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STRAIN DIFFERENCES IN TWO SPECIES OF *CALLOSOBRUCHUS* (COLEOPTERA: BRUCHIDAE) DEVELOPING ON SEEDS OF COWPEA {*VIGNA UNGUICULATA* (L.)} AND GREEN GRAM {*V. RADIATA* (L.)}

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ABSTRACT

Ovipositional behaviour, development period, and density effect on adult survival of C. *maculatus* strains from Indonesia, Nigeria, and Yemen, and C. *chinensis* strains from Indonesia and Kenya on cowpea and green gram were studied at 20°C and 70% relative humidity.

Variations on ovipositional behaviour were found among *C. maculatus* as well as among C. *chinensis* strains. Variations on developmental period were found only among *C. maculatus* strains. The developmental period of *Callosobruchus* spp. was shorter on green gram than that on cowpea. Density effect was remarkably found only on adult survival of *C. maculatus* Yemen strain. These results make useful contribution to the species biology, and have important implication if strains of these species are accidentally imported to countries, or when new legume crops are introduced.

INTRODUCTION

Beetles belonging to the family Bruchidae are the most important insect pests of stored legumes. Infestation by bruchids causes losses of weight, nutritional value and germination potential, and therefore the commercial value of the commodity may be reduced (Southgate 1978; Dick and Credland 1986). The most economically important and widespread bruchids species are the cowpea seed beetle, *Callosobruchus maculatus* (Fabricius), and the Adzuki bean beetle, *C. chinensis* (Linnaeus) (Southgate 1978; TDRI 1984).

The use of resistant varieties of cultivated legumes is one of the recommended control methods of bruchid infestations. Varietal resistance against *Callosobruchus* has been reported in cowpeas and chickpea (Dobie 1981; Raina 1971; Singh 1978).

However, there were variations reported in the response of geographically different strains of *C. maculatus* to a resistant variety of cowpea (Dick 1984; Dick and Credland 1986). Further studies on variation on geographically different strains of *C. maculatus* revealed the occurrence of differences in their biology and behaviour (Credland *et al.* 1986; Credland and Dick 1987; Credland 1986).

This study was made to seek more information on the occurrence of geographical variations among *C. maculatus* and *C. chinensis* strains, especially to compare strains from Indonesia (Asia) with those from other tropical countries.

MATERIALS AND METHOD

Three strains of *C. maculatus* viz. strain from the IITA cultures, Nigeria (labeled as IITA), Yemen, and Indonesia; and two strains of C. *chinensis* viz. strains from Indonesia and Kenya were used. They were obtained from the culture of ODNRI, Slough, UK.

Two types of seeds were used as hosts i.e. Californian black eyed cowpea *(Vigna unguiculata* (L.)) and Australian green gram *(Vigna radiata* (L.)).

Daily egg production

The surface area of cowpea seeds is larger than that of green gram. As oviposition of the species is assumed to be influenced by the surface area, the cowpea seed number used should therefore be different from that of green gram. After measuring the surface areas of both seeds, it was decided that the surface area of one cowpea seed was approximately equal to the surface area of three green gram seeds.

One kernel of cowpea or three kernels of green gram were introduced into a 2.5 cm diameter and 5 cm high glass tube. A pair of adults (age < 1 day) of each strain was added into the tube, and the tubes were covered with foam bungs. Twenty five replicates were made on both types of seed and all insect strains. The whole set of experiment was kept in the laboratory at 27°C and the R.H. at 70%.

The following day, the number of eggs laid by each female was counted and recorded, and egg-laden seeds were replaced by fresh seeds. The observation and seed replacement were done daily until the 10th day.

Effect of seed availability on egg production

Different numbers of seeds i.e. 1 (low), 3 (medium), and 10 (high) kernels of cowpea; and 3 (low), 9 (medium), and 30 (high) kernels of green gram were introduced into the tubes. One pair of adults (age < 1 day) of each strain was placed in each tube and the tube was covered with foam bungs. Twenty replicates were made of all treatments and all insect strains, and kept in the experimental room. Observation on the number of eggs laid by each female was done on the 7th day after treatment.

Developmental period

Eight to 10 adults of each insect strain were introduced into each petri dish containing cowpea or green gram seeds. The dishes were covered with the lids and kept in the experimental room for one night The adults were removed the following day. Under a binocular microscope, seeds bearing a single egg were taken out and individually put into tubes. The tubes were covered with foam bungs and kept in the experimental room. Twenty five replicates were made of all insect strains. Observation on adult emergence were started on the 20th day after treatment. Emergence of fresh adults was recorded daily until no more adults emerged.

