Research Article

Influence of intrinsic factors of cowpea seed varieties on the cowpea weevil, Callosobruchus maculatus (Coleoptera: Chrysomelidae)

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Abstract: In this study, the roles of proximate composition, mineral profile and heavy metal contamination were investigated on selected biological activities of Callosobruchus maculatus (F.) that were maintained on six cowpea cultivars. The levels of heavy metals; cadmium, lead and arsenic in six cowpea cultivars; Ife Brown, Ife BPC, Oloyin, Drum, Sokoto Local, and Niger White were determined by atomic absorption spectrophotometry while those of nutrient elements; zinc, sodium, calcium and potassium were determined using flame photometry. The proximate analyses and physico-chemical characterization of the cowpea cultivars were also performed. Callosobruchus maculatus was maintained on the cowpea cultivars and biological activities such as oviposition, adult emergence, percentage seed damage, and weight loss were monitored. The levels of heavy metals, mineral elements and proximate composition differed significantly among the cultivars investigated, and the values were comparable to those reported for cowpea cultivars in other studies. Of the cowpea cultivars, the Ife Brown was the most susceptible to the bruchid C. maculatus infestation while the Drum cultivar was the least susceptible. In conclusion, the results from this study show that the cowpea cultivars differed in the levels of toxic and mineral elements. While there were significant relationships between physical parameters (e. g. seed hardness), proximate composition (moisture contents and crude fibre) and adult emergence and oviposition, there was no strong evidence that the levels of heavy metals and mineral elements play significant role on the biology of C. maculatus.

Keywords: Cowpea cultivars, mineral elements, proximate composition, toxic elements, weevil susceptibility

Introduction

Cowpea Vigna unguiculata seeds provide cheap alternative source of dietary protein for humans and animals in many parts of the world especially in West and Central Africa, where they complement the mainly cereal and starchy tuber diets (Adedire et al., 2011a; Ofuya and Lale, 2001; Philips et al., 2003). The quest to improve the yield of cowpeas has led to development of various varieties of cowpeas, examples of available local varieties in Nigeria include IT80D-699, IT82 (e-18), Ife Brown, Ife BPC, IT870-9411, TVX3236, IT90K-277-2, IT870-9411, IT828-146, Oloyin, Drum, Sokoto local and Niger white cowpea, Olo-1, Olo-2, Banjara, Karadua, Manyan Fari, Kananado Fari, Kananado Yar and Akidi. These varieties differ in various physical and
Biological activities of C. maculatus on cowpea cultivars  

functional properties such as seed colour, texture, size, hardness and biochemistry, and these properties have been linked to play important role in resistance against pests and diseases (Agbogidi and Egho, 2012; Olapade et al., 2002).

The infestation of cowpea seeds by Callosobruchus maculatus (F.) accounted for the major loss of cowpea both in store and at fields (Adedire, 2001; Jackai and Daoust, 1986; Udo, 2005). The females of the bruchid lay their eggs on the seed surface which hatch into larvae within 5 days. The larvae bore through the seed coat into the underlying cotyledons where development continues until adult emergence; the adults are already sexually mature on emergence (Credland and Wright, 1989; Ofuya and Credland, 1995).

Technological advancement and increasing anthropogenic activities have resulted in the increased levels of heavy metals such as lead (Pb), cadmium (Cd), mercury (Hg) and chromium (Cr) and arsenic (As) in the environments (Li et al., 2014). When heavy metals are disposed into water, soil and air, they could be bioaccumulated by the crops (Fu et al., 2008) and could be harmful to humans when such crops are consumed (Balkhair and Ashraf, 2016; Satpathy et al., 2014). Studies on assessment of toxic elements in foods are important from public health point of view owing to various toxic effects that have been documented for these elements in humans (Adeyemi et al., 2016; Jarup, 2003). The objectives of the present study are i) to determine the proximate compositions, toxic elements (Cd, Pb and As) and mineral profiles (Ca, Na, Zn and K) of six cowpea cultivars that are common in major Nigerian markets, ii) to determine the correlation between toxic elements (Cd, Pb and As) and essential elements (Ca, Na, Zn and K) in different cowpea cultivars and biological activities (such as oviposition, hatchability and growth) of C. maculatus, and iii) to investigate the effect of cowpea cultivars on storage loss as well as oviposition and hatchability of C. maculatus.

