

Physicochemical Properties of Selected Varieties of Cowpea Seeds and Their Relation to The Infestation Potential by The Cowpea Bruchid, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae)

M.O. Abokersh¹, and E.M. Barakat²

¹Department of Zoology, Faculty of Science, Alasmarya Islamic University, Zliten, Libya.

²Department of Entomology, Faculty of Science, Ain Shams University, Abbasia, Egypt.

الخواص الفيزيوكيميائية لسلاسل بذور اللوبيا وعلاقتها بمعدل الإصابة بخنفساء اللوبيا الجنوبية

محمد عمر أبوكرش¹، عماد محمود بركات²

¹قسم علم الحيوان، كلية العلوم، الجامعة الأسمرية الإسلامية، زليتن، ليبيا

²قسم علم الحشرات، كلية العلوم، جامعة عين شمس، العباسية، مصر.

Abstract

Cowpea, *Vigna unguiculata* (L.) Walp is the most popular legume cultivated in tropical and sub-tropical countries because of its high nutritional value. The physicochemical properties of four local varieties of cowpea seeds (Cream 7, Kaha 1, Dokki 331 and Kafr El-Sheikh 1) in Egypt were studied. Length, major and minor diameter of seeds were in the range of 6.70-12.90 mm, 3.33-5.58 mm, and 3.18-4.65 mm while the grain weight of the seeds varied between 8.40 to 34.90 g. The results showed that Cowpea seeds contain a high value of crude protein in the range of 25.79 to 29.25%. Moisture, dry matter, fat, ash and crude fiber values were in the range of 8.57 to 10.07%, 89.93 to 91.44%, 0.79 to 3.18%, 2.72 to 3.73% and 1.92 to 3.37% respectively. Carbohydrate content varied between 53.56 to 57.36%. When *Callosobruchus maculatus*, the most destructive pest of stored leguminous seeds, was provided with these varieties of cowpea, the percentage of adults emerging differed with variety.

Keywords: Cowpea seeds, Physicochemical properties, *Callosobruchus maculatus*, Infestation potential.

الملخص

اللوبيا، *Vigna unguiculata* (L.) Walp هي البقوليات الأكثر شيوعاً المزروعة في البلدان الاستوائية وشبه الاستوائية بسبب قيمتها الغذائية العالية. تمت دراسة الخواص الفيزيائية والكيميائية لأربعة أصناف محلية من بذور اللوبيا (كريم 7، كاهي 1، الدقي 331، كفر الشيخ 1) في مصر. تراوح الطول والقطر الكبير والصغير للبذور ما بين 6.70-12.90 مم و 3.33-5.58 مم و 3.18-4.65 مم بينما تراوح وزن الحبوب بين 8.40 و 34.90 جم. أظهرت النتائج أن بذور اللوبيا تحتوي على نسبة عالية من البروتين الخام في حدود 25.79 إلى 29.25%. تراوحت قيم الرطوبة والمادة الجافة والدهون والرماد والألياف الخام من 8.57 إلى 10.07%، 89.93 إلى 91.44%، 0.79 إلى 3.18%، 2.72 إلى 3.73% و 1.92 إلى 3.37% على التوالي. تراوح محتوى الكربوهيدرات بين 53.56 إلى 57.36%. عندما تم تزويد هذه الأنواع من اللوبيا عند *Callosobruchus maculatus*، وهي أكثر الآفات تدميراً للبذور البقولية المخزنة، فقد اختلفت النسبة المئوية للبالغين الناشئين مع التنوع.

الكلمات الدلالية: بذور اللوبيا، الخصائص الفيزيائية والكيميائية، *Callosobruchus maculatus*، احتمالية الإصابة.

1. Introduction

Cowpea (*Vigna unguiculata* L. Walp.), an annual legume, is also commonly referred to as southern pea, blackeye pea, crowder pea, lubia, niebe, coupe or frijole. The Cowpea originated from Africa and is widely grown in Africa, Latin America, and Southeast Asia. It is chiefly used as a grain crop for animal fodder, or as a vegetable. In the developed world, cowpea is technologically processed into flour and used in various preparations such as protein concentrate and isolates for the formulation of animal feed. As a food crop it has many advantages including rapid and early growth, wide environmental adaptability, drought resistance, widespread acceptability, a broad range of genetic diversity, and ease of hybridization.