Adult survival from seeds bearing different number of eggs

To obtain cowpea seeds bearing 1, 2, 3, 4, and 5 eggs, the same procedure as in the **developmental period** experiment was applied. However, selection was done not only of seeds bearing 1 egg, but of those bearing 2, 3, 4, and 5 eggs as well. The replicate number of each treatment was designed to be inversely proportional to the egg density (Giga 1982). Thus, the number of eggs used were approximately the same in all treatments (Table 1).

Table 1. The design of the experiment on adult survival from seed bearing different number of eggs

No. of eggs/seed	1	2	3	4	5
No. of replicates	20	10	7	5	4
No. of eggs	20	20	21	20	20

Emergence of adults was observed and recorded daily starting from the 20th day after treatment.

RESULT

Daily egg production

Some females were found to lay eggs on the seeds and on the tube walls and so both were recorded and analyzed separately.

The general pattern of daily egg production of all strains on seed was the same. High number of eggs were laid during the early period of the female's life. The maximum number was reached either on the second or third day, and then it gradually decreased (Fig. 1).

The sum of 10-day egg production per female of all strains was analyzed by two factors analysis of variance. Oviposition on seed, on tube wall, or the total, over the period of 10 days was influenced by insect strain (Table 2).

C. maculatus IITA strain was found to have the highest fecundity followed by *C. chinensis* Indonesian, *C. maculatus* Yemen, *C. chinensis* Kenyan, and *C. maculatus* Indonesian strain (Table 2, column "Total"). Differences in the fecundity were found between each strain of *C. maculatus,* as well as between each strain of *C. chinensis* (Table 2 and Fig. 2).

C. chinensis Indonesian strain laid the highest number of eggs on seed, followed by *C. maculatus* IITA strain. Lower numbers were laid by *C. maculatus* Yemen and *C. chinensis* Kenyan strains with the lowest number laid by *C. maculatus* Indonesian strain (Table 2).

Large number of eggs on the tube wall were especially laid by *C. maculatus* IITA and Yemen strains. The rest of the strains laid very small number of eggs on the tube. The mean egg number was not significantly different from one another, but they differ significantly from those laid by *C. maculatus* IITA and Yemen strains.

Seed species did not affect oviposition of all strains over 10 days, either on seed, on tube wall or the total (Table 3).

Table 2. Means and standard errors of egg product ion of five strains of *Callosobruchus maculatus* over 10 days on seed, on tube wall, and the total

Insect strains	On seed	On tube wall	Total
Cm* IITA	63 .90 a ±4.57	17.02 a ±2.55	80.92 a ±4.98
Cm Yemen	45.90 b ±4.11	6.06 b ±1.35	51.96b ±4.67
Cm Indonesia	32.08 c ±2.08	0.50 c ± 0.30	32.55 c ±2.15
Cc* Indonesia	68.24 a ±1.86	0.47 c ±0.25	68.74 a ±1.91
Cc Kenya	38.33 bc ±3.20	0.02 c ± 0.02	38.35 be ±3.20

a, b and c indicate level of significance at 5%; relevant only down the column.

* Cm for *Callosobruchus maculatus*

Cc for *Callosobruchus chinensis.*

Table 3. Means and standard errors of insect oviposition over 10 days on seeds, on tube wall, and the total

Seed species	On seed	On tube wall	Total
Cowpea	48.91 NS ± 2.56	4.89 NS ± 0.96	53.80 NS ± 2.86
Green gram	51.08 NS ± 2.38	4.98 NS ± 1.09	56.06 NS ± 2.77

NS : Non significant.

