

Alterations Induced by Cowpea Weevil *Callosobruchus maculatus* F. (Coleoptera: Chrysomelidae) Infestation on Seed Germination Potential and Nutrient Quality of *Vigna aconitifolia* (Jacq.)

Abstract

Aims: To evaluate alterations induced by cowpea weevil *Callosobruchus maculatus* (*C. maculatus*) infestation on seed germination potential and nutrient quality of *Vigna aconitifolia* (*V. aconitifolia*).

Experimental design: The research was carried out in a complete randomized block design.

Place and duration of study: Department of Botany, University of Calabar, Calabar, Nigeria, between March and June, 2016.

Methodology: A mix of *V. aconitifolia* seeds was bought, infested seeds sorted from the non infested ones at different levels of infestation and kept for three months before planting to ascertain alteration induced by *C. maculatus* on seed germination potential. The other seeds were sundried, milled into powder and used for proximate, minerals and vitamins analysis.

Results: Significant ($P = .05$) alterations induced by *C. maculatus* on seeds germination ability and nutrient quality of *V. aconitifolia* were observed. These changes varied according to the severity of infestation. The germination potential of seeds was affected with significant reductions observed at all levels of infestation with respect to soil types compared to seeds before infestation (BI). Seed germination was the highest in sandy and loamy than in clay soil. Percentage germination observed on the eleventh day for seeds planted on clay, sandy and loamy soil were 61.7%, 87.1% and 66.7% (BI) compared to values of 4.2%, 6.1% and 5.3% respectively at severe infestation (SI). Results revealed that after infestation (AI) seeds of *V. aconitifolia* had significant decrease at SI level of 10.1% for moisture, 44.2% (ash), 25.5 % (fat), 18.0% (fibre) and 12.4% (carbohydrate) while protein had increase of 40.2%. P, Na, Cu and Ni showed decrease in content while K, Ca, Mg, Fe, Zn, Mn, Co depicted increase at all levels of infestation compared to seeds content BI. Lead was not detected. Significant decrease in vitamins A, B₁, B₂, B₃, B₅, B₆, B₉, C and biotin contents of the cowpea with decrease in severity of infestation was found. Vitamin E revealed increase with increase in severity of infestation.

Conclusion: *Callosobruchus maculatus* infestation damaged seeds resulting in a reduction in the germination ability and marketability of *V. aconitifolia* seeds with severe alteration in nutritional quality.

Keywords: Seed germination potential, *Vigna aconitifolia*, *Callosobruchus maculatus* infestation, Nutrient quality.

Introduction

The cowpea weevil, *C. maculatus* F. (Coleoptera: Chrysomelidae) is a devastating pest of *V. aconitifolia*.

The first instar larva burrows into the seed to start consuming it from the inside. All larval stages are spent inside the seed, where they remain concealed and protected as they feed. The last instar larva pupates inside the seed. After emergence from the pupa, the adult beetles chew its way out of the seed, leaving a characteristic round hole in the shell. Soon after adult (active, strong fliers) emergence from the pupa, they immediately begin their search for food. Insect pests need food, air and water to live. The best place for insect to live and grow is in stored grains because food, air and water are available in sufficient quantities. That is why some insect pests infest stored grains and pulses [1].

Callosobruchus maculatus is a cosmopolitan field-to-store pest ranked as the principal post-harvest pest of cowpea in the tropics and subtropics of the world [2], [3]. It posed a major threat to the production and storage of cowpea in developing countries [4]. Damage caused by cowpea weevils in seeds affected germination [5]. This pest causes substantial quantitative and qualitative losses evident by seed perforation or holes and reduction in weight, market value and germination ability of seeds [6]. *Callosobruchus maculatus* infestation significantly damaged seeds of *Vigna unguiculata* resulting in reduced seed germination and altered nutrient quality of stored beans [7]. Infestation of *Phaseolus lunatus* by the cowpea weevil, *Acanthoscelides obtectus* reduced seed germination with changes in the biochemical composition of seeds [8]. This damage caused by storage pests is threatening as global population growth is estimated to 9 billion persons inhabiting the Earth by 2050, a 60% increase in food production must be attained.

