Journal of Entomology and Zoology Studies
Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2019; 7(5): 156-162

Received: 09-07-2019
Accepted: 11-08-2019

Fakeerappa R Pattan
PhD. Scholar, Department of Entomology, UAS, GKVK, Bangalore, Karnataka, India

Correspondence
Fakeerappa R Pattan
PhD. Scholar, Department of Entomology, UAS, GKVK, Bangalore, Karnataka, India

Trade-off between fecundity and longevity in singly and multiply mated females of *Xylotrechus quadripes* Chevrolat (Cerambycidae: Coleoptera)

Fakeerappa R Pattan

Abstract
Mating behaviour, fecundity and longevity of female beetles of *Xylotrechus quadripes* was studied under laboratory conditions. In a single pairing event, the female approached and copulated multiply with the male (5.07 ± 1.49) and laid a batch of eggs. Following an egg laying bout the female mated again with the male and showed oviposition. This intermittent mating and egg laying continued throughout the lifespan of the female. Females that mated intermittently laid more eggs (140.72 ± 42.50) than mated singly (31.3 ± 6.08). On an average a female (intermittently) mated 12.2 times and was able to lay 140.72 ± 42.50 eggs. The maximum number of eggs per clutch was 19.3 ± 6.61 in the fourth clutch and 85.71% eggs were laid within six clutches. Later number of eggs per clutch decreased as did the frequency of mating. However, females mated multiply had significantly reduced longevity than virgins females.

Keywords: Intermittent mating; fecundity; longevity; *Xylotrechus quadripes*

1. Introduction

In general reproductive success of male increases with mating rate, while female requires one or few matings to maximize the reproductive success. Contrary to this, most of the female insects mate multiply. However multiple mating in short lived animals like insects is mysterious because cost of mating more than once (increased predation risk associated with searching for and mating males, searching an additional mate may incur energetic or time costs, *etc.*) usually appear to outweigh the benefits (Jennions and Petrie, 1999 [11]; Thornhill and Alcock, 1983 [20]).

There are two main hypothesized benefits of multiple mating to female: 1) material benefits and 2) genetic benefits (Reynolds, 1996 [16]). Material benefits means multiple mating directly enhances female fitness through increased number or size of eggs whereas genetic benefits brings indirect female fitness through increased genetic quality of insects. Material benefits may include nuptial gifts from male which are nutritional sources to female (Gwynne, 1997 [8]), a reduction in male harassment (Arnqvist, 1989 [1]) and replenishment of depleting sperms which are not sufficient enough to fertilize all eggs a female produce in her lifetime (Thornhill and Alcock, 1983 [20]). On the other side, genetic benefits may include opportunities to manipulate offspring paternity and to create more genetic variability among the offsprings by avoiding inbreeding (Birkhead, 1987 [2]: Ridley, 1993 [17]).

Females of coffee white stem borer (CWSB) mated multiply with same mounted male and laid batch of eggs then soon became sexually receptive by approaching another male to mate. This intermittent mating and laying eggs continued throughout their lifespan (up to 1 month). One of the possible explanations, for this kind of mating is that females increasing the reproductive success by laying more number of eggs. To test this hypothesis a series of experiments were carried out by varied number of matings and replaced the spent males (in an intermittently mating pairs) with virgin males then measured how this affected their fecundity. To avoid possible confounding effects of genetic benefit in our study, in all the treatments each female mated with a same male regardless of how often she mated (except in male replacement experiment). Our results clearly showed that females mated multiply has laid more number of eggs than female mated only once in the beginning of the experiment. In spent male replacement experiments females mated with new virgin males (on 12th day since first time mated) have laid more number of eggs than that of females mated intermittently with same male. This clearly indicated, multiple mating enhanced fecundity in CWSB.

But multiply mating females had significantly reduced life span than that of virgin and singly mated females.

