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The Biological Control of *Callosobruchus maculatus* (Fabricius) by *Dinarmus basalis* (Rendani) on Stored Cowpea (*Vigna unguiculata* Walp) Seeds

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Abstract: The control of *Callosobruchus maculatus* F. on cowpea seeds was studied in the laboratory using *Dinarmus basalis* Rendani as parasitoid. Five pairs of adult *C. maculatus* were allowed to oviposit onto 10 sets of cowpea seeds. Five pairs of *D. basalis* were introduced at 3 days intervals (beginning from the 6th day after oviposition) into each of the nine sets of seeds. No *D. basalis* was introduced into the tenth set (control). No significant differences ($p \geq 0.05$) were observed in the number of eggs laid per female *C. maculatus* in all the sets and this ranged from 37.80-41.20. No significant ($p \geq 0.05$) differences were observed on adult *C. maculatus* that emerged when *D. basalis* was introduced at the 6, 9 and 12th Day After Oviposition (DAO). Percent emergence was 0.59, 0.53 and 1.85% at 6, 9 and 12st day, respectively. *C. maculatus* emergence increased from 67.81% in the treatment that was inoculated on the 21st DAO to 74.48 % on the 30th DAO. The total *D. basalis* per female in each culture was 12.81 and 17.56% on the 6 and 15th day and decreased significantly to 11.78% when inoculation was done on the 30th day. Percentage mortality of *C. maculatus* increased from 25.52 to 99.48% on the parasitised culture compared to 25.03 in the control. The time of introduction of the parasitoid critically affected the mortality of *C. maculatus*. Thus, introduction of *D. basalis* during the larval stage of *C. maculatus* caused higher mortality of the pest and had no effect on adult *C. maculatus*.

Key words: Oviposition, inoculation, mortality, parasitoid, larval stage, adult

INTRODUCTION

The bruchid *Callosobruchus maculatus* is an important pest of stored cowpea (*Vigna unguiculata* L. Walp) with ample distribution in tropical and subtropical regions where this crop represents one of the main sources of protein in human diet (Singh *et al*., 2005). It is reported to be the principal storage pest of cowpea grain in sub-Saharan Africa (Taylor, 1981). Infestation of *C. maculatus* starts in the field and continues in storage (Messina, 1984). The insect infests cowpea before harvest and causes quantitative and qualitative losses to seeds in storage facilities (Shade *et al*., 1990;Caswell, 1984; Murdock *et al*., 1997) reported that during traditional post harvest storage in Nigeria, cowpea grain stored in pods for 8 months had 50% of the grain damaged by bruchids but when stored as grain, 82% had one or more holes. Female *C. maculatus* oviposits on the pods and larval development is entirely within the seeds (Messina, 1984; Stoll, 2000). The Hymenoptera *Dinarmus basalis* is an ectoparasitoid larvophagous species, which is also present in the graineries and represents 80-90% of the bruchid larvophagous parasitoids in the cowpea fields and the stores (Ouedraogo *et al*., 1996). According to Dugravot *et al.* (2002) *D. basalis* is an efficient natural enemy, which could be used for biological control. The larva of *C. maculatus* is parasitised inside the seeds by *D. basalis* (Dugravot *et al*., 2002; Gauthier *et al*., 2002). The objective of this study was to manage the population density of *C. maculatus* by introducing *D. basalis* at the time the pest is susceptible to the parasitoid.

MATERIALS AND METHODS

Cowpea seeds that were collected from cowpea store in Uselu Market, Benin City were placed in a beaker in an oven at 60°C for 1 h to disinfest them (Murdock and Shade, 1991). The experiment was carried out in the University of Benin laboratory under a temperature of 28±2°C and 70±10% RH. Cowpea seeds (200 g) were infested with five pairs (0-3 days old). The sexes of *C. maculatus* were determined by examining the pattern (Southgate *et al*., 1957). Females are usually dark and possess four elytral spots, while males are pale brown and less distinctly spotted. Furthermore, males have comparatively shorter abdomen and the dorsal side of the

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terminal segment is sharply curved downwards and inwards. In contrast, females have comparatively longer abdomen and the dorsal side of the terminal segment is only slightly bent downwards (Bandara and Saxena, 1995). The disinfested cowpea seeds were put in five jars with net mesh lids. The seeds in each jar were re-infested with *C. maculatus* and allowed to oviposit for 24 h before they were removed.

Cultures of *D. basalis* were reared from the stock collected from parasitised cowpea purchased at the Uselu Market, Benin City. Parasitoids were sieved out of the cowpea and the cowpea set aside for new emergence of the parasitoids.

