

know why the excision rate in cross A is significantly higher than in cross B. For the three *NA* stocks, the excision rates in cross A were significantly lower than the corresponding control rate (and also significantly lower than the control rate in cross B); however, the excision rates for the *NA* stocks in cross B were not lower than the rate for the cross B control. These observations indicate that *P*-element excisions in the male germ line are repressed, but only when the *NA* element is inherited maternally. Thus, regulation of *P* activity by *NA*, as by other telomeric *P* elements, involves a maternal effect, and an *NA* element that passes patroclinously from father to son loses its ability to repress hybrid dysgenesis.

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Influence of age on mating and fitness of *Drosophila melanogaster*.

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Abstract

The effect of age on mating time, remating duration, quantity of ACP transferred, fecundity, and productivity was studied within and between 7 and 26 days old *Drosophila melanogaster*. Mating time, accessory gland secretory protein transferred, fecundity, and productivity of 1st mating is more than the 2nd mating irrespective of age. The remating duration among young and old flies is almost similar, whereas it is more in different aged flies mating. Mating time, fecundity, and productivity are more in young flies compared to other combinations. In old flies mating time, fecundity, and productivity are more than the different aged flies. The fecundity is more if the female is young and productivity is more if the male is young in different aged flies mating. **Key words:** *Drosophila*, mating behavior, fecundity, productivity, age.

Introduction

Sexual reproduction occurs in a wide range of organisms. Reproductive capacity is particularly a good index of fitness in organisms that go through repeated cycles of rapid population growth, and it has evolved as a way for species to maximize their potential of survival. Mating is the most important and fundamental process of reproduction. Male fitness depends largely on the

number of matings and the average number of progeny sired from each mating; female fitness depends on the number of progeny production (Bateman, 1948; Singh and Singh, 2000, 2004).

Ageing is a multifaceted phenomenon that occurs in most of the species. In sexual reproduction, age is regarded as one of the most important factors for mate choice. It depends on genetic quality in sexual reproduction of insects. The effect of age has been studied on fitness parameters, such as mating choice, mating time, duration between matings, fecundity, productivity, and viability in different species of *Drosophila* (Moore and Moore, 2001; Anderson *et al.*, 1973). *Drosophila* is proving to be a powerful system for understanding the age dependent mating behavior.

Mating time and duration between matings are important components of *Drosophila* mating behavior. Mating time is the time from initiation of copulation till the time of release or departures of males and females. Mating time was observed in many species of *Drosophila*. It varies from 25 seconds (*D. polychaeta*) to 137 minutes (*D. acanthoptera*) (Markow, 1996). In *Drosophila* multiple mating is common in both natural and laboratory conditions. First and second mating has been studied. Whereas the male and female after 1st mating take some time to remate (2nd), it is called duration between matings. Male takes less time to remate than female in all the species. Remating has been studied in different species of *Drosophila*. It ranges between 1 hour to 5 days in some females, and some females (*D. differens*, *D. teteroneura*, *D. subobscura*, *D. sylvestris*, *D. acanthoptera*) rarely or never remate (Markow, 1996; Singh and Singh, 2004).

Accessory gland secretory proteins (ACPs) are transferred from male to female genital track during mating. It induces morphological and behavioral changes in mated females. ACPs are major components of seminal fluid; it contains about 80 proteins and peptides in *D. melanogaster* (Chen, 1984; Chapman and Davies, 2004). The synthesis of ACP increases up to 7 days. The quantity of secretion has been estimated in few species of *Drosophila*. It varies from 50 to 84% in *D. rubida* and *D. ananassae*, respectively, and 1/3 amount of the quantity is released during the 1st mating (Chen, 1984; Shivanna and Ramesh, 1995; Ravi Ram and Ramesh, 2001; Hiremani and Shivanna 2010).

Fecundity is the number of eggs laid per female after mating. It has been studied in different species of *Drosophila* on different aspects and it varies from 98 to 333 (Ashadevi and Ramesh, 1999; Harini and Ramachandra, 2007; Harini, 2010; Rezaei, 2012; Lushchak *et al.*, 2013; Pavkovic and Kekic 2013). Productivity is the number of progeny produced by each mated female. It has been observed in many species of *Drosophila* on different aspects. It varies from 55 to 500 in *D. hydei* and *D. melanogaster*, respectively (Markow, 1996; Ashadevi and Ramesh, 1999; Roopashree *et al.*, 2001).