2. Materials and Methods

2.1 Natural History of CWSB

In India, coffee has been at the receiving end of the onslaught by the white stem borer since several decades. Its preferential attack on Arabica (as against Robusta coffee) and the persistently high population density has had a telling blow on coffee in India; farmers have either abandoned cultivation of coffee or have shifted to the Robusta cultivation. CWSB grubs make tunnels in the main stem and thick primary branches of coffee plants and pupated beneath the bark (Plate 1). Adult emerge in two flight periods in India (Subramanian, 1934 [19]): the pre-monsoon flight period begin in April and extends to the end of May and the post-monsoon period starts from September to until the end of December. The pest causes substantial economic loss every year since infested plants have to be uprooted (they act as source infestation and further spread of the pest).

2.2 General Methods

CWSB infested coffee plants were uprooted and stored in the net house maintained at Chandrapore coffee estate, Mudigere, Karnataka (India). Adult beetles were collected from the net house and sexed by genital inspection later, placed in single-sex cultures on the day of emergence to prevent beetles from mating before the experiments (Plate 2). Beetles mate immediately after emergence but started laying eggs only after 24 hours of first mating (Visitpanich, 1994a [23]). Although female body size affects the fecundity (Thornhill and Alcock, 1983 [20]), the females of approximately equal size to different mating treatments were used. Mating successes were visually confirmed; copulation usually began within 10 min after placing them into mating box and lasts roughly 1 minute.

To facilitate CWSB to lay eggs in the plastic container (45cm x 30cm x 12cm), the mouth of the container was closed tightly with muslin cloth. Then the container was inverted on a sheet of white paper in such a way that muslin cloth covered mouth was resting on the sheet of paper. Females usually glue their eggs on the sheet by inserting her ovipositor through the pores of muslin cloth. So each day the sheet was changed to count the adhered eggs (Plate 3). The experiment 1-2 was conducted in November (2015) and experiment 3 was conducted in November (2016) within the laboratory maintained at 22-30 ^{0}C and ambient day length and humidity.

2.3 Experiment 1: Effect of no access to male after initial mating on fecundity

To test the effect of severely limiting access to male and matings on egg production. For this we set up two treatments: (1) females mated once at the beginning of the experiment (N = 10), (2) females kept continuously with a male and matings for the entire experiment (N = 25). Mating in the first experiment was visually confirmed and was not quantified the number of matings that occurred between female and male kept together. The number of eggs laid every day by the female was counted.

2.4 Experiment 2: Replacement of spent males (exhausted) with virgin male

Results from the experiment 1 showed that the females that mated only once at the beginning of the experiment had laid only one batch of egg (very rarely two) whereas female supplied continuously with male had laid more batches of eggs. In the intermittently mating pairs, fecundity is drastically reduced after 10 days or 11days since mating started. This may be because of males stop responding to female (Personal observation). Hence in the present experiment, replaced spent male with virgin male on the 12 day (N = 10) and thereafter number of eggs laid every day was counted.

Plate 1a: Extensive tunnelling by CWSB grubs inside the arabica stem

Plate 1b: Emergence hole of CWSB on coffee stem

Plate 1c: Adult beetle

Plate 1: Nature and symptom of damage by CWSB.

Plate 2a: Sexual dimorphism in CWSB. Male with generally blunt abdomen and female having pointed abdomen.