Cowpea seed ranges in size from very small wild types up to nearly 14 *in*. Seed shape is a major characteristic correlated with seed development in the pod. Seeds develop a kidney shape if not restricted within the pod. When seed growth is restricted by the pod the seed becomes progressively more globular. The seed coat can be either smooth or wrinkled and of various colors including white, cream, green, buff, red, brown, and black. Seeds may also be speckled, mottled or blotchy. Many are also referred to as 'eyed' (blackeye, pinkeye purple hull, etc.) where the white colored hilum is surrounded by another color (Giga and Smith, 1981).

A specific knowledge of the cowpea seed physical properties such as color, shape, porosity, volume, density, coefficient of static friction etc. and the chemical properties of fruit such as moisture, ash, crude fiber, protein, fat and carbohydrate contents is necessary for the selection of cowpea varieties that resist adverse storage conditions and abiotic and biotic stresses.

Insect pests are major constraint to cultivation and the family Bruchidae includes major pests of legume seeds. The cowpea weevil, *Callosobruchus maculatus* F. (Coleoptera: Bruchidae) is one of the most destructive pests of stored leguminous seeds. Infestations often begin in the field but the serious damage is done in the store where the insects spread from seed to seed until eventually up to half are holed and considerable losses of quality and market value are caused (Caswell, 1975). Because it is difficult to find suitable cheap methods of control, emphasis is being placed on developing acceptable new varieties that have a natural resistance to bruchids as well as a high yield.

Laboratory tests designed to compare the susceptibility to insects of stored seeds should give consistent results wherever and whenever used. The numbers of insects and the quantities of food used in these tests in order to discriminate between the susceptibility of varieties are crucial (Giga and Smith, 1981).

The objective of this study is to evaluate the level of cowpea infestation by cowpea bruchids, *C. maculatus* in proportion to the properties of cowpea variety. The rate of

emergence of adult insects is also studied for the different cowpea varieties. This study therefore sought to answer these questions: (1) Whether the outcome of larval penetration differed between seed varieties? and (2) Whether the physicochemical properties of a seed correlated with the seed varietal resistance?

2. Materials and Methods

2.1. Source of Seeds

Four local varieties of cowpea seeds, *Vigna unguiculata* (L.) namely: Cream 7, Kaha 1, Dokki 331, and Kafr El-Sheikh 1 were purchased from the Food Legume Research Section, Plant Protection Institute, Agriculture Research Center, Dokki, Giza, Egypt. Extraneous matter such as unhealthy seed, insect infested seed sand and chaff were removed from the samples before processing. Cowpea seeds were separately milled with an attrition mill (Model no ED-5) and sieved to a particle size of 1 mm. Flour samples were packaged in low density polyethylene bags and stored using covered plastic containers in a freezer at -18°C.

2.2. Determination of Physical Properties

2.2.1. Seed Color and Texture

The color of seeds was determined according to the method of Gomez *et al.* (1997). The color was effectively observed by placing 20 grain samples on a sheet of white paper. The difference in color of the outer coat of the grain was recorded. The external surface of the seed was examined whether it is smooth, rough, or wrinkled as described by Khare and Johari (1984).

2.2.2. Major and Minor Diameter

The major diameter of seeds was determined by the measure of cowpea in its greatest dimension, and the minor diameter was measured at the dorsal side as described by Dela and Khush (2000). The parameters were measured with a caliper (micrometer of high accuracy). The results obtained were recorded as average values of five determinations in millimeter.

2.2.3. Seed Weight

The seed weight was determined by weighing 100 randomly selected raw seeds of each variety (AOAC, 2000). Measurements replicated 5 times for each seed variety.

