International Journal of Zoology Studies
www.zoologyjournals.com
ISSN: 2455-7269
Received: 12-11-2021, Accepted: 27-11-2021, Published: 13-12-2021
Volume 6, Issue 6, 2021, Page No. 6-9

Male age influence on male mating ability in three geographical strains of *Drosophila malerkotliana*

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Abstract
Females may obtain greater direct benefits by mating with the male showing some types of signals, independently of the costs independent in such species. Five to six days old virgin females and a male (young/middle aged and older) were aspirated into an Elens-Wattaiux mating chamber and observed for an hour. Among three geographical strains males of Mysore strain had inseminated greater number of females in a given unit of time while males of Chamarajanagar strain had inseminated least number of females in a given unit of time.

Keywords: *Drosophila malerkotliana*, male age, male mating ability, remating

Introduction

In sexually reproducing organisms, each of the offspring has one father and one mother, so the average reproductive success is equal for both males and females. However, if a male gains a disproportionate share of reproduction, he will take away reproductive opportunities from other males, leading to a high reproductive variance among males. On the contrary female will not take away reproductive opportunities from other females, leading to a smaller variance in reproductive success. The higherthe reproductive variance, the stronger the effects of sexual selection. Studies have also suggested pathways through which females mated with preferred male may obtain direct benefits. Females may obtain greater direct benefits by mating with the male showing some types of signals, independently of the costs independent in such species. The benefits that male animals provide to females includes nutrients, body parts, and secretions (Gwynne, 1982; Sakaluk, 1984; Thornhill, 1976) [9, 15, 17] access to resources on a territory, including refuges, oviposition or nesting sites, and food (Howard, 1978) [11] more sperm, more viable sperm, or better fertilization ability (Drnevich *et al.*, 2001; Matthews *et al.*, 1997) [5, 14] a variety of products transferred in seminal fluid, including nutritive and defensive compounds (Iyengar and Eisner, 1999; Markow, 1988) [12, 13] male protection, including protection from harassment by other males and from predators (Borgia, 1979) [3] and male care of offspring, including care that frees females to engage in other activities and care that increases offspring fitness (Hill, 1991) [10]. Since these male contributions can affect female survivorship reproduction, offspring survivorship and reproduction. For two male contributions may refer to as direct benefits while later two male contribution may referred to as indirect benefits (Grether, 2010) [7].

Studies have also pointed out that females may also benefit from mating with certain males not because of the benefits they provide instead these males can impose lower costs on females. For example, females might risk damage from mating with certain male and she preferred to mate with males, which can lower the magnitude of damage (Grether, 2010) [7]. The present study has been undertaken in *D. malerkotliana*. It is a cosmopolitan species and belongs to a member of the *bipectinata* complex of the *ananassae* subgroup (Bock, 1971; Bock and Wheeler, 1972) [1, 2]. It has a wide ecogeographical distribution ranging from India through South east Asia and New Guinea to Fiji and Samoa in the Pacific (Bock and Wheeler, 1972) [1].

Materials and Methods

Experimental stocks: Progenies of 150 naturally inseminated iso female lines of *D. malerkotliana* collected separately at three geographical strains of Karnataka i.e. Mysore, Chamarajanagar and Bellur were used to establish experimental stocks. In each generation progeny obtained were mixed together and redistributed to 20 different culture bottles containing wheat cream agar medium each with 20 males and 20 females. These culture bottles were maintained them at 21 ± 1°C at a relative humidity of 70% using a 12: 12 h light: dark cycle. This procedure was continued for 2nd generations to allow them to acclimatize to the laboratory conditions. At the 3rd generation, synchronized eggs (± 30 min) were collected separately from each of three experimental stocks using Delcour's procedure (1969) [4].When adults emerged, virgin females and unmated males were isolated within 3 h of eclosion and were aged as required for the experiment.

Assigning of age classes to males; For obtaining males of different age classes before the start of experiment, longevity of male *D. malerkotliana* was studied by transferring unmated males into a vial containing wheat cream agar medium once a week and maintained them in above lab condition. This procedure was continued until their death and longevity was recorded. A total of 50 replicates was made and mean longevity was found to be 62±2days. In addition to this, mating activities of males were also studied from day 1st of their eclosion until 60th day. Results showed that showed least male courtship activities were found at 1st day, whereas from 2nd day and onwards (up to 46-47 days) male showed all the courtship activities and all of them mated with the female. At 47th day male showed least courtship activities and rarely mated with the female. Hence age classes assigned to males were 2-3 days for young, 24-25 days for middle aged and 46- 47 days for older males. The first set of flies emerged were allowed to age for 46-47 day (to obtain old males). When these flies reached 20th day the next set of new flies were isolated and were allowed to age for 24-25 days (to obtain middle aged males). When the second set of flies reached 20th day and the first set of flies reached 47th day, then the new set of flies was isolated and were aged for 2-3day (to obtain young males). This procedure helped us to culture all three male age classes young, middle aged and old and to conduct the experiment at the same time in same environment.