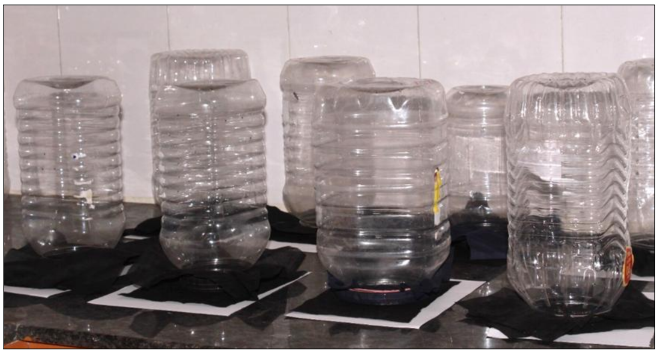

Plate 2b: General laboratory setup to study the fecundity, longevity *etc.*

Plate 3: Mating box provided with egg laying facility

2.5 Experiment 3: Mating frequency and clutch size in CWSB

To quantify how frequently a single female mates and lays eggs in a clutch, male and female beetles were allowed to mate in a mating box (N = 10). Every single mating and egg laying was visually confirmed. After completion of each mating and egg laying bout, the number of eggs were recorded in order to estimate number of eggs per clutch.

2.6 Experiment 4: Effect of multiple mating on female longevity

In order to test the hypothesis that multiple mating reduces adult longevity in female beetles. Three sets of females, (1) Female with male continuously (N = 50), (2) Female without males (virgin) (N = 25) and (3) Females that mated only once (N = 25) were maintained. Daily observations were made on the mortality of beetles. In all the three sets, beetles of similar size were selected.

2.7 Statistical Analysis

In most of the experiments, the number of eggs laid by females in each treatment were compared using t tests. Means (± SE) were reported using untransformed values for ease of understanding.

3. Results

3.1 Experiment 1: Effect of no access to male after initial mating on fecundity

Females kept continuously with males laid more number of eggs (140.72 ± 8.50) than that of females mated only once (31.3 ± 1.92) at the beginning of the experiment (Figure 1). Female mated once had laid only one batch of eggs (rarely two batch), wherein continuously mated female laid a batch of egg after every successive mating exhibited typical intermittent mating and egg laying behaviour (Personal observation).

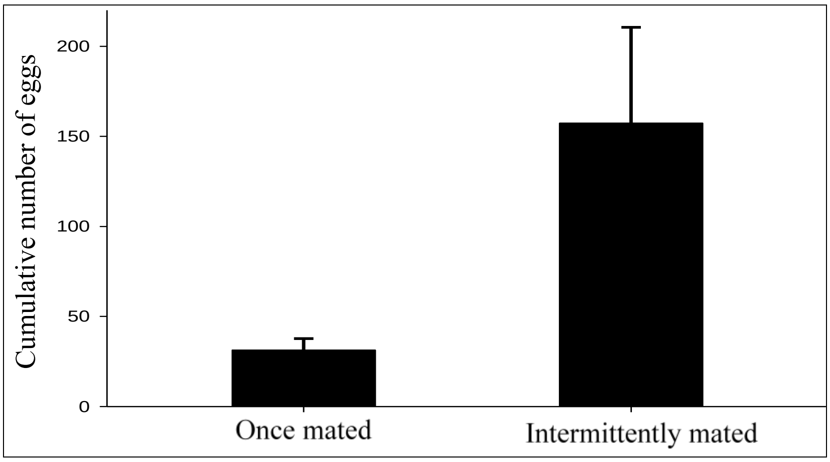

Fig 1: Cumulative number of eggs produced by once mated and intermittently mated female.

To determine when the difference in fecundity first occurred, average daily egg production in both the mating treatments was compared. The average daily egg production in both the mating treatments diverged significantly for the first time on the day 3rd (*t* test: *t* 10 = - 3.343, P < 0.01) (Figure 2) and thereafter. Clearly, females allowed to mate continuously laid more eggs than that mated only once at the beginning of the experiment.

