



Fungal Species Associated with Date Palm (*Phoenix dactylifera* L.) Fruit and Tiger Nut (*Cyperus esculentus* L.) Fruit in Lafia Metropolis, Nasarawa State, Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Author OOO designed the study, and performed the statistical analysis. Author IOO wrote the protocol, author DIO wrote the first draft of the manuscript. Authors VF and AO managed the analyses of the study. Authors OOO and IOO managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aims: Fungi are ubiquitous microorganisms that bring about large scale spoilage of plants and animal products. The resultant effects include contamination of products and disease conditions in colonized living host. The aim of the study is to enumerate and identify fungal species associated with date palm and tiger nut fruits in Lafia, Nasarawa State.

Methodology: Date fruit and Tigernut fruits samples (60 each) were collected from the six markets located in the Lafia metropolis. These were evaluated using the pour plate technique to determine fungal load in the samples.

Results: Results showed that Date palm obtained fungal population of $0.28 \times 10^4 \pm 7.1 \log_{10}$ CFU/g

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as the lowest in Koro Market and Tigernut fruits recorded $4.30 \times 10^4 \pm 3.0 \log_{10}$ CFU/g as the highest. Seven fungal genera were isolated in Date palm fruits, while 9 were obtained from Tigernut with genera *Aspergillus*, *Penicillium*, *Rhizopus*, *Mucor* found common to the two fruits. *Aspergillus* species obtained the highest percentage occurrence of 42.10% in Date fruits, while *Chrysosporium xerophilum* obtained 3.40% as the lowest in the fruits sampled

Conclusion: Findings from the study showed that Tigernuts and Date fruits sold in Lafia market presented heavy burden of fungal contaminations and these contaminants have the capacity of releasing harmful metabolites into the fruits thus making them unsafe for consumption.

Keywords: Fungi; fruits; contamination; metabolites; market.

1. INTRODUCTION

Date palm fruit (*Phoenix dactylifera* L.) is a food source readily eaten by man because of its nutritional, health and aesthetic benefits. The fruit contains carbohydrates (total sugars 44% - 88%), fats (0.2% - 0.4%), proteins (2.3% - 5.6%), fibers (6.4% - 11.5%), minerals and vitamins [1-3]. Date fruit apart from being taken as food, also has other important uses. It reportedly has antioxidant activity attributed to the presence of phenolic acids, flavonoids, anthocyanins, selenium [4]; anti-inflammatory potential [5]; protective effect on the gastric mucosa from stomach acid wearing [6]; capacity to decrease hepatic marker enzymes (ALP, lactate dehydrogenase, MDA) [7]; anti-microbial activities; and improve immune activities and to stimulate mRNA expression in cells [8,9]. The fruit also possesses antilipidic capabilities [10] and are able to prevent and control diabetes mellitus [11].

Tiger-nut (*Cyperus esculentus* L.) of the family Cyperaceae is a perennial crop found everywhere with such names as Aya, imumu, aki hausa, ofio in the sub African region [12]. The fruits are rich in protein, carbohydrate, sugars, and oil and fiber with capacity to alleviate flatulence, promote urine production, and ease menstrual pains [13,14]. Typical tigernut contains proteins, fats (oils), starch, glucose, fiber, minerals (sodium, calcium, potassium, magnesium, iron, copper, manganese, phosphorus) [15], and are consumed raw, soaked in water or mixed with roasted peanuts. The fruits can be processed into nut meals, extracts and flours, milks, tonic drinks, soap, body cream, and ice cream [13].

Aspergillus, *Penicillium*, *Fusarium* genera all filamentous fungi have been reported to colonize and infect stored fruits; and are also major sources of mycotoxins' production in stored fruits

[16-18]. Mycotoxins are toxic metabolites produced by fungi found contaminating crops and other agricultural products [19, 20]. These metabolites reduce food quality and bring about losses in capital; and impair animal and human health [21]. Abdel-Sater and Saber [22], Alghalibi and Shater [23], Iamanaka et al. [24] all reported the presence of various concentrations of ochratoxin A, zearalenone, and aflatoxin B1 in date fruits. A wide range of fungal species have been isolated from tiger nuts among which are *Aspergillus flavus*, *Aspergillus fumigatus*, *A. niger*, *A. terreus*, *Fusarium solani*, *Saccharomyces cerevisiae*, *S. fubiligera* and *Candida pseudotropicalis*, *Penicillium citrinum* [13].