Vigna aconitifolia (Jacq) is commonly called mat bean, moth bean, matki, Turkish gram or dew bean. The bean is widely consumed in Nigeria where it serves as a valuable source of dietary proteins, vitamins and minerals [9]. It is an important economic species in the diets of many societies. Due to the drought resistant quality of *V. aconitifolia*, its ability to combat erosion, its high protein content, the bean has been identified as a more significant food source in the future [10]. *Vigna aconitifolia* food products like food products of other species of *Vigna* exhibit many excellent nutritional attributes and these products provide a needed complement in diets comprised mainly of roots, tubers or cereals [11]. Seeds occupy a central place in the life of humans. Seeds serve as food, while

other seeds are important raw materials for the manufacturing of industrial chemicals and other products. When seeds are infested by *C. maculatus* remarkable changes in germination, nutrient content, reduction in grain weight and marketability occur [12], [7] resulting in great economic losses. The severity of these losses depends on the level of storage pest infestation. This study is designed to investigate changes induced by cowpea weevil on the germination ability of seeds, to estimate the nutrient content of *V. aconitifolia* before and after infestation. To account for the qualitative loss in cowpea during storage with a view to highlighting the deleterious alterations induced on stored cowpea after infestation by *C. maculatus*.

Materials and Methods

2.1 Seed collection

In this study, the damaged seeds of *V. aconitifolia* were purchased from the Watt Market Calabar, Calabar Nigeria. On seeds purchase, infested ones were sorted and taken to the Department of Zoology and Environmental Biology, University of Calabar for pest identification by a Professor of Entomology, Prof. E. E. Oku.

2.2 Seed preparation

The seeds were then grouped into different categories based on the level of damage and studied for germination potential and nutrient quality. Seeds before infestation with no emergence hole, were assigned group 1; seeds after infestation were assigned group 2 for slight infestation (SLI) 1-3 holes, group 3 for moderate infestation (MI) 4-5 holes and group 4 for severe infestation (SI) 6 holes and above. Each of the group of infested seeds was placed in a transparent glass jar covered with a net mesh size of 1 cm by 1 cm to enhance infestation continuity at $25\pm 2^{\circ}\text{C}$ and $70\pm 5\%$ relative humidity. The non infested control group was tightly sealed with a metallic lid. These seeds were then kept for a period of three months before they were used for further processes.

2.3 Experimental design

Pots used for planting were filled with different types of soil; clay, sandy and loamy obtained from different locations within the University Campus. The pots were grouped into two; group one for the planting of non infested seeds designated before infestation (BI) of *C. maculatus* and group 2 for the planting infested seeds from the different levels of infestation 2, 3 and 4 designated after infestation (AI).

2.4 Seed planting and germination

Infested and non infested seeds of *V. aconitifolia* were planted in clay, sandy and loamy soil. Three months post-storage seeds of infested from the different levels of *C. maculatus* infestation and non infested groups were placed superficially on the surface of the different soil types. Each soil type had 27 pots (3 replicates for each soil type repeated trice). A total of 270 seeds were planted on each soil types before infestation and after infestation. The pots were watered daily. Seed emergence started on the third day in all soil types. Germinated seeds count BI and AI was carried out daily for a period of twelve days and expressed as a percentage of the seeds planted. Germination percentage is an estimate of seed viability population calculated by the formula: $GP = \frac{\text{Seeds germinated}}{\text{Total seeds}} \times 100$ [13].

2.5 Preparation of samples for analysis

At the end of three months storage period, seeds of *V. aconitifolia* BI and AI at different levels of infestation were removed from their storage containers, checked for eggs with a low power (magnification x 10) dissecting microscope VT-11. The seeds were dissected to remove the larvae, pupae and adults of *C. maculatus* with a pair of forceps. After which seeds BI and AI were sun-dried for five days, ground into powder and used for nutrient quality analysis. Proximate and some mineral nutrients were analyzed by the method of Association of Official Analytical Chemists [14], Na and K by flame photometry. Vitamins were analyzed by AOAC [15].

2.6 Statistical analysis

Results obtained were subjected to analysis of variance (ANOVA) according to [16] to determine the level of significance BI and AI at different levels of infestation.

Results

3.1 Mean Germination of *Vigna aconitifolia* Seeds Infested by *Callosobruhus maculatus* at Various Levels in Three Different Soil Types.