Materials and Methods

Collections of sample and preparation

Six cowpea cultivars were used in this study; Oloyin, Drum, Sokoto local and Niger white cowpea obtained from Oba market in Akure metropolis (7.2571° N, 5.2058° E), Ife Brown and Ife BPC obtained from the Institute of Agricultural Research and Training (I. A. R. & T) Ibadan (7.3775° N, 3.9470° E). The samples were placed in polyethylene bags, and appropriately labeled for proper identification, and transported to the laboratory. In the laboratory, the samples were sorted to eliminate stones, chaff and other unwanted material. The samples were pulverized with mortar and pestle after which the pulverized samples were sieved using a sieve of 0.5mm mesh size to obtain very fine particles.

Proximate and metal analyses of the samples

The proximate compositions of the pulverized samples were determined using the AOAC methods of 2003. The parameters determined included ash content, moisture content, carbohydrates, protein, crude fat and crude fiber. The determination of levels of metals in the samples followed the standard procedures (AOAC, 2003). Approximately 5 g of the pulverized samples were carefully weighed out, and digested in acid mixture (perchloric acid and nitric acid), and placed on a hot plate set at 120 °C (increased gradually) until the samples were digested. The digested samples were diluted with distilled water appropriately in the range of the standards for each metal. The determination of levels of Cd, Pb, As and Zn in digested samples were performed using atomic absorption spectrophotometry (model 210 VGP) while the levels of Na, K and Ca were determined using flame photometry.

Laboratory culturing of C. maculatus

The parent stock of cowpea seed beetle, C. maculatus was obtained from established laboratory culture in the Biology Laboratory of the Federal University of Technology, Akure,
Briefly, cowpea seeds were disinfected by keeping in deep freezer at -20 °C for 3 days. The disinfested seeds were then air dried in the laboratory to prevent mouldiness before introduction of insects. They were then placed in Kilner jars and covered with muslin cloth. The jars were placed in insect rearing cages at ambient temperature of 30 ± 3 °C and 70 ± 5% relative humidity.

Physical and morphological characterization of cowpea cultivars

The physical characteristics that were determined in the cultivars included colour, length, breadth, width and seed hardness. The colours were observed visually while the length and width of the seeds were measured using the Vernier caliper. The weight per seed was measured using a Metler weighing balance machine by finding the average of 20 seeds. The hardness of the seeds was determined using a compression machine (Model: 200063 Milano, Italy).

Effect of cowpea cultivars on infestation and damage by *C. maculatus*

Approximately twenty gram (20g) of each cowpea cultivar was weighed into 250ml plastic containers and ten (10) copulating pairs of adult *C. maculatus* (2 to 3 days old) were introduced into each container. The containers were covered with tight lid that had been cut at the centre and sealed with muslin cloth for aeration. This was replicated three times. The infested cowpea seeds were left for 7 days in an insect cage in the laboratory during which the insects fed and laid eggs. On the day 7 of infestation, beetles were removed and discarded. The total number of eggs laid were counted and recorded. Twenty five days after infestation with beetles, the containers were checked daily for emerged adults. The emerged F1 adults were counted and recorded. Each trial replicate was terminated when no adult emergence was recorded for five consecutive days. The final weight of each cowpea varieties at the end of the experiment was determined, and the percentage weight loss was determined using the formula:

\[
\text{Weight loss (\%) } = \frac{\text{Difference in weight}}{\text{Initial weight}} \times 100
\]

The numbers of damaged cowpea seeds were evaluated by counting wholesome, bored or seed with bruchid emergent holes. Percentage seed damaged (bored seed and seeds with bruchid emergent holes) was calculated according to the method described by Odeyemi and Daramola (2000) as follows:

\[
\text{Seed damage (\%) } = \frac{\text{No of seeds damaged}}{\text{Total No of seeds}} \times 100
\]

Statistical Analysis

Data were subjected to analysis of variance (ANOVA) using SPSS 21.0 software and treatment means were separated using Tukey’s test. Data obtained from susceptibility studies were correlated with the heavy metals, proximate composition and mineral profiles of the cowpeas to determine the cause-effect factors. Statistical significance was assumed at p < 0.05.

