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Phytochemical Screening and Nutritional Quality of Yam Species Susceptible and Resistant to Damage by Yam Beetle, Heteroligus meles Bilb (Coleoptera: Dynastinae)

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Abstract: *Yam species including Dioscorea rotundata (white), D. cayenesis (yellow) and D. dumetorum (Trifoliate or bitter yam) were evaluated for damage caused by H. meles in biological sciences laboratory and undamaged and damaged tubers were evaluated for their phytochemical, mineral element and nutrient quantities in the tubers in order to determine possible traits for resistance, nutrient value and nutritional loss in susceptible species ,The phytochemical analysis ,proximate nutrient and elemental analysis were conducted in the biochemistry laboratory in Kogi State University Anyigba. The results of damage assessment showed significant (P<0.05) higher damage indices in D. rotundata (white yam) compared with D. Cayenesis,(yellow yam), and there was no damage on tubers of D. dumetorum (bitter yam) by yam beetle. The phytochemical results indicated the quantities of saponins, flavonoids, tannins, alkaloids and phenol present in all the tubers. However, D. dumetorum (yellow bitter yam) recorded significant (P<0.05) higher content of saponin (15mg/5g sample), flavonoid (15.7mg/5g samples) followed by D. cayenesis containing 17mg/5g saponin and 6.8mg/5g flavonoid. Thus, saponin and flavonoid are the major secondary metabolites in D. dumetorum resisting the feeding damage by H. meles. The results also showed significant reduction of CHO, Fat, Ca, Mg, Fe, Zn, vitamin C, vitamin A in damaged white yam that is most preferred and consumed species. This study suggests saponin and flavonoid as good sources of plant metabolites for control of H.meles.*

Keywords: *Yam species, Phytochemicals, damage, H. meles, Resistance, Susceptibility, nutrient loss.*

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I. Introduction

Yams, Dioscorea are among the most important staple food crops in the world particularly in the tropical and sub – tropical regions and play a central role in food economy in most west African countries especially Nigeria (FAO, 2006). There are over 600 species (IITA, 2006) but only a few are cultivated for food and medicine (IITA, 1992). Yam contributes more than 200 calories per person per day for more than 150 million people in West Africa (FAO, 2006). The species of yam cultivated in Nigeria include *Dioscorea rotundata* (white yam), *Dioscorea alata* (water yam), *Dioscorea cayenesis* (yellow yam), Dioscorea dumentorum (bitter or trifoliate yam), *Dioscorea esculenta* (Ike and Inoni, 2006, Onwueme, 1998). *Dioscorea rotundata* and *Dioscorea alata* are the most economical and popularly cultivated species of yam in Nigeria due to their high yielding qualities. However, *Dioscorea rotundata* is the most widely grown and preferred yam species in Nigeria. The production in Nigeria account for 70% of an annual production of 36.72 million metric tons in 2006.

The greatest constraint to increasing yam production in Nigeria are cost of seed yams for planting, pest and disease damage with average annual yield losses estimated at 25% (Nweke, et. al 1991). The most devastating insect pest identified in the southern Guinea savanna and forest zone are yam beetles, *Heteroligus meles* (Taylor, 1964, Emehute, 1991, Umeozor, 1998, Tobih et al, 2007, Okoroafor et. al, 2009). All edible yams are attacked by yam beetle except bitter or trifoliate yam,, *Dioscorea dometorum* (Emehute et al, 1998).
The objectives of this study were to assess the severity of damage by *Heteroligus meles* on *Dioscorea rotundata,* (white yam), *Dioscorea cayenesis* (yellow yam) and *Dioscorea dumentorum* (bitter or trifoliate yam) and evaluate their phytochemical concentration as sources of resistance to feeding by *Heteroligus meles*.
Secondly, to evaluate the nutritional quality of the three species and loss of nutrients in the white yam sold in the market for consumption.

II. Materials And Methods

The assessment of damage caused by *Heteroligus meles* on tubers of yam species was carried out in 2014 in the biological science laboratory while the phytochemical test, mineral element and proximate analysis of nutrients were carried out in the Biochemistry laboratory in Kogi State University, Anyingba, in the Southern Guinea savanna Ecological zone of Nigeria (Latitude 7° 36°N and Longitude 7° 12 East).