Of the two hundred and seventy seeds (270) of *Vigna aconitifolia* planted on clay, sandy and loamy soil, the number of seeds that germinated before infestations were 241, 269 and 268 respectively. Corresponding number of seeds that germinated after infestation at SLI, MI and SI on clay soil had number of germinated seeds of 123, 78 and 43, sandy soil had germinated seeds of 147, 98 and 60 while loamy soil had 131, 95 and 57 germinated seeds at each level of infestation. Before infestation seeds planted on the different soil types had higher germination ability when compared to seeds planted after infestation with highest seed germination potential observed with seeds planted on sandy soil followed by loamy soil while clay soil had the lowest seed germination potential

BI and AI. Significant ($P = .05$) alteration in mean germination was observed in seeds of *V. aconitifolia* after *C. maculatus* infestation (AI) when compared to seeds before infestation (BI). Reduction in seeds ability to germinate varied according to severity of infestation. Seeds with slight infestation (SLI) had the highest germination potential, followed by seed with moderate infestation (MI) while seeds with severe infestation (SI) had the lowest germination potential with respect to all soil types. Seeds of *V. aconitifolia* sown on sandy soil BI and AI showed highest mean germination potential, followed by seeds sown on loamy soil while seed planted on clay soil had the least germination potential. Seed germination began on the third day and ended on the eleventh day. Highest and lowest seed germination potential BI had values of 5.56 ± 0.12^d , 0.44 ± 0.11^a (clay soil), 6.67 ± 0.15^e , 6.67 ± 0.15^e (sandy soil) and 6.00 ± 0.03^e , 5.56 ± 0.10^d (loamy soil). Corresponding values AI at SLI, MI and SI were 1.89 ± 0.12^b , 2.56 ± 0.01^c (SLI), 2.11 ± 0.03^c , 0.22 ± 0.03^a (MI), 1.11 ± 0.33^b , 0.00 ± 0.00^a (SI) on clay soil, 2.89 ± 0.10^c , 0.33 ± 0.16^a (SLI), 2.22 ± 0.03^c , 0.22 ± 0.02^a (MI) and 1.44 ± 0.03^b , 0.11 ± 0.11^a (SI) on sandy soil while values for loamy soil were 2.22 ± 0.03^c , 0.33 ± 0.02^a (SLI), 1.78 ± 0.15^b , 0.22 ± 0.01^a (MI) and 0.67 ± 0.23^a , 0.11 ± 0.01^a (SI). From Table 1 percentage germination of seeds on different soil types was calculated. A trend of lowest percentage germination at initial period (day 3) and highest at later period (day 11) was observed in seeds BI and AI. A range in percentage germination values of seeds BI planted on clay, sandy and loamy soils were 0.44 to 60.7%, 1.9 to 87.1% and 1.9 to 66.7% respectively. A range in values of seeds AI at 2, 3 and 4 levels of infestation were 1.1 to 23%, 0.7-9.0% and 0.7 to 4.2% respectively for seeds planted on clay soil, 1.1 to 16.9%, 0.7 to 9.9% and 0.4 to 6.1% for seeds planted on sandy soil while loamy soil had a range in values of 1.1 to 15.4% at SLI, 0.7 to 9.8% at MI and 0.4 to 5.3% at SI.

Table 1: Mean Germination of *Vigna aconitifolia* Seeds Infested by *Callosobruchus maculatus* at Various Levels in Three Different Soil Types.

Mean germination on clay soil					Mean germination on sandy soil					Mean germination on loamy soil			
BI		AI			BI		AI			BI		AI	
GD	1	2	3	4	1	2	3	4		1	2	3	4
1	-	-	-	-	-	-	-	-		-	-	-	-
2	-	-	-	-	-	-	-	-		-	-	-	-
3	0.44 ± 0.11^a	0.33 ± 0.02^a	0.22 ± 0.03^a	0.00 ± 0.00^a	0.56 ± 0.11^a	0.33 ± 0.16^a	0.22 ± 0.02^a	0.11 ± 0.11^a		0.55 ± 0.01^a	0.33 ± 0.02^a	0.22 ± 0.01^a	0.11 ± 0.01^a
4	0.56 ± 0.12^a	0.44 ± 0.02^a	0.33 ± 0.03^a	0.22 ± 0.01^a	0.89 ± 0.10^a	0.56 ± 0.01^a	0.67 ± 0.02^a	0.44 ± 0.16^a		0.78 ± 0.03^a	0.67 ± 0.02^a	0.56 ± 0.02^a	0.33 ± 0.12^a
5	2.00 ± 0.11^b	1.11 ± 0.33^b	0.56 ± 0.02^a	0.33 ± 0.01^a	2.22 ± 0.12^b	1.33 ± 0.01^b	0.89 ± 0.01^a	0.56 ± 0.16^a		2.44 ± 0.01^b	1.11 ± 0.02^b	0.33 ± 0.13^a	0.56 ± 0.10^a
6	2.56 ± 0.10^b	1.56 ± 0.22^b	0.67 ± 0.01^a	0.33 ± 0.11^a	3.44 ± 0.10^c	1.78 ± 0.01^b	1.11 ± 0.03^b	0.67 ± 0.15^a		3.22 ± 0.06^c	1.67 ± 0.01^b	1.00 ± 0.01^b	0.67 ± 0.10^a
7	2.89 ± 0.12^b	1.67 ± 0.12^b	0.78 ± 0.33^a	0.44 ± 0.11^a	3.78 ± 0.10^c	2.00 ± 0.11^c	1.00 ± 0.12^b	0.67 ± 0.03^a		3.44 ± 0.06^c	1.89 ± 0.01^b	1.11 ± 0.01^b	0.67 ± 0.22^a