Most of the studies are made in different species and various aspects. The relationship between mating activities, quantity of ACP transferred during mating, and effect of age on fitness has not been analyzed. In view of this, the present work has been taken to study the relationships between above aspects in *D. melanogaster*.

Materials and Methods:

Drosophila melanogaster was obtained from *Drosophila* stock center, Department of Zoology, University of Mysore, Mysore. The flies were cultured in a standard wheat cream agar medium, prepared as per the procedure described by Shivanna *et al.* (1996) and maintained at a constant temperature of $22 \pm 1^\circ\text{C}$. The virgin males and females were aged up to 7 (young) and 26 (old) days and used for experiments.

Male and female flies were allowed to mate in a vial (9×2.5 cm). After initiation of mating, the 1st mating time was recorded. After 1st mating, the female was removed and another virgin

female was introduced. Time required for remating and 2nd mating time were noted as per the procedure described by Singh and Singh (2000, 2004). The mated and unmated males were dissected in an insect saline, accessory glands were isolated and fixed in 95% ethanol, and then the glands were washed with methanol and chloroform (1:1) and dried at 37°C in oven. The glands (5 pair) were dissolved in 40µl of sample buffer. Samples were prepared as per the procedure described by Shivanna and Ramesh (1995). Quantity of protein present in the samples was estimated using the procedure of Bradford (1976). Optical density of the solution was measured using a spectrophotometer at 595 nm. The quantity of protein present in the sample was calculated by extrapolation with BSA as the standard. The quantity of protein ejected during mating was calculated by subtracting the amount of protein of mated male from the amount of protein of unmated male.

The mated females were kept in separate vials; eggs were counted for a period of 30 days. Yeast was added to the vials containing larvae for feeding. After pupation of the larvae, the pupae were counted to know the productivity. All the experiments were conducted within young age, old age, and between young and old aged flies.

Table 1. Mean \pm SD of mating time, duration between mating, quantity of protein transferred, fecundity and productivity in *D. melanogaster* (♂- Male, ♀- Female).

Age of flies ♂ x ♀	Number of mating	Mating time (min)	Duration between mating (min)	% of Protein transferred	Fecundity	Productivity
7 x 7	I	20.4 \pm 1.21	27.4 \pm 10.88	42.67	429.0 \pm 32.80	261.4 \pm 21.41
	II	18.4 \pm 1.60		11.11	293.2 \pm 29.25	205.0 \pm 26.95
26 x 26	I	23.6 \pm 1.54	29.6 \pm 8.78	36.0	245.4 \pm 37.04	96.4 \pm 20.68
	II	16.6 \pm 1.33		19.0	214.0 \pm 28.30	65.6 \pm 10.31
7 x 26	I	18.6 \pm 1.46	50.2 \pm 11.07	39.12	188.0 \pm 25.5	56.4 \pm 13.01
	II	11.4 \pm 2.89		24.50	127.8 \pm 5.60	27.0 \pm 6.51
26 x 7	I	15.4 \pm 2.94	43.8 \pm 12.60	10.0	256.8 \pm 36.22	48.4 \pm 10.70
	II	14.2 \pm 1.74		6.0	206.0 \pm 16.99	18.0 \pm 6.28

Results

Table 1 reveals that the mating time of young flies is more than other combinations except 1st mating of old flies. First mating time is more than second mating time in all the combinations of mating. The remating duration of young flies and old flies is almost similar, whereas it is half of the duration of old and young flies mating. The quantity of ACP transferred in first time mating of young flies is four times more than the second time mating. Protein quantity of first mating is double that of the second mating when old flies were crossed. When old and young flies were crossed, the quantity of secretion transferred in second mating is slightly more than half the quantity of the first mating. The number of eggs laid by the female mated with second time mated male produced 2/3 of the female mated with bachelor male in all the crosses. Productivity/pupa produced by the female mated with second time male is almost 3/4 of the first female mated with bachelor male in young flies and old flies. When young male mated with old female, the pupa produced is 1/2 in the second mating than the first mating. Whereas old male mated with young female the pupa produced is less than 1/2 in the second mating than the first mating. The difference in number of egg and pupa of 1st and 2nd mating is highly significant (egg of 1st mating: $F = 9.718$, 2nd mating: $F = 6.6$. pupa of 1st mating $F = 10.73$, 2nd mating $F = 19.961$ { $df_1 = 3$, $df_2 = 116$ }).