2.3. Determination of Chemical Properties

Local cowpea varieties were analyzed for moisture, ash, crude fiber, protein, fat and carbohydrates. All determinations were carried out using standard procedures (AOAC, 2000). Data were presented as means \pm SD from triplicate determinations.

2.3.1. Tannins

Tannins were determined by using vanillin-hydrochloric acid method (V-HCl) as described by Price *et al.* (1978). About 2 g of fine ground sample was mixed with 50 ml methanol containing 1% HCl, shaken well, left for 24 hrs at room temperature, filtered and then completed to 50 ml of vanillin reagent (mixture of equal volumes of 4% vanillin solution and 8% HCl) and kept for 20 min. The intensity of the color produced was measured on Spectronic-20 spectrophotometer at 500 nm. A standard curve was established from catechin concentrations in methanol and then measured at 500 nm. The total tannins content was expressed as mg/100 g seeds.

2.3.2. Total Phenols

Total phenols were determined according to methods described by Gutfinger (1981). About 2 g of fine ground sample was mixed in 80% methanol and left for 24 hrs at room temperature and then filtered. The filtrated solution was completed to a constant volume with methanol. About 0.5 ml of the solution was mixed with 0.5 ml of Folin-Ciocalteu reagent, left for 3 min and then added to 1 ml of 10% freshly prepared sodium carbonate solution (w/v). The mixture was completed to 10 ml with distilled water and then measured at 720 nm on Spectronic-20 spectrophotometer. A standard curve was established from pyrogallol concentrations in methanol and then measured at 720 nm. The total phenols content was expressed as mg pyrogallol/100 g seeds.

2.4. Insect Culture

A stock culture of *Callosobruchus maculatus* (F.) was collected from infested cowpea seeds obtained from local grain shops and reared on cowpea seeds at room temperature (28-30°C and 65-75% RH) for several generations. Insects for use in experiments were established on different cowpea seed varieties.

2.5. Infestation of Seeds

Groups of 5 g seeds of the different varieties were distributed in small glass tubes. Bruchid females were allowed to lay eggs on seeds. Most of the observations were made using single seed. At least five replicates were prepared for each variety. Uninfected seeds were used as controls. Larval density was manipulated experimentally after first allowing one male and one female to mate and oviposit for two days. Peas and emerged adults were weighed individually.

2.6. Infestation Potential and Seed Varietal Resistance

Groups of 5 g seeds of the different varieties were distributed and kept in glass vials covered with muslin cloth and a single pair of newly emerged adults of *C. maculatus* was introduced into each of five vials for oviposition. Vials were maintained at rearing conditions (28-30°C and 65-75% RH). After 2 days (the oviposition period) the insects were discarded. The

numbers of eggs laid on the seeds of each variety were counted and observed for hatchability 5 days after removal of adult insects. Each vial was examined daily and when an adult emerged, its weight and developmental period were determined and recorded to evaluate developmental performance of *C. maculatus* on cowpea seeds. When emergence was complete, the weight of each damaged cowpea group was determined and percentage loss of dry weight was calculated.

2.7. Statistical Analysis

Percentage emergence and weight loss were analyzed by analysis of variance, using the angular transformation on the emergence data. ANOVAs and protected least significant difference (LSD) tests were used in each of the experiments. All analyses were performed using Super ANOVA, V 16.0 (Abacus Concepts, Inc., Berkeley, CA).

3. Results

3.1. Physical Characteristics

The results of the physical characteristics of cowpea seeds are presented in Table (1). Significant differences (5%) were observed among the cowpea varieties for all the physical properties. Colors of seeds include cream (Cream 7), white (Kaha 1), brown (Dokki 331), and brown (Kafr El-Sheikh 1). Cream 7 and Kaha 1 were smooth seeded. Dokki 331 and Kafr El-Sheikh 1 varieties had a rough seed coat. Grain weight ranged between 0.130 and 0.337 mg. Length of cowpea seeds varied from 6.75 mm (Kafr El-Sheikh 1) to 12.94 mm (Dokki 331). Major diameter of seeds ranged from 3.33 to 5.58 mm and minor diameter of cowpea seeds varied from 3.18 to 4.65 mm. The four varieties have calculated significant coat thickness. Test a thickness of seeds ranged from 0.080 to 0.154 mm.

