BioPublisher

Bioscience Methods 2014, Vol.5, No.2, 1-8
http://bm.biopublisher.ca

Research Article **Open Access**

Anti-nutritional factors in cowpea cultivars and their effects on susceptibility to *Callosobruchus maculatus* (Fab.) [Coleoptera: Bruchidae] infestation

Kayode David Ileke
Department of Environmental Biology and Fisheries, Faculty of Science, Adekunle Ajasin University, PMB 001, Akungba Akoko, Ondo State, Nigeria
Corresponding author email: kayodeileke@yahoo.com
Bioscience Methods, 2014, Vol.5, No.2 doi: 10.5376/bm.2014.05.0002
Received: 30 Mar., 2014
Accepted: 08 Sep., 2014
Published: 20 Nov., 2014

Preferred citation for this article:
Ileke, 2014, Anti-nutritional factors in cowpea cultivars and their effects on susceptibility to *Callosobruchus maculatus* (Fab.) [Coleoptera: Bruchidae] infestation, Bioscience Methods, Vol.5, No.2 1−8 (doi: 10.5376/bm.2014.05.0002)

Abstract Thirty-one cowpea cultivars from IITA in Nigeria were investigated for their antinutritional factors to determine their susceptibility to cowpea bruchid, *Callosobruchus maculatus* (Fab.) [Coleoptera: Chrysomelidae] infestation. Percentage seed damage and pest tolerance, Phytate, tannins and oxalate contents were determined using standard methods. The result showed significant variations in both % seed damage and pest tolerance among the cowpea cultivars tested. Cultivars MIT04K-399-1, MIT07K-299-92, IT96-610, MIT06K-281-1, MIT07K-187-24, MIT06K-121, MIT07K-304-9 and EIT07K-291-69 were the least susceptible to *C. maculatus* with 100% pest tolerance. The result of anti-nutritional factors clearly showed that cowpea seeds contain more phytate than oxalate and tannin. The mean phytate content in seeds was (3.10 mg/g) which was about three times higher than that of oxalate (0.78 mg/g) and five times higher than tannins (0.31 mg/g). Cultivar MIT04K-339-1 recorded the highest amount of phytate while cultivar MIT03K-337-6 recorded the lowest. The amount of oxalate ranges from 0.42 mg/g to 0.92 mg/g. Cultivar IT96-610 recorded the highest amount of tannin while cultivar MIT03K-337-6 recorded the lowest amount of tannin. This study revealed variations in the anti-nutrients composition among the cowpea cultivars. These variations contribute to the susceptibility of cowpea to *C. maculatus* infestation.
Keywords *Callosobruchus maculatus*; Cowpea seeds; Pest tolerance; Phytate; tannins; Oxalate; Susceptibility indices

Introduction

Cowpea, *Vigna unguiculata* (L) Walp is one of the most important food legume crops widely grown in semi-arid tropics as an inexpensive source of protein in both human diet and animal feed (Mahe et al., 1994; Ofuya, 2001). The cowpea bruchid, *Callosobruchus maculatus* (Fab.) has been recognized for decades as the major post harvest insect pest of cowpea seeds. It is a cosmopolitan species and a field-to-store insect pest of cowpea (Ofuya, 2001; Gbaye and Holloway, 2011). The huge post-harvest losses and quality deterioration caused by this insect is a major obstacle to achieving food security in developing countries such as Nigeria (Ileke et al., 2013).

Anti-nutritional factors are plant’s secondary metabolites which act to reduce food nutrient utilization (Soetan, 2008). Anti-nutritional factors affect susceptibility of grains to insect attack (Harborne, 1989). However, the presence of anti-nutritional factors commonly found in legumes is a major factor limiting the wider food use of these essential tropical plants (Liener, 1980). For instance, phytic acid and Oxalic acid reduce mineral bioavailability that leads to various mineral deficiency diseases e.g. anaemia (Gluthrie and Picciano, 1996), or form deleterious complexes with metal ions e.g. calcium-oxalate that leads to renal damage (Shukkur et al., 2006). Plants contain thousands of compounds which, depending upon the situations, can have beneficial or deleterious effects on organisms consuming them. These compounds, with the exception of nutrients, are referred to as ‘allelochemicals’ (Conn, 1979).

Anti-nutrients have been shown to possess pharmacological values. Tannins for examples, possess anticancer and cytotoxic properties (Koratkar and Rao, 1997; Das and Mahato, 1983; Schopke and Hiller, 1990; Wakabayashi et al., 1997). Tannins are complex polyphenol found widely in the plant kingdom (Haregman and Buther, 1978). Phytic acid's mineral binding properties is believed to prevent colon cancer by reducing oxidative stress in the lumen of the intestinal tract (Vucenik and Shamsuddin, 2003; Jenab and Thompson, 2000). The chelating effect may serve to prevent, inhibit, or even cure some cancers by depriving those cells of the minerals (especially iron) they need to reproduce (Klopfenstein et al., 2002). Antinutrients are found in almost all foods. However, their levels are reduced in most common food crops probably through selection during the process of domestication. Nevertheless, the large fraction of human diets that come from these crops raise concern about the possible effects of anti-nutrients on human health (Cordain, 1999). The possibility now exists to eliminate anti-nutrients entirely using genetic engineering, but since these compounds may also have beneficial effects, such genetic modifications could make the food crops more nutritious without the capacity to improve other aspects of human health (Welch, 2004).

