Dual Role of Mating in Egg Production and Survival in the Cricket, *Plebeiogryllus guttiventris* Walker

Mating besides serving the primary purpose of fertilization is also known to influence the fecundity of several insects[1]. However, interestingly enough, mating has been found to play a dual role with respect to egg production and survival in the cricket, *Plebeiogryllus guttiventris*. Under normal feeding conditions, females of this cricket when paired with males showed higher fecundity[2]. Mating specifically increased initially the oviposition which was followed by increase of egg production and rate of oviposition[3]. During starvation, on the contrary, females, if mated, were found to have produced less eggs than the virgins. This decreased egg production was associated with increased duration of survival of these mated females[4]. Thus mating during starvation ensured longer duration of survival by increased oocyte resorption that resulted in decreased egg 'production'[4]. It is of interest to discover whether both of these effects are exerted by the same component of mating stimulus, or whether 2 components are involved. In this paper we have examined this by selecting a substitute for mating and have found that 2 separate components of mating stimulus are responsible for the observed dual role. However, at present the nature of these components are not clear.

Materials and methods. Crickets from the stock-culture maintained in this laboratory[5] were selected. Nymphs of last instar were segregated into males and females, and adults emerging from these groups of nymphs were collected daily and used for the experiment after attaining the required age. Hence females were virgins until allowed to mate.

Castrated males were obtained by removing the 2 testes through the small lateral slits made on the 5th abdominal segment on the day of their emergence. Spermatophores from normal males were obtained by squeezing the abdomen gently and were then inserted into the female genitalia, thus artificial spermatophore transfer was accomplished. Normal mating was ensured by keeping a male with the female under observation until they mated.

During starvation, females were deprived of food but had a supply of distilled water in sand cups. Deaths were noted every 12 h. For each death in starved group, a control fed female of corresponding group was sacrificed to note the egg production during the same duration.

Eggs were collected by providing sterilized sand in small beakers of 5 ml capacity. At the end of the experiment, the females were dissected to note the number of eggs retained in the body. Total of eggs laid and retained in body formed the number of eggs produced. Other details of the experiments were similar to those described elsewhere[2,4].

In the first experiment 2 substitutes were selected to note their effect on egg production. In one, females were allowed to mate with castrated males and in the other, spermatophores were artificially transferred to the females. Virgin females and females mated with normal males formed the controls. For this experiment 30-day-old females, 10 for each treatment, were selected and egg laying was observed for 7 days. At the end of this period the experiment was terminated.

Results and discussion. Results of this experiment, presented in Table I, showed that neither of the substitutes for mating resulted in increased oviposition as noted for control mated females. Mating with castrated males resulted in higher egg production than that of virgin females ($p < 0.05$). Though this seems to contradict the observation for *Teleogryllus commodus*[6], the period of 7 days is not sufficient to assess the results with respect to egg production. It can be recalled in this connection that mating in *P. guttiventris* initially resulted in an increased oviposition within 1 week, which in turn led to the increased egg production that followed the subsequent weeks[7]. It is evident from these results that artificial transfer of spermatophore, and also possibly mating with castrated males, lacks that component of mating stimulus that triggers oviposition.

Selecting artificial transfer of spermatophore as the substitute for mating, its effect on survival during starvation was tested in the subsequent experiment. Here 30-day-old females were selected for starvation. A batch of these females and of control fed females received spermatophores artificially on day 4 of experiment. Duration of survival, egg production and oviposition were noted.

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Table I. Influence of mating with castrated males and artificial transfer of spermatophore on the egg production and oviposition of the cricket, *P. guttiventris*

Treatment	No.	No. of eggs produced (Mean ± SE)	Oviposition (%) (Mean ± SE)
Virgin females	10	125.1 ± 20.5^{a}	2.56 ± 1.3^{d}
Mated females	10	300.7 ± 49.6^{b}	65.89 ± 8.7^{e}
Females receiving spermatophores artificially	10	187.7 ± 43.4	7.20 ± 2.8^{f}
Females mated with castrated males	10	214.7 ± 40.6^{c}	6.51 ± 3.0^{g}

Comparison of means (t-test): $^{a-b}$, $p < 0.01$; $^{b-c}$, $p > 0.05$, NS; $^{a-c}$, $p < 0.05$; $^{d-e}$ and $^{e-g}$, $p < 0.001$; $^{d-f}$ and $^{d-g}$, $p > 0.05$, NS.