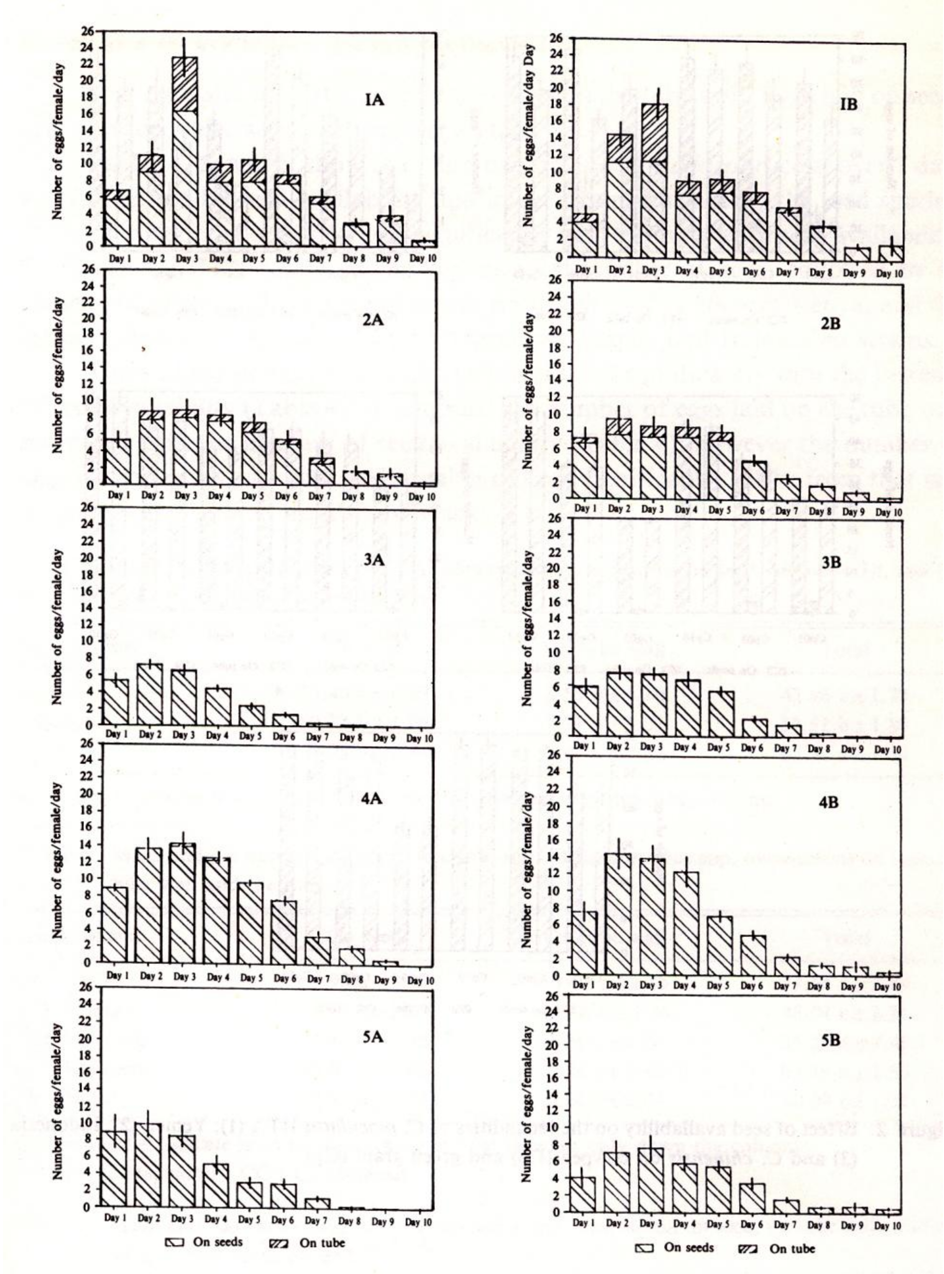

Figure 1. Oviposition over 10 days of *C. Maculatus* IITA (1), Yemen (2), Indonesia (3) and *C. Chinensis* on cowpea (Cp) and green gram (Gg)

