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Research Article

Delayed Mating: A Non-chemical Control Strategy for the Management of *Plodia interpunctella* (Hubner) (Lepidoptera: Pyralidae) Infesting Stored Products in Nigeria

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Abstract

Background and Objective: Management of *Plodia interpunctella* (Hubner) has been drifting away from insecticide-based approach to other control measures with broader potential capable of reducing their population. The impact of delaying mating on longevity and reproductive parameters was investigated. **Materials and Methods:** Moths were delayed for 1-5 days and the effects on egg laid (fecundity), egg viability, adult emergence and longevity of *P. interpunctella* were investigated at ambient temperature (28±2°C) and relative humidity (80±5%). **Results:** Delayed mating caused significant effect on moth fecundity ($F_{15, 32}$ = 3.905, p = 0.001), egg viability ($F_{15, 32}$ = 3.905, p<0.001), adult emergence ($F1_{5,32}$ = 4.665, p<0.001) and longevity of male ($F_{17, 36}$ = 7.349, p<0.001) and female ($F_{17,36}$ = 4.886, p<0.001) *P. interpunctella*, respectively. Fecundity, egg viability and adult emergence decreased with increasing delayed mating days. Highest fecundity (183.33) and viable eggs (50.00%) was however observed when there was no mating delay (<24 h) while lowest fecundity (31.67) and viable eggs (3.33%) was observed by delaying both sexes and only male for 5 days. Delaying females from mating however had a greater impact on egg laid and viability as opposed to delaying males. Average adult longevity ranged from 2-7 days for male and 3-7 days for female. **Conclusion:** Thus, mating disruption in *P. interpunctella* could provide an alternative non-chemical control method to the use of synthetic insecticides in the management of this insect pest in Nigeria.

Key words: Adult emergence, adult longevity, mating disruption, egg viability, fecundity

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Indian meal moth, *Plodia interpunctella* (Hubner) is a cosmopolitan pest capable of causing unwholesome damage on stored products such as dried fruits, nuts, cereals and a variety of processed food. Infestations of stored products by this insect pest have resulted in direct product loss and indirect economic costs through pest control costs, quality losses and consumer complaints[1]. The larva is the most destructive stage due to its feeding habit while other stages are non-feeding. Larvae of meal moth spin a web as they become fully grown and leave behind silken threads wherever they crawl. The webbing activity of the larva stage reduces the aesthetic value of the infected product and rendered it unfit and unattractive to consumers. The feeding activity of the larvae also decreases the weight of the affected products. *P. interpunctella* is therefore, a serious menace that could result in qualitative and quantitative losses of farm produce. The debilitating effect of this pest is particularly worsened in Tropical part of Western-Africa, particularly Nigeria due to poor and inadequate storage facilities. Low level of literacy among local peasant farmers on diverse means of controlling and minimizing infestation by this pest has further led to high loss of their stored products in Nigeria[2].

The use of synthetic and botanical insecticide has been advocated by various researchers for effective management of this infamous pest of stored products[3-5]. However, the overall acceptability of most synthetic insecticide has been greatly affected owing to widespread public concerns associated with their usage on human health and the environment coupled with the resistance of strains of *P. interpunctella* to several organophosphates and microbial insecticide[6-9]. Although, research work on the use of botanical pesticides has gained prominence over the years, myriads of problem such as relatively slow action, variable efficacy, instability in the environment, disagreeable odour, poor water solubility and inconsistent availability among others are threatening its long-term usage[10,11]. Management of *P. interpunctella* in recent years is therefore drifting away from insecticide-based approach to other control measures with broader potential capable of reducing their population.

Notable among the major factors that determine the viability of egg laid by any insects are rate of mating, humidity, temperature and light among others[12,13]. Several researchers have however opined that delayed mating in lepidopteran insect pest have detrimental effects on various life history traits such as fecundity, egg viability, eclosion and adult longevity[14-17]. Knowledge of reproductive biology is fundamental for understanding a species mating system and the implementation of behavioural-based control methods such as mating disruption[17]. The effect of delayed mating on reproductive performance of *P. interpunctella* has been investigated by few authors[12,18,19]. But in Nigeria, with the exception of study conducted by Mbata[13] where the impacts of temperature, relative humidity, light and mating on the oviposition of *P. interpunctella* were investigated, the effect of delayed mating on the fecundity, egg viability, adult emergence and longevity of *P. interpunctella* infesting stored products in the country have not been investigated. A good knowledge about the possible effect of delayed mating on reproductive performance and longevity of this species infesting stored products is needed in developing mating disruption devices. Thus, this study sought to investigate the effect of delayed mating on fecundity, egg viability, adult emergence and longevity of *P. interpunctella* infesting stored products in Nigeria.