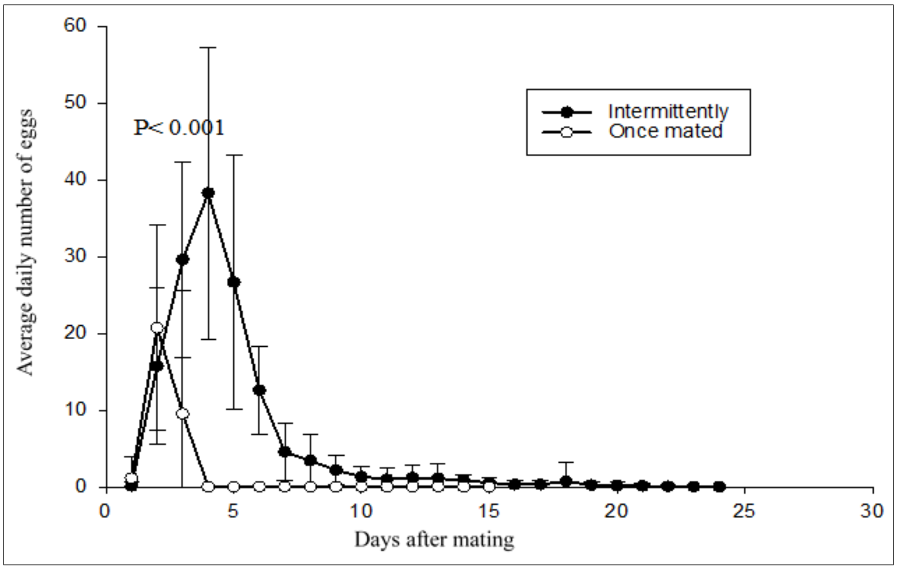

Fig 2: Average daily egg production by females (mated once and intermittently). Significant differences between the treatments started on 3rd day.

3.2 Experiment 2: Replacement of spent males (exhausted) with virgin male

Results from the experiment 1 showed that in the continuously mating pairs, fecundity is drastically reduced after 10 days or 11days since mating started. This may be because of males stop responding to female (personal observation). Hence replaced spent male with virgin male on the 12th day (N = 10) and thereafter we counted number of eggs laid every day. On the 12th day since mating started, female mated with new replaced (one day old virgin) male has laid significantly more number of eggs than that of female supplied with same earlier male (on the 12th day, *t* test: *t* 10 = - 3.845, P < 0.01) (Figure 3) and thereafter egg laying continued up to 15th day. Life time fecundity of female mated with replaced virgin male was 209.3 ±11.81 eggs as compared to female mated with same earlier male (140.72 ± 8.50) (Figure 3).

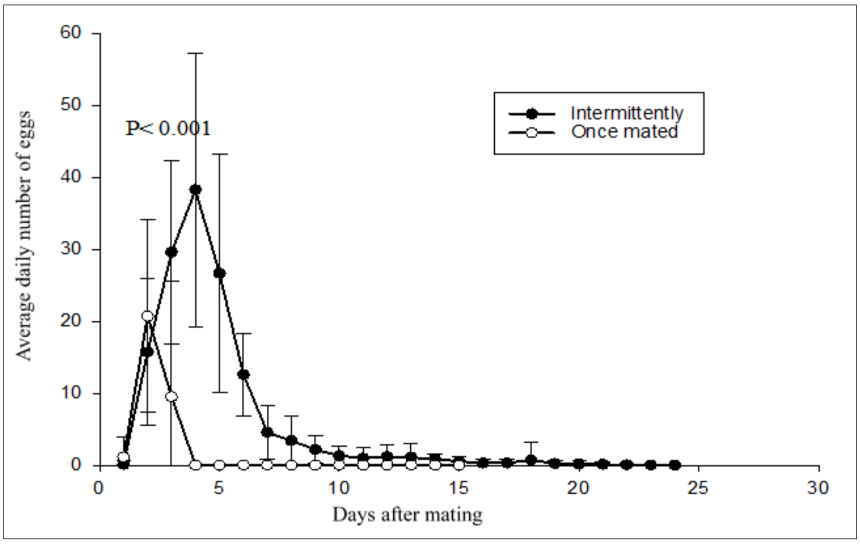

Fig 4: Average egg production in Experiment 3, females mated continuously.