2. MATERIALS AND METHODS

2.1 Sample Collection

Samples were collected from daily markets in Lafia, the capital of Nasarawa State, Nigeria. Date fruit samples were purchased from the six markets located in the metropolis - Mararaba, Shinge, Shabu, Koro, Lafia Old, and the Modern market. Ten samples were randomly collected and made into two composite samples per market. Dry date palm fruit samples were picked randomly from the seller to ensure good representation. The samples were cleaned by removing foreign materials and kept in polyethylene bags with labels then brought to the laboratory for analysis. Samples were collected around 7.30 am in the morning.

2.2 Isolation of Fungi

Date fruit seeds were mashed with mortar and pestle and 1 g dissolved in 9 mL distilled water, using the pour plate method it was inoculated on Sabouraud Dextrose Agar (SDA) incubated at 37°C for 72 to 96 h.

2.3 Morphological Identification of Fungal Isolates

The fungal species were characterized based on their morphological and cultural characters using taxonomic guides and standard procedures according to Dayan (2004). The morphological characteristics evaluated included; colony growth and colour. Microscopic fungal structures such as the fruiting structures and spores were examined using compound microscope with magnification of 100X.

3. RESULTS

3.1 Total Fungal Count

The total fungal counts of the samples were generally high as shown in Table 1. Date fruits collected in Shinge market had the highest contamination of $1.25 \times 10^4 \pm 5.6$ cfu/g, while the least was in Koro market within the metropolis with $0.28 \times 10^4 \pm 7.1$ cfu/g. These values were lower when compared with $4.30 \times 10^4 \pm 3.0$ cfu/g for tiger-nut collected in Shinge market and $4.25 \times 10^4 \pm 3.5$ cfu/g in Shabu market. From the result, Tiger-nuts were more contaminated than Date fruits.

3.2 Isolated Fungal Species

Nine fungal species were isolated and identified from date fruits collected in markets within Lafia as shown in Table 2. Genera *Aspergillus* and *Penicillium* had two species each isolated from the date fruits. Nineteen species of fungi belonging to nine genera were isolated from the tigernut samples. *Aspergillus flavus* occurred most followed by *A. niger*, while *Trichophyton concentricum* had the least occurrence from the samples collected. Seven *Aspergillus* species were isolated from Tiger-nut with a total of 45.12% occurrence rate, while *Rhizopus stolonifer* recorded the least occurrence with

4.76%. *A. niger* and *A. terreus* were the only species of *Aspergillus* isolated from Date fruit with total occurrence of 42.1% closely followed by *Penicillium* species with 18.86%. *Chrysosporium xerophilum* had 3.4% occurrence in Date fruits samples.

3.3 Markets and Fungal Species Isolated

Aspergillus niger (both in Tiger-nuts and Date fruits) and *A. terreus* were the only fungi found in all the markets where samples were randomly collected. Old market Lafia and the modern market had 9 fungal species isolated from tiger-nut samples collected from them which were the least fungal isolate from any market where Tiger-nuts were sampled, while Shabu market had the highest number of fungi species isolated. *Trichophyton concentricum* was found only in samples collected from Shinge market. *Saccharomyces cerevisiae* and *Chrysosporium xerophilum* were isolated only in Shinge and Shabu markets (Table 3), while *Penicillium allii* species were isolated in Mararaba market, Shinge market, and Old Market Lafia. The highest fungal species isolated from date fruit was in Shabu market as all the fungi were isolated from samples collected in the market except *Penicillium allii*, while Mararaba and Koro markets had only four fungi species isolated from date collected in the markets.

4. DISCUSSION

This study showed that date fruits and tiger nuts consumed in Lafia metropolis at the time of this study were contaminated with fungi. Fungal contamination recorded might not be unconnected with unsanitary and largely unhygienic nature of the selling conditions and the environment. These contaminations could have happened during or after harvesting, processing, storage or transition. Some important factors such as storage temperature, moisture

Table 1. Population of fungi isolated from date fruit samples expressed in log₁₀ CFU/g

Location	Date fruit (log ₁₀ CFU/g)	Tiger-nuts (log ₁₀ CFU/g)
Maraba market	$0.46 \times 10^4 \pm 4.5$	$2.71 \times 10^4 \pm 7.4$
Lafia old market	$0.36 \times 10^4 \pm 3.8$	$1.10 \times 10^4 \pm 9.3$
Shinge market	$1.25 \times 10^4 \pm 5.6$	$4.30 \times 10^4 \pm 3.0$
Modern market	$0.38 \times 10^4 \pm 1.6$	$1.96 \times 10^4 \pm 7.0$
Shabu market	$1.02 \times 10^4 \pm 9.5$	$4.25 \times 10^4 \pm 3.5$
Koro market	$0.28 \times 10^4 \pm 7.1$	$3.00 \times 10^4 \pm 8.5$