3.2. Chemical characteristics

The four varieties of cowpea seeds have approximately similar chemical properties of their cotyledon components, except the percentages of crude fiber, crude protein and contents of tannins (Table 2). They have insignificant difference in their moisture content, percentages of ash and total phenols. Brown seeded varieties have higher contents of crude fiber, crude proteins and carbohydrates than white seeded ones. White seeded varieties have higher tannin contents. There was significant difference ($P < 0.05$) in fiber content between Kaha 1 and other varieties.

Kaha 1 variety had the highest fat content of (3.16%) and Cream 7 had the lowest of (0.75%). There was significant difference ($P < 0.05$) between Dokki 331 and Cream 7 when compared to the other varieties. There was no significant difference ($P > 0.05$) in fat content between Kaha 1 and Dokki 331. However, there was a significant difference ($P < 0.05$) when Kafr El-Sheikh 1 variety was compared to the other samples.

Table 1. Some physical characteristics of cowpea, *V. unguiculata* seed varieties

Seed Variety	Seed color	Seed texture	Seed weight (g)	Seed length (mm)	Major diameter (mm)	Minor diameter (mm)	Helix color	Testa thickness (mm)
Cream 7	Cream	Smooth	0.130 ±0.004 ^a	8.41 ±0.36 ^a	4.03 ±0.10 ^a	3.33 ±0.09 ^a	White	0.080 ±0.002 ^a
Kaha 1	White	Smooth	0.270 ±0.032 ^a	9.81 ±0.42 ^a	4.34 ±0.18 ^a	3.48 ±0.11 ^a	Black	0.123 ±0.002 ^b
Dokki 331	Brown	Rough	0.337 ±0.033 ^b	12.94 ±0.37 ^b	5.58 ±0.24 ^b	4.65 ±0.17 ^b	Black	0.134 ±0.003 ^b
Kafr El-Sheikh 1	Brown	Wrinkled	0.226 ±0.018 ^a	6.75 ±0.32 ^a	3.33 ±0.16 ^a	3.18 ±0.10 ^a	White	0.154 ±0.004 ^c

The data reported are the mean ± SD, N = 5.

Means within a column followed by a same letter(s) are not significantly different from each other at 5% level of significance.

Table 2. Some chemical characteristics of cotyledons of cowpea, *V. unguiculata* seed varieties.

Seed variety	Moisture Content (%)	Ash (%)	Crude fiber (%)	Crude Protein (%)	CHO (%)	Fat (%)	Tannins*	Total Phenols**
Cream 7	9.39 ± 0.88 ^a	3.46 ± 1.01 ^a	2.33 ± 0.32 ^a	25.08 ± 3.49 ^a	53.53 ± 2.73 ^a	0.75 ± 0.20 ^a	21.72 ± 4.28 ^a	17.15 ± 2.64 ^a
Kaha 1	8.74 ± 0.91 ^a	3.67 ± 1.14 ^a	1.27 ± 0.09 ^b	25.95 ± 2.01 ^a	53.60 ± 3.92 ^a	3.18 ± 0.37 ^b	19.86 ± 2.13 ^a	16.80 ± 1.16 ^a
Dokki 331	8.80 ± 1.32 ^a	3.33 ± 0.52 ^a	3.44 ± 0.27 ^a	28.88 ± 1.95 ^b	57.79 ± 2.84 ^b	3.11 ± 0.79 ^b	10.71 ± 4.28 ^b	18.13 ± 2.00 ^a
Kafr El-Sheikh 1	8.41 ± 0.91 ^a	3.17 ± 1.14 ^a	3.59 ± 0.30 ^a	28.66 ± 3.05 ^b	53.65 ± 2.13 ^a	2.43 ± 0.99 ^b	10.61 ± 2.04 ^b	18.85 ± 2.32 ^a

The data reported are the mean ± SD from triplicate determinations.

Means within a column followed by a same letter(s) are not significantly different from each other at 5% level of significance.

