figures 1-6. The resulting curves are not bell shaped but skewed to the left indicating that most eggs are laid soon after mating, causing an early peak which then tapers off (Table 5). These results are of considerable importance in the management of the PBW. Since only the adult stage of the PBW is vulnerable to chemical control (larvae are protected in the bolls), chemical applications ideally should be made within 3 days of emergence for best suppression of populations.

Disease

The short staple boll-reared population collected from Yuma Co. was affected by a disease which caused approximately 50% mortality (equal numbers of males and females were killed). It was not possible to identify the disease and no symptoms appeared in larvae that pupated successfully. Nevertheless, sublethal, subsymptom level effects of the disease could have been responsible for the reduction

Table 10. Total eggs laid per day by female pink bollworms, by days afer emergence.

Days After Emergence	Yuma Co. Short Staple Blooms	Graham Co. Long Staple Blooms	Yuma Co. Short Staple Bolls	Graham Co. Long Staple Bolls	Graham Co. Long Staple Bolls Delayed Pairing	Graham Co. Long Staple Bolls Paired at Once	Pinal Co. Long Staple Bolls	Maricopa Co. Wheat Germ Diet Paired at Once	Maricopa Co. Wheat Germ Diet Delayed Pairing
1	---	---	---	--	--	--	--	---	---
2	---	---	---	15	15	--	2	136	---
3	372	517	215	24	24	--	2	1421	---
4	865	896	294	15	15	--	129	1880	---
5	1146	1139	535	9	9	--	190	1291	---
6	915	1021	405	94	76	18	72	1264	---
7	947	758	692	120	120	--	64	946	244
8	647	583	601	71	56	15	114	761	1246
9	857	673	572	50	50	--	112	856	745
10	812	765	580	63	63	--	82	873	658
11	674	924	418	293	293	--	102	815	825
12	544	310	456	151	126	25	122	472	856
13	462	295	475	84	57	27	65	512	779
14	413	173	263	175	134	41	83	349	461
15	153	121	195	218	116	102	47	303	451
16	72	21	171	160	96	64	114	196	265
17	100	14	108	168	117	51	59	173	178
18	29	---	50	84	31	53	13	103	118
19	19	---	26	34	23	11	28	77	84
20	13	17	22	34	6	28	49	80	86
21	---	---	20	35	37	--	26	44	58

Table 10.--Continued

Days After Emergence	Yuma Co. Short Staple Blooms	Graham Co. Long Staple Blooms	Yuma Co. Short Staple Bolls	Graham Co. Long Staple Bolls	Graham Co. Long Staple Bolls Delayed Pairing	Graham Co. Long Staple Bolls Paired at Once	Pinal Co. Long Staple Bolls	Maricopa Co. Wheat Germ Diet Paired at Once	Maricopa Co. Wheat Germ Diet Delayed Pairing
22	---	---	51	10	8	--	9	50	28
23	---	---	---	--	--	--	5	10	---
24	---	---	33	12	12	--	13	---	22
25	---	---	55	5	5	--	9	---	---
26	---	---	25	---	--	--	9	---	---
Totals	9040	8227	6262	1924	435	1489	1518	12662	7104

