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MYCOBIOTA OF POST-HARVEST SAMPLES OF KOLANUTS (*Cola acuminate*) AND TIGER NUTS (*Cyperusesculentus*) IN NIGERIA.

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ABSTRACT

A study was carried out to evaluate the mycobiota of fresh marketed seeds of *Cola acuminata* (Kolanut) and *Cyperusesculentus* (Tigernut) in Lafia, Nasarawa State, Nigeria. The serial dilution plate method was used and a total of 6 fungi belonging to 2 fungal classes and 3 genera were isolated from the washings of the plant materials.*Deuteromycetes* were the most frequently isolated fungi, having 100% and 75% percentage occurrences on Tiger and Kola nut seeds respectively; followed by *Zygomycetes* which were only isolated from Kola nuts (15%). Members of other fungal classes were not observed in the study. *Aspergillus* species were the most occurring on the plant materials (66.67%), followed by *Mucor* and *Penicillium* both with 16.67% relative abundances respectively. Kolanut had a higher fungal load (four fungi) compared to Tiger nut (two fungi). Fresh marketed Tiger and Kolanuts popularly consumed in the study area are frequently contaminated with fungi that could on the long run impact negatively on seed quality and pose serious health risks on the final consumers of the products.

Key Words: Mycobiota, Kolanut, Tigernut, Retail, Nasarawa State.

INTRODUCTION

Fungi are ubiquitous in the environment and are usually distributed by wind, rain, animals and humans (Brus *et al.*, 2005). So far, more than 100000 fungal species are considered as natural contaminants of agricultural and food products (Kacaniova, 2003). Their ability to successfully colonize plant products is enhanced by their capacity to grow and reproduce on a vast array of simple and complex food products, producing various metabolites in the process.

Contamination and consequently infection of different crop products by fungi may occur at four different stages, namely:

i). Prior to harvest while the fruits, seeds, tubers, nuts and other cultivated products are still on the parent plant;

ii). During harvest by poor handling and mechanical injury of plant tissues by farm workers; iii). During transportation and storage and finally iv). During marketing (Abdel-Hafez and Saber, 1993; Burdaspal *et al.*, 1990; Odebode, 1990).

The economic implications of fungal contamination of food products are enormous, ranging from reduced market value of deteriorated products to the health concerns of mycotoxicosis of the final consumers be they animals or humans (Galvano *et al.,* 2005; Zinedine *et al*., 2006).

The present study was carried out to evaluate the mycobiota(Formerly known as mycoflora) of retailed Kola and Tiger nuts in Lafia, Nasarawa State, Nigeria.

MATERIALS AND METHODS

Seemingly healthy Tiger and Kolanuts were purchased from vendors in Lafia Modern Market and brought in sterile polyethylene bags to the Botany Laboratory of Federal University, further examination.Collected nuts were identified on-site using morphological characteristics as reported by Leist and Jontis (2009). The serial dilution technique was used. 1 gram of surface tissues of unwashed seeds of fresh and healthy Tiger and Kolanuts were respectively scraped off using a sterile scapel and soaked separately in 9mls of sterile distil water. The stock suspension was thoroughly vortexed and serially diluted to a concentration of 10-4. 1 ml of the final concentration (10-4) was asceptically mixed with sterile molted Nutrient Agar (at 45^0C) impregnated with 0.01mls of streptomycinsulphate to inhibit growth of non-fungal microbes, and allowed to solidify. Inoculated media were incubated at room temperature (25-27^0C) for 7 days for spore germination and isolation of pure cultures as reported by Narayanasamy (2011). Cultural and microscopic characteristics of isolated fungi were used for identification in line with taxonomic keys of Samson *et al.,* (2010).

RESULTS

Results of cultural and microscopic characteristics of mycobiota of Kola and Tiger nut seeds (Table1) showed that a total of 6 fungi were isolated from the studied plants. Kolanut had the highest fungal load (four fungal isolates) compared to Tigernut (2 fungal isolates). Fungi isolated were molds belonging to the classes Deuteromycetes and Zygomycetes. Fungi belonging to the genus Aspergillus were the most occurring with four representatives, compared to Mucorand Penicilium with one fungal isolate respectively. Growth on Nutrient Agar showed various cultural characteristics mostly circular growth with entire margins.

Result of overall abundance of different fungal genera on evaluated nuts (Table 2) showed that members of the genus Aspergillus were the most abundant (66.67%) followed by both Mucor and Penicillium each with 16.67% abundance respectively.

Results of frequency of occurrence of different fungal classes on studied plants (Table 3) showed that only fungi belonging to the class Deuteromycetes(100%) were isolated from Tiger nuts while Deuteromycetes and Zygomycetes were the two fungal classes isolated from Kolanut. The Deuteromyceteswere most frequently isolated (75.00%) from Kolanut followed by Zygomycetes(15.00%).