* (mg catechin/100 g seeds).

** (mg pyrogallol/100 g seeds).

3.3. Infestation Potential of *C. maculatus* and Seed Varietal Resistance

Mean percentage emergence and mean development time of emergent adults of *C. maculatus* for each cowpea variety and the analysis of variance of the transformed data are given in Table (3). The statistical treatment of data of *C. maculatus* oviposition on four cowpea

varieties revealed significant differences on the total eggs production (fecundity). Cream 7 received the highest numbers of eggs. Mean number of eggs laid ranged from 49.5 in Kafr El-Sheikh 1 to 78.8 in Cream 7. Taking the larval penetration (%) and development of *C. maculatus* into consideration, the obtained results indicated that there were insignificant differences among the tested varieties. Adult emergence (%) of *C. maculatus* from different cowpea varieties ranged from 51.57% to 92.39%. Significant differences were detected between the four varieties in terms of susceptibility index. Kafr El-Sheikh 1 gave the lowest susceptibility index with *C. maculatus* and was considered moderately resistant (MR), compared to cream 7, which was susceptible (S).

Table 3. Susceptibility of cowpea, *V. unguiculata* seed varieties to *C. maculatus* infestation

Seed variety	Fecundity	Larval penetration (%)	Developmental time (days)	Adult emergence (%)	Susceptibility index (SI)	Loss in Seed weight (%)
Cream 7	77.4 ± 1.95 ^a	89.41 ± 0.03 ^a	25 ± 2.80 ^a	80.00 ± 16.0 ^a	7.61 (S)	27.34 ± 2.8 ^a (HS)
Kaha 1	69.2 ± 7.12 ^b	89.61 ± 0.05 ^a	29.0 ± 0.7 ^b	92.39 ± 4.0 ^a	6.78 (MS)	23.29 ± 6.95 ^b (MS)
Dokki 331	55.8 ± 5.31 ^c	89.60 ± 0.04 ^a	31.4 ± 0.9 ^c	78.94 ± 9.0 ^a	6.04 (MS)	15.90 ± 7.73 ^c (LS)
Kafr El-Sheikh 1	49.0 ± 2.92 ^d	88.55 ± 0.06 ^a	34.4 ± 3.05 ^d	51.57 ± 12.0 ^b	4.98 (MR)	10.10 ± 5.50 ^d (LS)
F-value	4.89	NS	3.92	5.08	-	3.86
LSD- 5%	4.64	-	0.43	1.27	-	4.78

The data reported are the mean ± SD, N = 5.

Means within a column followed by a same letter(s) are not significantly different from each other at 5% level of significance

NS = non significant, HS = highly susceptible, MS = moderately susceptible, LS = least susceptible.

Considering the total weight loss (%) by the pest *C. maculatus*, there were significant differences among the tested varieties in the terms of susceptibility index and weight loss (%). There was a marked low reduction in the percentage of weight loss of Kafr El-Sheikh 1 variety and was considered least susceptible (LS) compared to Cream 7 variety which considered highly susceptible (HS).

4. Discussion

Results of the present investigation indicated that there were significant differences among the tested cowpea varieties for both physical and chemical properties and some of them may play a role as tolerance factors. These differences in size and grain weight among cowpea varieties may be attributed to difference in genetic traits. However, the variations in the chemical compositions recorded in the varieties evaluated may be attributed to soil type, cultural practices, environmental condition and genetic factors.

The variation in cowpea color agrees with the findings of Bergmann *et al.* (1994), who reported variability in cowpea colors among different varieties. The color differences also have implication in the characteristics of the product produced from cowpea. Difference in color also might have quality implication where used as a composite flour.