Results

Proximate compositions of the cowpea cultivars

The proximate compositions of the cowpea cultivars used in this study are shown in Table 1. There were significant differences (p < 0.05) in the mean values for all the proximate analyses parameters determined in this study. The ash content of the cowpea ranged from 3.77% in Ife BPC to 3.88% in Drum. The highest moisture content and lowest moisture content in the cowpea cultivars were observed in cultivars Ife BPC (9.85%) and Drum (9.54%) respectively. The protein content ranged from 22.88% to 24.69% in Drum and Sokoto Local respectively. Sokoto Local had the highest carbohydrates content (59%) while the lowest (57.40%) carbohydrates content was observed in the Drum cultivar. The crude fibers of the cowpea cultivars ranged from 1.83% to 1.97%
in Niger White and Drum cultivars respectively. The highest percentage of crude fat content was recorded in Sokoto Local (2.59%) while Oloyin had the least (2.43%).

Concentrations of toxic metals and mineral elements in cowpea cultivars

The concentrations of toxic metals (Cd, Pb and As) and mineral elements (Zn, Na, Ca, and K) in the cowpea cultivars are presented in Table 2. The levels of Cd, Pb and As differed significantly (p < 0.05) among the cowpea cultivars. The highest Cd concentration was detected in Ife Brown (0.009 ± 0.002µg/g) while the least concentration was detected in the Niger White cultivar (0.003 ± 0.002µg/g). The highest concentration of Pb was detected in Oloyin cultivar (0.033 ± 0.003µg/g) while the Drum cultivar had the lowest Pb concentration of 0.025 ± 0.003µg/g. The Niger White cultivar had the highest As concentration of 0.024 ± 0.003µg/g while the lowest As concentration (0.011 ± 0.002µg/g) was detected in Ife BPC. The mean levels of Zn, Na, Ca and K differed significantly in the cowpea cultivars (p < 0.05). The highest levels of Zn (21.67 ± 0.020µg/g), Na (57 ± 20µg/g), Ca (810 ± 20µg/g) and K (16508 ± 60µg/g) were detected in Niger White cultivar while the lowest values of Zn (16.33 ± 0.020µg/g), Na (35 ± 10µg/g), Ca (580 ± 20µg/g) and K (13748 ± 40µg/g) were detected in Sokoto Local cultivar.

Table 1 Toxic metals and mineral profile of the cowpea cultivars.

<table>
<thead>
<tr>
<th>Cultivars</th>
<th>Cd</th>
<th>Pb</th>
<th>Zn</th>
<th>As</th>
<th>Na</th>
<th>Ca</th>
<th>K</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ife BPC</td>
<td>0.007 ± 0.002&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.031 ± 0.003&lt;sup&gt;b&lt;/sup&gt;</td>
<td>17.67 ± 0.020&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.011 ± 0.002&lt;sup&gt;d&lt;/sup&gt;</td>
<td>43 ± 30&lt;sup&gt;e&lt;/sup&gt;</td>
<td>713 ± 30&lt;sup&gt;f&lt;/sup&gt;</td>
<td>13866 ± 60&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
<tr>
<td>Sokoto Local</td>
<td>0.004 ± 0.002&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.029 ± 0.002&lt;sup&gt;e&lt;/sup&gt;</td>
<td>16.33 ± 0.020&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.018 ± 0.002&lt;sup&gt;b&lt;/sup&gt;</td>
<td>35 ± 10&lt;sup&gt;e&lt;/sup&gt;</td>
<td>580 ± 20&lt;sup&gt;e&lt;/sup&gt;</td>
<td>13748 ± 40&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
<tr>
<td>Oloyin</td>
<td>0.005 ± 0.003&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.033 ± 0.003&lt;sup&gt;c&lt;/sup&gt;</td>
<td>19.03 ± 0.015&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.016 ± 0.002&lt;sup&gt;b&lt;/sup&gt;</td>
<td>52 ± 30&lt;sup&gt;d&lt;/sup&gt;</td>
<td>727 ± 30&lt;sup&gt;e&lt;/sup&gt;</td>
<td>14512 ± 40&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
<tr>
<td>Drum</td>
<td>0.007 ± 0.002&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.025 ± 0.003&lt;sup&gt;d&lt;/sup&gt;</td>
<td>20.67 ± 0.030&lt;sup&gt;f&lt;/sup&gt;</td>
<td>0.021 ± 0.003&lt;sup&gt;b&lt;/sup&gt;</td>
<td>55 ± 20&lt;sup&gt;d&lt;/sup&gt;</td>
<td>757 ± 30&lt;sup&gt;e&lt;/sup&gt;</td>
<td>14624 ± 40&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
<tr>
<td>Niger White</td>
<td>0.003 ± 0.002&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.028 ± 0.003&lt;sup&gt;b&lt;/sup&gt;</td>
<td>21.67 ± 0.020&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.024 ± 0.003&lt;sup&gt;c&lt;/sup&gt;</td>
<td>57 ± 20&lt;sup&gt;d&lt;/sup&gt;</td>
<td>810 ± 20&lt;sup&gt;c&lt;/sup&gt;</td>
<td>16508 ± 60&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Units are presented in µg/g.