3.3 Experiment 3: Mating frequency and clutch size in CWSB

The adult female intermittently mated with the male and laid batches of eggs. This intermitted mating and egg laying continued throughout her lifetime. One successful copulation took 3 to 50 seconds duration. After 3-7 such copulations the female started laying eggs. On an average a female mated 12.2 times (N =10). The maximum number of eggs per batch was 19.3 ± 6.61 in the fourth clutch and 85.71% of the eggs were laid within six clutches and later the number of eggs per clutch decreased as did the frequency of mating (Figure. 4).

3.4 Experiment 4: Effect of multiple mating on adult longevity of female CWSB

The longevity of the virgin male and female beetles after emergence from the stem was 26.96 ±5.39 and 31.08 ± 6.21 days respectively. But female longevity decreased drastically when she mated continuously with male (1:1 sex ratio) and there was statistical significant difference between female mated continuously and virgin female. There was no statistical significant difference between virgin and once mated females (Figure. 5).

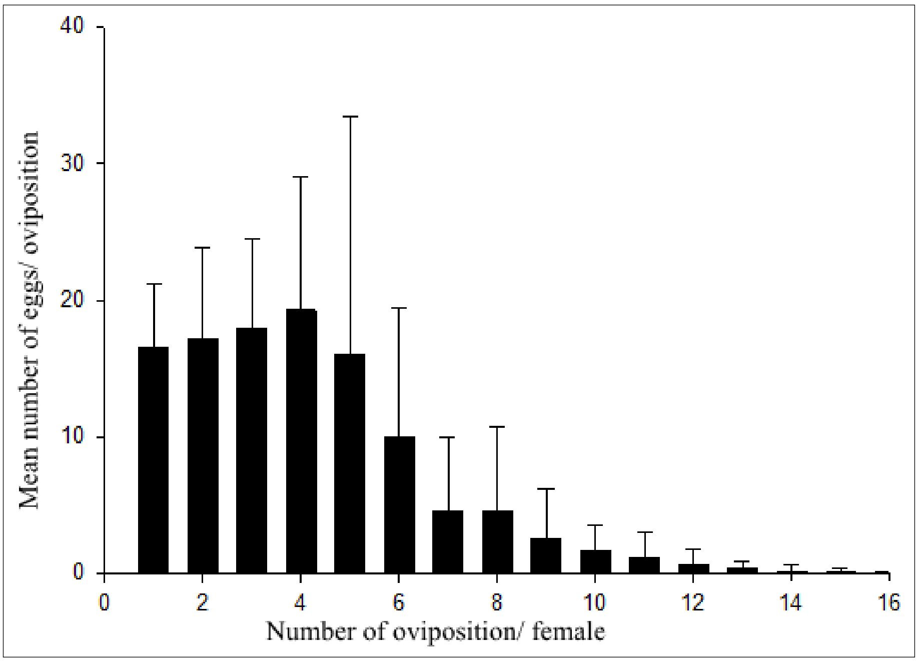

Fig 5: Average daily egg production with females mated intermittently with same male and replaced male. Significant differences between the treatments observed on the day of replacement (12^{th} day).

4. Discussion

From the above conducted experiments, it is clearly inferred that the females of CWSB mated multiply to increase reproductive fitness by laying more number of eggs. Females that have mated singly laid only one clutch of eggs (rarely two) whereas that have mated multiply has laid more clutch of eggs (12.2 on an average). Every time female has to lay a clutch of eggs means she has to mate with a male. The possible explanations, for this kind of intermittent mating and egg laying pattern is may be either i) female derives benefits from multiple mating or may be because of ii) life history strategies that make beetle to exhibit behaviour or may be both.

