

Fungal Infestations in Some Dry Fruits during Storage in Different Seasons

Madhulika Srivastava, Shail Pande*, Lalji Srivastava & Chhamta Srivastava

Department of Botany, M.G. Post Graduate College, Gorakhpur – 273001 (India)

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Abstract

Cuddapah almond (*Buchanania lanzan*), Coconut (*Cocos nucifera*) and Date palm (*Phoenix dactylifera*) are important and valuable food commodities. During storage the food commodities are spoiled by abiotic and biotic agents amongst whom fungi play important role. In the present study mycoflora of Cuddapah almond, Coconut, and Date-palm was studied using blotter method, dilution method and agar plate method during different seasons such as rainy, winter and summer. Osmophilic fungi were also isolated by increasing sugar concentration in CzapekDox agar medium. Altogether 14 fungal species were isolated from the kernels of *Buchanania lanzan*, 19 species from *Cocos nucifera* and 22 fungal species from *Phoenix dactylifera* during all three seasons.

Key words: Mycoflora, Cuddapah almond (*Buchanania lanzan*), Coconut (*Cocos nucifera*) and Date palm (*Phoenix dactylifera*).

Introduction

Cuddapah almond (*Buchanania lanzan*), Coconut (*Cocos nucifera*) and Date palm (*Phoenix dactylifera*) are important and valuable food commodities. During storage the food commodities are spoiled by biotic and abiotic agents. Among the microorganisms fungi play a significant role in deteriorating the aesthetic and nutritive value of stored food commodity (Christensen and Kaufmann, 1965; Neergard, 1977). Under storage conditions Cuddapah almond, Coconut and Date palm are susceptible to attack by fungi, insects and other microorganisms because they have a rich source of stored nutrients conducive for growth of numerous fungi such as *Aspergillus flavus*, *Aspergillus niger*, *Rhizopus*, *Penicillium* sp.

A total of nine fungi namely *Aspergillus flavus*, *A. niger*, *A. ochraceus*, *Curvularia lunata*, *Fusarium moniliforme*, *F. pallidroseum*, *F. solani*, *Penicillium citrinum* and *Rhizopus stolonifer* were recorded from the edible kernels of *Buchanania lanzan* (Punam Kumari Singh; Harsh N. S. K.; Khan S. N.; Pandey Rajeev, 2002). A significant variation in pattern of mycoflora incidence was observed in terms of source and season. Fungal infestation was highest during monsoon. Aflatoxins are the most common mycotoxins elaborated by different isolates of *A. flavus* obtained from *B. lanzan*.

Mycological investigation on spoilage fungi in the infected date fruits (*Phoenix dactylifera*, Linn.), sampled from Bayero University, Kano, Nigeria showed that the

most dominant isolated fungi were *Rhizopus* sp. (100%) and *Mucor* sp. (100%), followed by *Torula* sp. (40%), *Penicillium* sp. (30%), *Aspergillus* sp. (16.67%) and *Alternaria* sp. (13.33%) respectively. *Aspergillus* species appeared to be the most toxigenic fungi recovered from the dates. (Ibrahim, S. and Rahma, M. A, 2009). Another research in a total of 360 dried date palm (*Phoenix dactylifera*) fruits collected from Maiduguri metropolis for the detection of the presence of fungal species showed *Aspergillus niger* had 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%) (Colman, S., Spencer, T. H. I., Ghamba, P. E. and Colman, E. 2012). There is a need therefore to appropriately treat this fruit before consumption, to minimize possible mycotic infections.

Researches proved that, dehusked coconut fruits stored at 30°C deteriorated and the deterioration in nutritional composition was due to breakdown of protein and carbohydrate by the spoilage fungi. Further tests confirmed the ability of the isolated spoilage fungi to utilize the different carbohydrate and nitrogen sources as source of carbon and energy. *Aspergillus flavus* and *Aspergillus niger* were the principal fungal agents associated with the spoilage (P.N. Okolie, C.L. Obi and P.O. Uaboi-Egbenni, 2011).