Each value is a mean ± standard error of three replicates. Means within the same column followed by the same letter(s) are not significantly different at p < 0.05 using Tukey's test.

Table 2 Proximate composition of the cowpea cultivars.

<table>
<thead>
<tr>
<th>Cultivars</th>
<th>Ash</th>
<th>Moisture content</th>
<th>Protein</th>
<th>Carbohydrates</th>
<th>Crude fiber</th>
<th>Crude fat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ife Brown</td>
<td>3.85 ± 0.05&lt;sup&gt;e&lt;/sup&gt;</td>
<td>9.67 ± 0.02&lt;sup&gt;e&lt;/sup&gt;</td>
<td>24.64 ± 0.30&lt;sup&gt;e&lt;/sup&gt;</td>
<td>57.63 ± 0.11&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.92 ± 0.02&lt;sup&gt;e&lt;/sup&gt;</td>
<td>2.49 ± 0.03&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Ife BPC</td>
<td>3.77 ± 0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>9.85 ± 0.03&lt;sup&gt;d&lt;/sup&gt;</td>
<td>23.79 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>58.15 ± 0.12&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.88 ± 0.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.56 ± 0.02&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Sokoto Local</td>
<td>3.83 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.74 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>22.88 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>59.00 ± 0.13&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.95 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.59 ± 0.02&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Oloyin</td>
<td>3.79 ± 0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>9.65 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.57 ± 0.25&lt;sup&gt;c&lt;/sup&gt;</td>
<td>57.69 ± 0.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.86 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.43 ± 0.04&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Drum</td>
<td>3.88 ± 0.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.54 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>24.69 ± 0.02&lt;sup&gt;c&lt;/sup&gt;</td>
<td>57.40 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.97 ± 0.02&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.52 ± 0.02&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Niger White</td>
<td>3.86 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.77 ± 0.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>23.87 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>58.11 ± 0.05&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.83 ± 0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.54 ± 0.03&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Units are presented in percent.

Means within the same column followed by the same letter(s) are not significantly different at p < 0.05 using Tukey's test.

Physical and morphological characterization of cowpea cultivars

The physical and morphological characteristics of the cowpea cultivars are presented in Table 3. The length, width, weight and hardness of the cowpea cultivars differed significantly (p < 0.05). Two of the six cultivars (Sokoto Local and Niger White) were white in colour while the remaining (Ife Brown, Ife BPC, Oloyin and Drum) were brown. All the cowpea cultivars were opaque in appearance. The hardness of the cowpea cultivars ranged from 64.26 ± 3.64 N in Ife Brown to 164.22 ± 16.43 N in Drum cultivar. The Ife Brown cultivar had the highest...
mean values of length (14.22 ± 2.213 mm), width (9.18 ± 2.159 mm) and weight (0.41 ± 0.002 g) while the Sokoto Local cultivar had the lowest mean values of length (9.51 ± 2.561 mm), width (6.81 ± 1.204 mm) and weight (0.16 ± 0.001 g).

Effect of cowpea cultivars on oviposition and adult emergence and seed damage by *C. maculatus*

The data on oviposition and percentage adult emergence of *C. maculatus* cultured on the six cowpea cultivars are presented in Table 4. There were significant differences (p < 0.05) in the number of eggs laid by *C. maculatus* on the six cultivars studied. The trend in the number of eggs laid was Ife brown > Ife BPC > Niger White > Oloyin > Sokoto Local > Drum. The highest percentage of adult emergence was recorded in *C. maculatus* cultured on Ife Brown cultivar (77%) while the least was recorded in those cultured on Drum cultivar (43%). The extent of seed damage by *C. maculatus* quantified in terms of percentage weight loss and seed damage are also shown in Table 3. The percentages seed damage and weight loss were highest in Ife Brown cultivar (73% and 30% for seed damage and weight loss respectively) and least in Drum (18.7% and 2.5% for seed damage and weight loss respectively).