Phytic and oxalic acids are among the major anti-nutrients present in plant protein sources (Akande et al., 2010), both being anti-minerals. Phytic acid, also known as inositolhexakisphosphate (IP6), or phytate when in salt form is the principal storage form of phosphorus in many plant tissues, especially bran and seeds (Klopfenstein et al., 2002). It is not digestible to humans or non-ruminant animals, because these animals lack the digestive enzyme (phytase) required to remove phosphate from the inositol in the phytate molecule. On the other hand, ruminants readily digest phytate because of the phytase produced by microorganisms in their rumen (Klopfenstein et al., 2002). Phytate is well documented to block absorption of not only phosphorus, but also of other minerals such as calcium, magnesium, iron and zinc (Ramiel, 2010; Klopfenstein et al., 2002).

However, soaking, cooking, boiling and other food processing methods generally achieve significant reduction of the anti-nutrients (Udensi et al., 2005; 2007; Ekop et al., 2004; Ekop and Eddy, 2005). Thus, foods high in these anti-nutrients should be adequately processed to make them wholesome for consumers. In ruminants however, dietary oxalic acid can be degraded by rumen microbes into CO_2 and formic acid (Allison et al., 1990). The amount of antinutrients in food crops is highly variable and depends on factors including environmental condition, use of high-phosphate fertilizers in cultivation and genotypic variation (Offor et al., 2011). In this study, thirty-one cowpea cultivars were investigated for their anti-nutritional factors to determine their susceptibility to cowpea bruchid, *C. maculatus* infestation.

1 Materials And Methods

1.1 Sources of Experimental Insects and cowpea cultivars

Newly emerged adult *C. maculatus* used for this study were obtained from already existing culture in the Postgraduate Research Laboratory of the Department of Biology, Federal University of Technology, Akure, Ondo State, Nigeria. Insect rearing and the experiments were carried out at ambient temperature of 28$\pm$2^{o}C and 75$\pm$5% relative humidity.

The thirty one cowpea, *Vigna unguiculata* cultivars: MIT04K-339-1, MIT07K-291-92, MIT07K-211-108, MIT07K-292-10, EIT07K-303-1, EIT07K-243-1-10, EITO7K- 234-1-5, IT067-154-1, IT96-619, IT845-2246-4, IFE BROWN, MIT06K-128, MIT07K-188-49, MIT0K-835-45, MIT07K-318-33, MIT07K-309-44, MIT06K-281-1, MIT07K-187-24, MIT06K-124, MIT04K-219-2, MIT04K-321-2, MIT07K-304-9, MIT03K-337-6, MIT06K-121, MIT07K-194-3, MIT98K-503-1, EIT04K-221-1, EIT07-291-69, EIT07K-299-4, EIT03K-369-3 and EIT97K-499.35 used for this study were provided by the Cowpea Seeds Unit, International Institute for Tropical Agriculture, Ibadan, Oyo State, Nigeria.

1.2 Susceptibility of Cowpea Cultivars to *C. maculatus*

Twenty grammes of each cowpea cultivar was weighed into 250ml plastic containers and ten pairs of adult *C. maculatus* (2 to 3 days old) were introduced into each container. The containers were covered with tight lid that have been cut at centre and sealed with muslin cloth for aeration. This was replicated three times. The infested cowpea seeds was left for 7 days in an insect cage in the laboratory during which the insects fed and laid eggs. On day 7 of infestation, beetles were removed and discarded. Numbers of eggs laid by adult beetles were counted and recorded. Twenty five days after infestation with beetles, the containers were checked daily for emerged adults.

Each trial replicate was terminated when no adult emergence for five consecutive days. The developmental period were also recorded.

In calculating % weight loss, the contents of each container were sieved to remove dust, frass and any insect present within the seeds. The seeds were re-weighed and the % weight loss was determined as the difference between the initial and final weights of seeds in each replicate divided by the initial weight multiplied by 100 as described by Odeyemi and Daramola (2000).

$$\%\ \text{Weight loss} = \frac{\text{Change in weight}}{\text{Initial weight}} \times \frac{100}{1}$$

After re-weighing, the numbers of damaged cowpea seeds were evaluated by counting wholesome and bored or seed with bruchid emergent holes. Percentage seed damaged and pest tolerance were calculated according to the method described by Lephale et al. (2012) as follows:

$$\%\ \text{Seed damage} = \frac{\text{Number of seeds damaged}}{\text{Total number of seeds}} \times \frac{100}{1}$$

$$\%\ \text{Pest Tolerance} = \frac{\text{Number of undamaged seeds} - \text{Number of damaged seeds}}{\text{Number of undamaged seeds}} \times \frac{100}{1}$$

Susceptibility indices (SI) were calculated according to the method of Dobie (1974) and is given as;

$$\text{Index of Susceptiblity SI} = \frac{\text{Loge for number of F1 adult} \times 100}{\text{Time of emergence of 50\% F1 generation}}$$

1.3 Anti-nutritional composition of different cowpea cultivars

1.3.1 Oxalate

Total oxalate was determined according to Day and Underwood (1986) procedure. To 1 g of the ground powder, 75 ml of 15 N H_2SO_4 was added. The solution was carefully stirred intermittently with a magnetic stirrer for 1 h and filtered using Whatman No 1 filter paper. Twenty five ml of the filtrate was then collected and titrated against 0.1 N $KMnO_4$ solutions till a faint pink colour appeared that persisted for 30 seconds.