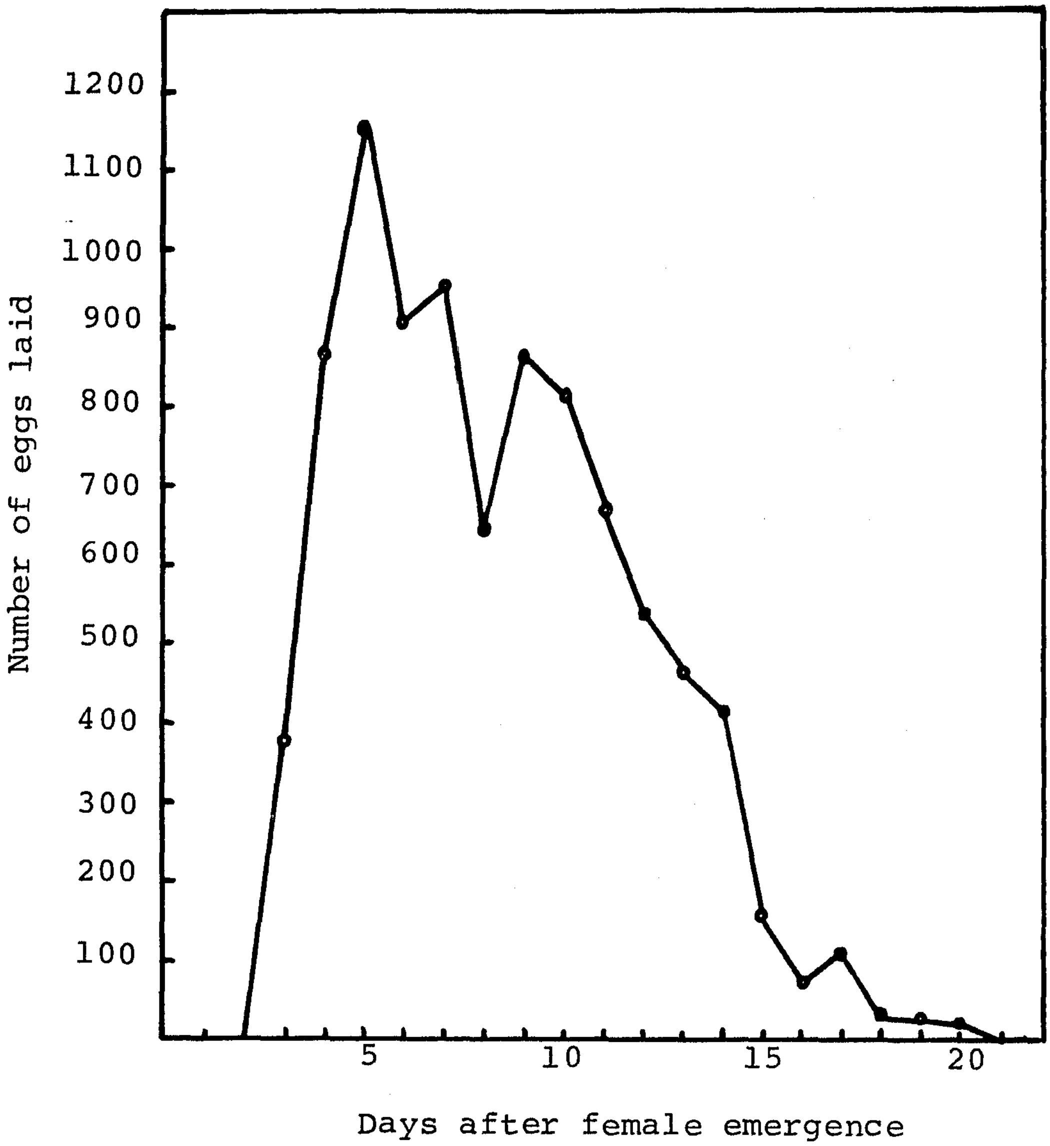

Figure 1. Egg production, by days after female emergence, for the Yuma Co. short staple bloom-reared population of PBW.