MATERIALS AND METHODS

The experiment was carried out in Biology Laboratory II, Department of Biology, Federal University of Technology Akure, Ondo State, Nigeria. The study lasted for four months from February-May, 2017.

Preparation of insect culture: The starter culture of *P. interpunctella* used in this study was obtained from infested maize sourced from Ilara-Mokin and Oba market, Akure, Ondo State, Nigeria. One hundred eggs of *P. interpunctella* were introduced into 1.5 L plastic containers containing 200 g of artificial diet of poultry feed, glucose and wheat flour in ratio of 40:20:40 (g) at ambient temperature ($28\pm2°C$) and relative humidity ($80\pm5\%$). Corrugated paper spools which served as pupation sites for wandering larvae were placed in rearing containers. Prior to adult emergence, pupae were removed from the spools and sexed according to characters described by Butt and Cantu[20]. To ensure virginity, male and female pupae were separated and placed in different plastic containers. Each container was later checked daily at 08.00 and 12.00 h local time for adult emergence. Thereafter, moths (0-12 h old) that emerged were used in this study.

Mating treatment: Newly emerged virgin females (unmated females) that emerged within 24 h were paired with virgin males (unmated males) that were delayed for: 0, 1, 2, 3, 4 and 5 days old (treatments 1-6). Newly emerged virgin male that emerged within 24 h were paired with virgin females that were delayed for 1, 2, 3, 4 and 5 days old (treatments 7-11).

Finally, virgin males of five different ages (1, 2, 3, 4 and 5 days old) were paired with virgin females of the same age (treatments 12-16). Control experiment were also set up using only three newly emerged virgin males (<24 h old) which were not paired with any female (treatment 17) and three newly emerged virgin females that were not paired with any male (treatment 18). Each treatment was replicated three times making a total of 54 replicates. Male and female were allowed to mate in transparent glass vials (3 cm diameter, 8 cm deep). After mating, the vials were opened in a flight chamber for the insects (30×20×25 cm) (L×W×H) to fly out and the number of eggs laid was counted daily between 8.00 and 12:00 am. All the vials were kept in an insect breeding cage (70×60×50 cm) (L×W×H). To estimate adult longevity, adults from the jars were checked daily until all the adults' moths died. Ten eggs were transferred to a moist filter paper that was overlaid inside the petri dishes. The total numbers of eggs that hatched was recorded using a dissecting microscope. Egg viability in each dish was calculated as the proportion of the total number of eggs that hatched out of the number of the eggs evaluated. To estimate the number of emergence, the eggs laid were introduced into a new culture media consisting of whole wheat grain, glucose and animal feeds. The culture media was formulated based on previous studies conducted by Akinneye *et al.*[21]. The total number of adult emergence from the total number incubated was counted and the average recorded.

Statistical analysis: A completely randomized design was used for this experiment and untransformed data on each of the life history trait i.e., number of eggs laid/fecundity, egg viability, adult (male and female) longevity and emergence] were checked for normality based on Shapiro-Wilk test[22] before being subjected to one-way analysis of variance (ANOVA). Means were separated using Tukey's Test where significant difference exist (p<0.05). Similarly, all data were subjected to linear regression to determine the relationship between mating delays (days) and each of the life history traits observed. All analysis were carried out using Statistical Package for Social Sciences (SPSS) 17.0 software.

RESULTS

Effect of delayed mating on fecundity and egg viability of *P. interpunctella*: Generally, moth fecundity and egg viability decreased as the delayed mating days increased except for male and female delayed (treatment 12-17) which showed erratic fecundity rate (Table 1). There was also a significant effect of delayed mating on moth fecundity ($F_{15,32}$ = 3.905, p = 0.001) and egg viability ($F_{15,32}$ = 3.905, p<0.0001). Regardless of the treatment, highest fecundity (183.33) and viable eggs (50.00) s were observed when there was no mating delay (<24 h). Lowest fecundity (31.67) was however observed when both sexes were delayed from mating for 5 days while the lowest viable eggs (3.33%) was observed when <24 h female was mated with male delayed for 5 days. Similarly, significantly higher fecundity and viable eggs was observed when there was no delayed mating (treatment 1) when compared to fecundity of both sexes delayed for 3-5 days and egg viability of both sexes delayed for 4 and 5 days respectively (treatment 12-16).