Collection of yam species

Three species of yam, *Dioscorea rotundata* (white yam), *Dioscorea cayenensis* (yellow guinea yam) and *Dioscorea dumetorum* (bitter or trifoliate yam) were randomly sampled from major yam sellers in Anyigba market. The bulk of fifty tubers sampled from ten sellers (five each) were separated into damage and undamaged except for tubers of *Dioscorea dumetorum* that were free of damage. Then, random selection of three out of ten damaged tubers of *Dioscorea rotundata* and *Dioscorea cayenensis* was done respectively, and were used for damage assessment in three replicate along side with the undamaged *Dioscorea dumetorum* .

Assessment of Damaged Tubers by H. meles

All the tubers were observed for yam beetle feeding and each identified beetle holes were counted and recorded following the method of Tobih , 2007 and Okoroafor et. al, 2009.

Depth and diameter of feeding holes:

The feeding holes were carefully measured using a mathematical set divider to trace the depth of feeding by *Heteroligus meles* and width of each hole which was placed on graduated ruler in centimetres (cm) after the method of Okoroafor et al, 2009 and 2010).

Phytochemical Analysis

Phytochemical Analysis was performed using 5g of healthy white , yellow and bitter yam damaged white yam was also used. Indophenols' method was used for the phytochemical test for alkaloid, tannin, saponin, flavonoid, phenol, Beta carotene, Vitamin C. it was also performed for white yam.

Determination of Mineral elements

This analysis was conducted for healthy white yam, yellow yam ,bitter yam and for damaged white yam. Mineral element composition was determined using absorption spectrophotometer (AAS) after acid digestion of the sample (A.O.A.C, 2010). This procedure was used to extract Sodium (Na), potassium (K), Magnesium (Mg), Iron (Fe), Zinc (Zn), Phosphorus (p) and Calcium (Ca).

Proximate analysis

The analysis was conducted for healthy and undamaged white, yellow, bitter yams .It was also conducted for damaged white Yam. Standard methods of the association of official analytical chemists (A.O.A.C, 2005) were used to determine the crude protein, crude fibre, and crude fat, total ash content. Carbohydrate content was determined using standard method by AOAC 2010 and was estimated as the remainder after accounting for moisture, Ash, fibre, fat, protein. 100 – (MC + Ash + fibre + fat + protein). The data collected on damage assessment, phytochemical, proximate nutrient analysis, elemental content and vitamins were subjected to analysis of variance (ANOVA) using SAS (2002) model and significant means were separated using Fishers LSD and ranked using DMRT. Data for damage and undamaged white yam were analysed using t – test.

Results

Results on table 1are mean number, depth and diameter of feeding holes on tubers bored by *Heteroligus meles*r. The mean number of holes on the tubers of *Dioscorea rotundata* was significantly ($p \leq 0.05$) higher (14 holes per tuber) when compared with the number on *Dioscorea cayenensis*. There was no feeding hole on the *Dioscorea domentorum*. The results showed high susceptibility of white variety compared with *Dioscorea cayenensis* and *dumetorum*.

Table 1: mean number, depth and diameter of feeding holes bored by *Heteroligus meles* on tuber of yam species.

Varieties	Mean no of holes/tuber	Mean depth/tuber (cm)	Diameter (cm)
White Yam	14.4^{a}	1.596^{a}	1.50^{a}
Yellow Yam	0.40^{b}	0.90^{b}	0.33^{b}
Bitter Yam	0.00^{b}	0.00^{b}	0.00^{b}
Prob of F	0.031	0.0001	0.018
SE±	2.21	0.201	0.201

Means in column bearing the same superscript do not differ significantly (P≤0.05) using DMRT, SAS (2000). The phytochemical analysis of these yam varieties indicated various concentrations of saponin, flavonoid, tannin, alkaloid, phenol, and the levels of saponin is significantly (P < 0.05) higher in *Dioscorea cayenensis* (17.2mg/5g sample) and *Dioscorea dumetorum* (15.8mg/5g sample) compared with *Dioscorea rotundata* (2.88mg/5g sample). Similarly, the concentration of flavonoid was significantly higher in *Dioscorea dumentorum* (15.9mg/5g) followed by *Dioscorea cayenensis* (6.81mg/5g) and very low in Dioscorea rotundata (4.21mg/5g). The results indicated low concentration of Tannin, alkaloid and phenol in all the yam varieties.