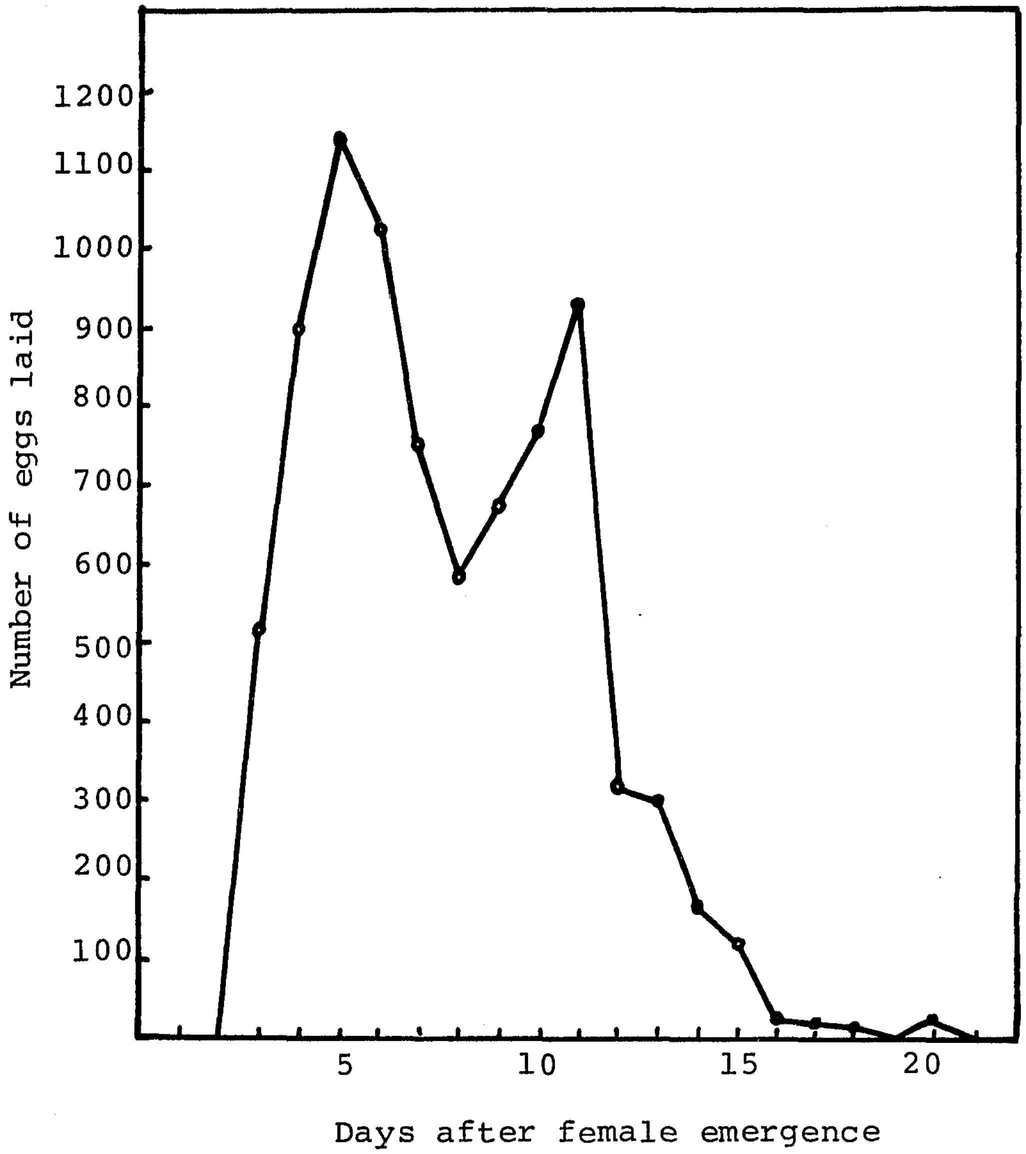

Figure 2. Egg production, by days after female emergence, for the Graham Co. long staple bloom-reared population of PBW.