Table 1: Effect of delayed mating on fecundity and egg viability of *P. interpunctella*

Male age (days)	Female age (days)	Mean number of egg laid/Fecundity	Mean viability (%)
<24 h	<24 h	183.33±5.70^{b}	50.00±5.77^{c}
1	<24 h	144.67±2.07^{ab}	40.00±5.77^{bc}
2	<24 h	142.99±3.16^{ab}	26.67±6.67^{abc}
3	<24 h	78.33±6.84^{ab}	20.00±1.15^{abc}
4	<24 h	75.33±6.89^{ab}	10.00±5.57^{ab}
5	<24 h	72.67±2.90^{ab}	3.33±3.33^{a}
<24 h	1	129.00±2.46^{ab}	40.00±5.77^{bc}
<24 h	2	129.00±3.25^{ab}	30.00±5.77^{abc}
<24 h	3	75.33±6.89^{ab}	26.67±3.33^{abc}
<24 h	4	68.00±1.53^{ab}	20.00±5.77^{abc}
<24 h	5	54.67±6.33^{a}	13.33±3.33^{ab}
1	1	87.67±1.27^{ab}	40.00±5.77^{bc}
2	2	46.00±3.51^{ab}	30.00±5.77^{abc}
3	3	70.66±1.49^{a}	23.33±8.82^{abc}
4	4	37.67±4.41^{a}	16.67±3.33^{ab}
5	5	31.67±5.93^{a}	13.33±8.82^{ab}
Female only		54.67±0.13^{a}	0.00±0.00^{a}

Means±SE (Standard error) followed by the same letter along the column are not significantly different (p>0.05) from each other according to Tukey's test

Effect of delayed mating on adult emergence of *P. interpunctella*: Delayed mating in *P. interpunctella* showed a significant effect (p<0.0001) on the adult emergence of this insect ($F_{15,32}$ = 4.665). Highest adult emergence was observed when *P. interpunctella* were not delayed from mating (<24 h) (treatment 1) (Table 2). Adult emergence however decreased with increasing delayed mating days for male, female and both sexes. Irrespective of the treatment, <24 h female mated with male delayed for 5 days produced eggs with the lowest adult emergence (3.33%) which was significantly lower (p<0.05) than the highest adult emergence (40.00%) observed in eggs of moths that were not delayed from mating (<24 h).

Table 2: Effect of delayed mating on emergence of adult *P. interpunctella*

Male age (days)	Female age (days)	Adult emergence (%)
<24 h	<24 h	40.00±1.15[d]
1	<24 h	36.67±3.33[cd]
2	<24 h	23.33±3.33[abcd]
3	<24 h	16.67±8.82[abcd]
4	<24 h	6.67±3.33[ab]
5	<24 h	3.33±0.33[a]
<24 h	1	33.33±3.33[bcd]
<24 h	2	23.33±6.67[abcd]
<24 h	3	20.00±0.00[abcd]
<24 h	4	13.33±3.33[abcd]
<24 h	5	10.00±0.00[abc]
1	1	33.33±3.33[bcd]
2	2	23.33±6.67[abcd]
3	3	13.33±3.33[abcd]
4	4	13.33±3.33[abcd]
5	5	6.67±0.67[ab]

Means±SE (Standard error) followed by the same letter along the column are not significantly different (p>0.05) from each other according to Tukey's test

Effect of delayed mating on adult longevity of *P. interpunctella*: Regardless of the treatment, there was significant effect (p<0.0001) of delayed mating on the longevity of male ($F_{17,36}$ = 7.349) and female ($F_{17,36}$ = 4.886) *P. interpunctella*. Highest male longevity (7 days) was however observed in virgin male and this was significantly higher (p<0.05) than the longevity of male that emerged from all the treatments except for those that emerged from treatment 5, 9 and 10 (Table 3). Approximately highest female longevity (7 days) was also observed in treatment 6, 9 and 10 and they were not significantly different (p>0.05) from each other. Of all the treatments, lowest male longevity (2-3 days) and female longevity (3-6 days) was observed in moths that emerged when both sexes were delayed from mating from 1-5 days (treatment 12-16) and no significant difference (p>0.05) existed among them.