Table II. Influence of artificial transfer of spermatophore on the duration of survival, egg production and oviposition of the cricket *P. guttiventris* during starvation

Treatment	No.	Duration of survival (days) (Mean ± SE)	No. of eggs produced (Mean ± SE)	Oviposition (%) (Mean ± SE)
Starved virgin females	20	6.1 ± 0.46^{a}	141.9 ± 18.3^{c}	0.0
Fed virgin females	18	—	194.4 ± 14.5^{d}	5.69 ± 4.8^{g}
Starved females receiving spermatophores artificially	20	8.2 ± 0.57^{b}	85.0 ± 20.9^{e}	11.72 ± 6.8
Fed females receiving spermatophores artificially	20	—	174.9 ± 33.0^{f}	15.99 ± 6.7^{h}

Comparison of means (t-test): $^{a-b}$, $p < 0.01$; $^{c-d}$ and $^{c-e}$, $p < 0.05$; $^{d-f}$ and $^{g-h}$, $p > 0.05$, NS.

Results of this experiment, presented in Table II, interestingly showed increased duration of survival and decreased egg production in starved females that had received spermatophores as against starved virgin females. These results further confirmed the earlier observation that females receiving spermatophores, under normal feeding conditions, did not show any difference in egg production or oviposition from those of virgin females.

Hence, it is apparent that such a transfer of spermatophore to the females, though lacking the component for triggering oviposition, could still increase survival duration during starvation. It is, therefore, obvious that 2 separate components are involved in expression of the dual role of mating, which in itself is unique and noted by us for the first time, in *P. guttiventris*. However, the exact nature of these components, at present, can only be speculated and hence is of interest for further studies[8].

Zusammenfassung. Bei der Kopulation der Grille *Plebeiogryllus guttiventris* werden mindestens 2 unabhängige Stimuli wirksam, welche Oviposition und Eiproduktion beeinflussen. Weder künstliche Spermatophorenübertragung noch Kopulation mit kastrierten Männchen stimuliert die Oviposition; letztere bewirkt aber erhöhte Eiproduktion. Künstliche Übertragung von Spermatophoren in hungernde, virgine Weibchen reduziert die Eiproduktion und bewirkt eine Lebensverlängerung.

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Induction of Flowering in *Lemna gibba* G3 by Aspirin

Several billion tablets of aspirin are used all over the world every year to relieve headaches and other pains, reduce fever and deal with a wide variety of ailments. The prevailing hypothesis is that in these disorders, the amount of copper in blood rises to two or more times the normal level. Simple removal of excess copper from blood does not relieve the pain or reduce the fever. Aspirin probably acts as a chelate, repairing this biochemical 'lesion' by picking up copper from the blood and returning it to the cells from which it was lost[1]. Studies by HILLMAN[2] and in our laboratory[3] demonstrated that copper somehow influences the photoperiodic sensitivity of a duckweed, *Lemna gibba* G3[2,3]. More recently, we have been able to induce flowering in *L. gibba* G3 by adding aspirin to the nutrient medium.

The plants of *Lemna gibba* G3 were aseptically cultured on M-medium[4] and 1/3 strength HUTNER's medium[5]. The methods for raising aseptic cultures, light and temperature conditions, determination of multiplication rate (MR) and evaluation of flowering were the same as described in our earlier papers[3,6]. Aspirin, manufactured by the Bayer Company, Division of Sterling Drug, Inc. New York, N.Y. 10016, was utilized in the present investigation.

The plants grew in 1/3 HUTNER's and M media. Incorporation of aspirin in both media influenced the MR of the fronds. In 1/3 strength Hutner's medium, the MR slightly declined at concentrations of 0.1, 1.0, 2.5 and 5 ppm but at higher concentrations (10, 20 and 25 ppm) no further decrease was observed. In M-medium, MR was not influenced by the presence of low amounts of aspirin (0.1, 1.0 and 5.0 ppm) but at 10 ppm and 20 ppm no growth occurred. The plants became yellow and died within 2 days after inoculation.

Besides its effects on MR, aspirin had pronounced influences on the growth and development of individual fronds. The effects were detectable within 4 days; magnitude of the effects varied with the concentration of aspirin. Plants were larger in 1/3 strength HUTNER's medium containing 0.1, 1.0, 2.5 and 5 ppm of aspirin. In M-medium an increase in the size of the fronds was apparent at 0.1, 1.0 and 2.5 ppm of aspirin. Higher concentrations (10, 20 and 25 ppm in HUTNER's medium, and 5 and 10 ppm in M-medium) resulted in a decrease in

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