8	3.00 ± 0.01 ^c	1.89 ± 0.12 ^b	1.00 ± 0.33 ^b	0.67 ± 0.11 ^a	3.33 ± 0.03 ^c	2.11 ± 0.12 ^c	1.11 ± 0.13 ^b	0.67 ± 0.03 ^a	3.67 ± 0.01 ^c	2.00 ± 0.12 ^c	1.11 ± 0.21 ^b	0.67 ± 0.23 ^a
9	3.33 ± 0.22 ^c	2.00 ± 0.01 ^c	1.56 ± 0.03 ^b	0.89 ± 0.03 ^a	3.67 ± 0.03 ^c	2.22 ± 0.13 ^c	1.78 ± 0.11 ^b	1.00 ± 0.06 ^b	3.44 ± 0.01 ^c	2.11 ± 0.03 ^c	1.78 ± 0.33 ^b	1.00 ± 0.01 ^b
10	5.33 ± 0.22 ^d	2.11 ± 0.33 ^c	1.44 ± 0.02 ^b	0.78 ± 0.01 ^a	5.56 ± 0.15 ^d	2.56 ± 0.13 ^c	1.89 ± 0.03 ^b	1.11 ± 0.06 ^b	5.56 ± 0.10 ^d	2.22 ± 0.03 ^c	1.78 ± 0.15 ^b	1.00 ± 0.22 ^b
11	5.56 ± 0.12 ^d	2.56 ± 0.01 ^c	2.11 ± 0.03 ^c	1.11 ± 0.33 ^b	6.67 ± 0.15 ^e	2.89 ± 0.10 ^c	2.22 ± 0.03 ^c	1.44 ± 0.03 ^b	6.00 ± 0.03 ^e	2.78 ± 0.03 ^c	2.11 ± 0.15 ^c	1.33 ± 0.13 ^b

- GD = Germination Days, - = No Germination, BI = Before Infestation (Non Infested = 1)
- AI = After Infestation (2= Slight Infestation SLI, 3 = Moderate Infestation MI, 4 = Severe Infestation SI).
- Means followed by the same superscript letters in each column are not significantly different, while means followed by different superscript letters are significantly different at ($P = .05$) according to Duncan Multiple Range Test.

3.2 Alterations in Proximate Composition of *Vigna aconitifolia* seeds Before and After *Callosobruchus maculatus* Infestation

Results of proximate nutrients of *Vigna aconitifolia* seeds before infestation revealed significant ($P = .05$) increase in moisture, ash, fat, fibre and carbohydrate. After infestation, these nutrients were found to decrease significantly. Percentage decrease observed at different levels of infestation (2, 3 and 4) were 1.8%, 6.8% and 10.1% for moisture, 22.6%, 29.0% and 44.2% for ash, 5.6%, 14.8% and 25.5% for fat, 2.0%, 8.6% and 18.0% for fibre and 3.5%, 6.5% and 12.4% carbohydrate. Protein increased significantly ($P = .05$) after infestation with percentage increase of 15.1%, 19.6% and 40.2% for infestation level 2, 3 and 4 respectively (Table 2). Decrease or increase in proximate nutrients varied according to levels of infestation with minimum (decrease or increase) observed at slight infestation (2) and maximum (decrease or increase) observed at severe (4) infestation level.