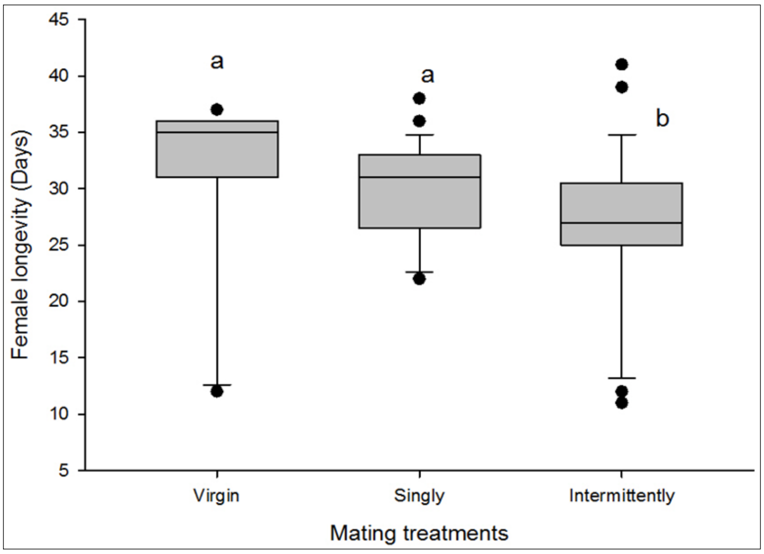

Fig 6: Female longevity in days (Means ± SD) produced in Experiment 4. Treatments that are not significantly different (Kruskal-Wallis method, α = 0:05) are indicated by the same letter.

4.1 Females derives benefits from multiple mating: Proximate reasons for polyandry

Multiply mating female gets the benefit of nuptial gift from males as nutritional source for the ovarian development (Gwynne, 1997 [8]). But this phenomenon has not been observed in this mating system where female approached male in the mating event and male just mated. Another benefit of multiple mating to female is reduction in the male harassment (Arnqvist, 1989 [1]). Again in this case, male are relatively smaller than female (Visitpanich, 1994a [23]) and no clasping structure to hold the female forcefully in the mating process. The proximate benefit the female can get from multiple mating is replenishment of depleting sperm supply (Thornhill and Alcock, 1983 [20]). To prove this, need to do dissection experiments to correlate the amount of sperm in the spermatheca, number of matured eggs in ovary and number of eggs laid recently. In *Tenebrio molitor*, Drnevich *et al.* (2001 [4]) did the dissection experiments to test same hypothesis and they observed from the dissection that i) Female received few or no sperm from the single mating as a result laid few or no eggs. ii) When sperm supply runs out, female started laying infertile eggs. iii) Then there is no correlation between sperm in the spermatheca and number of eggs laid – indicating very low sperm use efficiency. Probably these were the few among the many reasons (genetic benefit of polyandry) for which female *T. molitor* mating multiply.

In general, the act of multiple mating itself has some positive effects on the female fitness because it may directly stimulate the female egg production (Opp and Prokopy, 1986 [14]) and also mere presence of larger number of viable sperms in the female genital tract is known to stimulate egg production (Gromko *et al*., 1984 [7]). An ample and diverse supply of sperms may also increase the female fertility (Tregenza and Wedell, 1998 [21]). Apart from the above mentioned reasons, the large number of accessory gland substances, typically various proteins that are transferred to female with male ejaculates are known to stimulate egg production (Eberhard and Cordero, 1995 [6]; Eberhard, 1996 [5] and Klowden, 1999 [12]).

In further experiments, hoping to examine insemination rate, sperm use efficiency and response of female to depleting sperm supply and also the role of seminal fluid in the female receptivity of CWSB. But from the spent male replacement experiment, it may conclude that significant increase in the fecundity of 12 day old female was because of increased mating frequency with one day virgin male. This may probably indicated, sperms run out in the female spermatheca or short fall of viable sperms. Even though female had matured ovaries in the oviduct, she was not able to lay eggs is another evidence for sperms run out or lack of viable sperms (personal observation). Still this needs further investigations via dissection experiments to collect the direct evidences.