Several environmental factors like humidity and temperature during storage influence the infestation by

Table 1- Fungi isolated from kernels of *Buchanania lanzan* during different seasons.

Fungi Isolated	Rainy				Winter				Summer			
	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm
1	2	3	4	5	6	7	8	9	10	11	12	13
<i>Absidia spinosa</i>	-	+	+	-	-	-	-	-	-	-	-	-
<i>Aspergillus aculeatus</i>	-	-	-	-	-	-	-	-	-	+	-	-
<i>Aspergillus flavus</i>	+	+	+	+	+	+	+	+	-	+	+	-
<i>Aspergillus fumigatus</i>	-	+	-	-	-	-	-	-	-	-	+	-
<i>Aspergillus nidulans</i>	-	+	-	-	-	-	-	-	-	-	-	-
<i>Aspergillus niger</i>	-	+	+	+	+	+	+	+	+	+	+	+
<i>Aspergillus ochraceus</i>	-	-	-	-	-	-	-	-	+	-	-	-
<i>Aspergillus tamarii</i>	-	-	+	+	+	-	+	-	-	-	-	-
<i>Cladosporium herbarum</i>	-	-	-	-	-	-	-	+	-	-	-	-
<i>Eurotium amstelodami</i>	-	-	-	+	-	-	-	-	-	-	-	-
<i>Penicillium citrinum</i>	-	-	-	-	-	-	-	-	-	-	+	-
<i>Penicillium pupurogenum</i>	-	-	-	-	+	-	-	+	-	-	-	-
<i>Rhizopus nigricans</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Syncephalastrum racemosum</i>	-	+	+	-	-	-	-	-	-	-	-	-

Bl- Blotter method; Dil- Dilution method; Cz- Czapek’s –Dox agar medium; Osm- Osmophilic medium

Table 2- Fungi isolated from kernels of *Cocos nucifera* during different seasons.

Fungi Isolated	Rainy				Winter				Summer			
	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm
1	2	3	4	5	6	7	8	9	10	11	12	13
<i>Absidia spinosa</i>	-	-	+	-	-	-	-	-	-	-	-	-
<i>Alternaria humicola</i>	-	-	-	-	-	-	+	-	-	-	-	-
<i>Aspergillus flavus</i>	+	+	+	+	-	+	+	-	+	+	+	+
<i>Aspergillus fumigatus</i>	-	-	+	-	-	-	-	-	-	+	+	-
<i>Aspergillus nidulans</i>	-	-	-	-	-	-	-	-	-	+	-	-
<i>Aspergillus niger</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Aspergillus tamarii</i>	-	-	-	-	-	-	-	-	+	+	-	-
<i>Aspergillus terreus</i>	-	-	-	-	-	+	-	-	-	+	-	-
<i>Cladosporium herbarum</i>	-	-	-	-	+	+	+	+	-	-	-	-
<i>Mucor hiemalis</i>	-	-	-	-	-	-	-	-	-	+	-	+
<i>Nigrospora sphaerica</i>	-	-	-	-	-	-	+	-	-	-	-	-
<i>Paecilomyces varioti</i>	-	-	-	-	-	-	-	-	-	+	-	-
<i>Penicillium citrinum</i>	-	-	-	-	-	-	-	-	+	-	-	-
<i>Penicillium meleagrinum</i>	-	-	-	-	-	-	+	+	-	-	-	-
<i>Penicillium purpurogenum</i>	-	-	-	+	+	+	-	-	-	-	-	-
<i>Penicillium species</i>	-	-	-	-	-	+	-	-	-	-	-	-
<i>Rhizopus nigricans</i>	-	-	-	-	+	-	+	+	+	-	+	+
<i>Syncephalastrum racemosum</i>	+	-	+	+	-	+	+	+	-	-	+	-
<i>Trichoderma viridae</i>	+	+	-	+	-	-	-	-	-	-	-	-