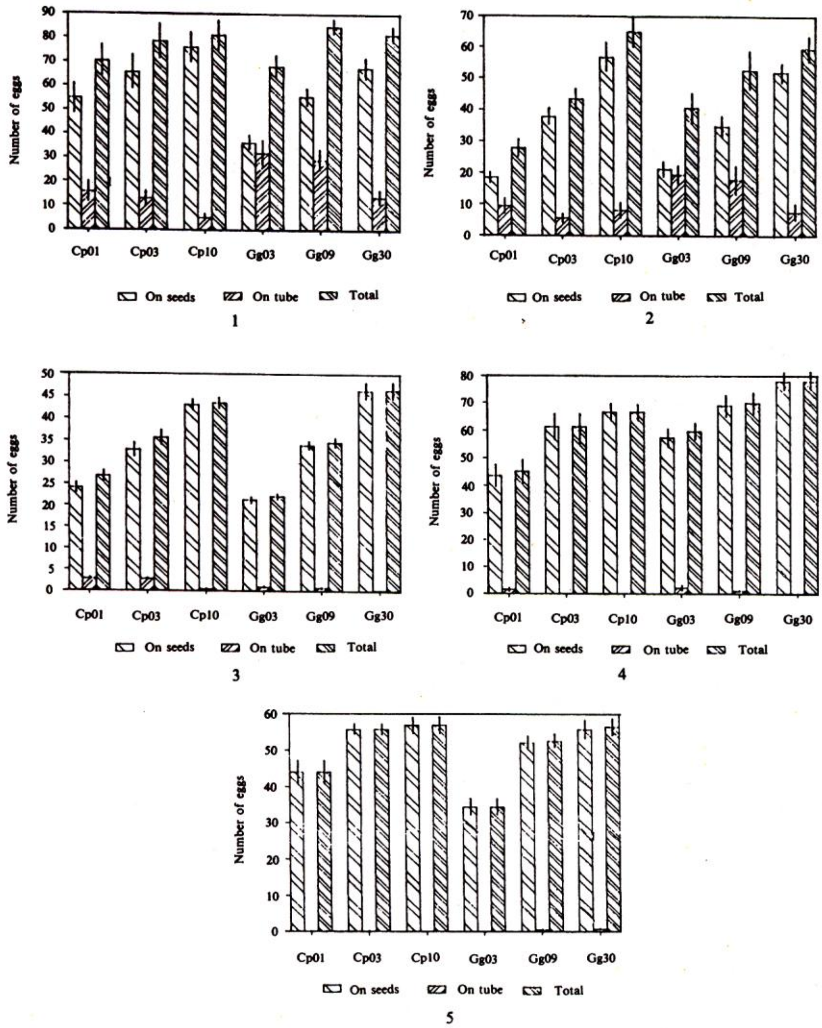

Figure 2. Effect of seed availability on the fecundities of *C. maculatus* IITA (1), Yemen (2), Indonesia (3) and C. *chinensis* on cowpea (Cp) and green gram (Gg).

Effect of seed availability on egg production

Some females were also found to lay eggs on the tube wall. Eggs laid on seeds and on tube walls were analyzed separately.

Tables 4, 5, and 6 show that the oviposition of *Callosobruchus* over 7 days was influenced by seed availability and insect strain, but not by the seed species. The total number of eggs increased significantly with the increase of seed availability. *C. maculatus* IITA produced the highest total number of eggs, followed by *C. chinensis* Indonesia. Lower number was produced by *C. chinensis* Kenya, and the lowest number was produced by *C. maculatus* Yemen and Indonesian strains.

The number of eggs laid on the seed increased significantly with the increase of seed availability (Table 4). In contrast, the number of eggs laid on the tube wall decreased with the increase of seed availability (Table 4), however the number of eggs laid on tubes with low seed number did not significantly differ from that laid on tube with medium number of seeds.

Table 4. Mean and standard errors of *Callosobruchus* spp. oviposition on seed, on tube wall, and the total at different seed availabilities

Seed number	On seed	On tube wall	Total
Low	35.42 c±1.50	8.55 b±1.14	43.96 c± 1.78
Medium	50.22 b±1.56	.31 b±1.03 3.	57.51 b±1.81
High	59.88 a ±1.46	63 a ±0.62	63.51 a±1.54

a, b, and c indicate level of significance at 5%; relevant only down the column.

Table 5. Mean and the standard errors of five strains of *Callosobruchus* spp. ovipositions on seed, on tube wall and the total

Insect strains	On seed	On tube wall	Total
Cm* IITA	58.98 a ±2.52	18.68 a ±1.85	77.64 a ±2.39
Cm Yemen	36.54 c ± 1.85	11.50 b ± 1.36	48.04 c ± 2.21
Cm Indonesia	34.10 c ± 1.48	1.21 c ±0.24	35. 29 d ± 1.48
Cc* Indonesia	63 .05 a ±1.92	0.84 c ± 0.32	63. 89 b ± 1.90
Cc Kenya	49.85 b ±1.31	0.24 c ± 0.07	50.09 c ± 1.33

a, b, c, and d indicate level of significance at 5%; relevant only down the column.