Our results indicated that the brown colored seeds (Kafr El-Sheikh 1 and Dokki 331) were less preferred for *C. maculatus*. Osuji (1976) have demonstrated that seed color, testa color, and texture did not seem to exert an overriding influence on pest susceptibility but may influence the oviposition behavior of *C. maculatus* females and other factors of the seed may determine the pre-adult development and the ultimate number of the progeny. In contrast, Khattack *et al.* (1987) stated that small-sized seeds of mungbean varieties were significantly less susceptible to bruchid infestation in terms of progeny production of *C. maculatus* than the large-sized seeds. Chavan *et al.* (1997) mentioned that smooth texture and dark colored seeds were preferred by *C. chinensis* and *C. maculatus* for oviposition than rough surface and white colored seeds. They added that mean developmental period (MDP) is increased by 10 times between the most and the least preferred cowpea lines.

The four varieties of cowpea have calculated significant coat thickness. Seed coat thickness may have a role in larval penetration. Larval survival during penetration of the seed coat is also affected by surface texture and structure, and larval development within seeds depends on quality and compactness of seed as well as the amount of food available (Nwanze and Horber, 1976). The amount of food available per developing larva is not the only explanation for increased susceptibility. In our experiment, numbers of eggs per seed were adjusted to ensure that each larva had the requisite amount of food yet there were still highly significant differences of mortality between varieties.

Plant breeders have increased yields by selecting for larger seeds, but large seeds are also preferred by bruchid pests for oviposition (Avidov *et al.*, 1965), particularly if smooth-coated and well-filled (El-Sawaf 1956; Srivastava and Bhatia 1959; Booker 1967; Nwanze and Horber 1976; Giga and Smith, 1981; and Chinma *et al.*, 2008).

The present investigation, regarding physical properties of different cowpea varieties and their relation to *C. maculatus* infestation has clearly demonstrated that Dokki 331 is the biggest tested variety in length of seeds when compared to other tested varieties. Although the

other varieties have approximately similar size, they were unlikely in their susceptibilities and hence seed size cannot be the main factor influencing the performance of the pest.

Regarding the chemical properties and their correlation to seed varietal resistance, Mueke (1986) found that the resistant cowpea seed varieties have more fiber content. Singh *et al.* (1995) found that the legume varieties that resist bruchids infestation were characterized by high protein contents. Similar results were obtained in the present investigation. Thus the resistance of pea varieties may correlate to high crude fiber and protein contents.

Tannins may affect the growth of insects in three main ways: they have an astringent taste which affects palatability and decreases feed consumption, they form complexes with proteins of reduced digestibility and they act as enzyme inactivators (Swain, 1977). The high levels of tannins in Cream 7 and Kaha 1 (white-seeded varieties of cowpea) were not effective barriers against *C. maculatus*. Moreover, a correlation was found between tannins content and oviposition and adult emergence. However, it could be inferred that no single tested phenotypic or chemical characters favored or inhibited the rate of oviposition and subsequent development to be determining the susceptibility of cowpea varieties to *C. maculatus* pest.

The weight loss means given in the present investigation having a statistically significant effect. Hence the inherent resistance of the cowpea varieties is expressed in the percentage emergence and consequently in the rate of buildup of an infestation rather than in damage caused by a given infestation. Our results in this respect are in accordance with the results of Mensah (1986) which indicated significant differences between the legume varieties in terms of weight loss percentages and susceptibility index. The reduction in the percentages of weight loss may be attributed principally to increased larval mortality observed particularly with the least preferred cowpea varieties.

Clearly the differences between varieties were significant biologically as well as statistically and according to the classification of Khare and Johari (1984), and Mensah (1986). The varieties Kafr El-Shekh 1 and Dokki 331 were notably the least susceptible to infestation by *C. maculatus* than the fairly susceptible varieties Cream 7 and Kaha 1.

References

- AOAC (2000). *Official methods of analysis*. Association of official Analytical chemists. Washington DC., 16th Edition.
- Avidov Z., Applebaum S.W., and Berlinger M.J. (1965). Physiological aspects of host specificity in the Bruchidae. II. Ovipositional preference and behaviour of *Callosobruchus chinensis* (L.). *Entomologia experimentalis et applicata*, 8: 96-106.
- Bergmann C.J., Gualberto D.G., and Weber C.W. (1994). Development of a high temperature. Dried. Soft wheat pasta supplemented with cowpea *Vigna unguiculata* (L.) Walp. Cooking. Quality, colour and sensory evaluation. *Cereal Chem.*, 71: 523-527.