DISCUSSION AND CONCLUSION

Fungi belonging to the class *Deuteromycetes*, mainly *Aspergillus* species were the major mycobionts of Tiger and Kola nut seeds in the study. According to Samson *et al.,* (2010), this class has been reported to contain many important toxigenic food and airborne contaminants. *Aspergillus* spp., are capable of mycelial growth and extensive sporulation upon a wide range of organic substrates, often causing appreciable deterioration of stored food materials (Sharma and Tripathi, 2008). Induced spoilage of Kolanuts and other crop products by *Aspergillus* spp. during post-harvest storage have been reported by several authors (Prakash and Raoof, 1989), grapes (Sharma and Vir, 1986; Sinha and Saxena, 1987; Adebajo and Popoola, 2003; Terna *et al.*, 2015).

The observed higher fungal load on Kolanut compared to Tigernut seeds can be partly due to the availability of a higher surface area on Kolanut seeds for spore deposition compared to the comparatively smaller seeds of Tiger nuts.Furthermore, the usually

high moisture content of Kolanuts (Agbeniyi and Ayodele, 2010) compared to Tiger nuts, are a better predisposing factor for nuts colonization by high moisture loving fungi such as *Aspergillus* spp., Mucor, etc. (Adebayo, 1966; Olunloyo, 1979; Agbeniyi and Fawole, 1999; Agbeniyi *et al*., 2000); a condition that is worsened by the use of peasant methods in processing, packaging and marketing of harvested nuts (Odebode, 1990). Adeniyi *et al*.,(2012) also reported the reluctance of vendors and consumers to discard fairly moldy nuts for their economic value, resulting in high re-contamination rates of healthier nuts.

Seed-borne fungi are responsible for deterioration of food reserves in seeds (Neegaard, 1977). The study revealed that the Tiger and Kola nuts popularly consumed in the study area are frequently contaminated with fungi that could on the long run impact negatively on seed quality and pose serious health risks on consumers of the products in the study area. Further research could focus on the determination of mycotoxin content of retail samples of Tiger and Kolanuts infected by various molds.

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Table 1. Cultural and microscopic characteristics ofmycobiota of Tiger and Kola nut seeds

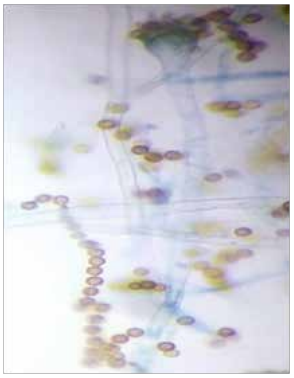
Name of crop	Isolate code	Cultural Characteristics on Nutrient Agar	Micromorphology	Identity of Isolate	Class
Tiger Nut	T1	Whitish-brown circular growth with convex elevation, entire margins and concentric surface	 Conidia globose, rough walled with black bars; phialides cylindrical tapering to a distinct neck	*Aspergillus nigervan Tieghem*	*Deuteromycetes*
Tiger nut	T2	Grayish-green coloured colony with wrinkled surface, raised elevation and undulating edges.	 Conidiophores hyaline, mononematous, terverticillate to quarter verticillate; stipes smooth, branches somewhat divergent; phialides are cylindrical tapering to a short neck; globose conidia.	*PenicilliumchrysogenumThom*	*Deuteromycetes*

Table 1 Contd.

Name of crop	Isolate code	Cultural Characteristics on Nutrient Agar	Micromorphology	Identity of Isolate	Class
Kola nut	K1 .	Olive green circular growth with concentric surface, flat elevation, and entire margins	 Conidiophores long and coarse; conidial heads radiate; phialides cylindrical tapering to a distinct neck; conidia globose with rough walls	*A. Flavus Link*	*Deuteromycetes*
Kola nut	K2	Dark brown to black circular growth with smooth surface, convex elevation, entire margins	 Conidial heads globose to radiate; conidiophores smooth, long and coarse; vesicles globose; phialides cylindrical, tapering to a distinct neck; conidia globose and rough-walled.	*A. carbonarius(Bainier) Thom*	*Deuteromycetes*
Kola nut	K3	Creamy-white growth with concentric surface, convex elevation, filamentous margins and.	 Phialides cylindrical with a short distinct neck; conidia colourless and globose	*A. candidusLink*	*Deuteromycetes*
Kola nut	K4	Creamy-white growth with concentric surface, convex elevation, filamentous margins.	 Short sporangiophores with circinnate branches and slightly encrusted walls; sporangiospores ellipsoidal	*Mucorcircinelloides Micheli: Fr.*	*Zygomycetes*

Table 2: Overall abundance of different fungal genera on evaluated nuts

Fungal Genus	% Abundance
Aspergillus	66.67
Mucor	16.67
Penicillium	16.67

Table 3. Frequency of occurrence of different fungal classes on evaluated nuts

Fungal Class	Percentage Occurrence	
	Tigernut	**Kolanut**
Deuteromycetes	100.00	75.00
Zygomycetes	0.00	15.00