* Cm : *C. maculatus*

CC : *C. chinensis*

Table 6. Mean and standard errors of *Callosobruchus* spp. oviposition on seed, on tube wall, and the total of two kinds of seed.

Seed kind	On seed	On tube wall	Total
Cowpea	49.29 NS ± 1.43	4 39 b ± 0.56	53 .67 NS ± 1.51
Green gram	47.72 NS ± 1.30	8 .60 a ± 0.95	56 .32 NS ± 1.45

a and b indicate the level of significance at 5%; relevant only down the column.

C. maculatus IITA laid the highest number of eggs on tube wall followed by *C. maculatus* Yemen. The rest of the three strains laid only a small number of eggs on tube wall and they did not differ significantly (Table 5). The number of eggs laid on the tube containing green gram was higher than that on tube containing cowpea (Table 6).

The effect of seed availability on egg production of individual strains on cowpea and green gram is shown in Figure 2. The effect of seed availability is remarkably seen in *C. maculatus* Yemen and Indonesian strains.

Development period

The results indicated that there were differences in the development rate among the insect strains (Table 7). The development period of *C. maculatus* IITA strain was the longest, and was significantly longer than the others. Development periods of *C. maculatus* Yemen and Indonesian strains did not differ significantly. The difference in the development period of the two strains of *C. chinensis* was not significant, however they were significantly shorter than the development periods of all C. *maculatus* strains (Table 7).

Seed species significantly influenced C. *maculatus* and *C. chinensis* development periods. Most of the insect strains developed faster on green gram than on cowpea, except the *C. maculatus* strain from Yemen (Table 8).

Table 7. Mean and the standard errors of development periods of 5 *Callosobruchus* strains

Insect strains	Development periods
C. maculatus IITA	32.25 a ± 0.40
C. maculatus Yemen	29.19 b ± 0.13
C. maculatus Indonesia	28.42 b ± 0.25
C. chinensis Indonesia	25.38 c ± 0.13
C. chinensis Kenya	24.90 c ± 0.13

Table 8. Mean and standard errors of development period of *Callosobruchus* on cowpea and green gram

Kind of seeds	Development period
Cowpea	28.19 a ± 0.30
Green gram	27.63 b ± 0.27

a and b indicate the level of significance at 5%.

Adult survival from seeds bearing different number of eggs

The results indicated that egg density, from 1 to 5 eggs per seed, did not influence significantly the number of adult survival of most strains, except on C. *maculatus* Yemen strain (Fig. 3). Remarkable decreases were noted on adult survival means of seeds bearing 4 and 5 eggs.

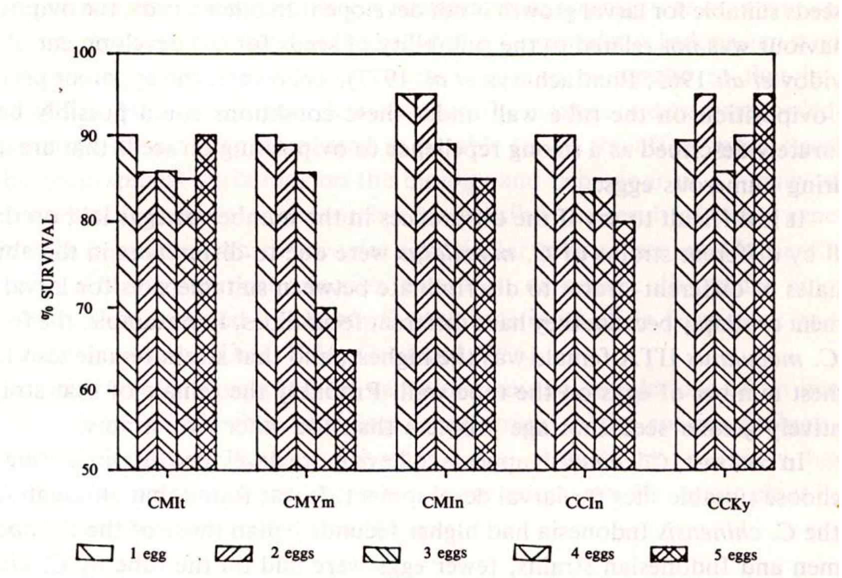

Figure 3. Adult survival from seeds bearing different number of eggs of 5 different *Callosobruchus* strains.