Values are Means of three triplicates and are expressed as Mean \pm Standard Deviation, at $P = 0.05$ there was no significant difference

content, and composition of aerial gaseous compounds can influence the growth of mold during storage [25]. Majority of the selling points are close to waste disposal points which encourages contaminations. This study showed that among the different locations where the fruits samples were collected from, Shinge market had the highest fungal load in the fruits sampled, followed closely by Shabu market which might not be unconnected with the dirty and dusty environment of the markets. Other sources of dates' fungal contaminations and infection might include insects' bites, wounds and presence of foreign matter such as sand, dust, and debris among others.

It was noted also, that date fruit and tiger nuts samples collected from wholesalers in the old and the new markets (result not shown) had lesser contamination compared to those bought from hawkers in Shabu, Shinge and Maraba. This variation could be because the wholesalers were situated in a particular position within the market unlike hawkers who move about from one point to another within the market and in the process have more access to contaminants. Constant exposure of the fruits at the point of sales could also aid deposition of spores, which

germinate at the storage temperature and humidity levels. Trucksess and Scott [26] and Piga et al. [27] explained that date fruits are susceptible to mold infection and the resultant mycotoxin production because of their high sugar content, method of harvest and drying conditions and moisture content of dried fruits. Drying, a post-harvest operation brings fruits and other agricultural produce in contact with soil which is a reservoir for microorganisms thus the contamination observed could have their source from the farm [28]. Insects wounds as explained by El-Juhany [29] could also increase fungal colonization. Wound creates easy route for fungal hyphae and spores to access internal recesses of plants and fruits.

The most identified fungi in this study were *Aspergillus* sp, *Penicillium* sp, *Rhizopus stolonifer* and *Saccharomyces cerevisiae* which agreed with the report of Colman [30] and Atia [31]. Findings showed that date palm and tiger nuts fruits are prone to contamination by *Aspergillus* and *Penicillium* species than other fungi species. Additionally, the hawkers do not store date fruits at refrigeration temperatures, further creating conditions required for the fungi to proliferate. *Aspergillus* grows on a wide range

Table 2. Fungal isolates and their percentage occurrence in Tiger-nuts and Date fruits collected in Lafia markets

S/N	Tiger nut samples		Date fruits samples	
	Fungal isolates	% Occurrence	Fungal isolates	% Occurrence
1	<i>Alternaria aborescens</i>	3.89	<i>Absidia corymbifera</i>	8.33
2	<i>Alternaria tenuissima</i>	3.11	<i>Aspergillus niger</i>	33.33
3	<i>Aspergillus acidus</i>	5.06	<i>Aspergillus terreus</i>	8.77
4	<i>Aspergillus carbonarius</i>	5.06	<i>Chrysosporium xerophilum</i>	3.4
5	<i>Aspergillus flavus</i>	14.01	<i>Mucor racemosus</i>	6.14
6	<i>Aspergillus niger</i>	11.67	<i>Penicillium allii</i>	7.02
7	<i>Aspergillus restrictus</i>	3.50	<i>Penicillium comenberti</i>	11.84
8	<i>Aspergillus terreus</i>	5.83	<i>Rhizopus stolonifer</i>	10.96
9	<i>Aspergillus wentii</i>	3.11	<i>Saccharomyces cerevisiae</i>	10.09
10	<i>Candida</i> sp.	6.61		
11	<i>Cryptococcus laurentii</i>	5.83		
12	<i>Fusarium</i> sp.	6.23		
13	<i>Mucor racemosus</i>	3.89		
14	<i>Penicillium solitum</i>	4.67		
15	<i>Penicillium</i> sp.	3.89		
16	<i>Rhizopus stolonifer</i>	4.76		
17	<i>Trichophyton concentricum</i>	1.95		
18	<i>Trichophyton</i> sp.	2.72		
19	<i>Trichophyton terrestre</i>	4.28		

Table 3. Fungal isolates from tiger-nuts and date fruits and markets where isolated