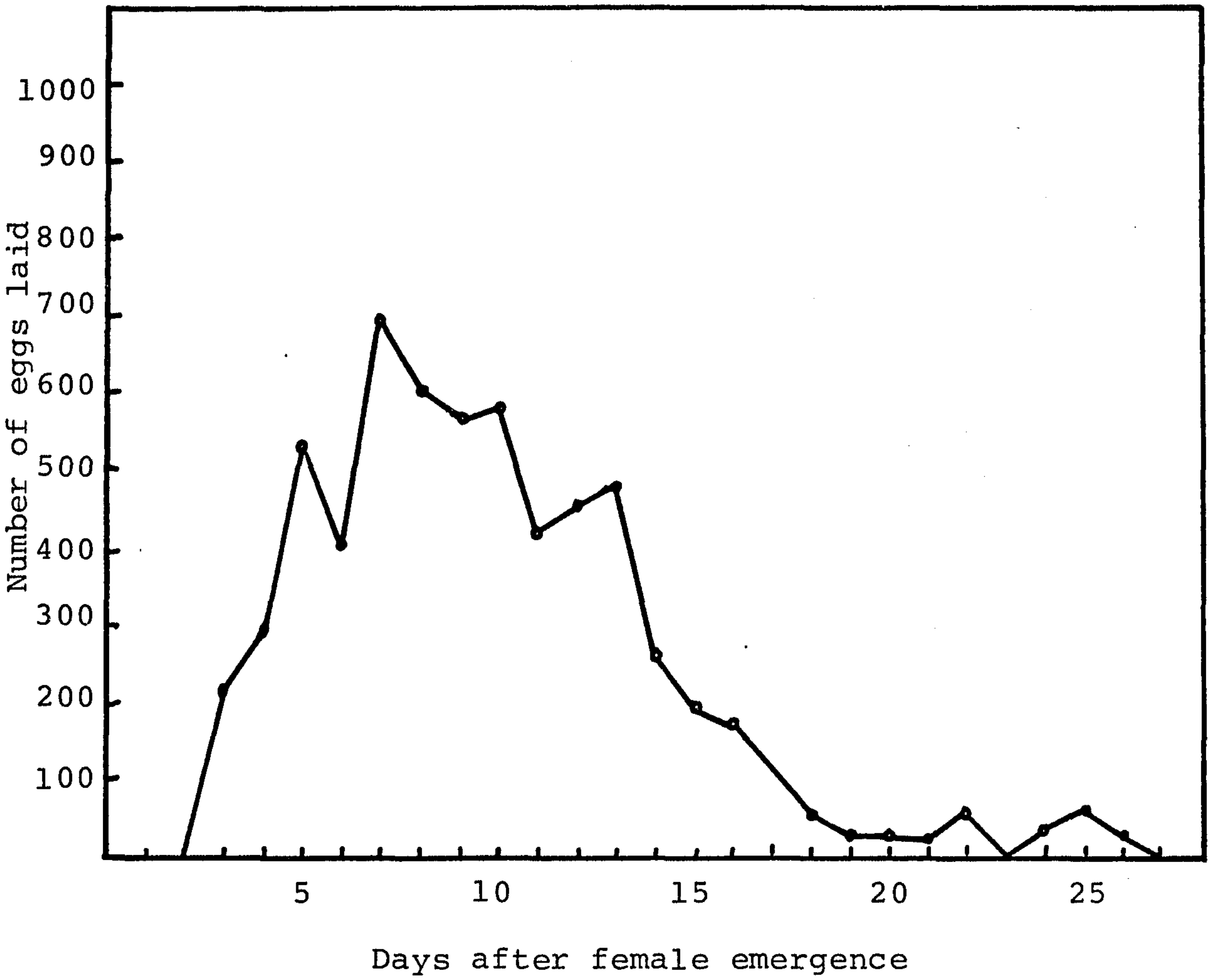

Figure 3. Egg production, by days after female emergence, for the Yuma Co. short staple boll-reared population of PBW.