1.3.2 Phytate

Phytate was determined using Reddy and Love (1999) method. four grammes of the ground sample was soaked in 100 ml of 2% HCl for 5 h and filtered. To 25 ml of the filtered, 5 ml 0.3% ammonium thiocyanate solution was added. The mixture was then titrated with Iron (III) chloride solution until a brownish-yellow colour that persisted for 5 min was obtained.

1.3.3 Tannin

Tannin was determined using the method of Trease and Evans (1978). One ml of the methanolic extract was treated with 5 ml Folin Dennis reagent in a basic medium and allowed to stand for colour development. The absorbance of the reaction mixture of each sample was measured at 760 nm spectrophotometrically.

1.4 Data Analysis

Data were subjected to analysis of variance (ANOVA) and treatment means were separated using the New Duncan's Multiple Range Test. The ANOVA was performed with SPSS 16.0 software (SPSS, 2007).

2 Results

2.1 Seed damaged and Pest tolerance.

There was no seed damaged in cultivars MIT04K-399-1, MIT07K-299-92, IT96-610, MIT06K-281-1, MIT07K-187-24, MIT03K-337-6, MIT07K-304-9 and EIT07K-291-69. The result revealed that the above cultivars had 100% pest tolerance. Cultivars IFE BROWN, MIT03K-337-6, MIT04K-219-2 and MIT07K-292-10 were the most susceptible cultivars to *C. maculatus* infestation with 18.98%, 20.03%, 29.04% and 30.94.63% pest tolerance respectively (Figure 1). Generally, it was observed that those cultivars that had least seed damage, had higher % pest tolerance. Consequently, % pest tolerance varied directly with number of undamaged seed.

2.2 Anti-nutritional contents of different cowpea cultivars

Table 1 presented the anti-nutritional contents of different cowpea cultivars. The result clearly showed that cowpea seeds contain more phytate than oxalate and tannin. The mean phytate obtained in this study (3.10 mg/g) is about three times higher than that of oxalate (0.78 mg/g) and five times higher than tannin (0.31mg/g). Cultivar MIT04K-339-1 (3.92mg/g) had the highest amount of phytate and cultivar MIT03K-337-6 (2.58 mg/g) had the lowest amount of phytate. The amount of oxalate ranges from 0.42mg/g to 0.92mg/g. Cultivar IT96-610 (0.56 mg/g) recorded the highest amount of tannin and cultivar MIT03K-337-6 (0.19 mg/g) recorded the lowest amount of tannin (Table 1).

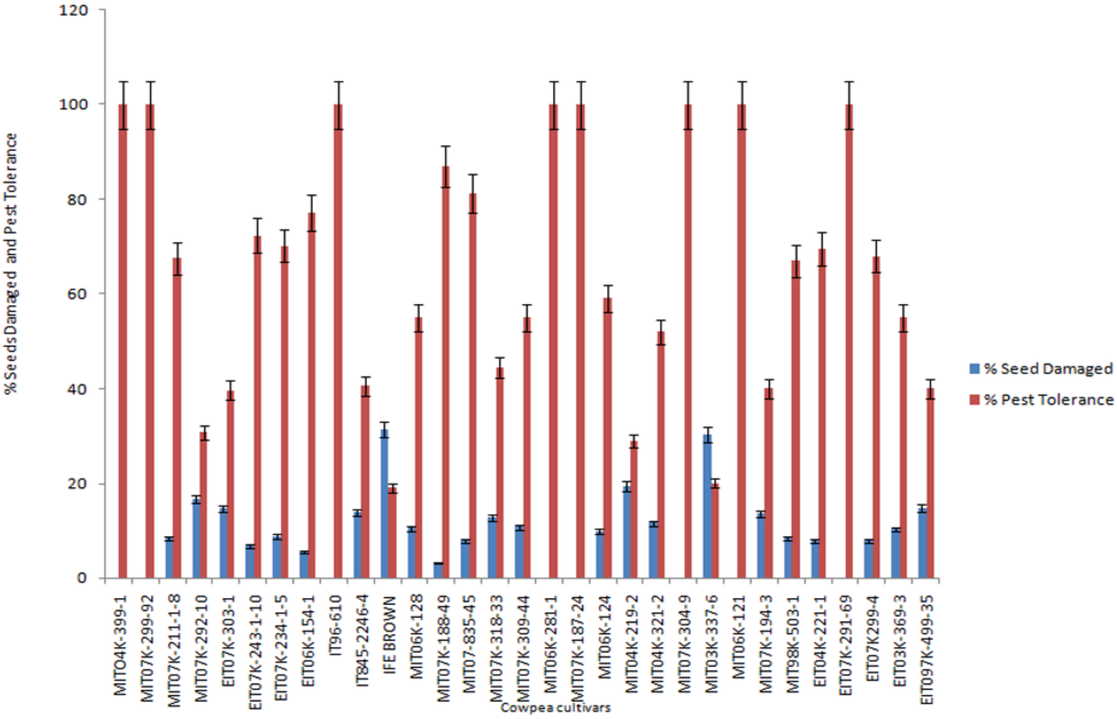

Figure 1 % Seed Damaged and Pest Tolerance by *C. maculatus* in cowpea cultivars

Table 1 Relative susceptibility of thirty one cowpea cultivars to *Callosobruchus maculatus*