Another interesting observation in a single pairing event, the female CWSB copulated multiply with the male (on an average 5.07 ± 1.49 times) and started moving forward in order to escape from the mounted male. This may be either because of females appears to have an internal mechanisms for assessing the amount of sperm conceived which are sufficient to lay one clutch of eggs or to avoid prolonged copulation by males (when given with multiple male in mating box). The rejecting mounted male by female is the important behaviour that help female to remate with yet another male. When given with multiple males, female prefer different male every time – indicating that female want to derive genetic benefit of polyandry (personal observation).

4.2 Life history strategies of CWSB.

Above all CWSB has very interesting life history strategies. There are two peaks of emergence in a year, one is in winter and other is in summer. There is protandry in the winter flight period (unpublished data). Immediately after emergence beetles started mating, but laid eggs only after 24 hours of pre-oviposition period (Visitpanich, 1994a [23]). Then after they mated intermittently and laid eggs in clutches and on an average a single female mated 12.2 times in their lifespan and deposited 85.71% of eggs within such six clutches. Then they lived as spent females. Provided potential males all the times in the mating event – female had significantly reduced lifespan than that of virgin. In contrary, availability of potential mates all the time may not be true situation in nature, because mate finding is tough task and requires much more time in complex multistoried cropping habitat of shade coffee. There must be much sophisticated life history strategies, that making female beetle to find a mate and also healthy host plants to lay eggs. There may be a relationship between these two events (host and mate finding) like in many birds male emerge first and build nest for female or select egg laying site by holding territories (Kokko *et al*., 2006 [13]).

For this statement there is an indirect evidence that, CWSB infestation is spreading from lower altitude area to high altitude, despite of the fact that the CWSB is not good flyer (Visitpanich, 1994a [23]). In 1838, when Stokes [18] reported it from Manjarabad Taluk in Hassan district where rainfall is less than 100 inches per annum and its infestations were rarely seen and not considered as serious pest in the area above 3500 feet altitude (Subramanian, 1934 [19]). Presently it has spread to high elevation mountain range of Karnataka (Chandra-Drona range) (Venkatesha, 1999 [22]). The point here is, early emerging male selecting healthy plant for female to lay egg and female approaching male in the mating event. There is a selection pressure on male to hold healthy plant every time to get female to mate and hence infestation is spreading to higher altitude. In order to connect all these dots, there must be an extensive study need to be carried out to answer the evolution of intermittent mating and egg laying, protandry with female mating multiply and field experiment to prove territory holding male behaviour.

4.3 Trade-off between polyandry and longevity

Multiple mating directly affect the female longevity in number of ways. The act mating itself is known to carry number of costs to females such as general time and energy costs (Daly, 1978 [3]; Thornhill and Alcock, 1983 [20]; Watson *et al.*, 1998 [24]) and also the physical injury (Parker 1979 [15]; Helvensen and Helvensen, 1991 [9]) and parasitic/pathogen infection (Hurst *et al.*, 1995 [10]). These costs may reduce the longevity of females.

5. Conclusion

In conclusion, CWSB female mated intermittently with a given male to increase fecundity with a cost of significantly reduced lifespan. But it’s very difficult to short list the proximate reason behind the evolution of intermittent mating and eggs laying because still lots of questions regarding life history strategies are needs to be answered.

6. Acknowledgement

Author is thankful to the Department of Agricultural Entomology, University of Agricultural Sciences, GKVK, Bengaluru and also R&D Café Coffee Day, Chandrapore estate and lab, Chikmagalur for providing necessary facilities to conduct these experiments. Author is also thankful to the Coffee Board, Balehonnur (Chikmagalur) for providing funds during the first year of the study period.

