

ASSESSMENT OF POST-HARVEST INSECT AND MOULD INFESTATION OF *VIGNA SUBTERRANEA*

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ABSTRACT

Aims: To screen four varieties of *Vigna subterranea* for post harvest insect and mould.

Study Design: The randomized complete block design having three replications for each variety was used to determine post harvest insect and mould infestation on the varieties of the nuts.

Place and duration of study: Department of microbiology, University of Calabar, Cross River State Nigeria was used for this research within the space of four months.

Methodology: Four varieties of *Vigna subterranea* in sack bags and those displayed on tables for sale were thoroughly mixed and randomly collected with a commercial grain collector from marketers in major markets in South Eastern Nigeria. Samples were properly labeled in a clean dry container and conveyed to the laboratory according. Samples were inoculated into sabourand dextrose agar (SDA) supplemented with 50mg of amoxicillin in 1000ml. Plates were incubated at room temperature (28°C) for 5-7 days to determine fungal growth.

Result: Cream black eye variety showed no resistance, while the Brown variety showed low resistance, speckled flecked spotted variety had a high resistance and Cream black eye variety had very high resistance to pest and mould attack. The total colony forming units (cfu) obtained, reduced as resistivity increased across the four varieties. *Callosobruchus maculatus* was identified as the insect that carried out the damage in some of the varieties of *Vigna subterranea*.

Conclusion: The level of resistivity to insect mould infestation varies across the four varieties of *Vigna subterranea*. Poor storage and management system contributed to the level of insect infestation amongst susceptible varieties.

Keywords: *Vigna subterranean*, post-harvest, insect, mould.

INTRODUCTION

The crop *V. subterranea* is a crop with many hidden potentials. Studies have shown that the crop has the capacity of contributing to food security in the world. The crop can be cultivated in regions of the world where rain is low. Besides, high disease resistance capacity has been found as one of the inherent potentials of the crop. Infact it's been noted as the most resistant pulse crop with high yield in regions with low rainfall (Akpala *et al.*, 2013). History has it that the crop originated from the north eastern states of Nigeria to the rest parts of the country, excluding the riverine and swampy zones of the country.

In line with the world's population demand for nutrient rich food stuff, *V. subterranea* is nutritionally endowed to combat malnutrition since it has been proven to contain a reasonable amount of some major food classes such as protein, carbohydrate, and lipids. About 63% carbohydrate, 6.5% oil and 19% protein is present in *V. subterranea* seed. No harm

is associated with consumption of nut at different stages of maturity (so far it's well processed) (Okonkwo *et al.*, 2010; Hillocks *et al.*, 2012; Aviara *et al.*, 2013). *Vigna subterranea* also has an appealing flavor which has highly influenced its demand from the local markets where producers sell them. It shares high nutritive value like other popularly consumed African legumes. *V. subterranea* is capable of growing in arid land where Ground nut (*Arachis hypogea*), maize (*Zea mays* L.) and sorghum (*Sorghum bicolor*) have failed (Hillocks *et al.*, 2012). According to Aviara *et al.* (2013), Nigerians are the highest producers of *V. subterranea* in Africa.

However, the good qualities of this nut, does not actually mean total absence of infestation by pest, as various extreme conditions may overpower the resistance of this crop. Researchers in some publications have disclosed that pest and disease are major problems affecting the high yield of *V. subterranea* in Africa (Aviara *et al.*, 2013; Akpalu *et al.*, 2013; Hillocks *et al.*, 2012). Poor processing and storage system are practices that contribute to post-harvest contamination and commercialization problems of bambara nut (Hillocks *et al.*, 2012).