Male age influence on percentage of male remating ability

To study male age effect on male remating. (Fig.1) 5-6 day old virgin female and a male (young/middle-aged/old) were aspirated into an Elens-Wattaiux mating chamber (Elens Wattaiux, 1964) [6] and observed for an hour. Flies which did not mate within an hour were discarded. If the flies mate, they were allowed to complete copulation and the duration of copulation was recorded (time between initiation to termination of copulation of each pair). Soon after mating the mated female was transferred to a vial containing wheat cream agar medium to check for larval activity. Mated male was once again provided with 5-6 days old virgin female and observation was continued up to an hour. If the second female that did not mate within the time was equally discarded. If mating occurred, the duration of copulation was recorded and mated female was transferred to a new vial to observe larval activity. A total of 50 trials was carried out separately for each of the three male age classes (young-50, middle aged-50, old male-50). The percentage of male remating ability was recorded.

Male age influence on male mating ability and male fitness

Five to six days old virgin females and a male (young/middle aged and older) were aspirated into an Elens-Wattaiux mating chamber and observed for an hour. Flies which did not mate within an hour were discarded. If the flies mated, they were allowed to complete copulation and duration of copulation was recorded (time between initiations to termination of copulation of each pair). Soon after mating mated female was transferred into a vial containing wheat cream agar medium every 24 hours. This was continued until the death of the female. Mated male was once again provided with 5-6 day virgin female and observation was continued up to an hour. If mating did not occur with second female within the time, then it was considered as male mated once in an hour. If mating occurred with the mated second female was allowed to complete copulation and the duration of copulation was recorded. Mated female was transferred to a vial containing wheat cream, medium once in 24hours and was continued up to the death of the second female. The total number of eggs laid and the progeny produced by male mated with the first and second female was recorded. Fifty male mated with one female in an hour and mated with two females in an hour were separately used for each male age class (young-50, middle aged-50, old male-50). Data of male mated with second female were added together to calculate duration of copulation, fecundity and fertility. Duration of copulation, eggs laid and the progeny produced data by male mated with one female within 1 hour, male mated with two females within 1 hour. Male mated with first female and second female was subjected to two way ANOVA followed by Tukey's post hoc test

Results

Average male mating ability of young, middle aged and older males of all the three geographical strains of *D. malerkotliana* is given in Figure 2. It was found that in all the three geographical strains middle aged male had inseminated more number of females in a given unit of time (1hour) than young and old males. Among three geographical strains males of Mysore strain had inseminated greater number of females in a given unit of time while males of Chamarajanagar strain had inseminated least number of females in a given unit of time (1hour). The mean male mating ability data subjected to Two-way ANOVA followed by Tukey's post hoc test showed significant differences in the mean male mating ability between male age, three geographical strains and interaction between geographical strains and male age (Table.1). Tukey's post hoc test showed that middle aged males inseminated significantly more number of females than those of middle aged and old males

Conclusion

Age in male insects may affect the reproductive activities including success in intra sexual counters, sperm precedence and female fecundity etc. However, male reproductive success depends on the number of females inseminated by a male in a given unit of time. Therefore, male remating ability forms one of the strategies for male fitness. Males of all the three age classes of *D. malerkotliana* remate within an hour suggesting that ability to inseminate more than one female becomes the inherent ability of male to increase his fitness. Singh and Singh (1988) [9] studied *D. ananassae* and noticed differences in male remating ability among different geographical populations. Guruprasd (2008) [8] also studied *D. bipectinata* are found differences in male remating in four geographical populations. In the present study in *D. malerkotliana* middle aged males have a greater percentage of remating than young and old males. This suggests that the age has significant influence on the percentage of male remating ability (Fig 1).The greater percentage of remating ability of middle aged male could be attributed to the occurrence of age specific variation in male fitness. In our study it was also noticed that in all the three male age classes all the males have not remated and this varies with different male age classes. This raises the question that if male remating indeed increases male fitness, then all the males have to undergo remating. In the present study copulation duration and progeny production of male mated with one female in 1 hour and male mated with two females in 1hour were assessed to rule out the implication of male remating in *D. malerkotliana.* Figure It was observed in this study that male mated with two females in an hour had copulated longer and laid a significantly greater number of eggs and progeny compared to male mated with one female in an hour (Fig 2 and table 1). This suggests that male remating is really advantageous and this enhances fitness. The result of our study also conforms to the result obtained from where it was observed that multiple mating in *D.*