Cowpea cultivars	Number of eggs laid	% adults emerged	Development time (days)	% seed damage	% Pest tolerance	% Weight loss	Susceptibility indices
MIT04K-339-1	4.1	0.0	0.0	0.0	100.0	0.1	0.00
MIT07K-299-92	5.0	0.0	0.0	0.0	100.0	0.1	0.00
MIT07K-211-1-8	12.5	63.9	24.6	8.5	81.2	5.6	2.86
MIT07K-292-10	29.9	70.3	22.8	16.6	73.4	9.4	3.11
EIT07K-303-1	19.5	71.8	24.7	14.7	74.8	6.2	3.02
EIT07K-243-1-10	13.6	66.0	27.0	6.8	82.7	5.8	2.92
EIT07K-243-1-5	14.9	60.6	26.9	8.8	81.9	5.8	2.97
EIT06-154-1	10.0	60.0	22.3	5.4	84.6	5.4	2.42
IT96-610	4.1	0.0	0.0	0.0	100.0	0.1	0.00
1T845-2246-4	25.2	79.6	27.8	13.9	75.1	10.2	3.05
IFE BROWN	35.4	79.2	28.0	31.5	62.4	10.9	6.72
MIT06K-128	13.5	74.2	23.9	10.5	79.3	6.0	2.43
MIT07K-188-49	7.8	51.4	24.9	3.2	88.2	5.1	1.87
MIT07K-835-45	18.3	76.5	27.1	7.8	82.3	6.1	2.56
MIT07K-318-33	21.0	71.3	26.8	12.7	77.2	8.5	3.01
MIT07K-309-44	15.9	81.7	25.8	10.8	79.0	6.1	2.79
MIT06K-281-1	4.3	0.0	0.0	0.0	100.0	0.1	0.00
MIT07K-187-24	4.0	0.0	0.0	0.0	100.0	0.1	0.00
MIT06K-124	19.7	66.1	25.1	9.9	80.3	6.1	2.86
MIT04K-219-2	26.0	73.2	26.8	19.4	72.8	10.1	4.67
MIT04K-321-2	22.1	68.0	27.3	11.6	78.5	6.6	3.08
MIT07K-304-9	4.0	0.0	0.0	0.0	100.0	0.1	0.00
MIT03K-337-6	33.7	77.1	28.0	30.3	63.4	10.4	5.57
MIT06K-121	6.3	0.0	0.0	0.0	100.0	0.2	0.00
MIT07K-194-3	27.0	63.1	25.9	13.5	75.6	9.4	3.32
MIT98K-503-1	19.7	55.9	26.9	8.3	82.1	6.2	2.95
EIT04K-221-1	17.7	50.8	26.9	7.8	82.3	6.1	2.74
EIT07K-291-69	4.1	0.0	0.0	0.0	100.0	0.1	0.00
EIT07K-299-4	13.1	53.3	27.8	7.8	82.3	5.7	2.84
EIT03K-369-3	20.7	62.9	28.0	10.3	79.9	6.1	3.12
EIT97K-499-35	26.9	66.8	27.7	14.8	74.2	10.0	3.43
SED	11.4	3.2	0.3	2.1	3.9	1.4	0.01
LSD (0.05)	21.6	7.5	NS	9.4	17.2	4.6	0.59

Note: Each value is a mean of three replicates

2.3 Relationship between index of susceptibility and anti-nutritional contents

There was a negative correlation between susceptibility index and anti-nutritional contents (Figures 2, 3 and 4). Cultivars of cowpea seeds with low phytate, oxalate and tannin contents tend to be more susceptible to infestation by *C. maculatus* than those with high phytate, oxalate and tannin contents (Table 2).

3 Discussion

Previous study by Ileke et al. (2013) has shown varietal resistance of thirty-one cowpea cultivars to cowpea bruchid, *Callosobruchus maculatus* (Fab.) [Coleoptera: Chrysomelidae] infestation. They reported that cultivars MIT04K-399-1, MIT07K-299-92, IT96-610, MIT06K-281-1, MIT07K-187-24, MIT06K-121, MIT07K-304-9 and EIT07K-291-69 were the most resistance cultivars tested, while cultivars IFE BROWN

