REPRODUCTIVE BIOLOGY OF *Cotesia flavipes* (CAMERON, 1891) (HYMENOPTERA: BRACONIDAE). I. INBREEDING EFFECT ON PRODUCTIVITY OF FEMALES MATED TO A SINGLE MALE[1]

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RESUMO

Biologia reprodutiva de *Cotesia flavipes* (Cameron, 1891) (Hymenoptera: Braconidae). I. Efeito do endocruzamento na produtividade de fêmeas cruzadas com um único macho.

A freqüência de acasalamento entre machos e fêmeas de *flavipes* com e sem parentesco próximo foi investigada, sendo registrado o tempo médio para o início da cópula e a duração do ato sexual. Também foi estimado o número médio de descendentes de fêmeas virgens, de cruzamento irmão x irmã e exocruzamentos. A origem dos cônjuges não afetou a proporção de acasalamentos nos dois grupos estudados. O sucesso de cópula foi de aproximadamente 69% nas duas situações experimentais.

Não houve diferença significativa entre os valores obtidos para o início da cópula de machos e fêmeas irmãos entre si e de indivíduos não aparentados. O mesmo ocorreu em relação aos valores médios de duração do ato sexual nos dois grupos testados. PALAVRAS-CHAVE: *Cotesia flavipes;* biologia reprodutiva; endocruzamento: produtividade.

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ABSTRACT

The mating frequency between males and females of *C. flavipes* with and eithout relationship was investigated, being registered the mean time for the beginning of copula and the mating duration. The mean number of descendants produced by virgin females, brother x sister crossings and non-endogamic matings was also estimated. The partners origin did not affect the mating sucess of *C. flavipes* couples from both groups. The mating frequency registered in these two experimental conditions was 69%.

No difference was registered in relation to the mean time for the beginning of copula between related and non-related partners. The same was observed in relation to the mean values of mating duration registered for both types of couples. KEY-WORDS: *Cotesia flavipes;* reproductive biology; inbreeding; productivity.

INTRODUCTION

C. flavipes is a larval parasitoid wasp which has been used in the biological control of the sugar cane borer *Diatraea saccharalis,* in many countries where sugar cane is cultivated in large scale (GIFFORD & MANN, 1967; ALAM *et al.*, 1971; BOTELHO *et al.*, 1980; MELO, 1984).

In Brazil, the great devastation of natural habitats in order to cultivate sugar cane, mainly in "cerrado" areas, produced a severe alteration in the native vegetation of many states. The excessive expansion of sugar cane plantations originated an unexpected increase in *D. saccharalis* populations, which then became a serious calamity for this culture.

Since then, many studies have been developed with *C. flavipes,* in order to obtain new informations wich make viable an improvement in the biological control of *D. saccharalis*.

These studies generally include massal rearing methods for *C. flavipes,* liberation of great number of parasitoid wasps in areas of sugar cane culture with different climatic conditions, estimative of the dispersion grade, host localization by the parasitoids, multiparasitism effects, food consumption by the sugar cane borer parasitized, level of parasitism in field conditions and other features. In spite of these important researches, little is known about the reproductive biology of *C. flavipes* until now.

This insect has gregarious habit and endogamic matings are frequent. Nevertheless, there are no information on the frequence of successful matings under controlled conditions, when the partners are not related and what happens when they are sibs. On the other hand, there are also no information on the relative viability of descendants from both endogamic and non-endogamic crossings.

The objectives of this research were:

1. To estimate the frequency of successful matings under controlled conditions, when the partners are brothers and sisters and when they are not related.

2. To estimate the number of descendants in these two mating conditions.

These informations are very important because it will be an essential subsidy to the correct planification for massal rearing of *C. flavipes*, aiming the biological control of *D. saccharalis*. An analysis of the progeny from virgin females was also carried out in this experiment.

