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Research Article

Prevalence of Toxigenic Mycoflora in Groundnut Cake (Kuli kuli) Sold in Niger State

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ABSTRACT

Deterioration of groundnut cake ('kuli kuli') a by-product of processed groundnut oil consumed widely in Nigeria is of great concern. Therefore, this study investigated the toxigenic mycoflora commonly present in kuli kuli sold in the markets in Niger state, Nigeria. A total of eighteen (18) samples were collected from 10 markets across each of the three agricultural zones of Niger state, namely; Bida, Mokwa (zone 1), Minna, Shiroro (Zone 2), Kotongora, and Kagara (zone 3) respectively. Isolation of associated fungi was done on PDA inoculated with 10^4 dilution factors and incubated at room temperature. A total of 166 fungal species were identified which include *Aspergillus*, *Penicillium*, *Rhizopus*, and *Fussarium*. The order of percentage occurrence (*A. niger* 27.11% being the highest and *F. oxysporum* 4.82% the least) was *A. niger* (27.11%)>*A. flavus* (19.88%)>*P. chrysogenum (16.87%)*>*A. parasiticus (11.45%)*>*Rhizopus spp.(10.84%)*>*A. fumigatus* (9.03%)>*F. oxysporum* (4.82%). The result indicates that majority of fungi isolated from kuli kuli sold in the markets in Niger state are toxigenic fungi. Therefore, improved management of these oil rich products from the farm, postharvest storage, and processing will enhance the high quality product to access a larger market and reduce the risk of health challenges that go with consuming contaminated kuli kuli.

Keywords: Toxigenic mycoflora; Prevalence; Groundnut; Distribution; Kuli kuli

INTRODUCTION

Groundnut cake (kuli kuli) is one of the products obtained from groundnut seeds after extraction of the oil. Groundnut cake is one of the most important staple food supplements for the majority of the Nigerian population. In Nigeria, it serves as an important source of food and constitutes an inexpensive source of protein, fat, minerals, and vitamins in the diets of rural populations, especially children ('kuli kuli) [1,2].

The chemical composition of groundnut seeds per 100 g edible portion as reported by USDA is moisture (6.5 g), carbohydrate (16.1 g), lipids (49.2 g), protein (25.8 g), dietary fiber (8.5 g), magnesium (168 mg), phosphorus (376 mg) and iron (4.6 mg). Groundnut kernels contain 40%-50% fat, 20%-50% protein, and 10%-20% carbohydrate and are rich in vitamin E, niacin, riboflavin, thiamine, folacin, calcium, phosphorus, magnesium, zinc, iron, and potassium. Groundnut cake is used as a feed supplement for livestock, as fertilizer, and for the preparation of kuli kuli and (traditional recipe in Nigeria) [3-5]. Kuli kuli is a groundnut based snack that is consumed by all age range and social class among Nigerian populace. It is also used as a major ingredient in poultry feed formulation.

The qualitative loss of groundnut cake (kuli kuli) can be attributed to biochemical changes in protein, carbohydrates, fatty acids, and vitamins due to fungal contaminations. Mostly, kuli kuli is vulnerable to microbial contaminants such as fungi (*Aspergillus*, *Fussarium*, and *Penicillium*) that produced mycotoxins which lead to significant economic loss.

Over the year, poor handling of groundnut cake has been responsible for contaminations leading to varying health challenges such as cancer and liver cirrhosis, weakened immune systems. Hence, this research studied the prevalence of toxigenic mycoflora in groundnut products (Kuli Kuli) sold in some markets in Niger state [6-8].

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MATERIALS AND METHODS

Collection of samples

A total of eighteen (18) samples of groundnut cake 50 g each from four major producing areas collected from 10 markets across each of the three agricultural zones in Niger state, namely Bida, Mokwa (zone 1), Minna, Shiroro (Zone 2), Kotongora, and Kagara (zone 3) respectively were individually homogenized. The samples were collected based on the rate of kuli kuli utilization by the residents and the population.

Preparation of Potato Dextrose Agar (PDA)

Thirty nine (39) grams of PDA (Hi media) was suspended in 1000 ml distilled water and heated to dissolve the powder completely, the medium was sterilized by autoclaving at 121°C for 15 minutes (Manufacturer's guide).

Isolation and identification of fungi contaminating Kuli kuili

One gram (1 g) of homogenized kuli-kuli was aseptically suspended into 9 ml of sterile distilled water in a test tube and vortexed properly. 1 ml was serially diluted up to fourth fold 10^4. 1 ml from the fourth dilution fold test tube (10^4) was transferred into a sterile petri plate. Twenty (20 ml) of Potato Dextrose Agar (PDA), to which 1 ml of streptomycin was added, and then poured into the petri dish incubated at 28°C ± 2°C for 3 days. After the third day, a single conidium was picked up with a sterile needle and viewed under microscopic observation, transferred individually to PDA plates, and incubated at ambient temperature. The monoculture was prepared and stored on PDA slants at 40°C ± 2°C. Subculture was made at regular intervals. The fungal isolate was identified using the fungal family of the world mycological monograph under microscopic observation [9-12].

RESULTS

Isolated and identified toxigenic mycoflora

A total of 166 fungal species were isolated and identified belonging to four genera. *Aspergillus niger* had the highest percentage of occurrence (27.11%) and the least was F.*oxysporum* (4.82%). The general order of occurrence was A.niger (27.11%)>*A.flavus* (19.88%)>*P.chrysogenum* (16.87%)>A.parasiticus (11.45%)>Rhizopus spp. (10.84%)>A. *fumigatus.* (9.03%)>F.*oxysporum* (4.82%), respectively as shown in Table 1 below [13-15].