**Table 3** Physical and morphological characteristics of the cowpea cultivars.

<table>
<thead>
<tr>
<th>Cultivars</th>
<th>Colour</th>
<th>Appearance</th>
<th>Length (mm)</th>
<th>Width (mm)</th>
<th>Weight (g)</th>
<th>Hardness (N)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ife Brown</td>
<td>Brown</td>
<td>Opaque</td>
<td>14.2 ± 2.21a</td>
<td>9.18 ± 2.16b</td>
<td>0.41 ± 0.002c</td>
<td>64.26 ± 3.64e</td>
</tr>
<tr>
<td>Ife BPC</td>
<td>Brown</td>
<td>Opaque</td>
<td>13.6 ± 1.79a</td>
<td>8.65 ± 1.79b</td>
<td>0.26 ± 0.001c</td>
<td>92.74 ± 9.73b</td>
</tr>
<tr>
<td>Sokoto Local</td>
<td>White</td>
<td>Opaque</td>
<td>9.51 ± 2.56a</td>
<td>6.81 ± 1.20b</td>
<td>0.16 ± 0.001c</td>
<td>119 ± 13.7d</td>
</tr>
<tr>
<td>Oloyin</td>
<td>Brown</td>
<td>Opaque</td>
<td>12.8 ± 3.11bc</td>
<td>7.93 ± 0.76b</td>
<td>0.37 ± 0.001d</td>
<td>113.72 ± 15.5e</td>
</tr>
<tr>
<td>Drum</td>
<td>Brown</td>
<td>Opaque</td>
<td>11.8 ± 1.41b</td>
<td>7.22 ± 1.35b</td>
<td>0.21 ± 0.001b</td>
<td>164.22 ± 16.44d</td>
</tr>
<tr>
<td>Niger White</td>
<td>White</td>
<td>Opaque</td>
<td>10.7 ± 1.76b</td>
<td>7.43 ± 1.47b</td>
<td>0.20 ± 0.001b</td>
<td>102.34 ± 15.1b</td>
</tr>
</tbody>
</table>

Each value is a mean ± standard error of ten replicates. Means within the same column followed by the same letter(s) are not significantly different at p<0.05 using Tukey's test.

**N** = Newton.

**Table 4** Oviposition, adult emergence and seed damage by *Callosobruchus maculatus* on different cowpea cultivars.

<table>
<thead>
<tr>
<th>Cowpea cultivar</th>
<th>Mean number of eggs laid</th>
<th>Adult emergence (%)</th>
<th>Seed damage (%)</th>
<th>Weight loss (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ife Brown</td>
<td>37.7 ± 1.45d</td>
<td>77.2 ± 2.35d</td>
<td>73.4 ± 4.48d</td>
<td>18.7 ± 2.22d</td>
</tr>
<tr>
<td>Ife BPC</td>
<td>35.3 ± 0.88d</td>
<td>70.8 ± 3.18c</td>
<td>62.8 ± 4.08d</td>
<td>9.95 ± 2.15c</td>
</tr>
<tr>
<td>Sokoto Local</td>
<td>23.7 ± 1.86b</td>
<td>52.1 ± 4.13ab</td>
<td>38.0 ± 4.97b</td>
<td>6.62 ± 1.19b</td>
</tr>
<tr>
<td>Oloyin</td>
<td>25.3 ± 0.89b</td>
<td>61.7 ± 2.54b</td>
<td>53.2 ± 3.22c</td>
<td>5.62 ± 1.66b</td>
</tr>
<tr>
<td>Drum</td>
<td>18.0 ± 1.16a</td>
<td>43.2 ± 6.81c</td>
<td>29.8 ± 3.53a</td>
<td>2.45 ± 0.61a</td>
</tr>
<tr>
<td>Niger White</td>
<td>30.0 ± 0.58c</td>
<td>60.0 ± 5.00d</td>
<td>59.4 ± 6.76c</td>
<td>9.33 ± 2.23c</td>
</tr>
</tbody>
</table>

Each value is a mean ± standard error of ten replicates. Means within the same column followed by the same letter(s) are not significantly different at p > 0.05 using Tukey's test.