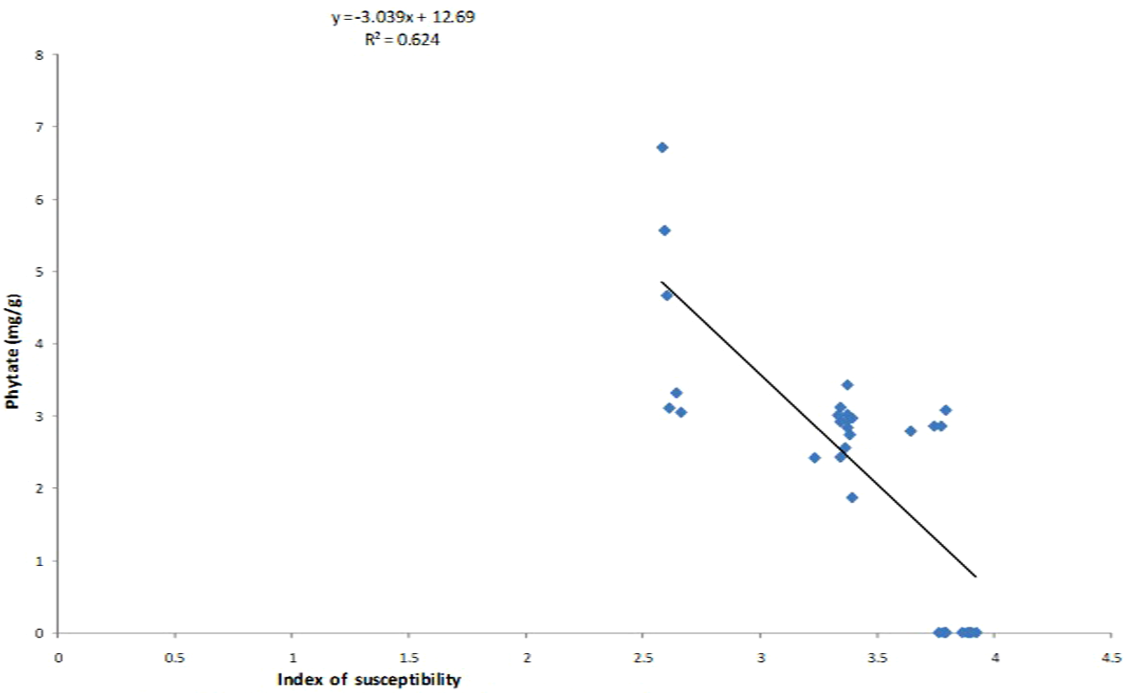

Figure 2 Relationship between index of susceptibility and phytate content of cowpea cultivars

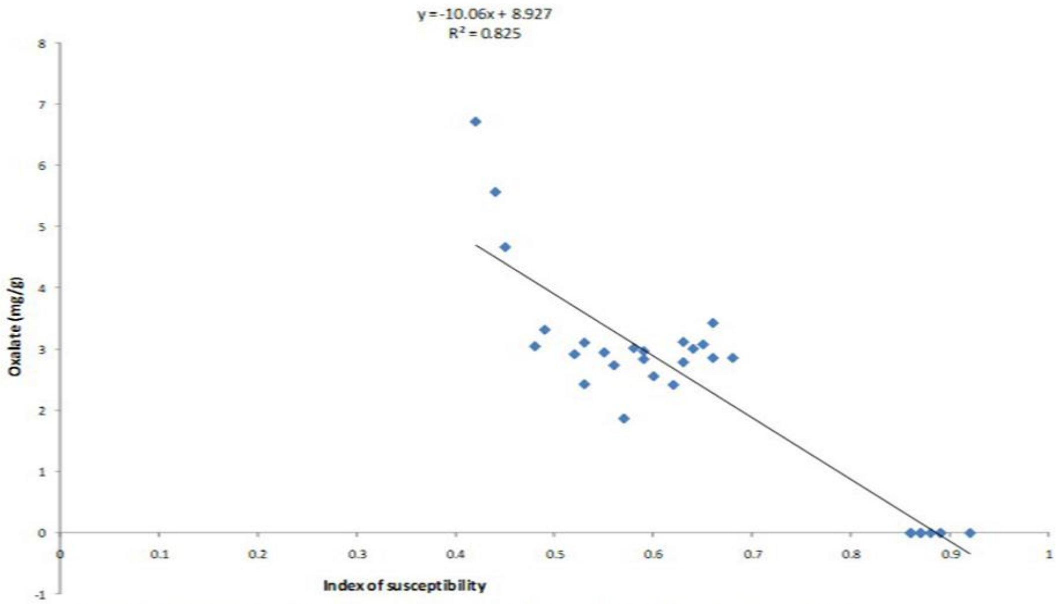

Figure 3 Relationship between index of susceptibility and oxalate content of cowpea cultivars

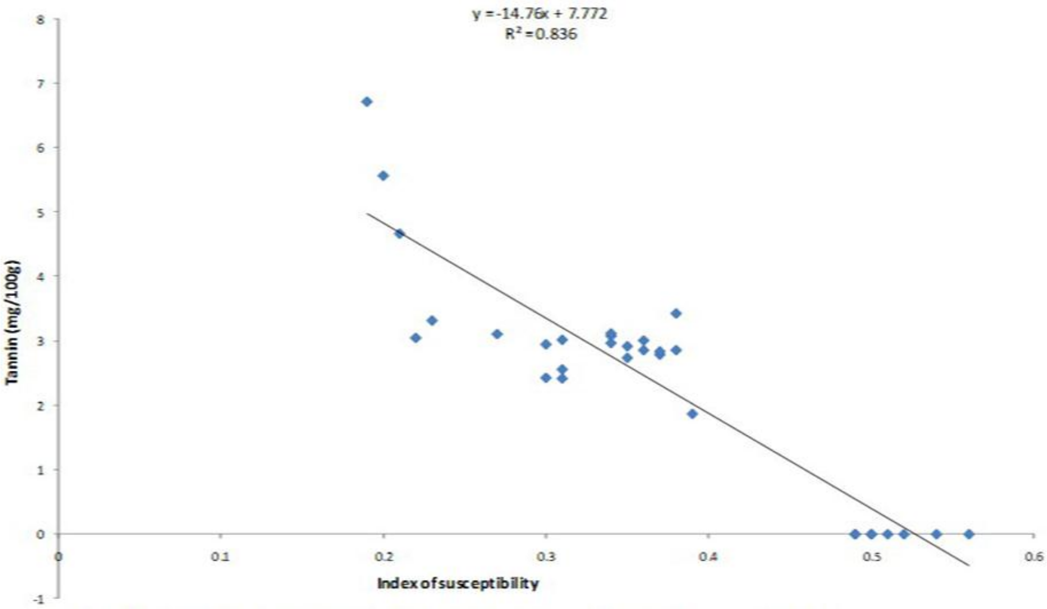

Figure 4 Relationship between index of susceptibility and tannin content of cowpea cultivars