7. References

1. Arnqvist G. Multiple mating in a water strider: Mutual benefits or intersexual conflict? Animal behaviour. 1989; 38(5):749-756.
2. Birkhead TR. Sperm Competition in Birds. Trends in ecology and evolution. 1987; 2(9):268-272.
3. Daly M. The cost of mating. American Naturalist. 1978; 112(986):771-774.
4. Drnevich JM, Papke RS, Rauser CL, Rutowski RL. Material benefits from multiple mating in female mealworm beetles (*Tenebrio molitor* L.). Journal of Insect Behaviour. 2001; 14(2):215-230.
5. Eberhard WG. Female Control: Sexual Selection by Cryptic Female Choice. Princeton University Press, Princeton, New Jersey, 1996, 3-43.
6. Eberhard WG, Cordero C. Sexual selection by cryptic female choice on male seminal products: a new bridge between sexual selection and reproductive physiology. Trends in Ecology and Evolution. 1995; 10(12):493-496.
7. Gromko MH, Newport ME, Kortier MG. Sperm dependence of female receptivity to remating in *Drosophila melanogaster*. Evolution. 1984; 38(6):1273-1282.
8. Gwynne DT. The evolution of edible “sperm sacs” and other forms of courtship feeding in crickets, katydids, and their kin (Orthoptera: Ensifera). The Evolution of Mating Systems in Insects and Arachnids. Cambridge University Press, Cambridge, 1997, 110-129.
9. Helversen DV, Helversen OV. Pre-mating sperm removal in the bush cricket *Metaplastes ornatus* Ramme (Orthoptera: Phaneropteridae). Behavioral Ecology and Sociobiology. 1991; 28(6):391-396.
10. Hurst GD, Sharpe RG, Broomfield AH, Walker LE, Majerus TO, Zakharov IA *et al*. Sexually transmitted disease in a promiscuous insect, *Adalia bipunctata*. Ecological Entomology. 1995; 20(3):230-236.
11. Jennions MD, Petrie M. Variation in mate choice: a review of causes and consequences. Biological Reviews of the Cambridge Philosophical Society. 1997; 72(2):83-327.
12. Klowden MJ. The check is in the male: male mosquitoes affect female physiology and behaviour. Journal of the American Mosquito Control Association. 1999; 15(2):213-220.
13. Kokko H, Gunnarsson TG, Morrell LJ, Gill JA. Why do female migratory birds arrive later than males? Journal of Animal Ecology. 2006; 75(6):1293-1303.
14. Opp SB, Prokopy RJ. Variation in laboratory oviposition by *Rhagoletis pomonella* (Diptera: Tephritidae) in relation to mating status. Annals of the Entomological Society of America. 1986; 79(4):705-710.
15. Parker GA. Sexual selection and sexual conflict. Sexual Selection and Reproductive Competition in Insects (Ed. by M. S. Blum & N. A. Blum). Academic Press, New York, 1979, 123-166.
16. Reynolds JD. Animal breeding systems. Trends Ecology Evolution. 1996; 11(2):68-72.
17. Ridley M. Mating frequency and fecundity in insects. Biological Reviews. 1993; 63(4):509-549.
18. Stokes H. Report of the Commissioner, Mysore. India, 1838.
19. Subramaniam TV. The Coffee stem borer. Mysore Department of Agricultural Entomology. Mysore, India, 1934, 11-12.
20. Thornhill R, Alcock J. The Evolution of Insect Mating Systems. Harvard University Press, Cambridge, MA, 1983, 300-490.
21. Tregenza T and Wedell N. Benefits of multiple mates in the cricket *Gryllus bimaculatus*. Evolution. 1998; 52(6):1726-1730.
22. Venkatesha MG. Why is white stem borer persistently a serious pest in arabica coffee plantations? Indian Coffee: Bulletin of the Indian Coffee Board. 1999; 63(1):11-14.
23. Visitpanich J. The biology and survival rate of the coffee stem borer, *Xylotrechus quadripes* Chevrolat (Coleoptera, Cerambycidae) in Northern Thailand. Japanese Journal of Entomology. 1994a; 62(4):731-745.
24. Watson PJ, Arnqvist G, Stallman RR. Sexual conflict and the energetic costs of mating and mate choice in water striders. American Naturalist. 1998; 151(1):46-58.