Relationship between mating delay, fecundity, egg viability, adult emergence and longevity of *P. interpunctella*: Table 4-6 show the relationship between mating delay and various life history traits of *P. interpunctella*. Negative slope of regression equation showed that moth fecundity, egg viability and adult emergence decreased with increasing delayed mating days for males, females and both sexes while positive slope of regression indicated that male and female longevity increased with increasing delayed mating days for males, females and both sexes (Table 4, 5 and 6). The only exception was observed in female longevity which decreased (Slope = -0.133) with increasing delayed

Table 3: Effect of delayed mating on longevity of adult of *P. interpunctella*

Male age (days)	Female age (days)	Male longevity (days)	Female longevity (days)
<24 h	<24 h	2.00±1.00[ab]	4.00±1.15[abc]
1	<24 h	2.00±0.00[ab]	5.00±0.00[bc]
2	<24 h	2.33±0.33[ab]	5.53±0.33[bc]
3	<24 h	3.00±0.00[bc]	5.67±0.33[bc]
4	<24 h	4.33±1.20[bcd]	6.12±0.58[bc]
5	<24 h	2.33±0.33[ab]	7.23±1.73[c]
<24 h	1	2.67±0.33[ab]	5.00±0.00[bc]
<24 h	2	3.33±0.67[bc]	5.00±0.57[bc]
<24 h	3	4.33±0.33[bcd]	6.67±1.20[bc]
<24 h	4	5.67±1.20[cd]	7.00±1.53[bc]
<24 h	5	2.67±0.33[ab]	6.00±058[bc]
1	1	1.67±0.33[ab]	4.67±0.33[bc]
2	2	2.67±0.33[ab]	4.67±0.67[bc]
3	3	3.00±0.00[bc]	6.33±0.33[bc]
4	4	3.33±0.67[bc]	3.67±1.20[ab]
5	5	2.00±0.00[ab]	3.33±0.33[ab]
Male only		6.67±0.33[d]	
Female only			5.67±0.33[bc]

Means±SE (Standard error) followed by the same letter along the column are not significantly different (p>0.05) from each other according to Tukey's test

Table 4: Relationship between male delayed and various life history traits of *P. interpunctella*

Variables	Slope	Intercept (Y)	R^2	df	F-value	p-value
Egg laid/fecundity	-23.600	198.600	0.440	1.16	12.552	0.003
Egg viability	-9.429	58.000	0.724	1.16	41.885	0.000
Male longevity	0.267	1.733	0.133	1.16	2.462	0.136
Female longevity	0.752	3.200	0.491	1.16	15.448	0.001
Adult emergence	-8.000	49.111	0.675	1.16	33.231	0.000

R^2: R-square, df: Degree of freedom

Table 5: Relationship between female delayed and various life history traits of *P. interpunctella*

Variables	Slope	Intercept (Y)	R^2	df	F-value	p-value
Egg laid/fecundity	-25.143	194.556	0.501	1.16	16.087	0.001
Egg viability	-7.048	54.667	0.724	1.16	42.042	0.000
Male longevity	0.381	2.111	0.164	1.16	3.140	0.095
Female longevity	0.505	3.844	0.246	1.16	5.233	0.036
Adult emergence	-6.095	44.667	0.610	1.16	24.976	0.000

R^2: R square, df: Degree of freedom

Table 6: Relationship between male and female delayed and various life history traits of *P. interpunctella*

Variables	Slope	Intercept (Y)	R^2	df	F-value	p-value
Egg laid/fecundity	-25.248	164.533	0.476	1.16	14.537	0.002
Egg viability	-7.429	54.889	0.633	1.16	27.851	0.000
Male longevity	0.524	1.911	0.074	1.16	1.281	0.274
Female longevity	-0.133	4.911	0.024	1.16	0.398	0.537
Adult emergence	-6.762	45.333	0.593	1.16	23.284	0.000

R^2: R square, df: Degree of freedom

mating days for both sexes (Table 6). Of all the life traits observed in this study, highest significant effect ($p<0.001$) of male delayed ($R^2 = 0.724$, $F_{1,16} = 41.885$), female delayed ($R^2 = 0.724$, $F_{1,16} = 42.042$) as well as male and female delayed ($R^2 = 0.633$, $F_{1,16} = 27.851$) was observed in egg viability of *P. interpunctella*. Male longevity was however the least affected with male delayed ($R^2 = 0.133$, $F_{1,16} = 2.462$, $p>0.136$) (Table 4) and female delayed ($R^2 = 0.164$, $F_{1,16} = 3.140$, $p>0.095$) (Table 5) while female longevity was the least affected when both sexes were delayed from mating ($R^2 = 0.024$, $F_{1,16} = 0.398$, $p>0.537$) (Table 6). Negative significant linear relationship however existed between male delayed and adult emergence (Slope = -8.000, $R^2 = 0.675$, $F_{1,16} = 33.231$, $p<0.001$) (Table 4), female delayed and adult emergence (Slope = -6.095, $R^2 = 0.610$, $F_{1,16} = 24.976$, $p<0.001$) (Table 5) as well as male and female delayed and adult emergence (Slope = -6.762, $R^2 = 0.593$, $F_{1,16} = 23.284$, $p<0.001$) (Table 6) of *P. interpunctella*.