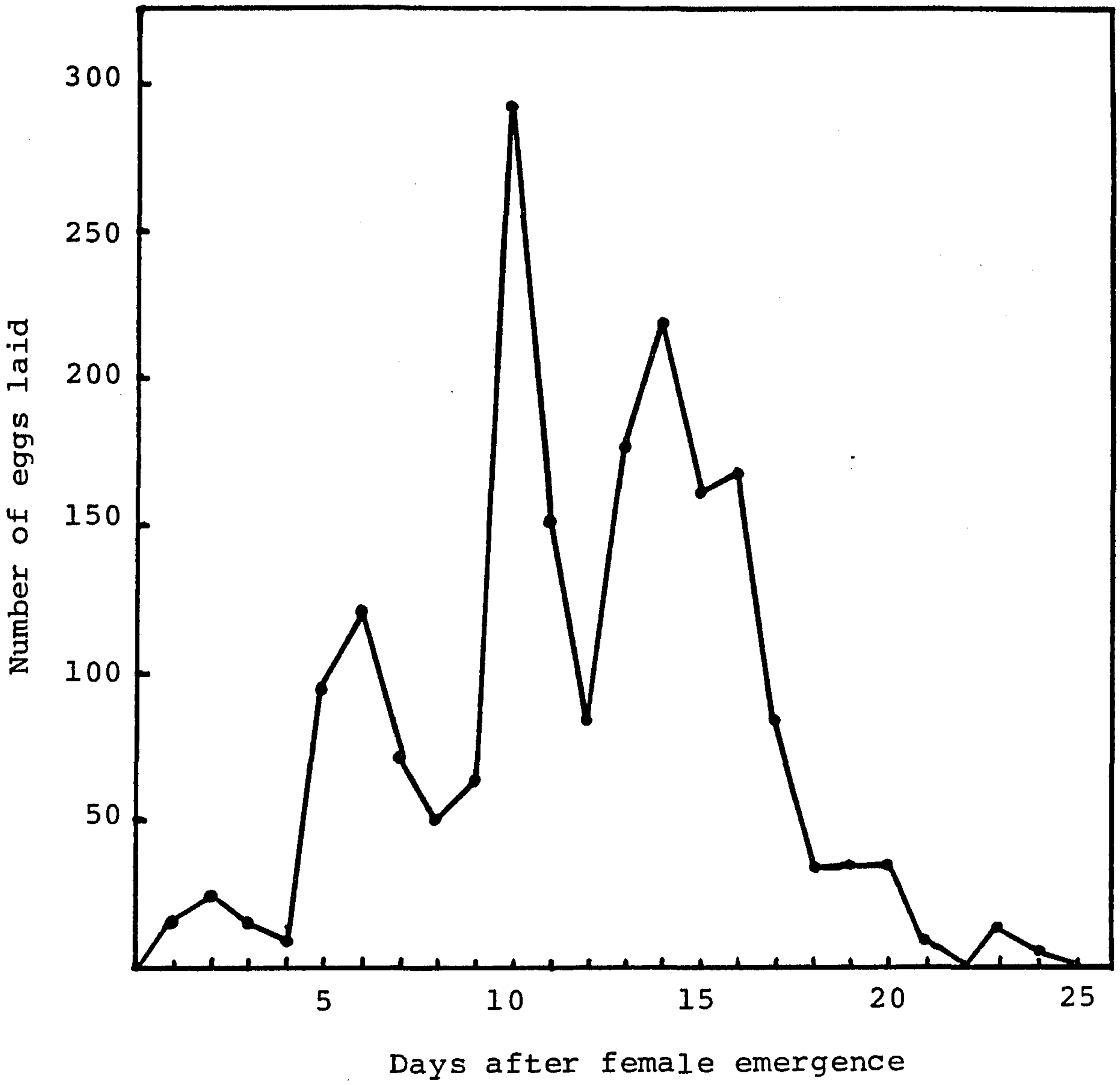

Figure 4. Egg production, by days after female emergence, for the combined Graham Co. long staple boll-reared populations of PBWs, including those females paired at once and those paired at 6 days.