**Correlation between toxic metals, mineral elements and activities of *C. maculatus***

The Pearson correlation tests between toxic metals, mineral elements, development of *C. maculatus* (oviposition and adult emergence), seed damage and cowpea weight loss are shown in Table 5. There was no significant correlation between the levels of toxic metals, mineral elements and biological activities (oviposition, adult emergence, seed damage and seed weight loss) of *C. maculatus*. However, there was a weak positive correlation between the mean level of Cd and adult emergence (r = 0.421). Also, a weak negative correlation was observed for the levels of As and adult emergence (-0.45), and oviposition (-0.445).
Correlation between hardness, proximate compositions and biological activities of *C. maculatus*

The Pearson correlation matrix between seed hardness, proximate compositions, development of *C. maculatus* (oviposition and adult emergence), seed damage and cowpea weight loss are summarized in Table 6. There was a negative significant correlation between seed damage and crude fibre (r = -0.59) while there was a positive correlation (r = 0.76) between seed damage and weight loss (Table 5). Seed hardness was strongly negatively correlated with adult emergence (r = -0.84), oviposition (r = -0.89) and weight loss (r = -0.73). The correlation matrix also showed that the moisture content of cowpea seeds was positively correlated with oviposition (r = 0.58) crude protein and fibre, and negatively correlated with parameters such as ash content.

**Table 5** Correlation between the toxic metals, mineral elements and biological activities of *Callosobruchus maculatus*.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Na (µg/g)</th>
<th>Ca (µg/g)</th>
<th>K (µg/g)</th>
<th>Cadmium (µg/g)</th>
<th>Lead (µg/g)</th>
<th>Zinc (µg/g)</th>
<th>Arsenic (µg/g)</th>
<th>Adult emergence</th>
<th>Oviposition</th>
<th>Weight loss</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium</td>
<td>0.867**</td>
<td>0.763**</td>
<td>-0.036</td>
<td>-0.393</td>
<td>0.795**</td>
<td>0.550*</td>
<td>-0.095</td>
<td>-0.188</td>
<td>-0.176</td>
<td></td>
</tr>
<tr>
<td>Calcium</td>
<td>1</td>
<td>0.764**</td>
<td>-0.199</td>
<td>-0.277</td>
<td>0.882**</td>
<td>0.478*</td>
<td>-0.124</td>
<td>-0.069</td>
<td>-0.233</td>
<td></td>
</tr>
<tr>
<td>Potassium</td>
<td>1</td>
<td>-0.403</td>
<td>-0.63**</td>
<td>0.784**</td>
<td>0.716**</td>
<td>-0.104</td>
<td>-0.031</td>
<td>-0.051</td>
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<tr>
<td>Cadmium</td>
<td>1</td>
<td>0.202</td>
<td>-0.356</td>
<td>-0.267</td>
<td>0.421</td>
<td>0.252</td>
<td>0.13</td>
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<tr>
<td>Lead</td>
<td>1</td>
<td>-0.386</td>
<td>-0.427</td>
<td>0.185</td>
<td>0.04</td>
<td>0.136</td>
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<tr>
<td>Zinc</td>
<td>1</td>
<td>0.657**</td>
<td>-0.467</td>
<td>-0.436</td>
<td>-0.534</td>
<td></td>
<td></td>
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<tr>
<td>Arsenic</td>
<td>1</td>
<td>-0.45</td>
<td>-0.445</td>
<td>-0.325</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Adult emergence</td>
<td>1</td>
<td>0.801**</td>
<td>0.653**</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oviposition</td>
<td>1</td>
<td>0.763**</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
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</tr>
</tbody>
</table>

*Correlation is significant at the 0.01 probability level (2 tailed). *Correlation is significant at the 0.05 probability level (2 tailed).

