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# **RESEARCH ARTICLE**

## SYNTHETIC ENERGY DRINK (BURN) EFFECTS ON MALE MATING ABILITY AND PROGENY PRODUCTION IN DROSOPHILA MELANOGASTER

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### ABSTRACT

Nutrient contents found in the diet have a significant influence on health benefits of an organism. Nowadays a number of synthetic energy drinks are in use; however the quality and quantity of nutrients found in these synthetic energy drinks are different. Present study has been undertaken in *Drosophila melanogaster (D. melanogaster)* to evaluate the effect of synthetic energy (Burn) on male mating ability and progeny production. It was noticed that larvae of *D. melanogaster* has shown increased rate of feeding in natural energy drinks compared to wheat cream agar medium and synthetic energy drinks. Males fed with natural energy drinks had inseminated significantly greater number of females in a given unit time and produced significantly greater number of progeny than those males fed with synthetic energy drinks. Thus this study suggests that synthetic energy drinks reduces male fitness in *D. melanogaster*.

Key words: D. melanogaster, Feeding Behavior, Male Mating ability, Progeny Production.

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# **INTRODUCTION**

In animals, reproduction is one of the important biological processes. It determines the fitness of an organism. In species of *Drosophila*, reproduction involves series of complex mating behavior such as mating latency, mating time, duration of copulation, fecundity, fertility which of all together constitute fitness of an organism (Anderson, 1983). In addition to this, remating of male or female is a strategy for improving the fitness which is more common polygamous populations (Abolhasan and Krishna, 2014). In species of Drosophila in each mating male transfers ejaculatory substance which includes accessory gland proteins and sperms to the mated females. Accessory gland proteins in mated female include egg laving or oviposition and also facilitate fertilizations of sperms into the eggs. Thus male which inseminate more females contribute more progenies to its next generation. According to Gromko (1992) multiple mating is widely believed to be advantageous to males and selection by males can produce a correlated response in females. Many factors are known to affect the male mating ability in spices of Drosophila such as size, age (Hegde and Krishna, 1997; Abolhasan and Krishna, 2014). Diet is another important factor which influences male mating ability and progeny production. Therefore present investigation has been undertaken in Drosophila to study the effect of synthetic energy drinks (Burn) on male mating ability and progeny production.

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### **MATERIALS AND METHODS**

Experimental stock used in the present investigation was Oregon K Strain obtained from Drosophila Stock Center, Manasagangotri, Mysore. Ten females were introduced into Drosophila culture bottle containing standard media (wheat cream agar media) which consist of 100 g of jaggery, 100 g of wheat powder, 8 g of Agar (Agar, boiled in 1000 ml of double distilled water and then addition of 7.5 ml of propionic acid in it). These flies were maintained at 22° C  $\pm$  1°C with a relative humidity of 70% in a 12 hour dark, 12 hour light cycle. Eggs were collected from these flies using Delcour's procedure (1969) and from which 100 eggs were separately placed into separate culture vials, containing wheat cream agar media, natural drink based media (Juice from each of the four fruit viz. apple, pomegranate, orange and banana are extracted separately. Measured quantity of 50 ml of each of such juice is mixed together along with 10 ml of vitamin B12 and 60 ml of carbonated water, finalizing a total volume of 270 ml for the further analysis and treatment) and synthetic drink based media (Burn). Flies obtained from these eggs were used in the present experiments.

**Quantification of food intake in larvae using dye method:** Ten Second instar larvae obtained from normal media were placed separately in a vial containing Normal / Natural / Synthetic energy drink based media treated with 2.5% (W/V) blue to eppendorf tube and frozen. These frozen larvae were homogenized by 200 ml of distilled water; a further 800 ml of distilled water was added. The absorbance was measured at 629 nm using calorie meter. The larvae which were not treated with blue dye were used as the blank. The amount of food taken was measured from the standard graph made from the serial dilution of a blue dye.

*Male mating ability:* To study male mating ability, a 2-3 days old male and a 5-6 days old virgin female were introduced into a mating chamber and observation was made for an hour. If the pair did not mate within an hour, it was discarded. If mating occurred, copulating pairs were allowed to complete copulation. Soon after copulation, the mated female was aspirated into a new vial containing wheat cream agar media for 2 hrs and observed until the death of the female. Total number of progeny emerged from mated females were recorded. Mated male was again aspirated into a new vial containing another 5-6 days old virgin female and were allowed to complete the copulation after which the mated female was transferred to a new vial to record progeny numbers as described above. This process was repeated and number of females inseminated by a single male in 1 hour was recorded and progeny production of all the mated female by single male was also recorded. Twenty five replicates were made separately for male obtained from natural energy/ synthetic energy drinks (Burn) / normal media.

### RESULTS

*Feeding behavior:* Larvae grown in natural energy drink based media had a significantly greater quantity of food intake than the larvae grown on synthetic energy drink based media. Fig. 1 shows the food intake by larvae in a specific unit of time (15 minutes), measured using the 'dye method'. The data obtained from feeding experiment was subjected to one way ANOVA followed by Tukey's Post Hoc Test using SPSS version 20.0, showed a significant difference in the amount of food intake (Table 1).

