

*Full Length Research Paper*

# Isolation and identification of fungal species from dried date palm (*Phoenix dactylifera*) fruits sold in Maiduguri metropolis

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**A total of 360 dried date palm (*Phoenix dactylifera*) fruits were collected from hawkers, shops and market places within Maiduguri metropolis for the detection of the presence of fungal species. Investigation was based on cultural, microscopically and biochemical tests. Of the 327 (90.83%) fungal isolates recovered on Sabouraud dextrose agar (SDA), *Aspergillus niger* had the highest percentage of occurrence (39.17%), followed by *Aspergillus flavus* (17.60%), *Mucor species* (16.67%), *Aspergillus fumigatus* (12.50%), *Trichophyton rubrum* (4.16%), while *Candida albicans* had the least percentage of occurrences of (0.83%). There is need therefore to appropriately treat this fruit before consumption, to minimize possible mycotic infection.**

**Key words:** Isolation, fungi, date palm, *Phoenix dactylifera*, Maiduguri.

## INTRODUCTION

Date palm (*Phoenix dactylifera*) is an erect palm with a height of 30.5 to 36.5 m (100 to 120 ft). The trunk is clothed from the ground up, with upward-pouring, overlapping, persistent woody leaf bases (Gepts, 2002). The fruit is oblong, measuring 2.4 to 5 cm in length, it contains hard seed which is deeply furrowed on the inside with fresh, sweet dark-brown, reddish or yellow-brown when ripened; the flesh may be of thin or thick skin (Ferrari, 2000). The fruit varies in size, colour and quality (Morton and Mannie, 1987). The scientific name "*Phoenix dactylifera*" was derived from "*Phoenix*", the legend bird of ancient Greece. The Phoenicians' dyed cloth with a purple colour from the murx shellfish; this colour was also called "*Phoenix*" possibly because it had such great appeal and value. The same colour was noted on the fruit of date; hence the date palm genus became "*Phoenix*"

and the specific name for "*dactylifera*" came from the shape of the fruit, "*dactylos*" being the ancient Greek word for "finger" (Geoff, 2002).

It was observed that the date palm fruits (*Phoenix dactylifera*) are mostly loaded with mixture of microbes; bacteria, molds and yeast (Atia, 2011) but people go ahead eating after clearing the inner environment, while some eat it whole irrespective of the state of the internal environment of the fruits (Naturland, 2002; Atia, 2011). The Agricultural industries sustained huge crop losses as a result of fungal diseases of fruits and plants (Christensen et al., 2007). Approximately, 80000 species of fungi have been described but fewer than 400 are of medically importance, and less than 50 species causes more than 90% of fungal infections of human and animals (Geo et al., 2007). The mycology with the highest incidence, *Candidiasis* and dermatophytosis are caused by fungi of the normal microbial flora and are highly adapted to survival on the human host (Lansing et al., 2002). Date palm fruits (*Phoenix dactylifera*) form a vital component of diet in Arabian Peninsula, especially Saudi

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**Table 1.** Sample distribution according to location.

Location	Number of samples
Post office	120
Custom area	120
Bulunkutu area	120
Total	360

Arabia and are well being consumed in many countries of the world, Nigeria not exceptional (Redmond, 2009). In Islamic countries, dates are among the religious first meals in breaking Ramadan fast and therefore largely consumed by muslims for religious and traditional purpose. However, they are attacked by various fungal species, thus causing their spoilage at ripening, as well as during storage and processing stages (Ibrahim and Rahma, 2009). Djerbi (1983) stated that the most common fungi causing date fruits spoilage are *Aspergillus* spp and *Alternaria* spp. These fungi secretes many kind of enzymes and poison that causes decay and loss of the nutritional value of the date and makes it unsuitable for consumption (Berbendi, 2000; Ibraheem and Klaef, 2003). Therefore, it becomes important to investigate both the inner and outer surface of the fruits, as regard the presence of mycotic agents, as well as its suitability for human and animal consumption.

So far, there is no available report on the isolation and identification of fungal species from dried date palm fruits around Maiduguri metropolis. Therefore, we undertook the study to ascertain that.

## MATERIALS AND METHODS

### Sample collection and preparation

A total of 360 date palm fruits samples were collected into sterile covered receptacles from shops, market places and hawkers in Maiduguri metropolis using aseptic techniques [Maiduguri is located in the North-Eastern part of Nigeria (11° 50 N and 13° 09 E), and estimated to have a population of 1,197,497 by 2007 (Maiduguri-Wikipedia, 2011)] for the determination of the presence of potential mycotic agents based on cultural, microscopically and biochemical test. The collected samples were then transported to the Bacteriology/Mycology laboratory, Federal College of Veterinary and Medical Laboratory Technology Vom, Plateau state for analysis. With gloved hands, the fruits were cut open by means of sterile pair of forceps and scalpel blades, the outer surface and the inner environment were scrapped aseptically.