Table 2 Anti-nutritional content in cowpea cultivars

Cowpea cultivars	Phytate (mg/g)	Oxalate (mg/g)	Tannin (mg/100g)
MIT04K-339-1	3.92±0.04a	0.89±0.04a	0.49±0.16a
MIT07K-299-92	3.89±0.06a	0.92±0.04a	0.54±0.11a
MIT07K-211-1-8	3.74±0.09a	0.68±0.17a	0.38±0.04a
MIT07K-292-10	2.61±0.17a	0.53±0.01a	0.27±0.02a
EIT07K-303-1	3.37±0.05a	0.58±0.0.1a	0.31±0.04a
EIT07K-243-1-10	3.34±0.06a	0.52±0.01a	0.35±0.03a
EIT07K-243-1-5	3.39±0.07a	0.59±0.04a	0.34±0.03a
EIT06-154-1	3.23±0.08a	0.62±0.17a	0.31±0.04a
IT96-610	3.78±0.09a	0.86±0.03a	0.56±0.11a
1T845-2246-4	2.66±0.17a	0.48±0.09a	0.22±0.03a
IFE BROWN	2.59±0.01a	0.44±0.09a	0.20±0.03a
MIT06K-128	3.34±0.05a	0.53±0.01a	0.30±0.04a
MIT07K-188-49	3.39±0.06a	0.57±0.01a	0.39±0.03a
MIT07K-835-45	3.36±0.07a	0.60±0.03a	0.31±0.03a
MIT07K-318-33	3.33±0.08a	0.64±0.09a	0.36±0.04a
MIT07K-309-44	3.64±0.09a	0.63±0.03a	0.37±0.04a
MIT06K-281-1	3.88±0.06a	0.89±0.04a	0.52±0.10a
MIT07K-187-24	3.90±0.04a	0.87±0.04a	0.50±0.08a
MIT06K-124	3.77±0.03a	0.66±0.04a	0.36±0.02a
MIT04K-219-2	2.60±0.11a	0.45±0.23a	0.21±0.08a
MIT04K-321-2	3.79±0.04a	0.65±0.07a	0.34±0.02a
MIT07K-304-9	3.86±0.08a	0.86±0.03a	0.49±0.16a
MIT03K-337-6	2.58±0.01a	0.42±0.15a	0.19±0.07a
MIT06K-121	3.79±0.09a	0.88±0.04a	0.51±0.09a
MIT07K-194-3	2.64±0.11a	0.49±0.08a	0.23±0.08a
MIT98K-503-1	3.37±0.08a	0.55±0.03a	0.30±0.04a
EIT04K-221-1	3.38±0.06a	0.56±0.01a	0.35±0.03a
EIT07K-291-69	3.76±0.09a	0.87±0.04a	0.50±0.08a
EIT07K-299-4	3.37±0.03a	0.59±0.02a	0.37±0.02a
EIT03K-369-3	3.34±0.09a	0.63±0.08a	0.34±0.02a
EIT97K-499-35	3.37±0.03a	0.66±0.03a	0.38±0.02a

Note: Each value is a mean ±standard error of three replicates was the most susceptible to *C. maculatus* infestation and this is followed by cultivar MIT03K-337-6 based on the following parameters assessed: oviposition, % adult emergence, weight loss and seed viability.

The results obtained on percentage seed damage and pest tolerance have further showed that cultivars MIT04K-399-1, MIT07K-299-92, IT96-610, MIT06K-281-1, MIT07K-187-24, MIT06K-121, MIT07K-304-9 and EIT07K-291-69 were resistant, while cultivars IFE BROWN was the most susceptible to *C. maculatus*. Damage inflicted by cowpea bruchid consists of the consumption of seeds, loss or conversion of nutrients, reduced germination of seeds and contamination with filthy materials composed of insect fragments, exuviae, excreta and moulds (Odeyemi, 2005).

Resistance could be anti-nutritional factors such as tannins, phytate and oxalate which had been reported by Singh and McCain (1963). Anti-nutritional factors are plant's secondary metabolites which act to reduce food nutrient utilization (Soetan, 2008). Anti-nutritional factors affect susceptibility of grains to insect attack. However, anti-nutrients is not an inherent feature of a compound but depends on the metabolic processes of the ingesting animal (Akande et al., 2010). The reason for anti-nutritional factors in plants seems to be as a way of storing nutrients or as a means of defence from destruction by insect pests and grazing animals (Harborne, 1989). It has been reported that tannins help in growth regulation and also protect the plants from predators like insects (Fasola and Egunyomi, 2005; Fasola et al., 2013). Phytate, oxalate and tannins contents were high in the resistance cowpea cultivars with more phytate than oxalate. The mean phytate obtained in this study was about three times higher than that of oxalate. This agrees with an earlier report by Afiukwa et al. (2011) who worked on variations in seed phytic and oxalic acid contents among Nigerian cowpea accessions and their relationship with grain yield.