**Table 6** Correlation between hardness, proximate compositions and biological activities of *Callosobruchus maculatus*.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Hardness</th>
<th>Moisture content (%)</th>
<th>Adult Emergence (%)</th>
<th>Crude protein (%)</th>
<th>Crude fat (%)</th>
<th>Crude fibre (%)</th>
<th>Ash content (%)</th>
<th>Carbohydrate (%)</th>
<th>Oviposition</th>
</tr>
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<tbody>
<tr>
<td>Hardness</td>
<td>1</td>
<td>-0.49</td>
<td><strong>-0.84</strong></td>
<td>0.037</td>
<td>0.059</td>
<td>0.44</td>
<td>0.266</td>
<td>-0.123</td>
<td>-0.89**</td>
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<td>Moisture content</td>
<td>1</td>
<td>0.46</td>
<td>-0.60**</td>
<td>0.447</td>
<td>-0.48</td>
<td>-0.508*</td>
<td>0.585*</td>
<td>0.583*</td>
<td>0.801*</td>
</tr>
<tr>
<td>Adult Emergence</td>
<td>1</td>
<td>0.119</td>
<td>-0.243</td>
<td>-0.46</td>
<td>-0.293</td>
<td>-0.069</td>
<td>0.979**</td>
<td>0.039</td>
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<td>Crude protein</td>
<td>1</td>
<td>-0.71**</td>
<td>-0.02</td>
<td>0.129</td>
<td>-0.634**</td>
<td>0.025</td>
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<tr>
<td>Crude fat</td>
<td>1</td>
<td>0.3</td>
<td>0.135</td>
<td>0.007</td>
<td>-0.417</td>
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<tr>
<td>Crude fibre</td>
<td>1</td>
<td>0.313</td>
<td>-0.178</td>
<td>-0.364</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Ash content</td>
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<td>0.01</td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td>Carbohydrate</td>
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<td>-0.187</td>
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<td></td>
<td></td>
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<td></td>
<td></td>
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<tr>
<td>Oviposition</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

*Correlation is significant at the 0.01 probability level (2 tailed). *Correlation is significant at the 0.05 probability level (2 tailed).

**Discussion**

The levels of heavy metals (Cd, Pb and As) in cowpea cultivars in the present study were similar to the levels that have been detected from cowpea cultivars in other parts of the world (Hao et al., 2011). Generally, heavy metals like Cd, Pb and As are toxic to plants with no known physiological importance; they have been shown to cause various adverse effects such as cellular damage, growth impairment, disruption of ionic homeostasis etc. (Kopittke et al., 2007; Piršelová et al., 2015; Yadav, 2010). Also, studies have shown that the presence of heavy metals on the substrate negatively affect the biological activities of insects (Schmidt et al., 1992; Diener et al., 2015). In this study however, there was no
significant correlation between the levels of heavy metals and considered biological parameters of *C. maculatus* except for weak negative correlation that was observed for As, adult emergence and oviposition. The detected levels of mineral elements (Zn, Na, Ca and K) were similar to the levels found in cowpea cultivars from other parts of the world. In a similar work that evaluated the levels of mineral elements in Brazilian cowpea cultivars, Santos and Boiteux (2015) reported range values of 27.5-34.8, 91-140.3, 620-1930 and 14350-18180µg/g for Zn, Na, Ca and K respectively. These values were quite similar to those detected in the cultivars analyzed in the present study. The levels of Zn, Pb, As and Cd in cowpeas could be attributed to untreated sewage water used for irrigation, and also to the plant’s natural way of loading heavy metals. For instance, zinc is loaded in seeds for storage at maturity and as a result zinc accumulates in the whole grain and seeds of some plants in unacceptable levels for either human or livestock consumption (Robson, 1993). The proximate analyses of the cowpea cultivars indicated that all the cultivars studied in this research were rich in proteins and carbohydrate. The average percentage protein and carbohydrate were 24 and 58% respectively. These values are quite high and comparable to the levels detected in other cowpea cultivars (Carvalho et al., 2012; Kaptso et al., 2008). Except for the protein and carbohydrate contents, the values of other proximate constituents such as crude fibre, crude fat, ash content and moisture content did not differ significantly from those reported for other cultivars (Kaptso et al., 2008). Overall, the seed length and weight of the cultivars investigated in the current study were higher than those reported for other Nigerian cultivars. For example, the Ife Brown cultivar had a weight of 0.41 g which was almost double the weight of the heaviest cultivar investigated by Olapade et al. (2002).

Stored food items especially grains are at risks of constant attack by storage pests, and *C. maculatus* has been recognized as an important storage pest of these items (Pessu and Umeozor, 2003; Umeozor, 2005). Consequently, efforts have been concentrated on production of cultivars that are less prone to attack by insect pests. The results of this study show that the Ife Brown cultivar was the most susceptible to *C. maculatus* infestation having the highest oviposition, adult emergence, percentage seed damage and weight loss. The susceptibility of Ife Brown cultivar to *C. maculatus* infestation could be attributed to its soft testa which could easily be damaged by the cowpea bruchid as demonstrated by its low hardness (Adedire et al., 2011b; Ashamo and Khanna, 2007).