Discussion

D. melanogaster is a domestic, cosmopolitan and most extensively characterized multi cellular organism used over more than 100 years for research. Since then it has been used for behavior, reproductive and developmental biology, ecology, evolution, population biology, genetics, and other studies.

Present study reveals that in *D. melanogaster* the first mating time is 2 minutes more than 2nd mating of 7 days old flies. In 26 days old flies it is 7 minutes more than 2nd mating, which is similar when young males mate with old females. It is 1 minute more in 1st mating than 2nd mating when young females mate with old males. These results are par with Pavkovic and Kekic (2009). Singh and Singh (2004) reported the contrasting result in *D. melanogaster*. Comparison of mating time of different age crosses showed significant variation. *Drosophila* females take more time to remate when compared to males (Markow 1996; Singh and Singh 2004). In the present study males have been given the chance to remate; it showed significant difference in remating duration when crossed with same and different aged females.

Accessory gland proteins contribute to important reproductive processes that lead to fertilization in species. Maximum accumulation of accessory gland takes place at 7 days in different species of *Drosophila*, such as in *D. melanogaster* (80.0%), *D. n. nasuta* (72.0%), *D. s. neonasuta* (70.0%), *D. rubida* (50.0%), and *D. pararubida* (54.20%) of *D. immigrans* group. *D. ananassae* and *D. varians* are about 84.84% and 58.13%, respectively (Chen, 1984; Ravi Ram and Ramesh, 2001; Hiremani and Shivanna, 2010). In the present study we selected 7 day old flies as the young age and 26 day old flies as the old age. The difference in transfer of protein in different crosses varies significantly with age of the flies.

Fecundity and productivities are the important components of fitness treaties and are increased with remating (Singh and Singh, 2004). Fecundity is usually considered as a female fitness component. It is affected by the presence of males and other factors, including male accessory gland protein in *D. melanogaster*, *D. ananassae*, *D. nasuta*, *D. albomicans* (Serradilla and Ayala, 1983; Chapman, 2001; Roopashree *et al.*, 2001; Harini, 2010; Rezaei, 2012; Lushchak *et al.*, 2013; Pavkovic and Kekic, 2013). To overcome the effects of different factors in the present study, standard conditions were maintained. Table 1 reveals that fecundity and productivity of young flies is more compared to other crosses. The fecundity and productivity of old flies is more than mixed fly crosses and less than young flies. The fecundity and productivity is more in 1st mating than 2nd mating in all the combinations, irrespective of age.

If same-aged flies mate with each other, mating time, fecundity, and productivity are more and remating duration is less compared to mixed flies. Mating time, duration between mating, quantity of ACP transferred, fecundity, and productivity are affected by the age of the mates.

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Influence of extremely low frequency magnetic field (50 Hz, 0.5 mT) exposure on fitness components of *Drosophila subobscura*.

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Summary

In addition to naturally occurring radiation, magnetic fields which are introduced by a man with the advanced development of industry and technology present an additional factor in environment that could have a significant influence on living systems. Impact of magnetic fields at different developmental stages of biological systems might induce changes on different organizational levels. In this study are presented developmental time, developmental dynamics, and viability of *Drosophila subobscura* after exposure of egg-first instar larvae developmental stage to extremely low frequency magnetic field (50 Hz, 0.5 mT). Exposure for 48 h at egg-first instar larvae developmental stage significantly shortens developmental time and increases viability of *D. subobscura*.

Introduction

Extremely low frequency magnetic fields (ELF-MFs, ≤ 300 Hz) derived from power lines as well as from the majority of household electrical appliances, represent one of the most important scopes for researching in magneto biology (Gandhi *et al.*, 2001; Gauger, 1985). ELF-MFs are in interaction with biological systems' tissues inducing electric fields and currents in them (Mathie *et al.*, 2003). Mostly, studies about effects of ELF-MFs on *Drosophila* are dealing with influence on reproductive behavior and fitness components. Ramirez *et al.* (1983) show decreased oviposition after exposure to pulsated ELF (100 Hz, 1.76 mT) and sinusoidal fields (50 Hz, 1 mT). In addition, there are found increased egg mortality and diminished adults' viability. *D. melanogaster* females and progeny exposed to ELF-MF show weakened oviposition in their subsequent generations (Gonet *et al.*, 2009). Also, Mirabolghasemi and Azarnia (2002) show a significant increase in the number of