Table 2: Comparison for Phytochemical Concentration in three species of yam

Varieties	Saponin	Flavonoid	Tannin	Alkaloid	T. Phenol
white Yam	2.88^{c}	4.21^{c}	0.00^{c}	0.34^{c}	0.02^{b}
Yellow Yam	17.21^{a}	6.81^{b}	0.01^{b}	0.76^{b}	0.06^{a}
Bitter Yam	15.8^{b}	15.69^{a}	0.02^{a}	0.97^{a}	0.00^{c}
LSD (P≤0.05)	0.035	0.02	0.00	0.035	0.00
SE±	2.88	2.19	0.00	0.11	0.01

Means in column bearing the same superscript (s) do not differ significantly (P≤0.05) using DMRT SAS (2000).
The calcium (1.92mg), magnesium (0.96mg) and Iron (0.64mg) content in *Dioscorea dumetorum* were significantly (P < 0.05) higher than the content in *Dioscorea cayenesis* and *Dioscorea rotundata*.

Table 3: Comparison of the Mineral Element Concentrations in Three Yam Species.

Varieties	Sodium	Calcium	Magnesium	Iron	Phosphorous	Zinc
White Yam	0.25^{c}	1.295^{c}	0.925^{b}	0.545^{b}	0.205^{c}	0.22^{c}
Yellow Yam	0.33^{b}	1.695^{b}	0.825^{c}	0.580^{ab}	0.390^{a}	0.255^{b}
Bitter Yam	0.39^{a}	1.920^{a}	0.960^{a}	0.640^{a}	0.30^{b}	0.275^{a}
LSD (P≤0.05)	0.00	0.11	0.02	0.06	0.03	0.02
SE±	0.02	0.12	0.03	0.02	0.03	0.01

Means in column bearing the same superscript do not differ significantly (P≤0.05) using DMRT, SAS (2000). The composition of vitamin A is significantly higher (P < 0.05) in *Dioscorea cayenesis* (8.02mg) followed by *Dioscorea dumetorum* (7.83mg) while vitamin C is significantly (P < 0.05) higher in *Dioscorea dumetorum* (1.75mg) and is comparable with yellow yam, *Dioscorea cayenesis* (1.27mg/5g), Lower concentrations in white yam, Dioscorea rotundata were recorded.

Table 4 Comparison of Concentration of Vitamin C and Beta- Carotene (vitamin A) in Three Yam Species

	Vitamin C	Beta-carotene(vitamin A)
White Yam	0.79^{b}	0.98^{c}
Yellow Yam	1.27^{ab}	8.02^{a}
Bitter Yam	1,75^{a}	7.83^{b}
LSD (P≤0.05)	0.553	0.076
SE±	0.18	1.46

Means in column bearing the same superscript do not differ significantly (P≤0.05) using DMRT, SAS (2000).
The comparison of proximate composition of nutrients in damaged white yam and undamaged is shown on table 5.

Table 5: Comparison of Proximate Composition of nutrients in Damaged and Undamaged White Yam.

	Moisture content	Ash	Crude	Fat	Protein	Carbohydrate	Vitamin C
Undamaged White	49.1±0.1	1.63±0.03	4.69±0.0	0.11±0.02	1.03±0.07	48.5±0.06	0.79±0.16
Damage White	54.3±0.15	1.54±0.15	4.38±0.8	0.54±0.02	1.40.0.09	37.9±0.13	0.32±0.00
‘t’ value	28.6	-3.09	-3.85	19.2	3.38	-40.06	3.06
Prob of ‘t’	0.001	0.09	0.06	0.03	0.08	0.006	0.09
Significance		NS	NS		NS		NS

The results showed that fat and carbohydrate contents of damaged white yam were significantly (P < 0.05) reduced as a result of feeding activities of the yam tuber beetles, *Heteroligus meles*. Similarly, there was significant (p ≤ 0.05) reduction in the Na, Ca, Mg, Fe, Zn content and vitamin C in the damaged tuber of white yam (Table 5). All the nutrients were significantly (P ≤ 0.05) reduced after feeding by the beetle. The results indicate 40% reduction in vitamin C.