Table 1: Percentage, frequency of fungi isolated groundnut cakes (kui kuli) samples collected in Niger state.

S/N	Fungus isolated	Frequency	Percentage %
1	*Aspergillus flavus*	33	19.88
2	*A. parasiticus*	19	11.45
3	*A.niger*	45	27.11
4	*A. fumigatus*	15	9.03
5	*Fussarium species*	8	4.82
6	*Penicillium species*	28	16.87
7	*Rhizopus species*	18	10.84
8	Total	166	100

Characteristics features of isolated fungi

***Aspergillus niger*:** Rapidly growth colonies on PDA with abundant submerged mycelium, carbon black/deep brownish black conidial heads. Non branched conidiophore with bulb end carries conidia like sun rays. Pin like black growth to pale yellow conidial on reverse Petri dish plate with at initial globose and then radiate well defined columns.

***Aspergillus flavus*:** Moderate to rapid growth colonies on PDA with pin like green growth, yellow/greyish green. Non branched conidiophore with bulb end carries conidia.

***Fussarium spp*:** Colonies appear brown in the center and with white edges, a White cottony colony with dense growth on PDA short crescent conidiophores, septate hyphae with abundant micro conidia, spindle like conidia, and multi cellular.

***Penicillium chrysogenum*:** Colonies are usually gradually to fast growing, green or green greyish color colonies with a white ring at the margin, sometimes white, mostly consisting of dense conidiophores. Brush like conidiophore carries conidia, conidiophores is hyaline, erect, branched, and penicillate at the apexes with 2-3 metula, 3-4 verticilatephialides, and catenulate conidia in each phialide, forming rather compact cylindrical (Figure 1).

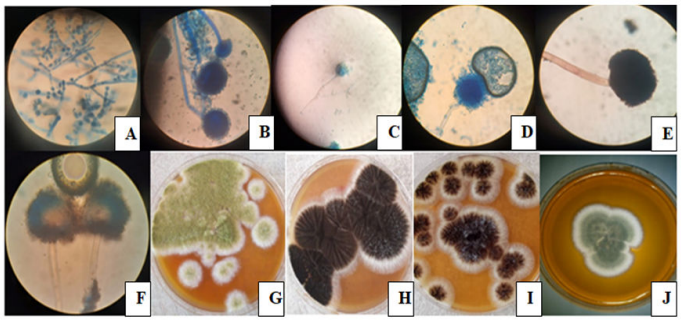

Figure 1: Photomicropha of isolated and identified toxigenic mycoflora A. *Fussarium species*; B. *A. parasiticus*; C. *P. chrysogenum*; D. *A. flavus*; E. *A. niger*; F. *A. fumigatus*; G. Pure culture of *A. flavus*; H. Pure culture of *A. fumigatus*; I. Pure culture of *A. niger*; J. Pure culture of *P. chrysogenum*.

DISCUSSION

The five fungal genera (*Aspergillus*, *Fusarium*, *Penicillium*, and *Rhizopus*) isolated from kuli kuli collected from different agricultural zones in Niger state revealed that this commodity is highly contaminated which confirm the earlier work of Vikas and Mishra who isolates nine species of fungi from the seeds of different varieties of groundnut during one storage year. The finding in this work is also in line with Chavan who reported the species of *Aspergillus*, *Penicillium*, *Fussarium*, *Rhizopus*, and *Alternaria* were commonly occurring postharvest molds in storage conditions.

The result from this study corroborates the work of Odeniyi, et al. who isolated *Aspergillus*, *Fussarium*, *Penicillium*, and *Rhizopus* species from Kuli kuli and other groundnut based products in Niger state. The mycoflora commonly associated with groundnut cake (Kuli kuli) samples belonging to the genera *Aspergillus*, *Penicillium*, *Rhizopus*, and *Fussarium* were found present in all samples studied. Oftentimes, *Rhizopus* species may also occur as a common saprophyte of many foods both at pre-harvest and post-harvest stages [16-20]. Boli, et al. recorded the occurrence of these species of fungi among others in their work on groundnut butter from Benin. These fungal species are reported to be commonly associated with the raw material (groundnut), amongst other materials, in their spore forms either in the field during plant propagation, nut storage, product preparation, packaging, or storage of finished product. They may also end up initiating deterioration of the food material and/or produce spores that assist their survival.

The presence of *Aspergillus* species such as *A. flavus* and *A. niger*, *Fussarium* species, *Penicillium* species, and *Rhizopus* in the kuli kuli samples pose a toxicological threat to the consumers since the majority of the strains of these fungal species have been reported to be toxigenic. *Rhizopus* is known to liberate a metabolite rhizonin A while aflatoxins, ochratoxins, fumonisins, trichothecenes, citrinin, and patulin are well produced during metabolism by the other above mentioned fungi. In 1990, Akano and Atanda reported the presence of these fungi and aflatoxins in kuli kuli from Ibadan, Oyo state, Nigeria after the incidence of deaths resulting from consumption of aflatoxin contaminated foods in Nigeria.

CONCLUSION

The result from this study shows a high incidence of fungal contamination from groundnut cake (Kuli Kuli) sold in major markets from Bida, Mokwa, Chanchaga, Shiroro, Kotongora, and Rafi in Niger state mostly toxigenic molds, *Aspergillus niger*, *A. flavus* and *P. chrysogenum* having the highest occurrence and their percentage occurrence has a direct effect on food values. Therefore, improved management of this high protein source and oil rich products from the farm, postharvest storage, and processing will enhance the high quality products to access a larger market and reduce the risk of health challenges that go with consuming contaminated kuli kuli.

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