melanogaster was advantageous to males and selection of males can produce a correlated response in females was equally observed that young, middle aged and old males *D. malerkotliana* remated males had greater productivity. Our study conforms to the study of in *Drosophila ananassae*. Furthermore, the present study is in consonance with on *D. malerkotliana and D. bipectinata*. Thus, it was noted that in species of *Drosophila* male remating ability is not only influenced by genetic and environmental factors. Figure 2 and Table 1 reveals that male investment to first and second mated females in 1 hour varies. Copulation duration of male with 1st female was found to be more or less the same. While male mated with 2nd female, the middle aged male copulated longer compared to old or young male that copulated with 2nd female. In contrast to this the number of eggs laid by male mated with first female was significantly greater compared to the male mated with 2nd female. This suggest male invest more in the1st female he encountered in 1 hour. From the results it can be seen that all age classes of males had inseminated and invested more towards fecundity in the 1st female than in the 2nd female that had mated. This result was found to be similar in males of all age groups, suggesting males to have invested more in 1 female than 2nd female.

Our results confirm the work of Singh and Singh in *D. ananassae* and Guruprasad in *D. bipectinata*. From our results and other studies it can be demonstrated that older males increase their fitness by remating effectively with a number of individuals. At the same time make effective investment in the number of sperms and other accessory gland proteins necessary for effective fecundity and fertility evidently seen. Figure 2 and table 1 showed that female of *D. malerkotliana* mated with middle aged male had produced the greatest number of progeny (fertility) compare to old age and young males. Further second mated females with middle and young male had higher fertility rate, whereas first female that mated with old male produced highest progeny number than male mated with 2nd female. These studies also suggest differentiates investment of male mated with 1St and 2nd females. Thus, these studies suggest that in *D. malerkotliana* male remating increases male fitness.

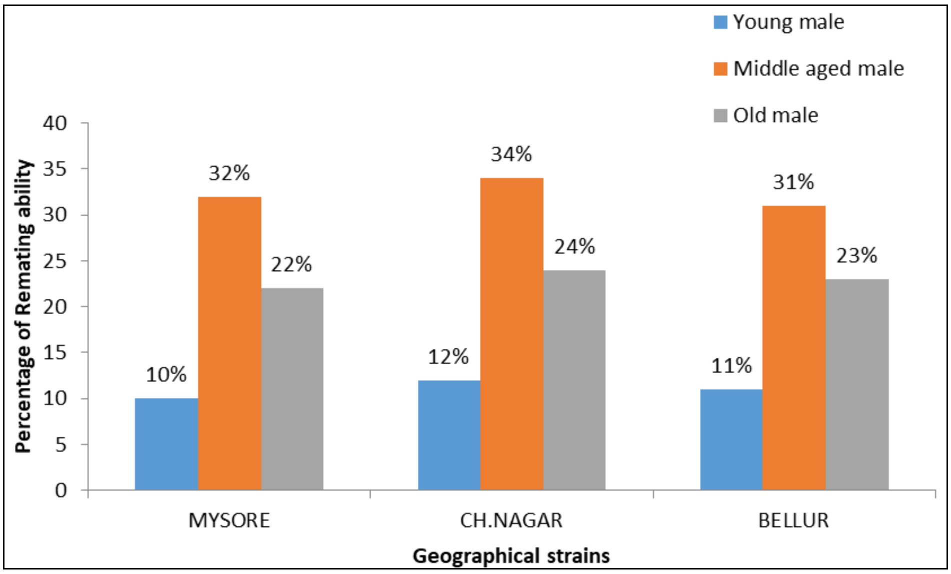
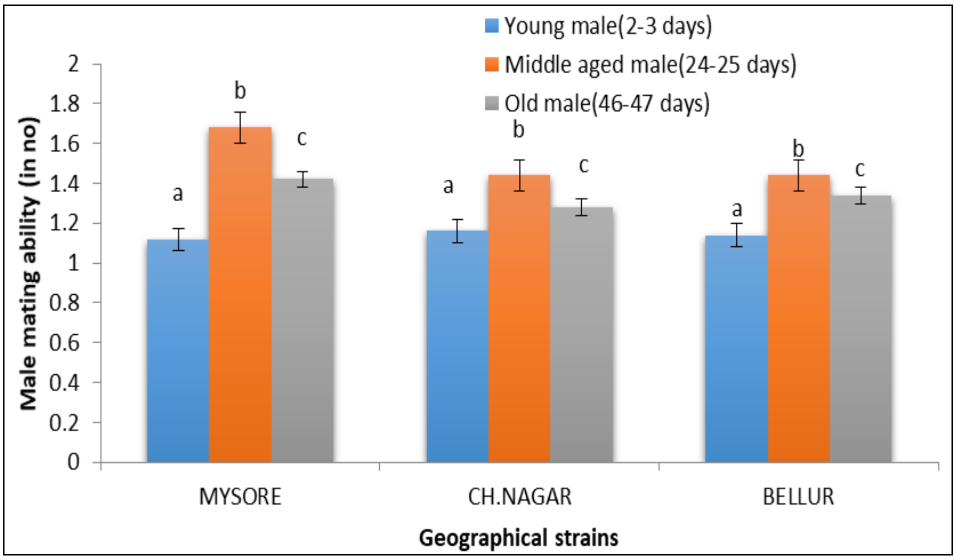