DISCUSSION

Some females, especially those of *C. maculatus* females, were found to lay eggs on tube surface. However, there seemed to be less preference for oviposition on tube wall because the number of eggs laid on the tube wall was usually large only when there was a shortage of seeds for oviposition, such as during the peak days of the ovipositional period (Fig. 1), or when there were few seeds for oviposition (Fig. 2).

Females of *Callosobruchus* spp. have been reported to control the successful development of their progenies by choosing an appropriate site for oviposition and distributing their eggs more or less equally over the available seeds. They avoid laying eggs on seeds with a rough surface and on seeds already bearing bruchid eggs if noninfested seeds are still available. If noninfested seeds are unavailable, the females will lay eggs on seeds already bearing eggs, but will choose the bigger ones first. The females can also detect small differences in egg density, and prefer to oviposit on seeds bearing smaller number of eggs (Yoshida 1961; Avidov *et al.* 1965; Nwanze *et al.* 1975; Messina and Renwick 1985a, b).

The discriminating ability of female *Callosobruchus* in uniformly distributing their eggs is highly developed, however, in some species the discrimination in choice of seeds suitable for larval growth is not developed. In other words, the ovipositional behaviour was not related to the suitability of seeds for the development of larvae (Avidov *et al.* 1965; Bhattacharya *et al.* 1977). Therefore, the apparent preference for oviposition on the tube wall under these conditions could possibly be more accurately described as a strong repellence to ovipositing on seeds that are already bearing numerous eggs.

It is difficult to say if the differences in the number of eggs laid on the tube wall by different strains of *C. maculatus* were due to differences in the ability of females of different strains to discriminate between suitable sites for larval development or simply because they have different fecundities. For example, the fecundity of *C. maculatus* IITA female was the highest, and that strain female also laid the highest number of eggs on the tube wall. Probably the female of that strain had relatively greater seed shortage problem than the other two strains.

In contrast, *C. chinensis* appears to have more developed discriminating ability to choose suitable sites for larval development. It was found that although females of the C. *chinensis* Indonesia had higher fecundity than those of the *C. maculatus* Yemen and Indonesian strains, fewer eggs were laid on the tube by *C. chinensis* Indonesia.

The number of eggs oviposited by females *C. maculatus* Indonesian and Yemen strain was suppressed when only a small number of seed was available. However, those oviposited by the other strains were not remarkably suppressed with the reduction of seed availability. Credland (1986) stated that the conditions that determine the maximum fecundity differ within and between strains. The reduction in female fecundity as a response to low seed availability is perhaps due to deterrence effects, chemically or physically, of eggs already laid on the seed (Messina and Renwick 1985a).

The development period of *Callosobruchus,* in this experiment was slightly shorter on green gram than that on cowpea. This suggests that green gram is a slightly better host for *Callosobruchus.* Giga and Smith (1978) reported that of the several pulses tested, including green gram and cowpea, green gram was the most favourable food species for oviposition, speed of development and survival of *C. maculatus.*

When suitable host seeds are infested at numbers above the population's optimum density, there is a reduction in the number emerging of adults due to mortality which primarily took place during the larval stage (Utida 1941; Mitchel

1975; Giga 1982; Dick 1984). The reduction in the number of adults produced due to high density was more pronounced in the *C. maculatus* Yemen strain than those of other strains used in this experiment. Variations in the effect of density on the number of adults produced have also been reported to occur among strains of *C. maculatus*. Strains from Brazil and Nigeria (IITA) can produce more than ten adults from a seed with numerous eggs, whereas a strain from Yemen rarely produced more than three (Dick 1984; Dick and Credland 1984; Credland *et al.* 1986). The density effect on the number of adults observed in *C. maculatus* Indonesian strain, *C. chinensis* Indonesian and Kenyan strains seems to be similar to that in *C. maculatus* IITA. However, higher densities than the maximum density recorded in this experiment should have been used to be able to see the effect more clearly.

The geographical variations on the biology and behaviour of *C. maculatus* and *C. chinensis* found in this experiment are possibly the result of either genetic evolution of a population which occupies a particular environment and is therefore subjected to that environment selection pressures; or genetic divergence among populations which is caused by chance fluctuations in its allele frequency; or the change in the gene pool (Dick 1984; Credland 1986).