The idea of storing grains and seeds for food purposes has made insect pest economically important to mankind (Harold, 1947). The ubiquity of insect pest in the tropics and subtropics is a great challenge to the storage of food stuffs. Loss of weight (due to qualitative and quantitative depletion), increase in mould growth, elaboration of toxins and loss of organoleptic qualities are some aftermath effects of insect pest activities on our food and feeds (William and McDonald, 1983). Insect and mould infestations on food products are great threat to food security in Nigeria and beyond. Food products especially nuts and grains are damaged both qualitatively and quantitatively. Post-harvest malpractices are the major reasons for insect and mould infestation of our stored food product. Farmers, traders and consumers store food products in conditions that support the growth of microorganisms and conducive for insects and rodents to feed and carry out their activities (Nwankwo *et al.*, 2015).

Global warming has also contributed to the increase in insect pest activity. Although increase in CO₂ due to this warming of the globe may result to vigorous growth of crops, the disaster associated with floods and droughts consumes all the benefits associated with growth. Therefore, seed production and storage drops drastically. Temperature increase will also encourage insect pest activities. More so, as the lower slopes of mountain peaks get warmer, plants, animals, and pest populations have started to migrate upward (William, 2007).

Loss of nutritional value of our food is an alarming effect of food contamination all over the globe. This low nutritional value of food results to malnutrition which is the greatest threat to the public health. Many developing countries where their climatic condition favors the proliferation of these moulds tend to suffer more of these dieting problems (Kouadio *et al.*, 2013).

The tones of food lost all over the world due to contamination of food stuffs is unbearable. About one billion metric tons of food stuff was estimated lost by the United Nations (UN) and Food and Agricultural Organization (FAO) per year. FAO has disclosed that 25% of the world's grains are contaminated with mycotoxins (Kouadio *et al.*, 2013).

Toxigenic moulds are one of the greatest threats associated with the contamination of food stuff by microorganisms. They produce secondary metabolites known as mycotoxins, which may be carcinogenic, teratogenic, genotoxic and hepatotoxic (Mohamed, 2011). Besides mycotoxin production, the ubiquity of toxigenic moulds is a challenge to control of mould contamination. They thrive in humid, warm and even damp environment. According to Anyanwu. (2007) over one thousand varieties of these moulds exist. Common occurring species of toxigenic moulds are those of *Aspergillus*, *Penicillium* and *Fusarium*. They contaminate food substrate of varying nutritional constituents; ranging from major food nutrients to minor ones. Foods rich in carbohydrates (e.g maize, rice, cassava by products and millet), Proteins (e.g Beans, wheat, fishes, etc) and other nutrients are not exceptional (Saravanakumar *et al.*, 2007). The fact that very little research effort has been directed towards the improvement of this crop is also a limiting factor (Aviara *et al.*, 2013).

2. MATERIAL AND METHODS / EXPERIMENTAL DETAILS / METHODOLOGY

Sample collection was done according to the method stated by Nwankwo *et al.* (2015) and Abriba *et al.* (2013) with some little modifications. *Vigna subterranea* in bags and those displayed on tables were thoroughly mixed and randomly collected with a commercial grain collector. Samples were stored in a clean cool and dried container for few days and replicated into four places to avoid loss of data and to minimize error. Afterward, samples were taken to the laboratory for analysis (Nwankwo *et al.*, 2015; Abriba *et al.*, 2013). Samples were exposed to pest and mould attack at room temperature with no form of preservative or pesticides added for four months.

Sabouraud Dextrose Agar (SDA) supplemented with amoxicillin was used to grow the mould. Each grain of *V. subterranea* was placed at the centre of the Petridishes containing SDA supplemented with amoxicillin and incubated at room temperature (30-32°C) for 5-7 days (Nwankwo *et al.*, 2015). Blocks of Pure cultures obtained with the aid of a cork borer from the plates were mounted on slides and stained with lactophenol cotton blue solution for examination under the microscope using ×40 objective lens. Standard mycological atlas was used to identify the moulds. The resulting spore heads and mycelia were compared with existing pictures in the atlas.