Fig 1: Percentage of Remating ability of male mated twice with femalewithin one hour in *D. malerkotliana*



Different letters on the bar graphs indicate significant difference by Tukey's post hoc test at 0.05 level.
Note: Male age –Young male (2-3 days) middle aged male (24-25days) old male (46-47days)

Fig 2: Male age influence on male mating ability of *D. malerkotliana*

Table 1: Two- way ANOVA of male age influence on male mating ability of *D. malerkotliana*

Parameter	Source	Sum of Square	df	Mean square	F-value
Male mating ability (in no)	Age	1.151	2	0.576	1.750^{NS}
	Strains	10.858	2	5.429	16.509*
	Interaction between strain and male age	1.302	4	0.326	0.990^{NS}
	Error	145.02	441	0.328844	
	Total	961	450		

*Significant at 0.001; NS- Non significant

Acknowledgements
The author is grateful to the Chairman, Department of Studies in Zoology, University of Mysore, for providing facilities to carry out this work

References

1. Bock IR, Wheeler MR. The *Drosophila melanogaster* species group University of Texas Publications,1972:7213:1-102.
2. Bock, I. R. Taxonomy of the *Drosophila bipectinata* species complex. Univ. Texas Publication,1971:7103:273-280
3. Borgia G. Sexual selection and the evolution of mating systems. In Sexual selection and reproductive competition (ed. M. Blum and A. Blum), 1979, 19-80. New York, NY: Academic Press.
4. Delcour JA rapid and efficient method of egg collecting. *Drosophila Information Service*,1969:44:133-134.
5. Drnevich JM, Papke RS, Rauser CL, Rutowski R. Material benefits from multiple mating in female mealworm beetles (Tenebrio molitor L.). *Journal of Insect Behavior*,2001:14:215-230.
6. Elens AA, Wattiaux JM. Direct observation of sexual isolation. *Drosophila Information Science*,1964:39:118-119.
7. Grether GF. The evolution of mate preferences, sensory biases, and indicator traits. *Advances in Study Behavior*,2010:41:35-76.
8. Guruprasad BR, Hegde SN, Krishna MS. Positive Relation between Male Size and Remating Success in Some Populations of *Drosophila bipectinata*, Zoological Studies,2008:47(1):75-83.
9. Gwynne DT. Mate selection by female ka- tydids (Orthoptera: Tettigoniidae): Concephalus ni- gropleurum. *Animal Behaviour*,1982:30:734-738.
10. Hill GE, Plumage coloration is a sexually selected indicator of male quality. *Nature*,1991:350:337-339.
11. Howard RD. The evolution of mating strat- egies in bullfrogs, Rana catesbeiana. *Evolution*,1978:32:850-871:1988.
12. Iyengar VK, Eisner T. Heritability of body mass, a sexually selected trait, in an arctiid moth (*Utetheisa ornatrix*). *Proceedings of National Academy of Sciences*, USA,1999:96:9169-9171.
13. Markow TA. Reproductive behavior of *Drosophila* 1828 S. Pitnick and F. Garcı´a Gonza´lez *Size and harm melanogaster* and *D. nigrospiracula* in the field and in the laboratory. *J. Comp. Psychol*,1988:102:16:173.
14. Matthews IM, Evans JP, Magurran AE. Male display rate reveals ejaculate characteristics in the Trinidadian guppy Poecilia reticulate. Proceedings of Royal Society of London Biological Science,1997:264:695-700.
15. Sakaluk SK, O'day H. Hoechst staining and quantification of sperm in the spermatophore and spermatheca of the decorated cricket, *Gryllodes supplicans* (Orthoptera: Gryllidae). *The Canadian. Entomologist*,1984:116**:**1585-1589.
16. Singh AK, BN Singh, An extreme linkage between inversions in *Drosophila ananassae*. Current Science,1988:57:400-402.
17. Thornhill R. Sexual selection and nuptial feed- ing behavior in Bittacus apicalis (Insecta: Mecop- tera). American Naturalist,1976:110:529-548.