The result revealed significant variations in the anti-nutrients among the cultivars. The amount of phytate, oxalate and tannins ranges from 2.58 mg/g to 3.92 mg/g, 0.42 mg/g to 0.92 mg/g and 0.21 mg/g to 0.56 mg/g respectively. These values compare well with 2.58 – 3.87 mg/g for phytate and for oxalate with mean values of 3.10 and 0.78 mg/g reported for ninety nine cowpea cultivars by Afiukwa et al. (2011). Cowpea seeds should always be adequately processed to avoid phytate-related health risks especially among individuals who depend largely on cowpea for protein. Thus, oxalate related problems are not likely to occur in healthy persons, except among individuals that consume large amounts on a long-term continuing basis and individuals with especial vulnerability to oxalates such as those with kidney disorders, gout and rheumatoid arthritis. Fortunately, most processing methods significantly reduce the anti-nutrients or totally eliminate some of them (Udensi et al., 2005 and 2007; Philips, 1993; Afiukwa et al., 2011). This study revealed variations in the anti-nutrients among the cowpea cultivars. These variations affect the susceptibility of *C. maculatus* infestation. With recent advances in biotechnology and plant breeding, it is possible to transfer desirable characters from resistant varieties in other to improve their resistance to cowpea bruchid. Alternatively, cultivars MIT04K-399-1, MIT07K-299-92, IT96-610, MIT06K-281-1, MIT07K-187-24, MIT03K-337-6, MIT07K-304-9 and EIT07K-291-69 with high degree of resistance to *C. maculatus* could be cultivated by farmers. This will go a long way in ensuring food security in Nigeria.

Acknowledgement

The author are grateful to Alh. Remi Adeleke, CGIAR, International Institute for Tropical Agriculture, Ibadan, Oyo State, Nigeria, for the supply of cowpea cultivars. The comments by three anonymous reviewers towards improving the quality of this research work are acknowledged.

References

Afiukwa C.A., Igwenyi I.O., Ogah O., Offor C. E., and Ugwu O.O., 2011, Variations in seed phytic and oxalic acid contents among Nigerian cowpea accessions and their relationship with grain yield. Continental Journal of Food Science and Technology, 5(2): 40-48

Akande K. E., Doma U. D., Agu H. O., and Adamu H. M., 2010, Major antinutrients found in plant protein sources: Their effect on nutrition. Pakistan Journal of Nutrition, 9 (8): 827-832 http://dx.doi.org/10.3923/pjn.2010.827.832

Allison M.J., Hammond A. C., and Jones R. J., 1990, Detection of ruminal bacteria that degrade toxic dihydroxy pyridine compounds produced from mimosine. Applied and Environmental Microbiology, 56: 590-594

Conn E. E., 1979, Cyanide and cyanogenic glycosides.In: Herbivores: Their interaction with secondary plant metabolites. Rosenthal, G.A. and Janzen, D.H. (eds.) A.P., New York pp. 387-412

Cordain L., 1999, Cereal grains humanity's double-edged sword. World Review on Nutritional Diet, 84: 19-73 http://dx.doi.org/10.1159/000059677

Day R. A., and Underwood A. L., 1986, Quantitative analysis. 5th ed. Prentice-Hall publication. :701

Das M. C., and Mahato S. B., 1983, Triterpenoids (Review). Phytochemistry, 22: 1071
http://dx.doi.org/10.1016/0031-9422(83)80198-8

Dobie P., 1974, The laboratory assessment of the inherent susceptibility of maize varieties to post harvest infestation by *Sitophilus zeamais* (Motsch.) (Coleoptera: Curculionidae) infesting field corn. Journal of Entomological Science, 21: 367-375

Ekop A. S., and Eddy, N. O., 2005, Comparative studies of the level of toxicants in the seed of Indian Almond (Terminaliacatappa) and African Walnut (Coula edulis). Chemistry Class Journal, 2: 74-76

Ekop A. S., Eddy N. O., and Udofia P. G., 2004, Effect of processing on the elemental composition of beans. Proceedings of 28th Annual Conference of Nigerian Institute of Food Science and Technology (NIFST) Ibadan, pp: 217-218

Fasola T. R., and Egunyomi A., 2005, Nigerian usage of bark in phytomedicine. Ethnobotany Research and Applications, 3: 73-77

Fasola T. R., Akinloye A. J., Tossou M. G., and Akoegninou A., 2013, The Phytochemical and Structural Make-Up of Regrown and Original Tree Barks Used in Ethnomedicine. World Journal of Agricultural Sciences, 9(1): 92-98

Gbaye O. A., and Holloway G. J., 2011, Varietal effects of cowpea, *Vigna unguiculata*, on tolerance to malathion in *Callosobruchus maculatus*. Journal of Stored Product Research, 47: 365-371
http://dx.doi.org/10.1016/j.jspr.2011.06.003

Guthre D. G., and Picciano M. F., 1995, Human nutrition.1stedn. WCB/ McGraw-Hill, New York. Pp 43-44

Harborne J. B., 1989, Biosynthesis and function of anti-nutritional factors in plants. Aspects of Applied Biology, 19: 21-28