DISCUSSION

This study investigated the effect of delayed mating on fecundity, egg viability, adult emergence and longevity of *P. interpunctella* infesting stored products in Nigeria. The fecundity of *P. interpunctella* was highest when there was no delayed mating and this is in agreement with the findings of Huang and Subramanyam[12] as well as Akinneye and Ashamo[16] where similar observation was reported for *P. interpunctella* and *Ephestia cautella* (Walker), respectively. The delay of male, female and both sexes resulted in significant reduction in the percentage of viable eggs of *P. interpunctella*. However, delaying females from mating had a greater impact on egg laid and viability than delaying males (F-value). Park *et al.*[23] had earlier attributed decreased viability in egg of *P. interpunctella* with age to reduction in the quantity and quality of accessory gland secretions and hormones transferred to females during copulation or total absence of these secretions resulting in decreased or no spermatophore formation. Also, egg production and their viability decreased in females mated with males delayed for 1-5 days. This suggests that sperm production in males decreased or degenerate with age, especially when they were delayed from mating. Reproductive potential of several moths has earlier been reported to decrease with their age[24-27].

While three percent of eggs produced by undelayed female mated with male delayed for 5 days were observed to be viable in this study, 100% non-viability was reported by Huang and Subramanyam[12] in their studies. Egg laid by virgin females failed to hatch and were similar to those laid by females delayed for 5 days. Significantly lower number of eggs laid by virgin females when compared to eggs laid by females that were not delayed from mating suggests that early mating in *P. interpunctella* increase the frequency of egg laid. Therefore, delaying female from mating could significantly reduce the rate of egg laying and hatchability and thus, helping to control the population of *P. interpunctella* attacking stored produce in Nigeria.

Average male longevity among treatments ranged from 2-7 days while female longevity ranged from 3-7 days. Mean adult longevity in this study revealed that adult unmated male moths were able to live for maximum of approximately 7 days while mated males live for maximum of approximately 6 days[12,28]. Highest mean female longevity of 7 days was observed in females mated with male delayed for 5 days and this showed that delayed male mating did not affect female longevity. The longevity of both male and female delayed for 5 days significantly reduced to 2 and 3 days, respectively. This is the lowest longevity reported so far for male and female *P. interpunctella* when they were delayed from mating. Prior to this study, the reported lowest longevity for male and female *P. interpunctella* was 4 and 5 days, respectively and this was when they were not delayed from mating[12]. The decrease in moth longevity with increased delayed mating days may be attributed to higher energy expended on search for opposite sex and intra-specific competition for space among species of same sex after emergence. However, because adults of *P. interpunctella* do not feed and were also delayed from mating, replacement of lost energy is impossible especially in females. Earlier studies have shown that females of few insect species that do not feed at adult stage usually re-mate primarily to acquire nutrient from seminal fluid to avoid being nutritionally challenged[29,30]. Consequently, this study has showed that delayed mating could reduce the life span of male and female *P. interpunctella*.

CONCLUSION

This study has clearly demonstrated that delayed mating in *P. interpunctella* significantly affect the reproductive performance of *P. interpunctella*. In fact, it has been shown that fecundity, egg viability and adult emergence decreased significantly with increased delayed mating days especially for both sexes at 5 days. The longevity of male and female *P. interpunctella* significantly reduced when both sexes were delayed from mating for 5 days. It could therefore be concluded that the approach used in this study could help in controlling *P. interpunctella* infesting stored products in Nigeria.

SIGNIFICANCE STATEMENT

This research work showed that fecundity, egg viability and adult emergence decreased significantly with increased delayed mating days especially when both sexes were delayed for 5 days. The longevity of male and female *P. interpunctella* was also significantly reduced to 2 and 3 days, respectively when both sexes were delayed for 5 days. Therefore, a technique that could encourage mating disruption in both sexes of *P. interpunctella* should be encouraged to reduce the destructive activity of this pest on stored products in Nigeria. This method is particularly beneficial because it is a non-chemical control method.

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