Table 6 Mineral Elements in Undamaged and Damaged White Yam

	Sodium	Calcium	magnesium	Iron	phosphorus	Zinc
Undamaged	0.25±0.01	1.29±0.02	0.93±0.01	0.55±0.01	0.21±0.01	0.22±0.00
Damage	0.18±0.01	1.09±0.03	0.71±0.02	0.22±0.01	0.17±0.01	0.10±0.01
't' value	6.71	7.20	11.93	29.1	3.13	25.00
Prob. of t	0.02	0.02	0.01	0.001	0.09	0.002

III. Discussion

The results obtained from this study showed that *Dioscorea rotundata* was the most susceptible to *Heteroligus meles* attack. Susceptibility of white yam to *H. meles* was reported by Tobih, et al, 2007, Okoroafor et al., 2009. The reason for significant damage on the white yam tubers than the yellow yam and bitter yam can be attributed to the low content of the secondary metabolites in white yam, for instance, low saponin (2.88mg), flavonoid (4.21mg) and alkaloid (0.34mg) in white yam.

The feeding by the beetle on Dioscorea cayenensis (yellow yam) was mild and this may be attributed to high saponin content and moderate concentration of flavonoid in the tuber while the non–feeding by *Heteroligus meles* on *Dioscorea dumetorum* may be due to the high content of both saponin and flavonoid, which implies that *Dioscorea dumetorum* naturally produced substantial toxic substances including saponin and flavonoid for defence mechanism against the yam tuber beetle, *Heteroligus meles* (Antixenosis). The defence mechanism of the white yam, *Dioscorea rotundata* against *Heteroligus meles* was weak due to low content of these bioactive principles (secondary metabolites). Thus saponin and flavonoid constitute the major trait for resistant ability of *Dioscorea dumetorum* and *Dioscorea cayenensis* to *Heteroligus meles*. The findings agree with the report of Thacker, 2002 that saponin and flavonoid are factors of resistance or antifeeding to insects feeding on plants.

The results on phytochemicals in this study showed that white yam recorded zero concentration of tannin compared with higher concentration of tannin in yellow yam , and bitter yam. This agrees with report of Arogba (2008), who reported that the more coloured or pigmented a variety, the higher the tannin content, although their content was low , their products and accumulation is one of the major mechanisms by which plants defend themselves against attacks by insects (Wink and Schimmer 1999).. The higher concentration of flavonoid in both yellow and bitter yam (trifoliate yam) agrees with the report of Harbone, 1988, that flavonoid constitutes 50% of all known phenolic compounds. Major Phytochemicals are phenolic compounds such as phenolic acids, flavonoids as well as many other polyphenols which function as antioxidants (Zielinski and Kozlowska, 2000). Plant phenols, as phytochemicals are known to possess a considerable range of bioactive properties which are not only broadly classified as nutritional but pharmacological, anti-microbial. Generally, food rich in potentially beneficial phytochemicals are acclaimed to promote overall health and prevent disease.

The feeding by *Heteroligus meles* resulted to 40% reduction in the vitamin C content of damaged tubers of white yam. The damaged tubers also recorded a low carbohydrate which implies low energy utilization by the body when consumed. This assumption is made based on the report of Suzanne, 2014, that 55% of total energy needed by the body is supplied by the carbohydrates. It was also reported that yam contributes more than 200 calories per person per day for more than 150 million people in West Africa (FAO, 2006). The results of this study recorded high content of calcium in white yam, but the content was reduced in the damaged yam.White yam is an important source of calcium for the body development. The body uses 99% of calcium for strong healthy bones, blood clothing and contributes to normal brain function (Alison, 2013). In this study, there was significant reduction of nutritional value of damaged white yam by the beetle as mineral elements, vitamin C and other nutrients were reduce..

IV. Conclusion

From this study, it has been deduced that saponin concentration of 15.8mg/5g sample and flavonoid concentration of 15.9mg/5g yam sample were possible sources of resistance in *D. dumetorum* that deterred *H. meles* feeding resulting to zero damage, while mild damage on *D. cayenesis* was due to high saponin 17mg/5g and moderate flavonoid 6mg/5g . The results also indicated significant reduction of nutrients in white yam damaged by H. meles. The findings present saponin and flavonoid as good sources for control of *H. meles* as they deter the beetle from feeding on *D. dometurum*.

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