fungi and aflatoxin production. Natural occurrence of aflatoxins in dried fruits such as dates has only been found in tropical and subtropical regions with high temperature and humidity which are suitable climatic conditions for growth of aflatoxigenic fungi (Shenasiet *al.*, 2002; Drusch and Ragab, 2003; Karaca and Nas, 2006). Besides aflatoxins, OTA a potent nephrotoxic and hepatocarcinogenic mycotoxin has been detected in many different food products including cereals, nuts, cocoa, spices and dried fruits from different geographical origin. This is due to the wide distribution of ochratoxigenic members of *Aspergillus niger* throughout the world. It is generally assumed that *Aspergillus* species, both AF’s and OTA producers, are more commonly

associated with commodities in warmer and tropical climates such as Mediterranean countries (Pitt and Hocking, 2009).Diener and Cole (1982) observed that when the seed moisture exceed 9% at the equilibrium humidity of 80% and 30° temperature, the chances of invasion by *Aspergillus flavus* increase drastically. Panasenko (1967) reported that even at constant relative humidity a temperature increase can stimulate fungal activities. The work was carried out to isolate and identify the fungal taxa that cause contamination of *Buchanania lanzan* , *Cocos nucifera* and *Phoenix dactylifera* in different seasons in the Eastern Uttar Pradesh region aimed at identifying a more reliable and safe methods of storing these commodities.

Table 3- Fungi isolated from kernels of *Phoenix dactylifera* during different seasons.

Fungi Isolated	Rainy				Winter				Summer			
	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm	Bl	Dil	Cz	Osm
1	2	3	4	5	6	7	8	9	10	11	12	13
<i>Absidia spinosa</i>	-	+	+	-	-	-	-	-	-	-	-	-
<i>Alternaria humicola</i>	-	-	-	-	-	-	+	-	-	-	-	-
<i>Aspergillus aculeatus</i>	-	-	-	-	-	+	-	-	-	-	-	-
<i>Aspergillus flavus</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Aspergillus flavipes</i>	-	-	+	-	-	-	-	-	-	-	-	-
<i>Aspergillus fumigatus</i>	-	-	+	-	-	+	-	-	-	-	-	-
<i>Aspergillus niger</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Aspergillus orhraceus</i>	+	+	+	+	-	+	-	-	-	-	-	-
<i>Aspergillus tamarii</i>	-	-	+	-	-	-	-	-	-	-	-	-
<i>Aspergillus terreus</i>	-	+	-	-	-	-	+	-	-	-	-	-
<i>Cladosporium herbarum</i>	-	-	-	-	+	-	+	+	-	-	-	-
<i>Curvularia lunata</i>	-	-	-	-	-	+	-	-	-	-	-	-
<i>Curvularia pallescens</i>	-	-	-	-	-	+	-	-	-	-	-	-
<i>Eurotium amstelodami</i>	-	-	-	+	-	-	-	-	-	-	-	-
<i>Penicillium chermisenum</i>	-	-	-	-	-	+	-	-	-	-	-	-
<i>Penicillium funiculosum</i>	-	+	-	-	-	-	-	-	-	-	-	-
<i>Penicillium meleargrinum</i>	-	-	-	-	-	-	+	+	-	-	-	-
<i>Penicilliu moxalicum</i>	+	+	+	+	-	-	-	-	-	-	-	-
<i>Penicillium purpurogenum</i>	-	-	-	-	+	+	+	+	-	-	-	-
<i>Penicillium varians</i>	-	+	-	-	-	-	-	-	-	-	-	-
<i>Rhizopus nigricans</i>	+	-	-	-	+	-	+	+	-	-	-	-
<i>Syncephalastrum racemosum</i>	-	+	-	+	-	+	+	+	-	+	+	+
<i>White mycelia sterilia</i>	-	-	-	-	-	+	+	+	-	-	-	-

l- Blotter method; Dil- Dilution method; Cz- Czapek'sDox agar medium; Osm- Osmophilic medium