3. RESULTS AND DISCUSSION

These varieties were Cream black eye variety (CBEV), Cream no eye variety (CNEV), Speckled flecked spotted variety (SFSV) and Brown variety (BV). These varieties had different levels of susceptibility and measurable resistance to post-harvest insect and mould infestation. The disparity in the level of resistivity could be due to some level of variation in some genetic constituents across the varieties. *Callosobruchus maculatus* (cowpea weevil) was identified as the insect

that carried out the damage across the varieties of *Vigna subterranea*. Within the space of four months, *Callosobruchus maculatus* successfully attacked three varieties (CBEV, SFSV and BV) out of the four varieties. The highest damage occurred in CBEV (Cream black eyed variety), followed by BV (Brown variety), thirdly by SFSV (speckled flecked spotted variety) and CNEV (Cream no eye variety) had no attack as represented in figure 1.



FIG. 1: Rate of insect infestation in four varieties of *Vigna subterranea* within the space of four months.

KEY:
M1-First month
M2- Second month
M3-Third month
M4- fourth month

Mould infestation was associated with all the varieties despite the high level of resistivity observed in Cream no eye variety. *Aspergillus niger*, *Penicillium species*, *Mucor species*, *Trichophyton species* and *Rhizopus species* were identified after mycological analysis of the samples. The total colony forming units (cfu) obtained from the readings per plate, indicated an increase in viable mould contamination as insect infestation increased.

ANOVA of Mean viable counts of moulds isolated from *V. subterranean* after six days

SOV	Degree of Freedom (DF)	Sum of square (SS)	Mean square (MS)	F-Value	F-Table (5%)	F-Table (1%)	
Total	15	3184.94					
Block	3	650.69	216.89	2.27			
Treatment	3	1674.69	558.23	5.85	3.86	6.99	
Error	9	859.56	95.50				

At P=0.05, there was no significant growth of moulds from *V. subterranea* after six days. However, at P=0.01, growth was significant.

The method of storage, poor hygiene and management of warehouses contributed to the level of insect infestation amongst susceptible varieties. *V. subterrene*a in sack bags and kept in ware houses were the most prevalent method of grain storage among traders in South Eastern Nigeria as shown in plate 1. However, grains were stored by local farmers in plastic gallons to prevent insect attack as shown in Plates 2.



Plate 2: *Vigna subterranea* stored in plastic gallons by local farmers of Mbu-Akpoti, Enugu State, to prevent insect mould attack.



Plate 2: Bambara nut stored in sack bags in New Enugu market Ware house Enugu, Enugu state.

Higher rate of insect infestation was observed among susceptible varieties stored in sack bags in the Ware house as compared to those stored in plastic gallons by rural farmers. This finding agrees with that of Earnest *et al.* 2014 which stated that seeds stored in plastic and glass had lower seed damage by insect. However, both pattern of storage were not safe enough for a healthy storage condition of *V. subterranea* to be achieved. The dirt in the storage houses, other food grains on the floor, presence of insects in other food seed in the same Ware house and some crushed grains of *V. subterranea* could attract rodents and fungal spores contamination.

4. CONCLUSION

Inherent properties of cream no eye variety is believed to have resulted to absolute resistance to insect attack. These properties will be of premium economic importance if discovered and incorporated into other varieties. The war against insect and mould infestation remains a constant battle until both citizens and government decides to wedge it. The Government and individuals must contribute meaningfully to see that the unbearable amount of food loss due to post harvest malpractice is curtailed. The benefits of awareness amongst rural farmers who are the major producers of *V. subterranea* can never be over emphasized. Farmers and traders of this food produce should be enlightened on the factors such as p^H , moisture content, temprature and other factors that encourage insect infestation and mould deterioration. Good hygiene practice and warehouse management system should be encouraged. Bambara nut should be harvested at the prime of maturity as early or late harvest may enhance postharvest losses. More so, there is need for frequent monitoring of stored bambara nut for adequate remedial measures to be taken before the losses appreciates.

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