S/N	Organism	Maraba	Old Mkt Lafia	Shinge	Modern Mkt	Shabu	Koro
1	<i>Alternaria aborescens</i>	-	-	+	+	+	-
2	<i>Alternaria tenuissima</i>	-	-	+	-	+	-
3	<i>Aspergillus acidus</i>	-	-	+	+	+	+
4	<i>Aspergillus carbonarius</i>	+	-	+	-	+	+
5	<i>Aspergillus flavus</i>	+	+	+	+	+	+
6	<i>Aspergillus niger</i>	+	+	+	+	+	+
7	<i>Aspergillus restrictus</i>	+	+	+	-	-	+
8	<i>Aspergillus terreus</i>	+	-	-	+	+	+
9	<i>Aspergillus wentii</i>	+	-	+	-	+	+
10	<i>Candida sp.</i>	+	+	-	-	+	+
11	<i>Cryptococcus laurentii</i>	+	+	-	-	+	+
12	<i>Fusarium sp.</i>	+	+	+	+	+	-
13	<i>Mucor racemosus</i>	+	+	-	-	+	+
14	<i>Penicillium solitum</i>	-	-	+	+	+	+
15	<i>Penicillium sp.</i>	-	-	+	+	+	+
16	<i>Rhizopus stolonifer</i>	-	-	+	-	+	+
17	<i>Trichophyton concentricum</i>	-	-	+	-	-	-
18	<i>Trichophyton sp.</i>	+	+	+	+	-	+
19	<i>Trichophyton terrestre</i>	-	+	+	-	+	-
Isolates from date fruits							
20	<i>Aspergillus terreus</i>	+	-	-	+	+	+
21	<i>Rhizopus stolonifera</i>	-	+	+	+	+	-
22	<i>Chrysosporium xerophilum</i>	-	-	+	-	+	-
23	<i>Absidia corymbifera</i>	-	-	+	+	+	+
24	<i>Aspergillus niger</i>	+	+	+	+	+	+
25	<i>Penicillium allii</i>	+	-	+	+	-	-
26	<i>Mucor racemosus</i>	-	+	-	-	+	+
27	<i>Penicillium comenberti</i>	+	+	+	-	+	-
28	<i>Saccharomyces cerevisiae</i>	-	-	+	-	+	-

of substrates including agricultural produce causing decay and contamination of commodities in the field and in storage; and grows at lower water content [32]. Field fungi such as *Fusarium* and *Alternaria* on the other hand contaminate grains before or during harvest operations. Post-harvest processes which include drying, storage and transportation steps also contribute to fungal contamination if not properly monitored and carried out under hygienic conditions. In complex cases, agricultural crops or produce may pass through the hands of merchants or third party drying facilities (since its moisture content is to be reduced) and held in storage for periods before finally arriving at the selling points. Produce are susceptible to fungal contamination at all times especially if the storage conditions are not strictly controlled.

Fungi belonging to the genus *Aspergillus* were present in high numbers in dates; the most predominant among them was the species *A. niger* with occurrence percentage of (33.33%),

followed by *Penicillium comenberti* (11.84%) and the least is *Chrysosporium xerophilum* (3.50%). Similar findings were reported by Ragab et al. [33], while Al Sheikh [34] isolated 10 species of *Aspergillus* of them *A. niger* predominated. Our results also agreed with Atia [31] who isolated *A. niger* as predominant fungi followed by *A. flavus* from the fruit. However, other dates contaminating fungal species such *A. flavus*, *P. chrysogenum* and *R. stolonifer* have been reported [30,33].

The presence of *Aspergillus*, *Penicillium*, *Rhizopus* were corroborated by the report presented by Udeozor and Awonorin [12] and Agbaje et al. [35]. Gambo and Da'u [36] observed that *Aspergillus flavus*, *Aspergillus niger*, *Fusarium sp* and *Candida sp* were the common fungi occurring at different frequencies in tigernuts that rendered it unwholesome with *A. flavus* being the highest occurring isolate which the findings of this study agreed with. Study of strains distribution showed that there is no

difference in the type of fungi obtained regardless of the location, meaning that fungal contamination to the date fruits sold in Lafia is mostly the same. So it could be deduced that the source of the fruits sold in the different market in Lafia is the same.

5. CONCLUSION

Tiger-nut and date fruits sold in markets in Lafia are contaminated with fungi especially those of health concerns (molds) that may secrete mycotoxins, as well as bring about economic losses, reduction in crop quality. It is necessary to sensitize the public on the dangers of fungal contaminations in seeds and fruits and their possible effect on human and animal health, and possible means of preventing and controlling this contamination. Majority of the effects of fungal infection could be prevented with good personal hygiene.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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