Ileke K. D., Odeyemi, O. O., and Ashamo M. O., 2013, Varietal resistance of some cowpea cultivars to cowpea bruchid, *Callosobruchus maculatus* (Fab.) [Coleoptera: Chrysomelidae] infestation. FUTA Journal of Research in Sciences, 9(1):72-81

Jenab M., and Thompson L. U., 2000, Phytic acid in wheat bran affects colon morphology, cell differentiation and apoptosis. Carcinogenesis, 21 (8): 1547-52
http://dx.doi.org/10.1093/carcin/21.8.1547

Koratkar R., and Rao A.V., 1997, Effect of soyabean saponins on azoxymethane-inducedpreneoplastic lesions in the colon of mice. Nutrition and Cancer, 27: 206-209
http://dx.doi.org/10.1080/01635589709514526

Klopfenstein T. J., Angel R., Cromwell G., Erickson G. E., Fox D. G., Parsons C., Satter L. D., and Sutton A. L., 2002, Animal diet modification to decrease thepotential for nitrogen and phosphorus pollution. Council for Agricultural Science and Technology, 21:175-188

Lephale S., Addo-Bediako A., and Ayodele V., 2012, Susceptibility of seven cowpea cultivars (*Vigna unguiculatus*) to cowpea beetle (*Callosobruchus maculatus*). Agricultural Science Research Journal, 2(2): 65-69

Liener L. E., and Kakade M. L., 1980, Protease inhibitors.In: The lectins: Properties, functions, and applications in biology and medicine. Academic Press, New York. Pp 527-552

Mahe S., Gausseres N., and Tome D., 1994, Legume protein for human requirement. Journal of Agriculture, 7: 15-17

Odeyemi O. O., 2005, Insect in action on impact assessment on food security. Inaugural lecturer series 42 delivered at the Federal University of Technology Akure, Ondo State Nigeria. 42pp

Odeyemi O. O., and Daramola A. M., 2000, Storage practices in the tropics: Food storage and pest problems. First Edition, Dave Collins Publication, Nigeria Vol. 1. 235pp

Offor C. E., Nweke F. N., Okaka A. N. C., Igwenyi I. O., and Onwe V.N., 2011, Analysis of the antinutrients levels in staple food crops in three different Local Government Areas of Ebonyi State, Nigeria. Continental Journal of Food Science and Technology, 5 (1): 26-30

Ofuya T. I., 2001, Pest of stored cereals and pulses in Nigeria. In:. Ofuya T. I. and Lale, N. E. S. Biology, Ecology and control of insect pests of stored Food legumes. Dave Collins publications, Nigeria. Pp 25-58

Philips R. D., 1993, Starchy legumes in human nutrition, health and culture. Journal of Human Nutrition, 44: 195-196

Ramiel N., 2010, Living with phytic acid, preparing grains, nuts, seeds and beans for maximum nutrition. Wise Traditions, 11: 27-29

Reddy M. B., and Love M., 1999, The impacts of food processing on the nutritional quality of vitamins and minerals. Advanced Experimental Medical Bioscience, 459: 99-106
http://dx.doi.org/10.1007/978-1-4615-4853-9_7

Schopke T. H., and Hiller K., 1990, Triterpenoidsaponins, Part 6. Die Pharamazie, 45: 313-342

Shukkur M. F., Abdul S. E., Karthik H. S., Ramasamy S., Nachiappa G. R., and Palaninathan V., 2006, Oxalate mediated nephronal impairment and its inhibition by C-phycocyanin: A study on urolithic rats. Molecular and Cellular Biochemistry, 284: 95-101
http://dx.doi.org/10.1007/s11010-005-9019-0

Singh D. N., and McCain P. S., 1963, The relationship of some nutritional properties of corn kernel to rice weevil infestation. Crop Science, 3: 259-261
http://dx.doi.org/10.2135/cropsci1963.0011183X000300030027x

Soetan K. O., 2008, Pharmacological and other beneficial effects of anti-nutritional factors in plants -A review. African Journal of Biotechnology, 7 (25): 4713-4721

SPSS, 2007, Inc. Statistical Package for Social Sciences. SPSS, Inc., Chicago, IL

Trease G. E., and Evans W. C., 1985, Pharmacognosy, 14th edn. London: W.B. Sanders Company

Udensi E. A., Onwuka G. I., and Oyewer C. R., 2005, Effect of autoclaving and boiling on some antinutritional factors in MucunaSloanie. Nigerian Food Journal, 23: 53-58

Udensi E. A., Ekwu F. C., and Isinguzo J. N., 2007, Antinutrient factors of vegetable cowpea (Sesquipedalis) seeds during thermal processing. Pakistan Journal of Nutrition, 6 (2): 194-197
http://dx.doi.org/10.3923/pjn.2007.194.197

Vucenik I., and Shamsuddin A. M., 2003, Cancer inhibition by inositol hexaphosphate (IP6) and inositol: from laboratory to clinic. The Journal of Nutrition, 133 (11): 3778-3784

Wakabayashi C., Hasegawa H., Murata T., Ichiyama M., and Saiki I., 1997, Expression of in-vivo antimetastatic effect of ginseng protopanaxtriolsaponins is mediated by their intestinal bacterial metabolites after oral administration. Chemical Abstract, 129(7): 44-57

Welch R. M., and Graham R. D., 2004, Breeding for micronutrients in staple food crops from a human nutrient perspective. Journal of Experimental Botany 55: 353-357
http://dx.doi.org/10.1093/jxb/erh064