MATERIAL AND METHODS

The parasitoid wasps *(C. flavipes)* and the borers *(D. saccharalis)* used in this research were gently furnished by COPERSUCAR laboratories of Sertãozinho - SP, between July and October, in 1989. Each group of *C. flavipes* pupae emerged from a same host was introduced in a bottle (10 cm^3) covered by net-lace, being maintained at 28°C and 12L - 12D photoperiodicity. *D. saccharalis* borers were maintained in the same experimental conditions and received artificial diet.

Experiment 1 - Effect of partners origin on mating success of *C. flavipes*.

C. flavipes pupae were individually introduced in small bottles (10 cm^3) 2 - 3 days before the emergence, in order to assure the virginity of males and females. The newly emerged wasps received diluted honey as food. Couples were formed by using a female and a male from a same cluster (brother x sister mating). Couples formed by non-related individuals were obtained by taking a male and a female from different clusters.

The mating success in these two experimental conditions was investigated, being registered the number of couples which effectively copulated, the mean time for the beginning of copula and the mating duration.

The couples were maintained at room conditions (temperature: 30,5° ± 1,5°C; light intensity: 500 LUX), being observed for one hour.

The results obtained in this experiment were analysed under two statistical treatments: the data concerning the number of matings registered in both groups (matings between related and non-related partners) were compared by using a test for differences in proportion of successful crossings (SPIEGEL, 1975).

The mean values of time to beginning the mating and duration of copula, in both experimental groups, were compared by using the *t* test.

Experiment 2 - Estimative of the number of descendants from endogamic and non-endogamic matings and virgin females.

Two days before the emergence some clusters of *C. flavipes* pupae were separated at random, being each pupa introduced in a bottle which received a drop of diluted honey over its net-lace covering (the honey is one of the best foods for newly emerged wasps).

After the emergence of each parasitoid wasp, initially maintained in an individual bottle, couples were formed by using a female isolated from a cluster of pupae and a male from another one. In this way, only non-endogamic matings were obtained.

The other clusters of *C. flavipes* pupae were maintained in their original bottles, in order to provide good conditions for occurrence of endogamic matings (brother x sister).

A third group of *C. flavipes* was formed by females maintained as virgin, in order to verify the occurrence of laying in this condition.

Twenty-four hours after the formation of couples, each mated female was introduced in a bottle (10 cm^3) containing one *D. saccharalis* borer of third larval instar. Each female of *C. flavipes* was removed from the bottle as soon as she applied the first stinging in the borer.

Each borer that received the stinging was immediately transferred to another bottle, provided with artificial diet.

Approximately ten days after the parasitism, the parasitoid larvae emerged from the host larvae. After pupation the number of parasitoids was registered.

The results obtained from *C. flavipes* females submitted to these three experimental conditions (virginity, non-endogamic and endogamic matings) were then compared by using the non parametric statistical analysis of Kruskal-Wallis, being estimated the least significant difference among the groups (CAMPOS, 1983).

RESULTS AND DISCUSSION

Tables I and II contain the data registered in the first experiment. The results concerning the effect of partners origin in mating success of *C. flavipes* are presented in Table I. As can be seen in this table, it was registered a mating success of approximately 69% in both experimental groups.

The test for differences in proportion of successful matings in the two groups furnished a non-significant value (Z = 0,001), indicating that the partners origin did not affect the mating success of *C. flavipes* couples.

The results concerning the mean time for the beginning of copula and mating duration are in table II.

The values of mean time for the beginning of copula registered for related and non-related partners of *C. flavipes* presented no statistical difference when compared by means of *t* test (t = 0,47, non significant at 5% level).

The same was observed in relation to the mean values of mating duration registered for both types of couples (t = 0,91, non-significant at 5% level).

Table III and IV contain the data registered in the second experiment.

Table III summarizes the results concerning the productivity of *C. flavipes* females submitted to the three experimental conditions established in this research (virginity, endogamic crossings and non-endogamic crossings).