- Booker R.H. (1967). Observations on three Bruchids associated with cowpeas in Northern Nigeria. *Journal of Stored Products Research*, 3: 1-15.
- Bressani R., and Elias L.G. (1984). *Legumes foods*. In: Altschul, A.M. (Editor), New protein food New York/London: Academic press, IA: 230-297.
- Caswell G.H. (1975). *The storage of grain legumes*. 9th Annual Conference of the Entomological Society of Nigeria, 9: 1-5.
- Chavan P.D., Singh Y., and Singh S.P. (1997). Ovipositional preference of *Callosobruchus chinensis* for cowpea lines. *Indian J. Ent.*, 59 (3): 29303.
- Chinma C.E., Alemode I.C., and Emelife I.G. (2008). Physicochemical and Functional Properties of Some Nigerian Cowpea Varieties. *Pakistan Journal of Nutrition*, 7(1): 186-190.
- Dela N.C., and Khush G.S. (2000). Rice grain quality evaluation procedure. In aromatic Rice (Ed). R.K.
- El-Sawaf S.K. (1956). Some factors affecting the longevity, oviposition and rate of development in the southern cowpea weevil, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). *Bulletin of the Society of Entomology*, 40: 29-95.
- Giga D.P., and Smith, R.H. (1981). Varietal resistance and intraspecific competition in the cowpea weevils *Callosobruchus maculatus* and *C. chinensis* (Coleoptera: Bruchidae). *Journal of Applied Ecology*, 18: 755-761
- Gomez M.I., Obilana A.B., Martin D.F., Madzramuse M., and Monyo E.S. (1997). *Manual of laboratory procedures for quality evaluation of sorghum and pearl millet*. ICRESTAT (International crop research Institute for the semi-Arid tropics) Patancheru 502324, Andhra Pradesh, India.
- Gutfinger T. (1981). Phenoles in olive oils. *J. Am. Oil. Chem. Soc.*, 58: 966-968.
- Khare B.P., and Johari R.K. (1984). Influence of phenotypic characters of chickpea (*Cicer arietinum* L.) cultivars on their susceptibility to *Callosobruchus chinensis* (L.). *Legume Res.*, 7(1): 54-56.
- Khattack S.U.K., Khatoon H.R., and Mohammad T. (1987). Relative susceptibility of different mungbean varieties to *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). *J. Stored Prod. Res.*, 23(3): 139-142.
- Mensah G.W.K. (1986). Infestation potential of *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) on cowpea cultivars stored under sub-tropical conditions. *Insect Sci. Applications*, 7(6): 78784.
- Mueke J.M. (1986). Varietal susceptibility Of cowpeas to *Callosobruchus maculatus* (F.). *East African Agric. Forestrt. J.*, 52(2): 10105.

- Nwanze K.F., and Horber E. (1976). Seed coats of cowpeas affect oviposition and larval development of *Callosobruchus maculatus* (F.). *Environmen. Entomol.*, 5: 213-218.
- Osuji F.N.C. (1976). A comparison of the susceptibility of cowpea varieties to infestation by *Callosobruchus maculatus* (F.). *Ent. Exp. Appl.*, 20: 209-217.
- Price M.I., Van Scoyoc S., and Butler L.G. (1978). A critical evaluation of the vanillin reagent as assay for tannins in sorghum grain. *J. Agric. Food Chem.*, 26: 1214-1218.
- Singh V.N., Pandey N.D., and Singh T.P. (1995). Relative resistance of gram varieties to *Callosobruchus chinensis* (L.) on the basis of biochemical parameters. *Indian J. Ent.*, 57(2): 77-82.
- Srivastava B.K., and Bhatia S.K. (1959). The effect of host species on the oviposition of *Callosobruchus chinensis* (L.). *Annals of Zoology*, 3: 37-42.
- Swain T. (1977). Secondary compounds as protective agents. *Ann Rev Plant Physiol.*, 28: 479-501.