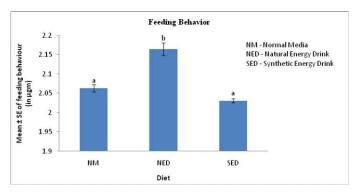


Fig. 1. Effect of Natural and Synthetic energy drink (Burn) on feeding behavior in larvae of *D. melanogaster* 

[Different letters on the bar graph indicates significance at 0.05 level by Tukey's Post Hoc Test]

**Male mating ability:** It was noticed that male obtained from natural energy drink had inseminated significantly greater number of females in 1 hour as compared to males grown in normal media and synthetic energy drink. Mean male mating ability of males grown in natural energy drinks, synthetic energy drinks and normal media are provided in Fig 2. The data obtained from feeding experiment was subjected to one way ANOVA followed by Tukey's Post Hoc Test using SPSS version 20.0, showed a significant difference in the amount of food intake (Table 1).

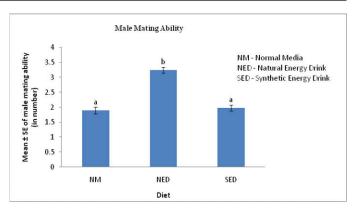


Fig. 2. Effect of Natural and Synthetic energy drink (Burn) on male mating ability in *D. melanogaster*.

[Different letters on the bar graph indicates significance at 0.05 level by Tukey's Post Hoc Test]

**Progeny Production:** It was noticed that progeny production was greater in females mated with males grown in natural energy drink than those females mated with males grown in synthetic energy drinks and normal media. Fig. 3 and table 1 provides data of mean progeny production of females mated by single male in 1 hr.

One way ANOVA followed by Tukey's Post Hoc Test showed that progeny production of females mated to single male obtained from natural energy drinks was significantly greatest compared to females mated with single male grown either in synthetic energy drink or normal media.

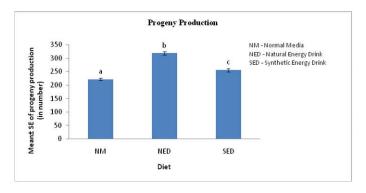


Fig. 3. Effect of Natural and Synthetic energy drink (Burn) on progeny production in *D.melanogaster*.

[Different letters on the bar graph indicates significance at 0.05 level by Tukey's Post Hoc Test]  $% \left[ \left( 1-\frac{1}{2}\right) \right] =0$ 

Table 1. One way ANOVA of feeding behavior, male mating
ability and progeny production in <i>D. melanogaster</i>

Parameters	Sum of Squares	df	Mean Square	F	Sig.
Feeding					
behaviour					
Between Groups	.486	2	.243	36.061	.000
Within Groups	.990	147	.007		
Total	1.476	149			
Male mating					
ability					
Between Groups	56.493	2	28.247	52.828	.000
Within Groups	78.600	147	.535		
Total	135.093	149			
Progeny					
production					
Between Groups	238177.333	2	119088.667	83.333	.000
Within Groups	210072.000	147	1429.061		
Total	448249.333	149			

### DISCUSSION

In an organism, the success of the species depends on the reproductive capacity of that species because reproductive capacity is an index of fitness that involves repeated cycles of rapid population growth. Thereby, organism evolved a way for species to maximize the reproductive potential (Turner and Anderson, 1983). Male reproductive success depends on the number of females they could inseminate in their lifetime and total number of fertile progeny produced. On the other hand, female reproductive success depends on the total number of eggs produced in its lifetime (Thornhill and Alcock, 1983). Male remating ability and progeny production in species of Drosophila varies between species, between different populations of same species (Parsons, 1973; Banerjee and Singh, 1978; Gromko and Pyle, 1978; Casares et al., 1998). It is known to influence by many factors such as male size, age, and other environmental factors (Abolhasan and Krishna, 2014). Male diet is also one of the main factors known to affect mating success and reproduction in species of Drosophila (Sisodia and Singh, 2012). The primary function of mating is to transfer sperms to mate females. Therefore each mating offers a male an opportunity to father offspring. In the present study (Fig. 1 and 2) males grown in natural energy drinks had inseminated significantly greater number of females in 1 hr compared to males grown in either in synthetic energy drink based media or normal media. This suggests that male grown in natural energy drink based media had greater mating ability as a result they showed greater reproductive capacity compared to males grown in synthetic energy drink based media and normal media.

Males grown in natural energy drinks based media had increased their fitness by mating with many mates and its high rate of mating are typically correlated with high rate of reproductive success. Our study confirms earlier studies of male mating ability in species of Drosophila (Gromko and Pyle, 1978; Turner and Anderson, 1983, Loukas et al., 1981, Levine et al., 1980; Koref- Santibanez, 2001; Abolhasan and Krishna, 2014). They found that male which inseminates greater number of females in given time had greater reproductive capacity and produced greater number of progeny. They also showed that male traits such as size and age could influence male mating ability and progeny production in species of Drosophila. In the present study it was noticed that male diet is also important factor known to affect the male mating ability and progeny production. Thereby male diet had influence on reproductive success in Drosophila melanogaster. In the present study, Fig. 3 and table 1 also showed that females inseminated by males grown in natural energy drinks based media had produced significantly greater number of progeny than those females inseminated by males grown in synthetic energy drink based media and normal media. That it was noticed that in species of Drosophila male fitness is therefore related to the number of females inseminated and preventing the females from remating.

Thus these studies in *Drosophila melanogaster* suggest that male nutrition plays an important role in male mating ability and progeny production. Males grown on natural energy drinks based media had significantly greater reproductive success compared to males grown in synthetic energy drinks and normal media.

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