The Pearson correlation coefficients between seed hardness, development of cowpea seed bruchid, seed damage and weight loss in this study were negative and significant at p < 0.01. This established a strong relationship between cowpea seed hardness, oviposition, adult emergence, seed damaged and weight loss. There have been reports that the hardness of the grain strongly influenced the number of eggs that are deposited on the grain (Dobie, 1974; Prasad et al., 2015). However, Ashamo and Khanna (2006) observed that seed hardness in paddy did not correlate with susceptibility to *Sitotroga cerealella* whereas amylase content did. Aside from seed hardness, other physical characteristics such as texture, size and thickness of the grain have been linked to resistance to *S. oryzae* in rice cultivars (Dobie, 1974; Prasad et al., 2015). It has been shown that the size of the grain is strongly related to resistance; larger grains supply more food and space for insect growth while smaller grains or grains with less mass offer more resistance to pest infestation than larger grains (Lehale et al., 2012; Singh et al., 1974). In addition, in a comprehensive review, Dordas (2008) pointed out the role of essential nutrients in the susceptibility of plants to pest and diseases. These nutrients, which were not reported in this work, could favour rapid development of the bruchids. Ashamo (2001) and Arnason et al. (1993; 2004) reported that increase in phenolic and ferulic acid contents were responsible for the resistance of *S. zeamais* to maize grains.
Biological activities of C. maculatus on cowpea cultivars

J. Crop Prot.

Other factors which may be responsible include low phytate, saponin and tannin contents which were not considered in this research (Ileke, 2014).

In conclusion, the results from this study show that the nutritive values of Nigerian cowpea cultivars investigated in this study are comparable to those of cowpea cultivars from other parts of the world with respect to proximate composition and mineral profiles. Of the cowpea cultivars, the Ife Brown was the most susceptible to the bruchid C. maculatus infestation having highest oviposition, adult emergence, percentage seed damage and percentage weight loss while the Drum cultivar was the least susceptible. While there were significant relationships between physical parameters (e.g. seed hardness), proximate composition (moisture contents and crude fibre) and adult emergence and oviposition, there was no strong evidence that the levels of heavy metals in the present study are high enough to negatively impact the development of C. maculatus.

References


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Saudi Journal of Biological Sciences, 23: S32-S44.
Biological activities of C. maculatus on cowpea cultivars


بررسی عوامل ذاتی مؤثر در ارقام بذر لوپیا چشم‌پیلنلی روی سوسک چهارنقاطهای حیوانات

*Callosobruchus maculatus* (*Coleoptera: Chrysomelidae*)

تمیزتوب ای اروتلولو، زورف آ آدیس و کریس او آدیدر

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دریافت: 3 بهمن 1396، پذیرش: 6 خرداد 1397

چکیده: در این پژوهش نشان می‌دهد که ارقام بذر لوپیا چشم‌پیلنلی در حیات حیوانات مورد بررسی قرار گرفت. سطح فلزات سنگین شامل کادمیوم، سرب و آرسنیک در شش رقم لوپیا چشم‌پیلنلی توصیف دستگاه اسپکتروفتوترم جذب انجام مورد طالعه قرار گرفت. آمار عادی مانند روزی، سدیم، کلسیم و پتاسیم توسط دستگاه فلیم فتوترم (عکس‌برداری شما) مورد بررسی قرار گرفت. همچنین، شناسایی کلیه شیمیایی ارقام لوپیا چشم‌پیلنلی توسط آزمایشگاه فیزیولوژی سوسک‌های حیوانات حیات بذر لوپیا چشم‌پیلنلی، با بررسی میزان حضور کلسیم، پتاسیم و آرسنیک در داخل و خارج بذر، که این میزان در این نوع بذر مورد بررسی قرار گرفت، میزان فلزات سنگین و عناصر معدنی در بین ارقام دریافت می‌شود. در این سه تجربه، که در آزمایشگاه Dr um Brown حساسيت بالای آزمایشگاه در این نوع بذر مورد بررسی قرار گرفت، میزان عناصر معدنی چشم‌پیلنلی را به حضور افزایش یافته داد. رابطه میان دریافت و حساسيت افزایش یافته نشان می‌دهد. این میزان در این نوع بذر مورد بررسی قرار گرفت، میزان عناصر معدنی چشم‌پیلنلی را به حضور افزایش یافته داد.

واژگان کلیدی: ارقام لوپیا چشم‌پیلنلی، عناصر معدنی، حساسیت به آفت

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