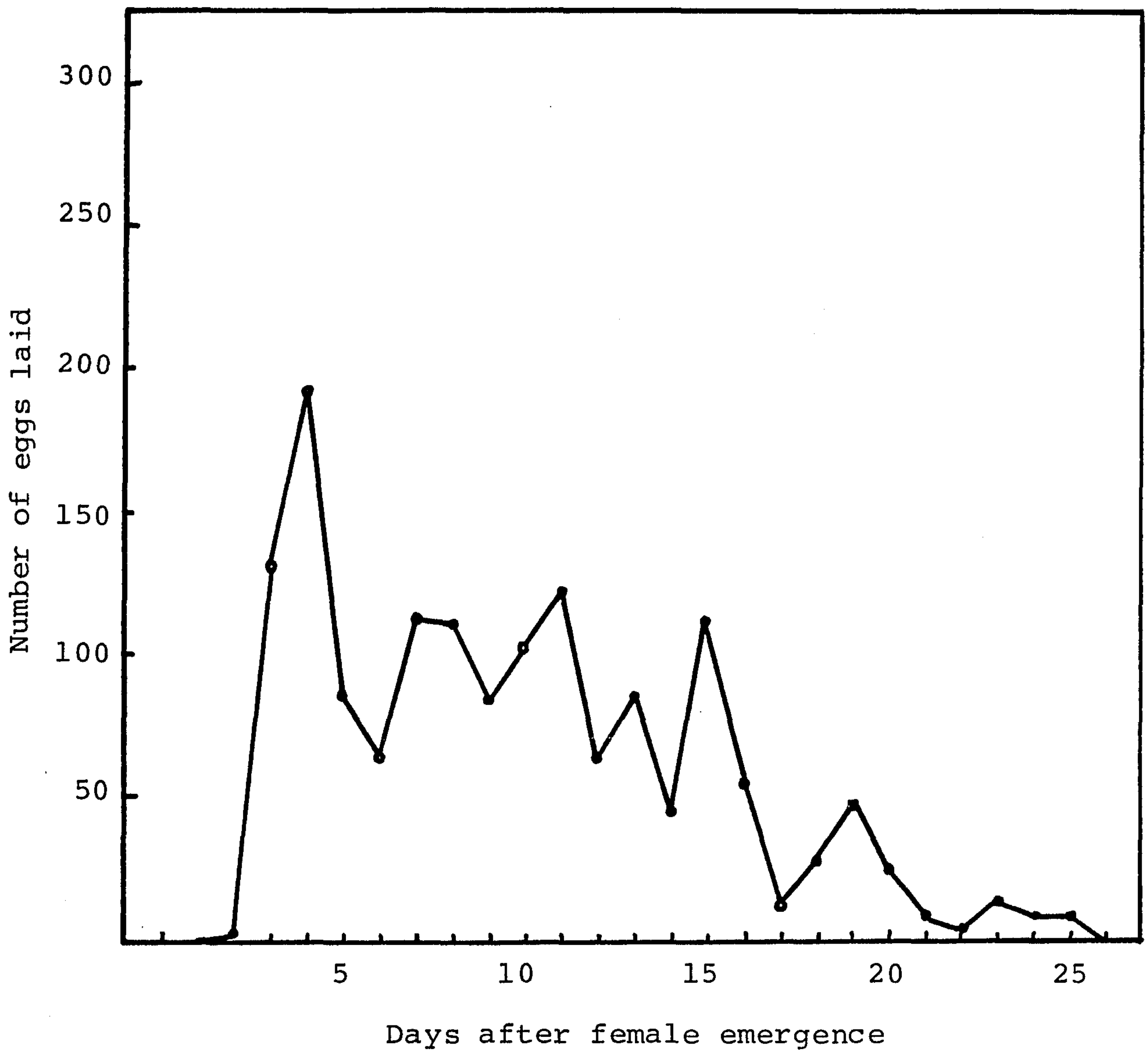

Figure 5. Egg production, by days after female emergence, for the Pinal Co. long staple boll-reared population of PBWs.

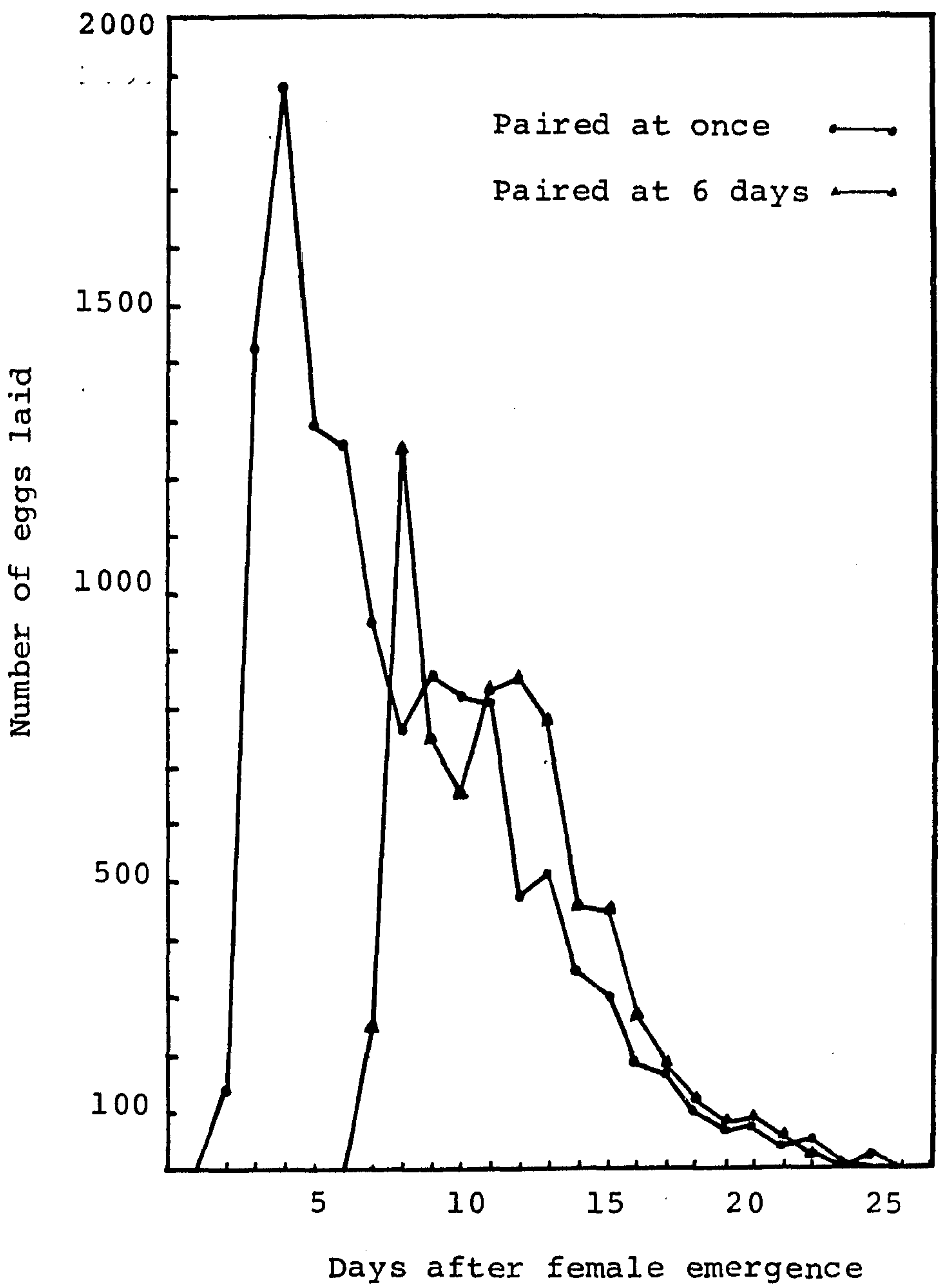

Figure 6. Egg production, by days after female emergence, for the artificial diet reared population of PBW.