Table 2: Alterations in Proximate Composition of *Vigna aconitifolia* seeds Before and After *Callosobruchus maculatus* Infestation

g/100 g							
Proximate nutrients	Before infestation		After infestation				
	1	2	%	3	%	4	%

Moisture	65.60 ± 0.1 ^c	64.40 ± 0.1 ^c	1.8	61.12 ± 0.01 ^c	6.8	58.99 ± 0.1 ^c	10.1
Ash	3.10 ± 0.1 ^a	2.40 ± 0.1 ^a	22.6	2.20 ± 0.2 ^a	29.0	1.73 ± 0.01 ^a	44.2
Protein	23.61 ± 0.01 ^b	27.18 ± 0.02 ^b	15.1	28.23 ± 0.1 ^b	19.6	33.10 ± 0.02 ^b	40.2
Fat	14.10 ± 0.1 ^b	13.30 ± 0.1 ^b	5.6	12.01 ± 0.02 ^b	14.8	10.50 ± 0.01 ^a	25.5
Fibre	3.50 ± 0.1 ^a	3.34 ± 0.02 ^a	2.0	3.20 ± 0.01 ^a	8.6	2.87 ± 0.01 ^a	18.0
Carbohydrate	55.36 ± 0.02 ^c	53.44 ± 0.01 ^c	3.5	51.77 ± 0.1 ^c	6.5	48.51 ± 0.2 ^c	12.4

139 • 1 = Non infested, 2 = Slight infestation (SLI), 3 = Moderate infestation (MI), 4 = Severe infestation (SI)
140 • Means followed by the same superscript letters in each column are not significantly different, while means followed by
141 different superscript letters are significantly different at ($P=0.05$) according to Duncan Multiple Range Test.

142 3.3 Alterations in Mineral Content of *Vigna aconitifolia* Before and After *Callosobruchus maculatus* Infestation

143 *Callosobruchus maculatus* infestation of *Vigna aconitifolia* led to significant ($P = 0.05$) increases and decreases in mineral nutrients.
144 Before infestation *V. aconitifolia* had higher amount of P, Na, Cu and Ni but after infestation these mineral nutrients were decreased
145 significantly. Mean value for Na before infestation was 17.42 ± 0.02^d g/100 g and values after infestation of 15.80 ± 0.1^b for slight
146 infestation, 14.02 ± 0.01^b for moderate infestation and 12.91 ± 0.01^b for severe infestation. A similar trend was observed with P, Cu
147 and Ni. Results revealed that K, Ca, Mg, Fe, Zn, Mn and Co were higher in amount after infestation from one level of infestation to
148 another. Percentage increases in infested seeds for Zn, K and Mn at different levels (SLI, MI and SI) of infestation depicted values of
149 34.8%, 65.7% and 78.7% (Zn), 6.5%, 11.3% and 13.5% (K) and 7.7%, 15.4% and 30.8% (Mn) (Table 3). Lead was not detected in
150 seeds of *V. aconitifolia*.

151 Table 3: Alterations in Mineral Content of *Vigna aconitifolia* Before and After *Callosobruchus maculatus* Infestation

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Mineral nutrients	g/100 g						
	Before infestation			After infestation			
	1	2	%	3	%	4	%
Potassium (K)	15.10 ± 0.1 ^b	16.08 ± 0.03 ^b	6.5	16.92 ± 0.1 ^b	11.3	17.14 ± 0.2 ^b	13.5
Phosphorus (P)	296.01 ± 0.1 ^d	257.89 ± 0.1 ^e	12.9	231.53 ± 0.02 ^e	21.8	211.86 ± 0.1 ^d	28.4
Sodium (Na)	17.42 ± 0.02 ^b	15.80 ± 0.1 ^b	9.3	14.02 ± 0.01 ^b	19.5	12.91 ± 0.01 ^b	25.9

Calcium (Ca)	87.70 ± 0.02 ^c	92.40 ± 0.02 ^d	5.4	93.01 ± 0.02 ^d	6.1	95.12 ± 0.02 ^d	8.5
Magnesium (Mg)	83.42 ± 0.02 ^c	86.42 ± 0.02 ^c	3.6	87.00 ± 0.1 ^c	4.3	88.10 ± 0.1 ^c	5.6
Iron (Fe)	60.80 ± 0.01 ^c	72.30 ± 0.1 ^c	18.9	73.03 ± 0.02 ^c	20.1	74.76 ± 0.1 ^c	23.0
Zinc (Zn)	2.30 ± 0.1 ^a	3.10 ± 0.1 ^a	34.8	3.81 ± 0.1 ^a	65.7	4.11 ± 0.1 ^a	78.7
Copper (Cu)	10.80 ± 0.1 ^b	9.80 ± 0.1 ^a	9.3	8.44 ± 0.2 ^a	21.9	7.39 ± 0.1 ^a	31.6
Manganese (Mn)	0.13 ± 0.01 ^a	0.14 ± 0.01 ^a	7.7	0.15 ± 0.02 ^a	15.4	0.17 ± 0.1 ^a	30.8
Cobalt (Co)	0.12 ± 0.02 ^a	0.14 ± 0.01 ^a	16.7	0.15 ± 0.1 ^a	25.0	0.16 ± 0.1 ^a	33.3
Nickel (Ni)	1.20 ± 0.02 ^a	1.12 ± 0.03 ^a	6.7	1.04 ± 0.3 ^a	13.3	0.97 ± 0.01 ^a	19.2
Lead (Pb)	ND	ND	ND	ND	ND	ND	ND