The data of productivity registered in this research were compared by means of a Kruskal-Wallis analysis being obtained a value statistically significant, indicating a difference between the number of descendants produced. A multiple comparison test applied latter revealed that the number of descendants produced by virgin females did not differ in relation to that registered for *C. flavipes* females mated to their own brothers. Nevertheless, there was a significative difference between the values obtained for these two groups of females and the values registered for *C. flavipes* females mated to non-related males, which produced a number of descendants significatively greater (Table IV).

WRIGHT (1921) stated that in a population with high level of inbreeding there is a progressive increasing of homozygotical allelic combinations for almost all loci considered. As the populations generally present lethal, semi-lethal and deleterious genes, the homozygosis of these genes certainly will produce a great rate of mortality if the inbreeding is maintained by many consecutive generations. According to FALCONER (1964), the whole population physiology is changed as to the fertility, physiological efficiency and so on, if it is submitted to a high level of inbreeding. Falconer called this effect by "inbreeding depression". It is explained by the fact that in a diploid population, recessive deleterious genes are maintained at low frequency by selection-mutation balance or a weak heterozygote advantage and few homozygotes for the deleterious genes occur (CROW & KIMURA, 1970). A rapid increase in inbreeding elevates the frequency of individuals homozygous for the deleterious alleles, with a consequent decline in mean fitness. On the other hand, if mating is confined to small groups of individuals by virtue of their location of birth, such that males do not compete at large, a "weak" male might be able to mate, simply because it is the only male with access to a particular group of females (BORGIA, 1980). This particularity is known as "local mate competition", and has been studied with respect to sex ratio selection by HAMILTON (1967). Local mate competition usually involves the mating of sibs, and so may be associated with inbreeding.

A second complication from inbreeding concerns the possible deleterious effect of mating with close relatives. The initial uniparental males may face complications in meiosis causing them to produce aneuploid sperm. These can fertilize eggs in many especies, but the progeny produced would like die. Under inbreeding, uniparental males preferentially mate with sisters or other closely related females, and these are the females producing uniparental sons. The aneuploid sperm preferentially decrease the fecundity of these females, so the mothers of uniparental sons would be at a disadvantage (BULL, 1983).

In the present research, in spite of the fact that only one generation of brother x sister matings was established, the number of descendants from endogamic matings was significatively smaller in relation to that one registered for non-endogamic matings.

As each *C. flavipes* female usually copulates only once in the field or in laboratory conditions (ARAKAKI & GANAHA, 1986; CUMAGAY, 1990), the partners origin is very important in relation to the number of descendants produced.

So, the results obtained in this research can be useful in order to help improving the massal rearing of *C. flavipes*, furnishing therefore better conditions to increase the efficiency in the biological control of *D. saccharalis*.

TABLE I - Mating success of *Cotesia flavipes* couples formed by related males and females (brothers and sisters) and non related partners.

Partners Relationship	Number of Couples Observed	Number of Successful Matings	Copula Success(%)
Brothers and sisters	123	85	69.10
Non-related partners	110	76	69.09

TABLE II - Mean time for the beginning of copula and mating duration of *Cotesia flavipes* couples formed of related (brothers and sisters) and non-related partners.

Partners Relationship	Number of Couples Observed	Mean time for the beginning of copula (in minutes)	Mating Duration (in seconds)
Brothers and Sisters	51	11.43	11.84
Non-Related Partners	46	12.65	11.21

TABLE III - Productivity of *Cotesia flavipes* females submitted to different experimental conditions.

Experimental Conditions	Descendants Produced by Mated or Virgin Females	Mean Number and Standard Deviation
Endogamic Crossings (Brother x Sister) (n=20)	623	31.15 ± 19.38
Non-Endogamic Crossings (n=20)	963	48.15 ± 19.19
Virgin Females(n=18)	552	30.66 ± 13.74
TOTAL	2 138	-

TABLE IV - Kruskal-Wallis analysis concerning the data registered in the experiment 2.

Group	Average Rank	LSD I	LSD II	LSD III
I - Endogamic Crossings	38.98	-		
II - Non-Endogamic Crossings	24.15	13.11	-	
III - Virgin Females	24.92	13.11	12.79	-

H ajusted = 9.64** (df=2)

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