Five g of the disinfested cowpea seeds were weighed into ten different specimen bottles (labelled A-J) with fine nylon net lids of about 1×1 mm mesh size and these were replicated four times. Cowpea seeds in each of the bottles were infested with five pairs (females and males) of the newly emerged *C. maculatus* (0-3 days old) and were allowed to oviposit for 24 h. The total eggs laid per female was taken 3 days after oviposition.

Introduction of *D. basalis* to cowpea infested with eggs of *C. maculatus*: Each of the nine bottles (labelled A-I) containing the infested cowpea except the control was infested with five pairs (males and females) of *D. basalis* at three days intervals beginning from the 6th day after oviposition. No *D. basalis* was introduced into the tenth bottle (labelled J) and this served as control. Treatments were replicated four times and set-aside on a laboratory bench arranged in completely randomized block design.

Data collection: The number of eggs laid by *C. maculatus* per female was recorded. Emergence of *C. maculatus* and *D. basalis* per female were recorded. Analysis of variance was carried out on the total number of *D. basalis* in each culture. The percentage mortality and emergence of adult *C. maculatus* was calculated based on the mean number per female that emerged relative to the total number of eggs laid per female. Data were taken cumulatively from 25-38 DAO and the means were used for analysis of variance. The Fisher's least significant difference was used to separate the means where applicable.

RESULTS AND DISCUSSION

Emergence of *C. maculatus* was observed from the 25th DAO until 38th day when no emergence was observed. *D. basalis* emergence was observed from the 12th DAO. Results (Table 1) indicated no significant differences ($p \geq 0.05$) in the mean number of eggs laid per female *C. maculatus*. Significant differences ($p \leq 0.05$) occurred in adult *C. maculatus* that emerged after inoculation of the parasitoid. The first six days after inoculation, significantly lower *C. maculatus* emerged compared with the cultures that were inoculated later. The highest emergence of *C. maculatus* was observed with the 27th DAO, although there were no significant differences in emergence between the 27th, 30th and the control. Percent mortality was significantly higher in the cultures that were inoculated earlier at the 6th to 18th DAO (i.e., the larval stages) compared with the control and cultures inoculated later at 21st to 30th DAO (when *C. maculatus* has reached adult stage). This corroborates the report by Ouedraogo *et al.* (1996) and Stoll (2000) which indicated that the eggs hatch in about six days after oviposition. There are four larval instars, which last about 20 days. Percent mortality of *C. maculatus* had positive relationship with the parasitoid. The results showed two-phased relationship between the *D. basalis* and *C. maculatus*. The first is from the 6-18th DAO when mortality of *C. maculatus* was high and this time corresponds to the larval stage of *C. maculatus* while from 21-30th DAO, the pest had reached adult stage

Table 1: Effect of *Dinarmus basalis* on the population density of *Callosobruchus maculatus*

Inoculation of *Dinarmus basalis* (days after oviposition) in each sets (A-J)	No. of eggs laid/female *Callosobruchus maculatus*	*% Emergence of *Callosobruchus maculatus*/female	*% Mortality of *Callosobruchus maculatus*/female	*Total No. of *Dinarmus basalis* in each culture
6 (A)	37.80	0.59	99.41	12.81
9 (B)	39.35	0.53	98.48	13.21
12 (C)	41.15	1.85	98.15	15.99
15 (D)	38.60	3.67	96.33	17.56
18 (E)	39.50	11.78	88.22	14.17
21 (F)	41.10	67.81	32.20	13.38
24 (G)	39.45	69.78	30.23	12.78
27 (H)	38.50	75.32	24.68	12.70
30 (I)	39.55	74.48	25.03	11.78
Control (J)	39.90	74.96	25.52	-
LSD (p = 0.05%)	NS	4.98	4.98	0.64

- = No *Dinarmus basalis* was included; * Each value is mean of cumulative sampling for 19 days hence were no more susceptible to the larvophagous parasitoid. The population of adult *D. basalis* increased during the larval stages of its host and decreased as the later got to adult stage. Thus, the time of introduction of the parasitoid was a crucial factor that affected the mortality of *C. maculatus* in the study. Introduction of adult *D. basalis* during the larval stages of *C. maculatus* caused higher mortality of the pest but no effect on the adult *C. maculatus*.

This study has confirmed reports by other authors that *D. basalis is* an efficient parasitoid of *C. maculatus* and can cause significant reduction in the numbers of its prey. The report by Kristina *et al*. (2005) that the bean damage can be reduced approximately by 35-96% using *D. basalis* as control agent can also be confirmed in this study since direct damage to cowpea seeds was not directly observed in this study, it can be deduced that since the result showed that up to 99.41% mortality of *C. maculatus* was achieved, only about 0.59% was left to damage the grains.

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