in fecundity. The Yuma Co. boll-reared population was collected too early for diapause to have been a factor, and the field the bolls were collected from was a control check for chemical application studies, and consequently not treated. Because no chemical applications were made the infestation level was approximately 100%, a condition often considered advantageous for the spread of an infectious disease.

Finally, similar sublethal effects of a disease have been shown in insects. Veber and Jasic (1961) observed a highly reduced fecundity in female *Bombyx mori* (L) as a result of the sublethal effects of microsporidian disease *Nosema bombycis* Naegeli. Smirnof and Chu (1968) also found a reduction in fecundity caused by a microsporidian infection. In this case fecundity of the larch sawfly, *Pristiphora erichsonii*, was reduced up to 50% by the microsporidion, *Thelohania pristiphorae*.

Importance in Agriculture

As part of pest management programs for the PBW, boll sampling for larvae and sex pheromone trapping of the adults are used to monitor populations, and thus predict their growth, and/or determine the need for suppression. It is in the interpretation of trapping and sampling results that this study contributes.

As mentioned earlier, knowledge of sex ratios are important in pheromone trapping programs. Since only males are trapped, determining the reproductive population in the field relies on the sex ratio. This study showed that the sex ratio for all populations was 1 to 1. Consequently each male caught in a trap implies an equal number of females in the field, independent of cotton variety, fruiting form or the time of season.

The results of the log linear model analysis showed there were differences in the numbers of females that laid eggs. In terms of a sampling program this means that because the numbers of females that laid eggs was significantly lower in the late season long staple boll-reared populations, the same sample in the late season would indicate fewer females that are going to lay eggs than the same sample in the early season. When this is combined with the knowledge that females which laid viable eggs, laid the same number of viable eggs for all the populations, it could be concluded that for a whole population, fewer viable eggs were being produced by a late season boll-reared population. Since the same percentage of females laid eggs in the early season long and short staple bloom-reared populations, trap catches in the early season would indicate the same number of viable eggs whether long or short staple cotton was sampled.

CONCLUSION

Early in the season there was no difference in fecundity between long staple and short staple bloom-reared PBWs. Late in the season, however, long staple boll-reared moths laid significantly fewer eggs than short staple boll-reared moths. In short staple cotton there was no difference in fecundity between bloom-reared and boll-reared moths, but in long staple cotton the bloom-reared population laid significantly more eggs than the boll-reared population. From these data alone it could be argued that some nutritional factor in long staple cotton bolls is detrimental to PBW egg production. However, when one considers viable eggs laid by females that laid some viable eggs, then all the populations display similar fecundities. This suggests late season long staple bolls are sufficiently nutritious and that the reduced egg laying must be due to some other factor.

The effects of seasonal larval diet and variety of cotton on PBW longevity and pupal duration were also examined. Boll-reared moths were found to have longer longevities than bloom-reared moths. Short staple pupal durations were the same for the bloom-reared and boll-reared populations, but long staple boll-reared pupae took longer to develop than bloom-reared pupae.

Finally reproduction as measured by viable egg production was evaluated. Statistical tests using a log linear model demonstrated that both the variety and fruiting form were factors in determining the viable egg production of the pink bollworm populations.

APPENDIX A

EGG PRODUCTION PER FEMALE

Yuma Co. short staple bloom-reared population

Female No.	Total Eggs	Viable Eggs	Female No.	Total Eggs	Viable Eggs
1	122	112	33	389	358
2	6	0	34	296	290
3	197	194	35	35	34
4	119	93	36	231	227
5	175	169	37	280	278
6	19	2	38	195	186
7	39	7	39	287	256
8	13	0	40	223	218
9	4	0	41	229	229
10	33	32	42	21	8
11	287	247	43	78	73
12	5	5	44	3	0
13	206	192	45	4	0
14	406	401	46	181	180
15	200	190	47	26	0
16	178	168	48	38	32
17	388	351	49	541	535
18	14	0	50	64	64
19	4	0	51	7	0
20	7	1	52	25	18
21	105	99	53	23	23
22	186	176	54	136	130
23	335	232	55	114	114
24	24	0	56	171	163
25	7	0	57	162	145
26	6	0	58	7	0
27	193	175	59	29	26
28	46	10	60	85	61
29	177	168	61	11	10
30	41	11	62	109	107
31	6	0	63	70	65
32	161	160	64	55	55