- 1 = Non infested, 2 = Slight infestation (SLI), 3 = Moderate infestation (MI), 4 = Severe infestation (SI)
- Means followed by the same superscript letters in each column are not significantly different, while means followed by different superscript letters are significantly different at ($P = .05$) according to Duncan Multiple Range Test.

3.4 Alterations in Vitamins Content of *Vigna aconitifolia* Before and After *Callosobruchus maculatus* Infestation

All the vitamins investigated BI and AI were significantly ($P = .05$) depleted by *C. maculatus* infestation with the exception of vitamin E which was higher in seeds AI. Vitamins content of *V. aconitifolia* seeds were significantly ($P = .05$) altered by *C. maculatus* infestation compared to vitamin content of seeds BI. This alteration resulted in a decrease in vitamins content of *V. aconitifolia*. The highest percentage reduction was found in severely infested seeds and the lowest in slightly infested seeds. Vitamins C, A, B₁, B₂, B₃, B₅, B₆, B₉ and biotin were observed to decrease with severity of infestation. Reductions observed in some vitamins after infestation at SLI (2), MI (3) and SI (4) were 24.5%, 27.7% and 31.3% for vitamin A, 29.4%, 43.6% and 58.7% for vitamin C and 15.0%, 30.0% and 75.0% for vitamin B₂. A similar pattern of decrease with severity of infestation was also found in vitamins, B₁, B₃, B₅, B₆, B₉ and biotin. Vitamin E content of seeds revealed increase with severity of infestation when compared to that of seeds before infestation. Increase observed AI were 3.7% for slightly infested seeds, 4.9% for moderately infested seeds and 22.0% for severely infested seed (Table 4).

Table 4: Alterations in Vitamins Content of *Vigna aconitifolia* Before and After *Callosobruchus*

maculatus Infestation

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Vitamins	Before infestation (BI)			After infestation (AI)			
	1	2	%	3	%	4	%
Ascorbic acid Vit. C (mg/100 g)	9.84± 0.01 ^c	6.95 ± 0.01 ^c	29.4	5.55± 0.2 ^b	43.6	4.06 ± 0.2 ^c	58.7
Vitamin E (mg/100 g)	1.64 ± 0.01 ^b	1.70 ± 0.1 ^b	3.7	1.72± 0.1 ^b	4.9	2.00± 0.1 ^b	22.0
Biotin (mg/100 g)	1.79 ± 0.02 ^b	1.52 ± 0.02 ^b	15.1	1.28 ± 0.01 ^b	28.5	1.01 ±0.01 ^b	43.6
Vitamin A (µg/dl)	102.75± 0.01 ^d	77.54± 0.01 ^d	24.5	74.33± 0.1 ^c	27.7	70.61± 0.1 ^d	31.3
Thiamin (Vit.B ₁) (µg/dl)	154.29± 0.01 ^d	116.44 ±0.01 ^e	24.5	100.08±0.1 ^d	35.1	93.01± 0.1 ^e	39.5
Riboflavin (Vit. B ₂) (µg/dl)	0.2 ± 0.1 ^a	0.17 ± 0.01 ^a	15.0	0.14 ± 0.01 ^a	30.0	0.05 ± 0.02 ^a	75.0
Naicin (Vit. B ₃) (µg/dl)	3.0 ± 0.1 ^b	2.8 ± 0.2 ^b	12.5	2.22 ± 0.02 ^b	30.6	2.00 ± 0.03 ^b	37.5
Patothenic acid (B ₅) (µg/dl)	0.6 ± 0.02 ^a	0.3 ± 0.3 ^a	16.0	0.45± 0.01 ^a	25.0	0.3 ± 0.03 ^a	50.0
Vitamin B ₆ (µg/dl)	0.5 ± 0.02 ^a	0.39 ± 0.3 ^a	22.0	0.30 ± 0.01 ^a	40.0	0.24 ± 0.01 ^a	52.0
Folate (B ₉) (µg/dl)	6451 ± 0.01 ^f	6112 ± 0.1 ^f	7.0	5734 ± 0.02 ^e	11.1	5401 ± 0.2 ^f	19.3