The occurrence of geographical variations among populations of an insect species should be noted when studying or referring to the species biology or behavioral characteristics. Attention should also be given to the possibility that a less important species population might become a serious pest if a better plant host species or variety were introduced to the area.

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REFERENCE

AVIDOV, Z., S.W. APPLEBAUM, and M.J. BERLINGER, 1965. Physiological aspects of host specificity in the Bruchidae II: Ovipositional preference and behavior of *Callosobruchus chinensis* (L.). Ent. Exp. & Appl. 8: 96-106.

BHATACHARYA, A.K., P.K. PATHAK, and S.P. SHAH, 1977. Oviposition and development of *Callosobruchus chinensis* (Linn) (Coleoptera : Bruchidae) on several host species. Bull. Grain Technol. 15: 38-41.

CREDLAND, P.P. 1986. Effect of host availability on reproductive performance in *Callosobruchus maculatus* (F) (Coleoptera : Bruchidae). J. Stored Prod. Res. 22: 49-54.

CREDLAND, P.P. and K.M. DICK. 1987. Food consumption by larvae of three strains of *Callosobruchus maculatus* Coleoptera : Bruchidae). J. Stored Prod. Res. 23: 31-40.

DICK, K.M. 1984. Bionomic variation among populations of Southern cowpea weevil, *Callosobruchus maculatus* and their responses to different varieties of the primary host. Ph.D. thesis, Bedford College. 305p.

DICK, K.M. and P.F. CREDLAND. 1984. Egg production and development of three strains of *Callosobruchus maculatus* (F.) Coleoptera : Bruchidae). J. Stored Prod. Res. 20: 221-227.

DICK, K.M. and P.F. CREDLAND. 1986. Variation in the response of *Callosobruchus maculatus* (F.) to a resistant variety of cowpea. J. Stored Prod. Res. 22: 43-48.

DOBIE, P. 1981. The use of resistant varieties of cowpea *(Vigna unguiculata)* to reduce losses due to post harvest attack by *Callosobruchus maculatus. In:* The Ecology of Bruchids Attacking Legumes (Pulses): 185-192. (ed. V. Labeyrie). Dr. W. Junk Publisher. London.

GIGA, D.P. 1982. The comparative biology of four *Callosobruchus* species with particular reference to competition in *C. rhodesianus* and C. *maculatus*. Ph.D. thesis, University of Reading.

GIGA, D.P. and R.H. SMITH, 1987. Egg production and development of *Callosobruchus rhodesianus* Pic and *Callosobruchus maculatus* (F.) (Coleoptera : Bruchidae) on several commodities at two different temperatures. J. Stored Prod. Res. 23: 9-15.

MESSINA, F.J. and J.A.A. RENWICK, 1985a. Ability of oviposition seed beetles to discriminate seeds with differing egg loads. Ecological Entomology 10: 225-230.

MESSINA, F.J. and J.A.A. RENWICK, 1985b. Mechanism of egg recognition by cowpea weevil *Callosobruchus maculatus.* Entomol. Exp. Appl. 37: 241-245.

MITCHEL, R. 1975. The evolution of oviposition tactics in the bean weevil. *Callosobruchus maculatus* (F.). Ecology, 56: 696-702.

RAINA, A.K. 1971. Comparative resistance to three species of *Callosobruchus* in a strain of chickpea *(Cicer arietinum* L.). J. Stored Prod. Res. 7: 213-216.

SINGH, S.R. 1978. Resistance to pests of cowpea in Nigeria. *In:* Pests of Grain Legumes: Ecology and Control: 267-279. (ed. Singh *et al.).* Academic Press. London.

SOUTHGATE, B. J. 1978. The importance of Bruchidae as pests of grain legumes, their distribution and control. *In:* Pests of Grain Legumes: Ecology and Control: 219-229. (ed. Singh *et al.).* Academic Press. London.

TDRI. 1984. Insect and Arachnid of Tropical Stored Products : Their biology and identification (ATraining Manual). Tropical Development and Research Institute. Slough.

UTIDA, S. 1941. Studies on experimental population of the azuki bean weevil *Callosobruchus chinensis* (L.) I: The effect of population density on the progeny population. Mem. Coll. Agr. Kyoto Imp. Univ. 48: 1-31.

YOSHIDA, T. 1961. Oviposition behaviour of two species of bean weevils and interspecific competition between them. Mem. Fac. Lib. Arts and Educ., Miyazaki Univ. 11: 41-65.