Yuma Co. short staple bloom-reared population--Continued

Female No.	Total Eggs	Viable Eggs	Female No.	Total Eggs	Viable Eggs
65	58	58	72	85	82
66	50	50	73	140	139
67	337	328	74	5	0
68	25	25	75	153	146
69	59	59			
70	274	269			
71	20	19	48 females laid no eggs		

Graham Co. long staple bloom-reared population

1	99	70	27	30	0
2	320	313	28	37	37
3	9	0	29	94	94
4	21	17	30	4	0
5	254	247	31	49	49
6	3	0	32	129	123
7	4	0	33	64	64
8	115	109	34	175	170
9	185	181	35	228	221
10	364	356	36	137	131
11	45	38	37	245	241
12	163	126	38	68	59
13	4	0	39	29	22
14	201	197	40	105	97
15	178	176	41	196	189
16	211	208	42	74	66
17	17	17	43	164	155
18	221	220	44	11	5
19	2	0	45	166	159
20	295	278	46	80	74
21	178	173	47	241	235
22	92	87	48	33	30
23	4	0	49	9	0
24	44	44	50	63	61
25	143	141	51	33	31
26	169	163	52	100	97

Graham Co. long staple bloom-reared population--Continued

Female No.	Total Eggs	Viable Eggs	Female No.	Total Eggs	Viable Eggs
53	139	135	66	6	0
54	69	68	67	97	96
55	373	356	68	114	114
56	79	70	69	32	27
57	7	0	70	52	52
58	23	23	71	19	18
59	170	166	72	87	63
60	148	146	73	84	79
61	3	0	74	75	64
62	141	137	75	186	181
63	92	92	76	160	158
64	23	0			
65	143	141	30 females laid no eggs		

Yuma Co. short staple boll-reared population

1	289	288	22	294	387
2	5	3	23	12	0
3	27	8	24	139	137
4	4	4	25	49	43
5	183	155	26	374	366
6	108	99	27	68	65
7	117	109	28	36	31
8	26	12	29	453	444
9	249	239	30	24	24
10	93	50	31	21	18
11	361	361	32	202	195
12	32	30	33	120	102
13	263	256	34	5	0
14	142	129	35	48	29
15	71	71	36	53	53
16	59	52	37	321	310
17	39	36	38	21	0
18	230	219	39	67	65
19	187	182	40	332	316
20	25	0	41	209	185
21	236	233	42	45	43

Yuma Co. short staple boll-reared population--<u>Continued</u>

Female No.	Total Eggs	Viable Eggs	Female No.	Total Eggs	Viable Eggs
43	84	84	51	11	10
44	27	12	52	14	13
45	3	3	53	3	3
46	3	3	54	231	221
47	2	0	55	4	0
48	14	0			
49	124	119			
50	3	3	42 females laid no eggs		

Graham Co. long staple boll-reared population

1	33	0	26	176	176
2	28	0	27	13	0
3	24	0	28	3	0
4	103	0	29	225	216
5	264	255	30	6	0
6	114	0	31	15	10
7	4	0			
8	5	0			
9	8	0	53 females laid no eggs		
10	163	0			
11	38	0			
12	7	0			
13	11	0			
14	178	175			
15	15	14			
16	54	0			
17	30	0			
18	87	87			
19	193	187			
20	2	0			
21	9	0			
22	7	7			
23	97	93			
24	4	0			
25	8	0			

Pinal Co. long staple boll-reared population

Female No.	Total Eggs	Viable Eggs	Female No.	Total Eggs	Viable Eggs
1	234	234			
2	20	0			
3	205	200			
4	5	5			
5	47	47			
6	283	283			
7	17	0			
8	189	187			
9	4	0			
10	108	108			
11	2	0			
12	66	0			
13	23	14			
14	3	0			
15	32	0			
16	24	0			
17	6	0			
18	37	37			
19	75	0			
20	25	0			
21	40	38			
22	65	0			
23	8	0			

19 females laid no eggs

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