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176 1 = Non infested, 2 = Slight infestation (SLI), 3 = Moderate infestation (MI), 4 = Severe infestation (SI)

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Discussion

179 The effect of pest on storage products (maize, wheat, cowpea etc) has been widely studied. This research investigated the effect of
180 *C. maculatus* infestation on the seed germination potential and nutrient quality of *V. aconitifolia* under different types of soil. Results
181 of this research revealed that before infestation seeds had high mean and percentage germination than after infestation. The
182 damage caused by *C. maculatus* feeding affected germination of *V. aconitifolia* seeds because before infestation seeds showed
183 higher mean and percentage germination. This result is similar to report that control seeds of all legume hosts showed a high
184 frequency of germination while infestation by a single *C. maculatus* larva significantly reduced the frequency of germination of each
185 host [16]. In reference to soil type; seeds germination count was highest in sandy soil followed by loamy and lowest in clay soil. Dry
186 sandy soil is most suitable for the production of moth bean, and the bean can tolerate a variety of soil types [18]. Sandy and loamy
187 soils are more effective for the germination of *A. obtectus* infested and non infested seeds of Lima bean (*P. lunatus*) than clay soil [8].
188 The lowest mean and percentage germination of infested and non infested seeds of *V. aconitifolia* (moth bean) on clay soil may be
189 attributed to poor aeration since clay soils are composed of lot of tiny mineral particles which reduces the air spaces in the soil.

Germinating seeds require enough air to enhance the process. Alteration resulting in seed reduction germination potential caused by *C. maculatus* infestation may be attributed to partial embryo damage induced by the pest. Germination of Infested seeds did not fail because seed endosperm and embryo were not completely damaged, though the germinated seedlings of infested seeds were not studied and comparison made with the non infested ones.

The germination percentage is an indicator of the ability of the seed to emerge from the soil to produce a plant in the field under normal conditions. The loss of a seed's ability to germinate is an indication of loss of viability which involves decrease in seed vigour and other physiological changes. Seeds can only fulfill its biological role if viable. It is important to note that physically uniform seed of an adapted variety will be useless if it is low in germination and vigour or if it fails to germinate when planted. Seed are physiologically built to germinate so germination is a must for seeds. This is why germination, particularly high percentage of it is an essential technical specification for seeds [19]. The destruction of *V. aconitifolia* seeds in storage by *C. maculatus* reduces seeds germination potential causing undue stress of repeated planting, time wasting and money spent in the process by farmers. Reduction in germination of moth bean seeds affected yield as only little quantities of seeds germinated. It takes a germinated seed to grow, mature and reproduce food.

Seeds are the primary basis for human sustenance. Plants require seeds for reproduction so that they do not die off and become extinct. Plants are living things that operate in the natural cycle of seed, plant and fruit. Life on earth depends on plants. Without seeds there would be no vegetation. Man requires seeds for food production. The quality of seed is important in agriculture. Agricultural production depends on a lot of factors. The seed is the basic catalyst of efficiency of all other factors. Modern crop production and agricultural science also confirms that without seed quality we won't have a successful agricultural production. Seed quality is that one which has a genetic purity, physical purity, is healthy and has good physiological condition in accordance with standards prescribed for seed certification [20]. Food security therefore is dependent on seed security of farming communities or growers.

Results revealed a decrease in moisture, ash, fat, fibre and carbohydrate content with severity of infestation. The moisture content of the seed is an important factor in storage. The lower the temperature and relative humidity, the longer the seeds can be safely stored. The decrease in these proximate nutrients may be attributed to metabolic activities of the pest as it utilizes these nutrients for growth and other activities. The feeding damage of *C. maculatus* affected these proximate nutrients because before infestation seeds had higher quantity of these nutrients. These findings agree with previous reports of decrease in moisture, and carbohydrate contents of *V. unguiculata* infestation of *C. maculatus* [7]. Similar results of decrease in moisture, ash, fibre, fat and carbohydrate with an increase in protein content of *Phaseolus lunatus* seeds infested by *Acanthoscelides obtectus* [8]. Protein content of seeds increased with severity of infestation. This increase may be due to the eggs, egg cases, excretory products left after the removal of larval, pupal and adult stages of *C. maculatus* before analysis.