### Cultural technique

The samples were then inoculated aseptically into sabouraud dextrose agar (SDA) medium containing 5 g of Streptomycin and 4 mega of Penicillin per litre, respectively (Mackie et al., 1996). Pair cultures were made at body temperature of 37°C and room temperature of ± 25°C, for the period of 2 to 4 weeks with constant checking (Ochei and Kolhalhar, 2000; Baker et al., 1998). Macroscopic examination was carried out to observe the growth

characteristics, colour, presence and absence of pigmentation on the media (Patrick et al., 1995). Microscopic examination was carried out using the lacto phenol cotton blue to observe the colonial morphology and the spore formation (Bello, 2002). Gram staining technique was one of the backgrounds of microbiology carried out to confirm for a large blue or purple colony indication of yeast or yeast-like fungi (Mukhtar and Huda, 2005).

### Biochemical test

Urease test was carried out to confirm the presence of *Trychophyton rubrum* as described by Mahamoud et al. (1996). Fermentation test, a process by which carbohydrate is fermented by yeast to produce alcohol and carbon dioxide by gas production (Bulayan and Thomas, 2002), was positive for *Candida*.

### Confirmatory test for *Candida albicans*

Germ tube test was carried out to distinguish *Candida albicans* from other *Candida* spp by their formation of germ tubes (Abayomi, 2007). Assimilation test was carried out to confirm the *Candida albicans* as described by Iyer et al. (2002), Abayomi (2007) and Kenchappa and Sreenivasmurthy (2003).

## RESULTS

Out of the (360) total number of the date fruits samples collected for this study, 327 were positive for fungal organisms and took part in the study, whereas 33 were negative and was discarded. Distribution of the date fruits according to their location shows that custom area, Bulunkutu area, and Post office area was with 120 samples from each location (Table 1). Total of six different species of fungi were isolated, namely; *Aspergillus niger*, *A. flavus*, *A. fumigatus*, *Mucor* spp, *Trychophyton rubrum* and *Candida albicans*. Of these six different fungal species (90.83%) occurrence in the study, *Aspergillus* species had the highest percentage of occurrence (69.17%), with the least percentage of occurrence (0.83%) seen in *Candida albicans*. The highest percentage of fungi isolated was seen in the Post office area (33.33%), Bulunkutu area has (30.00%), and the least (27.50%) of the fungal isolates was seen in Custom area (Table 2). Table 3 shows the percentage occurrence of the three different species of *Aspergillus* isolated; *A. niger* had the highest percentage occurrence (39.18%), followed by *A. flavus* (17.50%), and *A. fumigatus* (12.50%) has the least percentage occurrence.

## DISCUSSION

Out of the 327 fungal isolates recorded in this study, *A. niger* had the highest percentage of occurrence (39.17%), while *C. albicans* had the least percentage of occurrences (0.83%). This findings agree with that of Saffron (2006), who reported that the fruit of date palm was highly infected by filamentous fungi. Salalh and

**Table 2.** Distribution of fungal isolates and their percentage (%) occurrence.

Fungal isolate	Custom area	Post office area	Bulunkutu area	Total
<i>Aspergillus</i> spp.	69 (19.17)	93 (25.83)	87 (24.17)	249 (69.17)
<i>Mucor</i> spp.	24 (6.67)	15 (4.17)	21 (5.83)	60 (16.67)
<i>Trichophyton</i> spp.	6 (1.67)	9 (2.50)	0 (0.00)	15 (4.16)
<i>Candida</i> spp.	(0.00)	3 (0.83)	0 (0.00)	3 (0.83)
Total	99 (27.5)	120 (33.33)	108 (30.00)	327 (90.83)

**Table 3.** Distribution of *Aspergillus* species isolated according to their location and percentage (%) occurrence.

Fungal isolate	Custom ar	Post office area	Bulunkutu area	Total
<i>Aspergillus niger</i>	51 (14.17)	60 (16.67)	30 (8.33)	141 (39.17)
<i>Aspergillus flavus</i>	12 (3.33)	27 (7.50)	24 (6.67)	63 (17.50)
<i>Aspergillus fumigatus</i>	6 (1.67)	6 (1.67)	33 (9.17)	45 (12.50)
Total	69 (19.17)	93 (25.83)	87 (24.17)	249 (69.17)

Nawal (1997) reported that *A. niger* had high occurrence isolates. Hashem (2009) reported *Aspergillus* spp. as the predominate genus. Other fungal infected organisms isolated included *A. flavus* (17.50%), *Mucor* spp (16.67%), *A. fumigatus* (12.50%), and *T. rubrum* (4.16%). This does not agree with the findings of Ibrahim and Rahma (2009), who reported *Rhizopus* spp as the dominate isolate; EL-Deeb et al. (2006) in their study, could not isolate any of the above fungal organisms. These differences could be attributed to variance in geographical location.

Results of this study have shown that date palm fruits are much more prone to contamination by *Aspergillus* spp than other fungal species. It is very possible therefore that there could be some active principles in the fruits which could favour colonization by this organism. From the public health point of view, the contamination of date palm fruits by moulds is significant because of the presence of mycotoxins which can cause severe poisoning, emesis, diarrhoea, prostration and even death (Abdulla, 2008). It was observed that the fruits being hawked around, sold in shops or market places were neither covered nor protected in any way from dust or atmospheric contamination, and when consumed without any form of clearing or washing could predispose such unsuspected consumers to imminent mycotic infection. In the cause of isolation, it was observed that some fungi, especially mucor, grew faster at 37°C, which is the total optimal human body temperature; this could hence increase its adaptability and potential in causing disease in both man and animals.

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