Materials and Method

Samples of Cuddapah almond, endosperm of coconut and Date palm were procured from local market in pre-sterilized glass containers during rainy (July-October), winter (Nov-Feb) and summer (March-June) seasons. After each collections, they were studied for their associated mycoflora by standard blotter, agar plate and dilution plate method (Annon 1996). For isolation of osmophilic fungi sugar concentration in the Czapek Dox agar medium as recommended by Mehrotra and Basu (1975), was increased to 20% so as to allow the growth of osmophilic fungi, if any, associated with different commodities. The identification of fungal isolates was made with the help of available literature (Raper and Thom 1949, Raper and Fennel 1965, Gilman 1967, Booth 1971, Ellis 1971, 1976).

Data regarding total number of colonies in each plate, the species present and the number of colonies of each species were noted. For calculating the % occurrence following formula was used.

$$\% \text{ Occurrence on commodity} = \frac{\text{No. of colonies of a particular fungal species in all plates}}{\text{Total no. of colonies of all the fungal in all the plates on a commodity}} \times 100$$

Results and Discussion

Table -1 reveals that 14 species were isolated from the kernels of *Buchanania lanzan* during all three seasons. Nine, seven and seven species were isolated during rainy, winter and summer seasons respectively. *Aspergillus flavus*, *A. niger* and *Rhizopus nigricans* were found associated with the kernels in all the seasons while fungi like *Aspergillus fumigatus*, *A. ochraceus* and *A. tamarii* were restricted to any two of the seasons. *Aspergillus aculeatus*, *A. nidulans*, *Cladosporium herbarum*, *Eurotium amstelodami*, *Penicillium purpurogenum*, *P. citrinum* and *Syncephalastrum racemosum* infested the kernels in any one of the seasons. *Aspergillus niger* was the most frequent fungus followed by *Rhizopus nigricans* during all the three seasons. The third frequent species was *Aspergillus flavus*. However *Penicillium purpurogenum*

and *Aspergillus fumigatus* showed almost equal frequency ranking third during winter and summer respectively.

Table-2 reveals a total of 19 species encountered on the endosperm of *Cocos nucifera* during all the three seasons. The total number of species recorded in rainy, winter and summer were 6, 11 and 11 respectively. *Aspergillus flavus*, *A. niger* and *Syncephalastrum racemosum* were invariably found in all the seasons while *Rhizopus nigricans* was not isolated during rainy season. However, rest of the fungi were found associated in any one of the seasons only. *Aspergillus niger* ranked first followed by *Aspergillus flavus* during all three seasons; though the per cent occurrence of *Penicillium meleagrinum* and *P. purpurogenum* was slightly more than *Aspergillus flavus* during winter. *Syncephalastrum racemosum* was the third frequent fungus during winter and rainy seasons; though the per cent occurrence of *Cladosporium herbarum* was equal to that of *Syncephalastrum racemosum* during winter.

Table 3 exhibits that all 22 species infested the dried fruits of *Phoenix dactylifera* during the three seasons but the number of species varied with the seasons; 14, 16 and 3 during winter, rainy and summer respectively. *Aspergillus flavus*, *A. niger* and *Syncephalastrum racemosum* were associated during all the seasons while *Aspergillus fumigatus*, *A. ochraceus*, *A. terreus* and *Rhizopus nigricans* during two seasons viz. rainy and winter. Rest of the species were restricted either to rainy or to winter only. *Aspergillus niger* was most frequent species followed by *A. flavus* in all the three seasons. Besides *A. ochraceus*, *P. oxalicum*, *P. purpurogenum*, *Cladosporium herbarum* and *Syncephalastrum racemosum* were some frequent species isolated in rainy, winter and summer seasons.

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