Alterations induced by *C. maculatus* on mineral nutrients were significantly different in seeds of *V. aconitifolia* after infestation compared to those of seeds before infestation. Infestation led to significant increase in K, Ca, Mg, Fe, Zn, Co and Mn and decrease in P, Na, Cu and Ni contents of *V. aconitifolia*. These alterations were observed to vary according to severity of infestation. A decrease and increase in some of these nutrients has been reported in *V. unguiculata* infestation by *C. maculatus* [7], in Lima bean (*Phaseolus lunatus*) infested by *Acanthoscelide obtectus* Say [8].

Nutrients are components in food that an organism uses for survival and growth. Some nutrients can be stored in the body like fat-soluble, while others are required more or less continuously. Poor health can be caused either by a lack of required nutrients or, in extreme cases, too much of a required nutrient. This has been observed with water and salt (both absolutely required) but will cause illness or even death in excessive amounts [21]. Changes induced by *C. maculatus* infestation resulted in decrease in content of P, Na, Cu and Ni essential nutrients required for proper functioning of the body. Phosphorus is required component of bones; essential for energy processing [22], Na is a common electrolyte in food not found in dietary supplements, despite being needed in large amounts, Cu is a required component of many redox enzymes including cytochrome c oxidase, Ni is present in urease. The reduction in these nutrients present insufficient amount for adequate body functions. Increase observed in K, Ca, Mg, Fe, Zn, Co and Mn after *C. maculatus* infestation of *V. aconitifolia* should be attended to as excessive consumption of these nutrients is also problematic to the body. Some of these nutrients are structural elements but many play a role as electrolytes [23] while others play a catalytic role in enzymes. Potassium is a common electrolyte required for heart and nerve health, Ca is another common electrolyte, but also needed structurally for muscle and digestive health, bone strengthening, some forms acidity, may help clear toxins, provides signaling ions for nerve and membrane functions, Mg is needed for processing ATP and related reactions (bone building, causes strong peristalsis, increases flexibility and alkalinity), Zn is needed for carboxypeptidase, liver alcohol dehydrogenase, and carbonic anhydrase enzymes, Co is required for vitamin B₁₂ coenzymes biosynthesis and Mn is required for the processing of oxygen. The number of nutrients known to be required by man according to the words of Marion Nestle, is almost incomplete [24].

Vitamins were also severely altered by *C. maculatus* infestation of *V. aconitifolia*. Vitamins depletion varied with severity of infestation. Results revealed reductions in all the vitamins investigated with the exception of vitamin E that showed increase after *C. maculatus* infestation. Deficient amount emanating from reductions and excesses from increase present serious health consequences. Deficiencies in vitamins result in disease conditions such as goiter, scurvy, osteoporosis, impaired immune system, cell metabolism disorders, certain forms of cancer, premature aging symptoms and poor psychological health among many others [25].

Apart from serving as a food crop moth bean also provide some form of environmental protection; its widespread tap roots and low-lying leaf-covering on the soil surface helps the soil to retain moisture. The strong roots enable the soil to stay and thus, help in

preventing soil erosion. Not only this, but the dense cover also help in weed control. This will not be possible if the seeds failed to germinate and grow. It takes a growing plant to protect the environment. Plants are environmental purifiers.

Conclusion

Alterations induced by *C. maculatus* affected seed germination with a resultant decrease in food production. Plants account for over 80% of the human diet and nutrition. In view of the critical role played by these nutrients; increases in these nutrients (protein, vitamin E, K, Ca, Mg, Fe, Zn, Co and Mn) induced by *C. maculatus* infestation of *V. aconitifolia* should be checked in order not to exceed acceptable levels, while attention should also be paid on decrease (moisture, ash, fat, fibre, carbohydrate fat, P, Na, Cu, Ni, vitamin A, C, B₁, B₂, B₃, B₅, B₆ and B₉) to avoid the consumption of low amount so as to prevent nutritional diseases resulting from deficiencies of these nutrient elements. Man obtained these elements by feeding on plant-based food since the body cannot synthesize all the nutrients needed for normal body function. There is therefore the need for routine check of *V. aconitifolia* and improved hygienic conditions in storage houses.

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